Flight activity of longhorn beetles *Monochamus sartor* and *M. sutor:* Attractiveness of insect and tree produced volatiles

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Abstract

The pine wood nematode (*Bursaphelenchus xylophilus*), causal agent of pine wilt disease, is transmitted by longhorn beetles of the genus *Monochamus* (Col., Cerambycidae). Trapping vectors is one important measure for monitoring and control of pine wilt disease. Lures have been developed for *M. galloprovincialis*, the main vector in Europe. Flight behaviour of two potential vectors *M. sartor* and *M. sutor* was studied regarding pheromone, kairomone and weather influence. From July to August 2012, multiple funnel traps with three different combinations of attractants were installed in a mountainous spruce forest in Dürrenstein Wilderness Area, Lower Austria.

Traps baited with the commercial lure Galloprotect 2D® (SEDQ, Spain), which consists of a *Monochamus* aggregation pheromone compound and two bark beetle pheromone components, caught lowest numbers of female and male *M. sartor*. Highest catch was attained by addition of the host tree volatile α-pinene; the increase in males was statistically significant. Further addition of smoke volatiles did not enhance captures. Due to lower *M. sutor* catch, no significant differences in response to the lures were established.

Catches of *M. sartor* and *M. sutor* were significantly correlated with weather factors mean and maximum air temperature. No flight occurred when mean temperatures fell below 15 °C. Captured beetles were marked, released and recaptured to determine dispersal behaviour. Four *M. sartor* and five *M. sutor* of the total released 308 and 85 were recaptured. Within a mean time of seven days (maximum of 14 days), released beetles spread in all directions up to the most remote trap in 390 m distance, traversing spruce stands as well as open areas.

This experiment gave first insight into flight activity of two potential pine wood nematode vectors in mountainous Austria, as well as into their reactions to traps and lures, developed for *M. galloprovincialis*.

Abstract

Der Kiefernholznematode (*Bursaphelenchus xylophilus*), Verursacher der Kiefernwelkekrankheit, wird von Bockkäfern der Gattung *Monochamus* (Col., Cerambycidae) übertragen. Der Fang der Vektoren stellt eine wichtige Maßnahme für das Monitoring und die Bekämpfung der Kiefernwelke dar. Für den Hauptvektor in Europa *M. galloprovincialis* wurden bereits Lockstoffe entwickelt. Die Flugaktivität der beiden potenziellen Vektoren *M. sartor* und *M. sutor* wurde hinsichtlich des Einflusses von Pheromonen, Kairomonen und Witterungsfaktoren analysiert. Zu diesem Zweck wurden von Juli bis August 2012 Vieltrichter-Fallen mit drei verschiedenen Lockstoffkombinationen in einem montanen Fichtenbestand im Wildnisgebiet Dürrenstein, Niederösterreich eingesetzt.

Fallen mit dem kommerziellen Lockstoff Galloprotect 2D® (SEDQ, Spanien), welcher aus einer *Monochamus*-Aggregationspheromon-Verbindung und zwei Borkenkäfer-Pheromon-Komponenten besteht, erzielten die geringste Fangzahl *M. sartor* Weibchen und Männchen. Der höchste Fang wurde durch Zugabe des wirtsbaumbürtigen, volatilen α-Pinen erreicht. Hierbei war der Anstieg gefangener Männchen statistisch signifikant. Weiterer Zusatz von Rauch-Lockstoffen steigerte das Fangergebnis nicht. Aufgrund des geringeren Fangs von *M. sutor* wurden keine signifikanten Unterschiede in der Reaktion auf die Lockstoffe festgestellt.

Die Anzahl gefangener *M. sartor* und *M. sutor* korrelierte signifikant mit den Witterungsfaktoren mittlere und maximale Lufttemperatur. Bei mittleren Lufttemperaturen unter 15 °C erfolgte kein Flug. Zur Untersuchung des Ausbreitungsverhaltens wurden gefangene Käfer markiert, freigelassen und wiedergefangen. Vier *M. sartor* und fünf *M. sutor* der insgesamt freigelassenen 308 bzw. 85 wurden wiedergefangen. Nach Freilassung verbreiteten sich diese innerhalb einer durchschnittlichen Zeitspanne von sieben Tagen (maximal 14 Tagen) in alle Richtungen bis hin zur entferntesten Falle in 390 m Distanz. Dabei überquerten sie sowohl Fichtenbestände als auch Freiflächen.

Dieses Experiment lieferte einen ersten Einblick in die Flugaktivität zweier potenzieller Überträger des Kiefernholznematoden im montanen Österreich sowie in ihre Reaktion auf Fallen und Lockstoffe, die für *M. galloprovincialis* entwickelt wurden.

Content

1 Introduction

Pine wilt disease (PWD), lethal wilting of pine trees (*Pinus* spp.), caused by feeding of the pine wood nematode (PWN) *Bursaphelenchus xylophilus* (Steiner and Buhrer 1934) Nickle 1970 (KIYOHARA and TOKUSHIGE, 1971) is currently in expansion. Therefore, PWD is worldwide considered as a threat to forest ecosystems (WEBSTER and MOTA, 2008).

The first occurrence of PWN in Europe was recorded in 1984, when it was imported to Finland with wood chips (RAUTAPÄÄ, 1986). Consequently the European and Mediterranean Plant Protection Organization (EPPO) listed PWN as quarantine pest A1 in 1985 (EPPO, 1986). In 1999, both PWN and PWD were detected in Portugal on the Setúbal Peninsula from where they spread to the Coimbra and southern Alentejo region in the inland (MOTA et al., 1999, FVO, 2010). Thus, EU research projects started in order to prevent a further expansion of PWD as well as to increase studies of PWN and its vector. Resuming the research project PHRAME from 2007, the REPHRAME (Research Extending Plant Health Risk And Monitoring Evaluation) project for detection and control of the PWN started in 2011.

PWN is transmitted by longhorn beetles of the genus *Monochamus,* populating coniferous trees all over the world and cut timber as secondary and technical pest insects, respectively. Therefore, control of the vector is one crucial point in preventing further spread of PWD. Understanding its flight habits and developing efficient traps for monitoring are prerequisites. Besides the main vector in Europe *Monochamus galloprovincialis*, PWN can be transmitted by *Monochamus sartor* and *M. sutor* (HELLRIGL, 1974, LINIT, 1988, AKBULUT and STAMPS, 2012). Both are native to Austria, mainly living on spruce (*Picea abies*) in higher elevations, but also occurring on pine trees (HELLRIGL, 1974). Although spruce is not the main host of PWN, it can survive and reproduce there as in a reservoir without killing the tree (FUTAI and SUTHERLAND, 1989). In this context, for risk assessment and monitoring of a possible PWD-spread in Austria, studies on flight activity of *M. sartor* and *M. sutor* are required. Contributing to REPHRAME, a field experiment researching on the chemical ecology of these two species was conducted in summer 2012, supported by phenological analyses of beetle emergence and flight period.

In the protected wilderness area Dürrenstein, Lower Austria, flying beetles were captured with multi-funnel traps, using three different combinations of attractants. Responses of *M. sartor* and *M. sutor* to the commercial lure Galloprotect-2D® (G2D), consisting of a

Monochamus aggregation pheromone compound (undecyloxy-ethanol) and two bark beetle pheromone components (ipsenol, methyl-butenol), were tested. The lure was developed in Spain for trapping of *M. galloprovincialis* (SEDQ, 2012). In a second treatment G2D was combined with the host tree volatile α-pinene to analyse a potential synergistic effect. The third blend additionally applied smoke volatiles. Besides preferences for the tested lures, concerning interspecific differences and dissimilarities between male and female beetles, flight distances were evaluated by marking and recapturing beetles. Additionally, flight orientation of bycatches towards attractants was analysed, especially regarding other cerambycids and potential carriers of PWN.

2 State of knowledge

2.1 Pine wilt disease

2.1.1 Historical overview

From its native range in North America (STEINER and BUHRER, 1934), PWN was accidentally introduced to Nagasaki in the south of Japan in 1905. By the 1980s, it had spread northwards over the whole country, only Aomori and Hokkaido in the north were not affected by the disease (FUTAI, 2008). In 1969, Tokushige and Kiyohara discovered PWN as causal agent of PWD (KIYOHARA and TOKUSHIGE, 1971). In 1982 and 1988, PWN reached China and Korea and expanded from the southeast to northwest in both countries (SHIN, 2008, ZHAO, 2008). Contemporaneous, PWD was rediscovered in Missouri, USA in 1979 and Manitoba, Canada in 1983. Therefore in 1985, nationwide investigations were conducted in Canada to establish the distribution area of PWN, ascertaining its presence all over the country except Prince Edward Island (SUTHERLAND, 2008). Moreover, PWN occurs in Mexico as secondary pest, where it was detected in 1992 for the first time (DWINELL, 1993).

A nematode infestating Maritime pine (*Pinus pinaster)* in south-western France in 1979 was first diagnosed as *B. xylophilus* on the basis of morphological comparisons with the Japanese PWN (BAUJARD et al., 1979). However, further investigations showed that wilt of *P. pinaster* was rather initiated by an intermediate nematode strain morphologically closely related to *B. mucronatus*, but more aggressive and able to hybridise with *B. xylophilus* (DE GUIRAN and BOULBRIA, 1986). Importations of wood chips from North America to Finland accidentally introduced PWN to Europe in 1984. Two consignments contained infested wood originating from Alabama, USA and Matane, Canada (EPPO, 1984). In order to prevent an outbreak of PWD, the consignments were destroyed and Finland decided to ban soft wood from countries with PWN occurrence (RAUTAPÄÄ, 1986). Moreover, EPPO included PWN in “A1 list of pests recommended for regulation as quarantine pests” (EPPO, 2011a) in the following year (EPPO, 1986).

No further occurrence of PWN or PWD was recorded in the EU region until 1999, when both were found in Portugal. The initial disease outbreak occurred on the peninsula of Setúbal to the south of Lisbon. Molecular biological analyses showed an agreement with the American PWN (MOTA et al., 1999). After unsuccessful attempts to eradicate PWD, it expanded to Central Portugal and was introduced to Madeira in 2009 (FVO, 2010). The conditions in the island of Madeira were favourable, leading to a rapid establishment of PWN (FONSECA et al., 2012). In November 2008, the first discovery of PWN in Spain was reported from the province of Cáceres, where it was eradicated soon afterwards (EPPO, 2010a). However, in 2010 new infestations occurred in Galicia, north of Portugal (EPPO 2010b, ABELLEIRA et al., 2011). Besides these cases, in January 2012 a third one was detected in a distance of about 600 m from the Portuguese border and is currently under eradication (EPPO, 2012).

The first find of PWN in Central Europe was made in Switzerland in May 2011, when coniferous bark for garden decoration originating from Portugal contained *B. xylophilus*. Those concerned bark consignments were not sold, preventing a distribution to nature. As a consequence of this incidence, the Swiss authorities decided to treat coniferous bark from Portugal generally as risk product, which requires laboratory examinations before sale (EPPO, 2011b, BAFU, 2011). Besides these detected finds, there is always a high risk of unrecognised PWN introduction, since low amounts of nematodes in wood or wood products are difficult to discover (BRAASCH et al., 2001).

2.1.3 Pine wood nematode

The PWN was first described as *Aphelenchoides xylophilus* by STEINER and BUHRER (1934). They found it associated with blue-stain fungi in longleaf Louisiana pine (*Pinus palustris*) cut in Orange, Texas in 1929. After further investigations, Nickle classified PWN as *Bursaphelenchus xylophilus* within the family Parasitaphelenchidae, superfamily Aphelenchoidoidea, order Aphelenchida in 1970 (NICKLE, 1970, KANZAKI, 2008). In Japan, PWN was described as *Bursaphelenchus lignicolus*, which was later recognised as a synonym for *B. xylophilus* according to morphological criteria and mating experiments (MAMIYA and KIYOHARA, 1972, NICKLE et al., 1981).

Host trees

Japanese scientists established PWN as causal agent of PWD in 1969 in the aftermath of pine tree death. Tree mortality was first suspected to be related with insect damage by Cerambycidae and Curculionidae (KIYOHARA and TOKUSHIGE, 1971). As shown in Table 1, native pine species as well as the European *P. pinaster* are severely susceptible for PWD in Japan and Korea, whereas foreign *Pinus* spp. from North America are resistant (KIYOHARA and TOKUSHIGE, 1971, MAMIYA, 1976, SHIN, 2008, FUTAI, 2008).

Table 1: Resistance of pine species (*Pinus* spp.) in Japan and Korea (KIYOHARA and TOKUSHIGE, 1971, MAMIYA, 1976, SHIN, 2008, FUTAI, 2008)

Susceptible tree species		Resistant tree species	
Japan	Korea	Resistant (exotic)	Highly resistant (exotic)
P. densiflora *P. thunbergii* *P. luchuensis* *P. pinaster* (exotic)	*P. densiflora* *P. thunbergii* *P. koraiensis*	*P. rigida* *P. elliottii* *P. caribaea*	*P. banksiana* *P. echinata* *P. palustris* *P. pungens* *P. strobus* *P. taeda*

A nationwide survey in Canada in 1985 showed that almost all pine species and some other conifers can be considered as host trees (SUTHERLAND, 2008). In the USA, at least 20 pine species as well as other coniferous trees are preferred hosts (WINGFIELD and BLANCHETTE, 1983, KINN, 1986). Table 2 gives an overview of the PWN populated tree species spectrum in North America. Beyond that, *Pinus estevesii* is a suitable host for PWN in Mexico (DWINELL, 1993). According to examinations in the United States, PWN only acts as a primary pathogen in exotic trees, which are not habitat-adapted and consequently stressed (WINGFIELD et al., 1982, WINGFIELD, 1983). Plantations of introduced *Pinus sylvestris* in Illinois were severely attacked due to their susceptibility resulting from abiotic stress. No weakening was observed because of insect or fungi infestation. Even healthy young trees were populated by PWN, similar to the incidence of PWD in Japan (MALEK and APPLEBY, 1984). Contrary to this, pine species and conifers native to North America are predominantly resistant (WINGFIELD et al., 1984), explaining the non-epidemic occurrence of PWD in natural coniferous forests of the USA (DROPKIN et al., 1981). However, greenhouse inoculation tests proved differences in susceptibilities to PWN among these native pines, reaching from highly resistant Virginia pine (*P. virginiana*) with secondary occurrence of PWN, to very susceptible slash pine (*P. elliottii* var. *elliottii*), where PWN constitutes a primary pathogen. The likewise investigated eastern white, loblolly (*P. taeda*) and pond pine (*P. serotina*) have an intermediate susceptibility (DWINELL, 1985). These findings of DWINELL (1985) conflict with results of inoculation tests reported from MAMIYA (1976). Greenhouse experiments do not necessarily reflect the situation of established trees in forests: Red pine seedlings, killed under greenhouse conditions, did not die during field observations. Furthermore, comparing the effect of PWN on red (*P. resinosa*), jack (*P. banksiana*) and

Austrian pine (*P. nigra*) in Minnesota and Wisconsin with its impact known from native Japanese pines shows that the former were not such susceptible (WINGFIELD et al., 1986).

Table 2: Host tree species in Canada and the USA (WINGFIELD and BLANCHETTE, 1983, KINN, 1986, SUTHERLAND, 2008)

<table>
<tr><th colspan="3">Host tree species</th></tr>
<tr><th></th><th>Canada</th><th>USA</th></tr>
<tr><td rowspan="7"><i>Pinus</i> spp.</td><td colspan="2"><i>P. banksiana</i></td></tr>
<tr><td><i>P. contorta</i></td><td><i>P. nigra</i></td></tr>
<tr><td colspan="2"><i>P. ponderosa</i></td></tr>
<tr><td colspan="2"><i>P. resinosa</i></td></tr>
<tr><td colspan="2"><i>P. strobus</i></td></tr>
<tr><td colspan="2"><i>P. sylvestris</i></td></tr>
<tr><td></td><td>In total:
20 species of <i>Pinus</i></td></tr>
<tr><td><i>Pseudotsuga</i> spp.</td><td><i>P. menziesii</i></td><td></td></tr>
<tr><td><i>Abies</i> spp.</td><td colspan="2"><i>A. balsamea</i></td></tr>
<tr><td rowspan="2"><i>Larix</i> spp.</td><td colspan="2"><i>L. laricina</i></td></tr>
<tr><td></td><td><i>L. decidua</i></td></tr>
<tr><td rowspan="3"><i>Picea</i> spp.</td><td><i>P. rubens</i>
<i>P. mariana</i></td><td></td></tr>
<tr><td colspan="2"><i>P. glauca</i></td></tr>
<tr><td></td><td><i>P. pungens</i></td></tr>
<tr><td><i>Cedrus</i> spp.</td><td></td><td><i>C. atlantica</i>
<i>C. deodara</i></td></tr>
</table>

The mortality rate of pine species depends on the respective populations of PWN, differing in aggressiveness between geographical regions (DROPKIN et al., 1981, FUTAI and SUTHERLAND, 1989).

In contrast to Japan, PWN was found on trees or tree parts without causing wilt symptoms in the USA. These trees were stressed by pathogens and insects in advance or died because of these infections. A coincidence of infestation by wood borers and PWN was established (WINGFIELD et al., 1982). Furthermore, PWN is provably able to survive and reproduce in dead trees and cut logs after transmission, offering a temporary habitat for PWN in absence of appropriate living host trees (WINGFIELD, 1983, WINGFIELD and BLANCHETTE, 1983).

Preferred habitats of PWN within the host tree are trunk and branches, less frequent terminal twigs. Cones and needles are unsuitable for population (MALEK and APPLEBY, 1984). PWN enters the resin canals, feeding on epithelial cells, whereupon oleoresin flow stops (MAMIYA and KIYOHARA, 1972, DROPKIN et al., 1981). Japanese red pine (*P. densiflora*) seedling inoculation experiments showed an immediate penetration of resin canals, cambium and cortex, leading to death of the affected tissues (MAMIYA, 1980). The whole process of tree dying occurs within 30 to 50 days in one vegetation period, i.e. from mid July to the beginning of August when first internal signs are ascertainable until late

August to October (MAMIYA and KIYOHARA, 1972, MAMIYA, 1976). Inoculation experiments in pine stands determined the highest and fastest mortality of trees from June to August, when the latent period was shortest, compared to lower mortality in spring and no deterioration of tree vitality in autumn until the end of December (KIYOHARA and TOKUSHIGE, 1971).

Feeding of PWN is possibly not the only factor inducing wilt symptoms in trees. Nematode transmitted bacteria producing toxins can have an additional effect on PWD onset. The pathogenicity depends on bacterial species, differing regionally in association with PWN, resulting in susceptibility of different tree species (HAN et al., 2003, VICENTE et al., 2012). HAN et al. (2003) isolated highly pathogenic biotypes I and II of *Pseudomonas fluorescens* and one less pathogenic species of *Pantoea*, colonising the surface of PWN and confirmed that sterilised PWN themselves are not lethal, using seedling inoculation experiments with *Pinus thunbergii*. Beyond that, *Pinus pinaster* inoculation studies of 35 bacterial isolates from different PWN resulted in bacteria functioning as supporting agents of nematode reproduction and virulence (VICENTE et al., 2012).

Besides the consumption of living plant tissue, PWN is mycophagous on blue stain fungi, e.g. of the genera *Ceratocystis* and *Ceratocystiopsis*, ensuring feeding in dead wood (STEINER and BUHRER, 1934, WINGFIELD, 1987).

Life cycle

PWN life cycle consists of two different forms – propagative and dispersal form. The propagative type grows without vector presence and does not serve the purpose of dissemination. The first (J1) of four juvenile stages still develops inside the egg. Feeding starts with the eclosion of J2, continuing over J3 and J4; eventually leading to tree death. Following copulation, the females oviposit. Laboratory studies showed that successful development of PWN requires a minimum temperature of 9.5 °C (MAMIYA, 1975, AIKAWA, 2008). The dispersal cycle starts with the formation of third and fourth instar dispersal juveniles J_{III} and J_{IV}, which are morphologically different from J3 and J4 of the propagative type. Dispersal J_{III} are considered as survival stage with high starvation resistance, whereas J_{IV} dauerlarvae are able to tolerate dryness. Both dispersal stages are closely related to the development of the cerambycid vector beetles, transmitting PWN to new host trees (MAMIYA, 1975, MAMIYA, 1976, ISHIBASHI and KONDO, 1977).

***Bursaphelenchus* spp.**

Worldwide, various species of *Bursaphelenchus* populate pines and other coniferous trees, without leading to PWD due to low pathogenicity. In comparison to *B. sexdentati*, *B. anamurius* and *B. vallesianus, B. mucronatus* is the most pathogenic species, according to seedling experiments in greenhouse. However, killing seedlings does not equate to an ability to kill older trees. Generally, seedling species is a more important factor in susceptibility than nematode species (DAYI and AKBULUT, 2012). Of all *Bursaphelenchus* species, *B. mucronatus* shows the highest morphological and biological affinity to *B. xylophilus*. But besides its slight capability of killing trees unlike *B. xylophilus*, its natural distribution area is larger and not expanding as rapidly as the range of *B. xylophilus* (MAMIYA and ENDA, 1979).

2.2 *Monochamus* spp.

2.2.1 Distribution and host trees

The genus *Monochamus* belongs to the subfamily Lamiinae within the coleopteran family Cerambycidae (BENSE, 1995). It is distributed throughout the holarctic, in excerpts shown in Table 3 (HELLRIGL, 1971, EVANS et al., 1996). In Central Europe, four species of *Monochamus* are found: *M. sartor* (Fabricius, 1787), *M. sutor* (Linnaeus, 1758), *M. galloprovincialis* (Olivier, 1795) and *M. saltuarius* (Gebler, 1830), all of them exclusively developing in conifers. All *Monochamus* – apart from the African species – are secondary insects in cut trees or weakened living trees. Especially recently cut timber or trees freshly killed by abiotic factors such as storm, snow, lightening, heat, drought or forest fire as well as trees infested by other insects are suitable for *Monochamus* development given the availability of bark (TRÄGÅRDH, 1929, HELLRIGL, 1971, RASKE, 1972, HELLRIGL, 1974).

The distribution area of *M. sartor* in higher elevations (800 to 1000 m, up to the timberline) is bounded by the southern slopes of the Alps up to Trentino in the south and extending to Scandinavia in the north. Eastern France forms the border in the west and Poland as well as western Ukraine in the east. Old spruce (*Picea*) of large diameters is considered as main host, whereas pine (*Pinus*) and fir (*Abies*) are rarely colonised. With a body length of 19 to 35 mm, *M. sartor* is the largest of the Central European sawyers. The smaller *M. sutor* (15 to 24 mm) is more widely spread over North, Eastern and Central Europe to the Pyrenees. Thin wood or tops of spruce are mainly infested in Central Europe and the Russian area. However, in Scandinavia, where gradations frequently occur, pines are preferred, particularly lower stem parts with thick bark (TRÄGÅRDH, 1929, HELLRIGL, 1971). But also tree tops remaining in the forest after harvest serve as breeding sites (BAKKE and KVAMME, 1992). Almost exclusively attacking pine trees, *M. galloprovincialis* causes sudden gradations in Central Europe. Spruce is only a minor host. In general, upper stem parts and branches or lower stem parts of younger trees serve as breeding site. The beetle is similar in size to the previous with a length of 12 to 25 mm. Its range, extending from Northern Africa and Asia Minor over whole Europe, continues to Siberia. *M. saltuarius* is found in Central and Eastern Europe as far as Japan, attacking weakened branches and tops of older spruce trees (80 to 120 years). Rarely, vigorous tree branches are attacked. Reaching 11 to 20 mm body length, it is the smallest indigenous Central European sawyer (HELLRIGL, 1971, HELLRIGL, 1974).

Indigenous *Monochamus* species in Central Europe are of marginal interest in the forest, if only acting secondarily there. But they cause damage in cut logs or wood stores, getting a higher economic importance as technical pest insects (HELLRIGL, 1974). With discovery of their potential vector function referring to PWN, the European *Monochamus* beetles' significance increased. Especially *M. galloprovincialis* is considered as main vector in Europe due to its preferred host tree genus *Pinus* (SOUSA et al., 2002). In the USA, PWN is most often transferred by indigenous *Monochamus scutellatus, M. mutator* and *M. carolinensis*, rarely by *M. marmorator* which only populates balsam fir (WINGFIELD and BLANCHETTE, 1983, LINIT et al., 1983). The main vector for PWN in Japan is *Monochamus alternatus* (MAMIYA and ENDA, 1972).

Table 3: *Monochamus* species of the northern hemisphere, host trees, geographical range (BENSE, 1995, EVANS et al., 1996)

Asia		
***Monochamus* species**	**Main hosts**	**Geographical distribution**
M. alternatus Hope	*Pinus, Cedrus, Abies, Picea, Larix*	Japan, Korea, Taiwan, Hong Kong, Laos, China
M. grandis Waterhouse	*Abies*, *Picea*	Japan
M. nitens Bates	*Pinus*	Japan
M. subfascianus Bates	*Ginkgo biloba*	Japan
M. tesserala White	*Pinus*	Japan, China

North America		
***Monochamus* species**	**Main hosts**	**Geographical distribution**
M. carolinensis Olivier	*Pinus*	US, Canada, Mexico
M. clamator LeConte	*Pinus contorta*	US, Canada
M. marmorator Kirby	*Abies, Picea*	US, Canada
M. mutator LeConte, syn. *M. maculosus* Haldeman	*Pinus*	US, Canada
M. notatus (Drury)	*Pinus strobus*	US, Canada
M. obtusus Casey	*Pinus, Abies, Pseudotsuga*	US, Canada
M. rubigeneus Bates	*Pinus*	US, Mexico, Guatemala, Honduras
M. scutellatus Say spp. *scutellatus*	*Pinus, Picea, Abies, Larix*	Eastern North America (incl. Mexico)
M. scutellatus spp. *oregonensis* LeConte	*Picea*	US, Canada
M. titillator (Fabricius)	*Pinus, Abies, Picea*	US, Canada

Asia + Europe		
***Monochamus* species**	**Main hosts**	**Geographical distribution**
M. galloprovincialis (Olivier)	*Pinus*	North Africa, south & central & north Europe, Russia (European), Siberia
M. saltuarius (Gebler)	*Picea*	Japan, China, Siberia, Lithuania, central & eastern Alps, middle & east Europe & south to Italy
M. sartor (Fabricius)	*Picea, Pinus*	Mid-Europe (France to Ukraine)
M. sutor (Linnaeus)	*Pinus, Picea, Larix*	China, Siberia, Russia (European), Georgia, Scandinavia, Baltic states, central & east Europe: Pyrenees, Alps
M. urussovii (Fischer), syn. *M. rosenmuelleri* Cederheim	*Abies, Larix, Picea, Pinus*	Japan, China, Siberia, Russia (Caucasus), Finland, Poland

Besides their phytophagous lifestyle, sawyer larvae are facultative predators of associated bark beetle larvae, feeding on the same substrate – phloem of recently died trees – in temporal coincidence. Both this foraging behaviour and competition on Scolytinae are summarised as intraguild predation of Cerambycidae, which *Monochamus* larvae benefit in gaining extra food as well as all available resources. However, killing and ingestion of competing bark beetles is not the only carnivorous way of feeding; cannibalism is observed in several sawyer species. But they perform neither inter- nor intraspecific predation actively, instead is mutual avoidance always preferred. Only direct contact induces feeding of other insects. Generally, *Monochamus* spp. larvae develop more rapidly when nourishing on insect protein than on plants only, explaining the advantages of an occasional carnivorous behaviour (HELLRIGL, 1971, DODDS et al., 2001).

2.2.2 Life cycle

The two life cycles in Figure 1 illustrate the close association between *Monochamus* vector beetles and PWN. *B. xylophilus* dispersal form is induced by the presence of beetle larvae in the wood (ISHIBASHI and KONDO, 1977). The various sawyer species (*Monochamus* spp.) worldwide undergo a similar life history (OCHI, 1969, MAMIYA and ENDA, 1972, HELLRIGL, 1974).

Starting in autumn and continuing in winter, sawyer larval development within the host tree takes place. Simultaneously, adult nematodes reproduce and J1, J2 stages develop (HELLRIGL, 1974, AIKAWA, 2008). When *Monochamus* larvae build pupal chambers, PWN switches from the propagative to the dispersal developmental cycle. J_{III} juveniles assemble around these pupal chambers. Beetle pupation hormones are responsible for moulting of PWN to J_{IV}, demonstrating the nematode-vector-relationship (MAMIYA, 1976, ISHIBASHI and KONDO, 1977). In early summer, J_{IV} dauerlarvae – the transmitted stage of PWN – enter the tracheal system of eclosing beetles, preferring the thoracic segments, particularly the metathorax. Besides this internal contamination, PWN dauerlarvae aggregate under *Monochamus* elytra (MAMIYA and ENDA, 1972, WINGFIELD and BLANCHETTE, 1983, LINIT et al., 1983).

Newly emerged beetles feed on young needles and bark of twigs of vigorous trees to become sexually mature. The feeding wounds enable PWN to invade these trees. *Monochamus* beetles' maturation feeding is therefore considered as primary pathway of transmission (MAMIYA and ENDA, 1972, MAMIYA and ENDA, 1979, NAVES et al., 2007). Transmission reaches its peak during the first six weeks, particularly in week two, after beetle eclosion. Generally, males and females transmit PWN with equal success. Transfer ends in the ninth week, although beetles still carry nematodes.

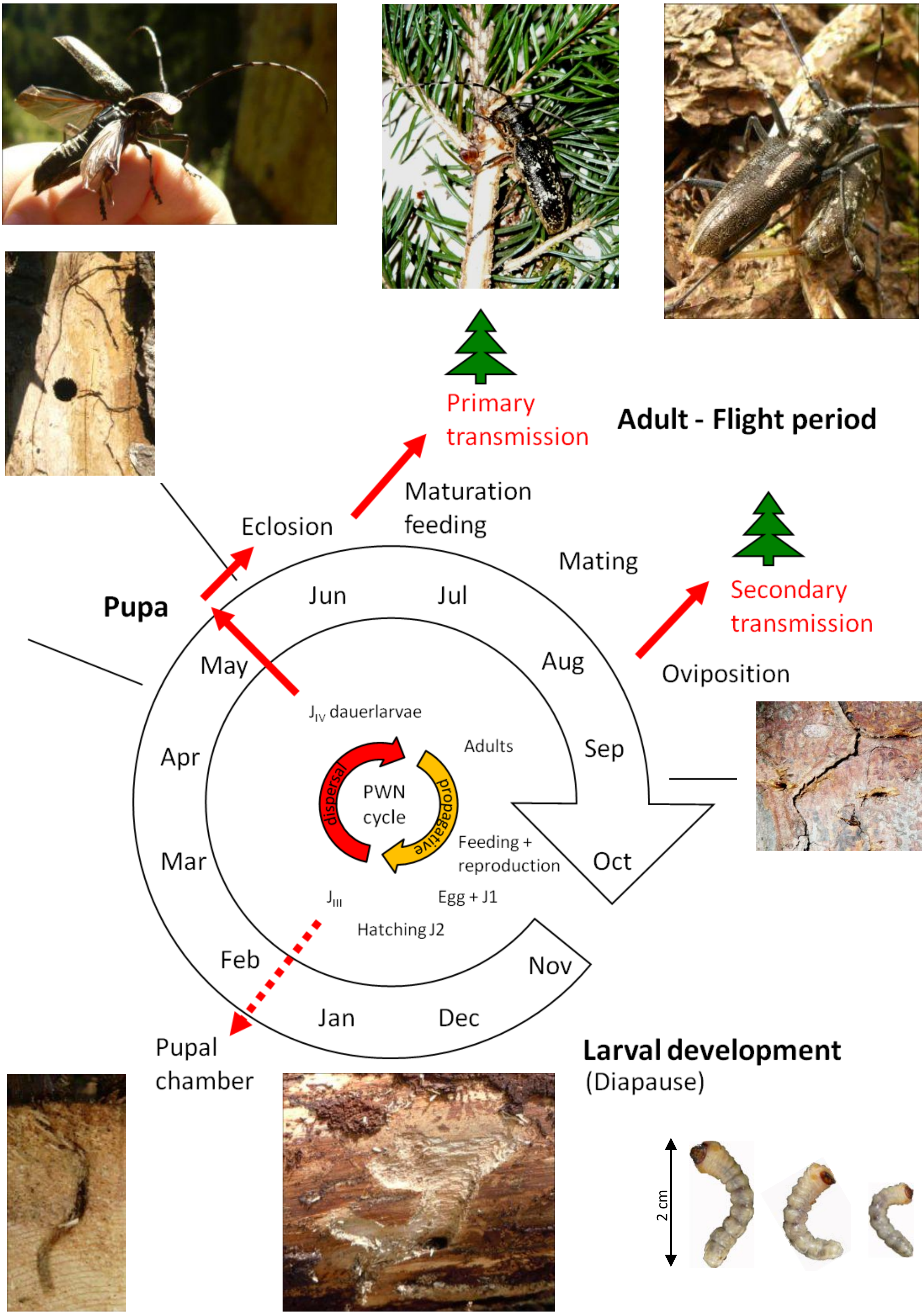

Figure 1: Life cycle of *Monochamus* spp. (outer circle) and pine wood nematode (PWN) *Bursaphelenchus xylophilus* (inner circle, red: dispersal, yellow: propagative cycle); transmission way of PWN to new host trees (red arrows): from arrangement around sawyer pupal chamber, entering eclosing beetles, infecting trees through feeding wounds during beetle maturation feeding (primary transmission) and through slits for *Monochamus* egg deposition (WINGFIELD et al., 1984), modified, own photos.

Transmission by maturation feeding is independent from the vectors' longevity and feeding intensity (LINIT et al., 1983, NAVES et al., 2007). However, ambient temperature correlates with the number of transferred nematodes. Cool temperatures hinder PWN leaving the sawyer due to slowed movement, consequently reducing transmission efficiency (JIKUMARU and TOGASHI, 2000).

After reaching sexual maturity, feeding serves as uptake of nutrients and continues throughout the adult lifetime. The beetles copulate; subsequently females oviposit on weakened host trees, again transferring PWN as secondary transmission (OCHI, 1969, HELLRIGL, 1974, WINGFIELD and BLANCHETTE, 1983, AIKAWA, 2008). Induced PWD is not the principal cause of tree death in this case. The trees function as reservoirs for PWN (WINGFIELD, 1983).

These routes of transfer demonstrate the significance of sawyers in the life cycle of PWN as well as their central role in PWD epidemics. Beetles are loaded with high numbers of *B. xylophilus* on average 15 000 (MAMIYA and ENDA, 1972), up to 289 000 at most per single beetle (KOBAYASHI et al., 1984), transmitting 96 nematodes at maximum during oviposition per pair of beetles. The amount of successfully transferred PWN during egg deposition is mainly influenced by the vectors' lifespan and oviposition rate as well as their load with nematodes. Total load is directly correlated with vector weight; in contrast, size has no impact (WINGFIELD and BLANCHETTE, 1983, LINIT et al., 1983).

Oviposition behaviour and larval development of the European *Monochamus* species is similar. Period of egg deposition extends from June to September, depending on the time of beetle eclosion. Before oviposition, female beetles gnaw slits across the stem, wounding the bark as entrance way of PWN. Each day, females lay one to six eggs deeply into the phloem, in total 45 to 120. One or two weeks later, the larvae emerge, first feeding on the phloem, continuing on the sapwood surface, later tunnelling into the wood. Feeding on phloem continues. At the end of this hook shaped tunnel into the sapwood, the pupal chamber is created. Larval development of feeding larvae can be finished within three and a half to six months, plus genetically-fixed winter dormancy period, which lasts up to four to five months in case of larval development completion in autumn. Number of instars depends on species, as four are reported from *M. sutor* and five from *M. urossovi* (BAKKE and KVAMME, 1992). On the contrary, *M. sartor* reared in laboratory requires ten larval stages. Fully grown larvae, so-called prepupae, stop feeding and prepare the pupal chamber (HELLRIGL, 1971). Under unfavourable conditions, larvae undergo a diapause of several months up to one year, explaining a prolonged life cycle of two years, while sawyers generally are univoltine (HELLRIGL, 1971, BAKKE and KVAMME, 1992, NAVES et al., 2008). After a two to three weeks long pupal stage, young beetles hatch boring out of the stem through a circular borehole (OCHI, 1969, HELLRIGL, 1974). Lifetime of adult *Monochamus* spp. varies between two and 180 days, but females tend to grow older than males (TOGASHI and SEKIZUKA, 1982).

Despite its coordination, the relationship between *Monochamus* spp. and PWN cannot be considered as truly mutualistic. Undoubtedly, PWN benefits from its transport by sawyers, which is essential for colonising new hosts (MAMIYA and ENDA, 1972, LINIT, 1988). The vectors profit by availability of dying or dead trees as breeding habitats through PWD – however, only in case of introduced exotic trees or in areas not belonging to PWN's native range (IKEDA et al., 1980a, IKEDA and ODA, 1980, JIKUMARU and TOGASHI, 2000, NAVES et al., 2008). On the other hand, nematode population of beetles shortens their lifespan to a certain extent, depending on the sawyer's individual nematode load (NAVES et al., 2007), due to limitations of the gaseous exchange when the tracheae are filled with nematodes. Likewise, the females' fecundity diminishes with an increase of carried nematodes, leading to less deposited eggs when the amount of nematodes exceeds 10000 (TOGASHI and SEKIZUKA, 1982). Generally, sawyers do not depend on a nematode association and can complete their life cycle without trees dying from PWD. This suggests that PWN has adapted to *Monochamus* spp. in evolution and not vice versa (MAMIYA and ENDA, 1972, MAMIYA, 1976).

2.2.3 Host and mate finding behaviour

The occurrence of *Monochamus* spp. depends on its discovering ability of suitable survival conditions, from landscape-scale over host tree-scale to microhabitat choice (SAINT-GERMAIN et al., 2004). Host seeking behaviour is controlled by visual and chemical cues. The beetles orientate towards their host trees by recognition of the dark-coloured stem silhouette (DE GROOT and NOTT, 2001, MCINTOSH et al., 2001). Further, approximately three weeks after eclosion, they start reacting to volatiles of deteriorating trees or cut logs, such as oleoresins, terpenes and ammonia. Both males and females are equally attracted to these substances, signalising preferences for weakened but still fresh trunks (HELLRIGL, 1971, IKEDA et al., 1980b, KOBAYASHI et al., 1984). Moreover, as other cerambycids, *Monochamus* spp. orientate towards pheromone components of intraguild insects, which indicate suitable hosts and presence of mates (ALLISON et al., 2012).

Host finding – host volatiles

Suitable weakened trees emit the bicyclic monoterpenes α-pinene, β-pinene, camphene and sabinene, acyclic β-myrcene and the monocyclic monoterpenes α-terpinene, limonene, β-phellandrene, γ-terpinene, ρ-cymene and terpinolene as well as ethanol, regardless whether the trees have been felled or infested with PWN (IKEDA et al., 1980a, IKEDA and ODA, 1980, IKEDA et al., 1980b). Furthermore, *Monochamus* are attracted to diterpenes (pimaral) and sesquiterpenes (juniperol, longifolene), which are released by healthy pine trees (SAKAI and YAMASAKI, 1990, PAJARES et al., 2004).

Host volatiles in general have relatively low attractiveness for sawyers as evidenced for *M. galloprovincialis*, but they can act as synergistic components. A blend of ethanol and turpentine (major components α-pinene, β-pinene, limonene, camphene and longifolene), can increase the luring effect for *M. galloprovincialis* (PAJARES et al., 2004). In fact, ethanol itself further enhances attraction to other host odours, which was shown in experiments with *M. alternatus* (IKEDA et al., 1980a). However, by process of elimination, the key role of α-pinene in synergism before other host stimuli was established for *M. galloprovincialis* (IBEAS et al., 2006).

Host finding – forest fire

Typical for all softwood-colonising *Monochamus* species is their attraction to fire injured trees (HELLRIGL, 1971). Such recently killed or weakened trees release smoke volatiles probably luring sawyers up to distances of several kilometres. This behaviour is distinctive in *M. sutor* in Scandinavia, but also in *M. urossovi* and *M. galloprovincialis*, reaching high population densities. Intensity of fire damage on trees directly correlates with the extent of subsequent beetle infestation (TRÄGÅRDH, 1929, HELLRIGL, 1971, MEYER, 2002). *M. scutellatus* responds to burnt hosts, however notably colonising the edges of stands affected by fire with direct contact to healthy host trees. Favouring such habitats is based on the combination of different food requirements of adults and larvae. Moreover, large tree diameters are preferred because of their thicker bark, since water containing cortex after fire secures the survival conditions for the sawyers' larval instars (MEYER, 2002, SAINT-GERMAIN et al., 2004). Responses to fire odours are also known from other Cerambycidae such as *Arhopalus tristis* and *A. ferus*, evidencing that longhorn beetles are physically adapted to post-fire habitats as the constitution of their antennal receptors shows (SUCKLING et al., 2001, SANTOLAMAZZA-CARBONE et al., 2011). Smoke distributes over a larger area than host volatiles, attracting sensitive insects over long distances (SAINT-GERMAIN et al., 2004). The buprestid genus *Melanophila* exclusively colonises fire damaged trees, detected within distances of 50 kilometres. Their infrared receptors as well as highly sensitive antennae, operating up to odour concentrations of parts per billion, localise suitable breeding material. Perception mainly orientates towards 2-methoxyphenol (guaiacol) derivates. General fire detection capability of a species varies depending on its habitat requirements, since *Phaenops cyanea* (Buprestidae) and *Ips typographus* are less able to recognize guaiacol, compared with M*elanophila* spp. (SCHÜTZ et al., 1999).

Host finding – non-host volatiles

Sawyers' host seeking is further controlled by non-host volatiles, e.g. Conophthorin originating from bark of angiosperms repelling *M. scutellatus* and *M. clamator* (ALLISON et al., 2004). Negative response ensuing in avoidance of inappropriate trees is the consequence, likewise proved for *A. tristis,* which is deterred by green leaf substances also reducing oviposition (SUCKLING et al., 2001).

Host finding – bark beetle volatiles

Furthermore, *Monochamus* spp. colonise the same habitat as bark beetles. The occurrence of *M. sartor* is linked to the presence of *Trypodendron lineatum* and *Ips typographus*. *M. sutor* is often found together with *Pityogenes chalcographus*, *Pityophthorus pityographus*, *Polygraphus poligraphus* and less frequent with *Pityophthorus exsculptus*, *I. typographus* and *Xyloterus lineatus* within same tree parts. Also *M. sartor* is known to populate these trees simultaneously, but prefers the lower trunk (HELLRIGL, 1971). Therefore, the incidence of Scolytinae indicates appropriate breeding conditions for sawyers (ALLISON et al., 2003). Thus, they also orientate towards bark beetle (*Ips* spp.) pheromone components such as ipsenol, ipsdienol, *cis*-verbenol and methyl-butenol, besides host volatiles. This kairomonal response was observed for male and female *M. scutellatus* and male *M. clamator* in Canada as well as for both sexes of *M. galloprovincialis* in Spain (ALLISON et al., 2003, PAJARES et al., 2004). Particularly those bark beetles (*Ips* spp.), which are able to kill trees finally; lead *Monochamus* to appropriate host trees. On the contrary, *Dendroctonus* spp. infested trees mostly do not die and consequently do not provide breeding material for sawyers. In principle, *Monochamus* are exclusively associated with scolytine species requiring similar larval conditions regarding the constitution of host substance (ALLISON et al., 2003, ALLISON et al., 2004). But not only aggregation pheromone components evoke reactions of sawyers, also bark beetle repellents such as verbenone signalise breeding trees for *Monochamus*. These completely colonised trees offer an additional food source to sawyers, optionally nourishing on scolytine larvae (DODDS et al., 2001, IBEAS et al., 2006). Following bark beetle attractants is a congenital behaviour of sawyers, since they generally react to pheromones produced by bark beetles not populating the sawyers' current habitat, as proved for *M. galloprovincialis* responding to ipsenol in experiments (IBEAS et al., 2006).

In contrast, weakened trees can be infested by *Monochamus* spp. before bark beetles colonised the trees. This was observed on wounded spruce trees (*Picea abies*) with relatively low diameters after an avalanche (BLACKWELL, 2011).

Mate finding

Male and female beetles encounter for copulation, because both are equally attracted to host trees in spatial and temporal coincidence. During the initial step mature males, at first present on the host tree (NAVES et al., 2008), lure other males and particularly females by producing a short distance volatile aggregation pheromone compound. Studies of *M. sutor* (REPHRAME Periodic Report, 2012)*, M. galloprovincialis, M. alternatus, M. carolinensis, M. titillator, M. scutellatus scutellatus* and *M. notatus* ascertained that males of all these species release and females react to the compound 2-undecyloxy-1-ethanol (so-called "Monochamol"). However, for *M. notatus,* only responses and no production were established until 2012. This evidences the occurrence and attractancy of identical pheromone compounds within the genus *Monochamus* (PAJARES et al., 2010, TEALE et al., 2011, ALLISON et al., 2012, FIERKE et al., 2012). The structural alliance of 2-undecyloxy-1-ethanol to the dialkyl esters, produced by *Monochamus leuconotus* and *Anoplophora glabripennis*, demonstrates the common base of lures inside the lamiin tribe Monochamini (WICKHAM et al., 2012). Attraction of female and male *M. carolinensis* and *M. titillator* to (2*R**,3*R**)-2,3-hexanediol combined with α-pinene might indicate the function of the previous as pheromone component, as it is for Prioninae and Cerambycinae (ALLISON et al., 2012).

Subsequently, in the second step male antennae recognise approaching females through body touch taking the contact pheromone (HELLRIGL, 1971, KIM et al., 1992). Moreover, uptake of these hydrocarbons in the females' cuticle occurs by licking and accompanied contact with labial and/or maxillary palpi (IBEAS et al., 2009). Mounting and copulation of *M. alternatus* are elicited mainly by ether and faintly by benzene fractions of the female extracts (KIM et al., 1992). Spanish studies of *M. galloprovincialis* established, that most abundant cuticular hydrocarbons are present in both sexes. Only n-hexacosane was more plentiful in the females' body surface, probably playing a major role in recognition and copulation start-up (IBEAS et al., 2009). Besides olfactory cues, antennal- and mouth part-cuticle contact, in some cases visual perception is also involved in female identification (HELLRIGL, 1971, KIM et al., 1992).

Although long range pheromone production is not proved for females of *Monochamus*, female beetles of *A. glabripennis* emit such odours for male attraction. The contact pheromones oxidise under ozone and photo-irradiation to volatile substances. Cuticular alkenes, initiating the copulation process in males, constitute precursors of linalool oxide as well as the aldehydes heptanal, nonanal and hexadecanal, which are ultimately antennally active and perceptible over long distances (WICKHAM et al., 2012).

Oviposition stimulants

Linked to host and mate finding, oviposition behaviour of cerambycid beetles is also directly influenced by volatile chemical substances. However, the same tree produced compounds responsible for the discovery of breeding and feeding substrate do not automatically induce oviposition. Especially ethanolic tree compounds as well as the synergistically acting flavanoids D-catechin and 2,3-trans-dihydroquercetin-3'-O-β-D-glucopyranoside cause oviposition of *M. alternatus* (ALLISON et al., 2004). Furthermore, *M. alternatus* females respond to the sesquiterpene juniperol, the diterpene pimaral and in particular to a combination of these (SAKAI and YAMASAKI, 1990).

M. sutor females prefer laying eggs in fire injured pines with a tree damage proportion of more than 50 %. Stems scorched all around are more attractive than those burnt on one side. Severity of fire damage directly correlates with beetle attack intensity, leading to deposition of up to 100 eggs per stem meter in cases of proliferation (TRÄGÅRDH, 1929). Similar reactions to burnt trees are established in *A. tristis* (SUCKLING et al., 2001). Egg deposition is moreover controlled by avoidance of already infested stems or stem parts preventing larval competition and cannibalism. Therefore, females mark their filled eggs slits with secretions repelling conspecific females (PEDDLE, 2000, ALLISON et al., 2004).

2.2.4 Application of *Monochamus* spp. chemical ecology in trapping

Employing host and mate finding behaviour of *Monochamus* spp. in monitoring and pest management, considering control of PWD in particular, led to the development of suitable trap types and commercial lures. Generally, traps with a vertical black shape imitating the prominent stem silhouette are recommended to catch wood-colonising Cerambycidae and Buprestidae, according to DE GROOT and NOTT (2001, 2003). Black-coloured multi-funnel, cylinder, single-vane and cross-vane traps differ in efficacy depending on sawyer species (DE GROOT and NOTT, 2001, MCINTOSH et al., 2001, PAJARES et al., 2004). Increased capturing results of *M. scutellatus* and *M. mutator* were established with black traps in Canada, whereas *M. notatus* did not prefer black over clear traps (DE GROOT and NOTT, 2001). Despite high capturing results of *M. scutellatus* and *M. mutator* with crossvane traps compared to low catch with multi-funnel traps in Canada (MOREWOOD et al., 2002, DE GROOT and NOTT, 2003), to trap European *Monochamus* species particularly in mountainous regions the latter trap type is more suitable. Besides the stability in strong winds and space-saving transport, which are essential in trapping experiments in mountains, high *Monochamus* spp. trap catches were attained with multiple funnel traps in Europe. For trapping of *M. galloprovincialis* in Spain and Portugal, both Teflon coated multi-funnel and crossvane traps were adequate. Compared to standard traps, the Teflon film seemed to be an important factor to gain highest beetle amounts (Hoch, pers. comm., 2012, REPHRAME Periodic Report, 2012). Further tests showed no difference in trapping efficiency between new and one year old Teflon coated traps (REPHRAME Periodic Report, 2012). Generally, durability of the Teflon film after repeated trap use in the field is not known (REPHRAME Periodic Report, 2012). The manufacturer ECONEX® gives the operation of traps up to seven years.

For successful *Monochamus* spp. trapping, escape-proof dry collection cups either with insecticide or enlarged depth and Teflon film or water containing collection cups are required (PAJARES et al., 2004). German studies showed the escape of all trapped *M. galloprovincialis* within one hour after capture, when standard dry receptacles were employed (HIELSCHER pers. comm., 2013).

Traps are provided with operative, but cost-efficient attractants consisting of host volatiles combined with bark beetle components and the sawyer aggregation pheromone compound. High trap catches are induced by adding bark beetle pheromones, especially ipsenol (2-methyl-6-methylene-7-octen-4-ol) and ipsdienol (2-methyl-6-methylene-2,7-octadien-4-ol), to the host tree volatiles α-pinene and ethanol, although ipsenol has a stronger attracting effect (ALLISON et al., 2003, PAJARES et al., 2004). Additions of *cis*-verbenol and 2-methyl-3-butenol can further increase the number of catches. However, using a blend consisting of more than three components is uneconomic and not practicable in pest management. Spanish investigations of the kairomone orientated flight activity of *M. galloprovincialis* resulted in a most effective mix of α-pinene, ipsenol and 2-methyl-3-butenol (IBEAS et al., 2006). An addition of the *Monochamus* aggregation pheromone compound 2-undecyloxy-1-ethanol synergises the efficacy of the whole mixture (PAJARES et al., 2010). For practical application of the sawyers' chemical ecology, commercial lures are available (Table 4).

Table 4: Commercial lures for sawyer beetles (EBNER, Witasek, pers. comm., 2012, SEDQ, 2012, Alpha Scents, 2013)

State	**Company**	**Product**	**Target species**	**Content**
Austria	Witasek	MG-Kombi® (successor to Gallowit®)	*M. galloprovincialis*	• Host: ethanol, α-pinene • Bark beetle: ipsenol, ipsdienol, methyl-butenol
Spain	SEDQ	Galloprotect 2D®	*M. galloprovincialis*	• Bark beetle: ipsenol, 2-methyl-3-buten-2-ol • *M. galloprovincialis* pheromone compound: 2-undecyloxy-1-ethanol
		Galloprotect Pack®	*M. galloprovincialis*	• G2D + α-pinene
USA	Alpha Scents	Monalt®	*M. alternatus*	Not specified by manufacturer
		Monoch®	Longhorn beetles	• Host: α-pinene, ethanol • Bark beetle: ipsenol

2.3 PWN association with other Coleopteran species

Besides *Monochamus* spp., also other beetle species are known to carry PWN (Table 5). According to experiments of WINGFIELD and BLANCHETTE (1983), smaller cerambycids contained less PWNs.

Some of these coleopteran species show the same reaction to host volatiles like *Monochamus* spp. The blend of monoterpenes α-pinene, β-pinene, camphene, sabinene, β-myrcene, α-terpinene, limonene, γ-terpinene, ρ-cymene, terpinolene combined with ethanol attracted *Spondylis buprestoides*, *Arhopalus rusticus* and the curculionid *Shirahoshizo* spp., besides *M. alternatus* in Japan (IKEDA et al., 1980a).

During the EU-project PHRAME relationships between beetles and *Bursaphelenchus* species were determined in Spain and Portugal; ensuing in two cerambycid, seven bark beetle and one weevil species in Spain as well as five scolytine and one other curculionid species in Portugal (PHRAME, 2007). In principle, all insects with an ability to carry nematodes might be regarded as possible vectors for PWN (JURC et al., 2012).

Table 5: Coleopteran species in association with *Bursaphelenchus xylophilus* (MAMIYA and ENDA, 1972, WINGFIELD and BLANCHETTE, 1983, LINIT et al., 1983, LINIT, 1988, PHRAME, 2007)

Species	**Hosts**
Cerambycidae	
Acaloptera fraudatrix	*Pinus*
Acanthocinus griseus	*Pinus*
Amniscus sexguttatus	*Pinus, Picea*
Arhopalus ferus	*Pinus*
Arhopalus rusticus	*Pinus*
Arhopalus rusticus obsoletus	*Pinus, Picea*
Asemum striatum	*Pinus, Abies*
Corymbia succedanea	*Pinus, Abies, Picea*
Neocanthocinus obsoletus	*Pinus*
Neocanthocinus pusilus	*Pinus*
Spondylis buprestoides	*Pinus*
Uraecha bimaculata	*Pinus*
Xylotrechus saggitatus	*Pinus*
Buprestidae	
Chrysobothris spp.	*Pinus*
Scolytinae	
Hylastes ater	*Pinus*
Hylurgus ligniperda	*Pinus*
Ips acuminatus	*Pinus*
Ips mannsfeldi	*Pinus*
Orthotomicus erosus	*Pinus*
Pityogenes bidentatus	*Pinus*
Tomicus piniperda	*Pinus*
Other Curculionidae	
Hylobius sp.	*Pinus*
Hylobius pales	*Pinus*
Pissodes approximates	*Pinus, Picea*
Pissodes validirostris	*Pinus*

2.4 EU protective measures and projects

Generally, PWD occurrence worldwide depends on five factors: climate, topography, nematode pathogenicity, vector biology and distribution of susceptible tree species. Areas with mean summer temperatures above 20 °C and elevations below 700 m are acutely endangered (RUTHERFORD and WEBSTER, 1987, RUTHERFORD et al., 1990). Even if appropriate host trees are available in these regions, PWD will not break out in absence of PWN and its vectors, showing the situation in Japan before 1905. Suitable abiotic preconditions as well as vector and PWN presence only result in occasional incidences of PWD in North America due to lacking susceptible trees. Summarised, PWD occurrence is always linked to stressed, exotic trees in PWN's native range as in the US and Canada or indigenous trees in warm climates, where PWN is introduced, as in Asia and South West Europe. Hence, PWD outbreaks are linked to human activity: inadequate sylviculture, transport and trade (RUTHERFORD et al., 1990).

Owing to the establishment of PWN in Portugal and Spain and the consequent risk of expansion to other European countries, the EU instituted missions and projects for study, monitoring and pest management of PWD. From 2002 to 2007, PHRAME (Plant Health Risk and Monitoring Evaluation) was conducted, pursuing the object of developing enhanced quarantine pest risk analysis methods according to the model system of *B. xylophilus* in Portugal. Resuming PHRAME, in 2011, the EU-project REPHRAME (http://www.rephrame.eu) with increased research started in order to create: improved techniques for detection, control and eradication of pine wood nematode in support of EU Plant Health policy. Furthermore, the Council of the EU included PWN and foreign *Monochamus* species in directive 2000/29/EC to prevent their introduction and spread within the Community (EC, 2000). Based on this, the European Commission promulgated the implementing decision 2012/353/EU against dissemination of PWN within the Union (EC, 2012).

Contributing to REPHRAME, several research institutions in EU member states carry out work packages treating currently open questions on PWD. The Austrian Federal Research and Training Centre for Forests, Natural Hazards and Landscape (BFW) is inter alia responsible for work packages concerning vector phenology and improved vector trapping. Flight period and capacity under climatic impacts are determined for *M. galloprovincialis*, but also for *M. sartor* and *M. sutor*. Field studies of trapping, colouring and recapturing beetles including tests of different trap types and synthetic lures are required. Especially, investigating chemical ecology of two mainly spruce colonising sawyers *M. sartor* and *M. sutor* constituted the aim of this thesis.

In the course of a PWD expansion to Central Europe, approximately 4.5 % (ca. 152 000 ha) of Austria's managed forest area (total: 3 367 000 ha) would be endangered only regarding PWN's main host pine (*Pinus sylvestris*). In consideration of all further coniferous trees as potential hosts and reservoirs of PWN, ca. 59 % (approximately 1 987 000 ha) would be periled, including *Picea* with 50.8 %, *Abies* 2.4 %, *Larix* 4.6 % and other conifers 1.2 % (RUSS, 2011). The major vector *M. galloprovincialis* is known to transfer *B. xylophilus* in Portugal and Spain (SOUSA et al., 2002). However, also Central European specimens carry nematodes (*B. mucronatus* and *Diplogasteroides haslacheri*) as evidenced by studies in Germany (MEYER, 2002), leading to the conclusion that indigenous representatives of *M. galloprovincialis* in Austria are suitable vectors of PWN. Furthermore, *M. sartor*, *M. sutor* and *M. saltuarius,* naturally populating conifers in Austria, function as potential PWN transmitters in case of PWD occurrence (HOYER-TOMICZEK and TOMICZEK, 2005). Both high risks of introduction of these four sawyer species into new habitats and their settlement ability would increase the dissemination speed of PWD (EVANS et al., 1996, MEYER, 2002). Therefore, detailed knowledge of their biology and chemical ecology are required for successful surveillance and control. In this context, data on flight activity of *M. sartor* and *M. sutor,* particularly concerning the attractiveness of insect and tree produced volatiles, were carried out.

3 Material and methods

3.1 Study area Dürrenstein

The study area (Figure 2) is situated in Dürrenstein Wilderness Area in the southwest of Lower Austria bounding on Styria enclosing the Dürrenstein Mountain (1878 m) as part of the Northern Limestone Alps. The local climate is characterised by 3.9 °C annual mean temperature as well as 2300 mm precipitation per year.

Figure 2: Study area Hundsau and the Dürrenstein mountain in the Dürrenstein Wilderness Area, two avalanche swaths, dead spruce trees were killed by spruce bark beetle attacks during the last years.

Containing unspoiled nature with a rare species complex, Dürrenstein area was classified as nature reserve in 2002 and Austria's first wilderness area since 2003, according to management guidelines of the International Union for Conservation of Nature and Natural Resources (IUCN). In 1875, Albert Rothschild started to protect the forest areas on the reverse side of the Dürrenstein mountain chain, later called after him primeval forest "Rothwald", to date without any human influence. The study site "Hundsau", which is located on the Dürrenstein south slope, was subjected to forest management until the 1950s before being incorporated in the nature reserve. Approximately 2500 ha nature were conserved in course of the EU LIFE-project from 1997 to 2001, extending to additional 1000 ha in 2013 (BLACKWELL, 2011, WILDNISGEBIET DÜRRENSTEIN, 2012). This whole area belongs to category I of IUCN classification: strict nature reserve (Ia), wilderness area (Ib). The first protects biodiversity, geomorphologic and geological features in the sense of conservation under strongly limited or without anthropogenic influence, but with purpose of serving as study and monitoring area. A strict nature reserve requires a comprehensive spectrum of expected indigenous species in all corresponding expected native ecosystems. Any natural processes must run automatically without human intervention. In contrast, a wilderness area is mostly larger and does not strictly limit or prohibit human activities. Use is allowed as long as natural cycles are not disturbed. The area must not be affected by modern infrastructure and excessive anthropogenic activity. Slight eco-tourism and scientific research are desired in order to offer insights into the functionality of almost pristine nature (DUDLEY, 2008).

The typical natural vegetation in Dürrenstein Wilderness Area consists of beech-fir-spruce-forests (*Fagus sylvatica, Abies alba, Picea abies*) up to the timberline, partially mixed with hardwood, e.g. ash (*Fraxinus excelsior*), sycamore (*Acer pseudoplatanus*) and whych-elm (*Ulmus glabra*) on more humid slopes. Pure spruce stands are of small size, not dominating the vegetation. In 2009, two avalanches of dust and wet snow formed large swaths into the forest in Hundsau. Stand damages at the edges and weakened trees were the consequence, leading to a bark beetle (*Ips typographus*) attack on spruce (BLACKWELL, 2011, WILDNISGEBIET DÜRRENSTEIN, 2012). The deteriorating trees provided breeding habitats for *Monochamus* spp., which were observed at high abundance in recent years (BLACKWELL, 2011, HOCH, pers. comm., 2012).

3.2 *Monochamus* spp. emergence from logs

In order to determine the time of beetle emergence and beginning of flight, *Monochamus* infested logs from the study area Hundsau were brought to BFW on 10 May 2012. For that, one tree in an approximately 115-year-old, recently died pure spruce (*Picea abies*) stand (Figure 3) was felled. The tree showed fresh larval feeding tunnels on cortex and sapwood surface as well as oval holes formed by larvae boring into the wood, but lacked circular emergence holes of adult beetles. Activities of a *Monochamus* population in Hundsau were already noticed by HOCH (pers. comm., 2012) and BLACKWELL (2011) in 2009 and 2010. The felled tree was cut into 0.5 m logs for easier handling from bottom to top up to 0.2 m diameter, since thinner stem parts are not colonised by *M. sartor* and *M. sutor* larvae (HELLRIGL, 1971).

Figure 3: *Monochamus* spp. infested spruce stand in Hundsau, Dürrenstein Wilderness Area

Those logs with highest infection probability, in total seven (numbered D1 – D7), according to outer signs were transported to BFW for control of beetle emergence. The stem parts were stored outside and covered with a plastic net (Figure 4), to prevent the escape of hatching *Monochamus*. Nets possibly could not prevent the escape of other insects; due to their installation they were not completely tight. Therefore, only emergence of *Monochamus* spp. was recorded. Indications of larval activity, using the amount and colour of shavings, as well as beetle eclosion were recorded. After surveillance of the eclosion period from 1 June to 26 September 2012, logs remained in their outdoor storage before they were carefully chopped and sawn in April 2013, in order to check for larvae, pupae or adults. Living objects were put back into the tunnel and chopped stem parts were put together again for development completion in the breeding cage.

To establish the relation between weather conditions and hatching time of adult beetles, daily mean air temperature values were taken from the Central Institute of Meteorology and Geodynamics (ZAMG), Vienna. Furthermore, temperatures were measured hourly by a data logger fixed on one stem part from 15 June to 31 October 2012.

Figure 4: *Monochamus* spp. infested logs from Hundsau, Dürrenstein Wilderness area, with nets against beetle escape at BFW, Vienna

3.3 Trapping *Monochamus* spp.

3.3.1 Trap arrangement

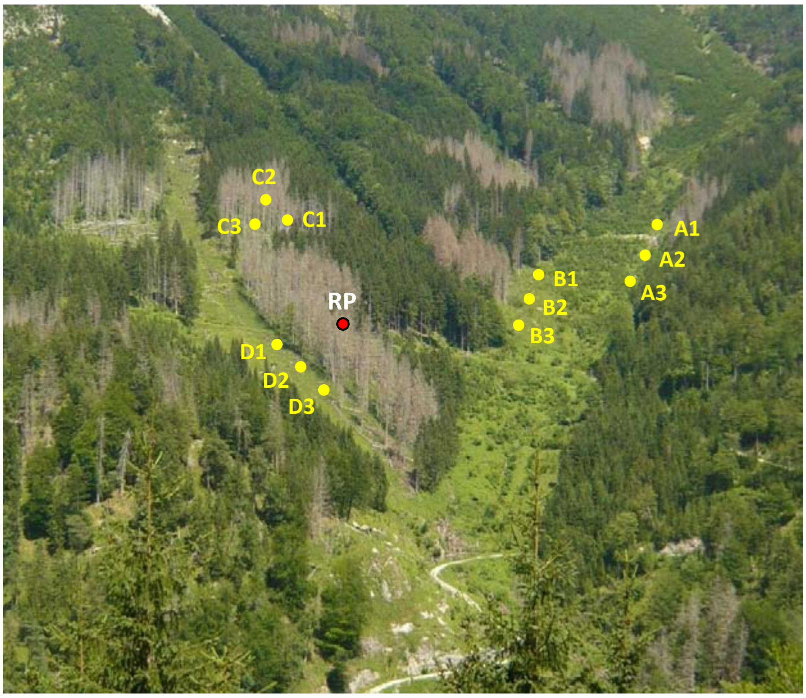

Figure 5: Randomised blocks (A – D) of traps and central point for *Monochamus* release (RP) in Hundsau, Dürrenstein Wilderness Area

The attractiveness and effect of insect and tree produced volatiles on flight behaviour of *M. sartor* and *M. sutor* were analysed in Hundsau study area from 10 July to 20 August 2012. During this period, 12 ECONEX® multiple funnel traps were installed in 4 groups (A, B, C, D) with each 3 traps in randomised block design on open space for sufficient visibility of the prominent trap silhouette (Figure 5, Table 6). The first trap groups A, B were placed on a former avalanche swath down the slope. Group C formed a triangle in a dead open spruce stand, whereas D was installed alongside the second avalanche swath. The trap groups altogether surrounded the spruce stand, which supplied material for beetle eclosion surveillance (vide 3.2). Traps were placed approximately 50 m apart within trap groups. Moreover, the traps were arranged with minimum distances of 50 m to bark beetle traps, which were installed for another experiment in the area.

Table 6: Coordinates of trap sites at study area Hundsau

	Latitude (N)	Longitude (E)	Altitude (m)
A1	47° 46' 34"	15° 02' 48"	978
A2	47° 46' 33"	15° 02' 45"	963
A3	47° 46' 33"	15° 02' 44"	960
B1	47° 46' 34"	15° 02' 40"	933
B2	47° 46' 34"	15° 02' 39"	926
B3	47° 46' 32"	15° 02' 37"	914
C1	47° 46' 38"	15° 02' 35"	950
C2	47° 46' 39"	15° 02' 34"	956
C3	47° 46' 37"	15° 02' 32"	938
D1	47° 46' 34"	15° 02' 27"	893
D2	47° 46' 33"	15° 02' 27"	877
D3	47° 46' 32"	15° 02' 24"	872

In coordination with other REPHRAME partners, ECONEX® multi-funnel traps were employed (Figure 6). They were mounted on 2.5 m wooden poles on 9 July. The traps consisted of a black top, 12 black overlapping funnels and a white collection bucket of 0.1 m in diameter. All parts were equipped with a Teflon film on their surface for more efficient capture. Since *Monochamus* spp. trapping was combined with a mark-recapture experiment and to avoid massive removal of sawyers from the wilderness area, only dry receptacles without insecticide were used. To prevent insect escape, extended receptacles with 0.2 m depth and net bottom draining water were employed. Trap size reached a total of 1.1 m in height and 0.3 m in diameter. Starting with trap installation, fresh spruce twigs with needles and bark for beetle feeding were provided in the collection cups and renewed at each trap control. Traps were emptied every three to four days.

Figure 6: ECONEX® 12-unit multi-funnel trap in Dürrenstein Wilderness Area

3.3.2 Applied attractants

All traps were activated on 10 July 2012 by placing the lures on the first or second funnel from top in northward alignment, avoiding direct solar irradiation. Each trap in a group contained one of three combinations of attractants (Table 7), resulting in four replicates. In one treatment, the commercial lure Galloprotect 2D® (G2D), produced by SEDQ in Spain, was employed solely. It consists of the *Monochamus galloprovincialis* aggregation pheromone compound and two bark beetle pheromone components acting as kairomones. This test established whether the specific pheromone compound – in combination with bark beetle substances – is able to lure other species (*M. sartor, M. sutor)* within the same genus, since the attractancy of 2-undecyloxy-1-ethanol was already proven for *M. alternatus* and three North American sawyers (TEALE et al., 2011). In a second treatment, G2D was combined with the host tree volatile α-pinene (the combination was obtained as Galloprotect Pack®, SEDQ). The third blend additionally applied smoke volatiles, which

Table 7: Natural origin and ingredients of employed attractants

Code	**Origin**	**Ingredients**
G	Sawyer	2-undecyloxy-1-ethanol
	Bark beetle	ipsenol, 2-methyl-3-buten-2-ol
A	Host tree	α-pinene
S	Smoke	2-methoxyphenol, 4-methyl-2-methoxyphenol, 4-vinyl-2-methoxyphenol, Iso-eugenol (4-(1-propenyl)-2-methoxyphenol), Vanillin (4-carboxy-2-methoxyphenol), Eugenol ((4-(2-propenyl)-2-methoxyphenol)

were newly produced by D. R. HALL (University of Greenwich, UK). They have been used in tests with *M. galloprovincialis* and *M. sutor* in Spain and UK within the REPHRAME project (PAJARES and HALL, pers. comm., 2012). These four categories of attractants – intraspecific, bark beetle, host and smoke volatiles – cover all substances that have been shown to positively influence *Monochamus* behaviour (vide 2.2.3).

10-day periods

Trap re-randomisation was achieved by clockwise trap rotation every ten days within each group, avoiding habituation effects of beetles to the site of lures, generating four catching periods (I – IV) (Table 8). In order to prevent contamination of traps and a mix of odours, the whole traps were moved between rods. Due to less favourable weather conditions for *Monochamus* flight in period I, the same lure arrangement was repeated in period IV. Both G2D and α-pinene remained attractive during the entire trapping period according to the manufacturer. Smoke volatiles had to be renewed every 14 days (HALL, pers. comm., 2012).

Table 8: Arrangement of attractants to trap sites in Dürrenstein Wilderness Area at date of installation or change from 10 July to 20 August 2012, G – G2D, A – α-pinene, S – smoke volatiles

	10 July	**19 July**	**30 July**	**09 August**
	I	**II**	**III**	**IV**
A1	G + A	G + A + S	G	G + A
A2	G + A + S	G	G + A	G + A + S
A3	G	G + A	G + A + S	G
B1	G + A + S	G + A	G	G + A + S
B2	G + A	G	G + A + S	G + A
B3	G	G + A + S	G + A	G
C1	G	G + A + S	G + A	G
C2	G + A + S	G + A	G	G + A + S
C3	G + A	G	G + A + S	G + A
D1	G + A	G	G + A + S	G + A
D2	G	G + A + S	G + A	G
D3	G + A + S	G + A	G	G + A + S

3.3.3 Weather influence

Weather influence on flight activity in Hundsau was ascertained from 10 July to 20 August 2012. Air temperature as well as precipitation were measured by a meteorological station in Hundsau in the southeast of the study area and processed by J. PENNERSTORFER (Institute of Forest Entomology, Forest Pathology and Forest Protection, University of Natural Resources and Life Sciences, Vienna). For examination of weather impact, sum of beetles captured in all traps was calculated for 3-4-day periods, consistent with trap control dates. Moreover, arithmetic means of temperature and precipitation were determined for each period. Maximum temperature resulted from the highest value within one period. Kendall's tau correlation analyses of mean and maximum air temperature, precipitation and number of captured *M. sartor* and *M. sutor* beetles were computed with IBM® SPSS® Statistics, version

21. In course of a 5-hour trapping observation on 19 July 2012, hourly and daily mean, maximum and minimum air temperature values were taken.

3.3.4 Statistical analyses

In order to check lure preferences of *Monochamus* spp., statistical analyses were performed using IBM® SPSS® Statistics, version 21. The number of captured beetles was summed up for 10-day trapping periods. Since the dependent variable "number of captured beetles" was not continuous and capturing data were not normally distributed, as proved by Kolmogorov-Smirnov-Test, Kruskal-Wallis-Test (with $\alpha = 0.05$) was applied to examine differences in lure attractancy. To localise the differences between the three groups of attractants, pairwise Mann-Whitney-Tests as follow-up tests were applied (with $\alpha = 0.017$) for each test to control for type I errors.

3.3.5 Mark, release and recapture

At each trap control, caught *Monochamus* beetles were determined to species and sex. After recording in lists, they were individually marked, using water-resistant paint markers (Marabu brilliant painter, Marabu GmbH, Germany) in four colours according to four trap groups. Trap number as well as capturing date were marked on pronotum and elytra, respectively (Figure 7). Negative impacts on the beetles' constitution through marking with touch-up pens were not observed in preceding laboratory tests (HOCH, pers. comm., 2012). Marking beetles with lacquer paint is a usual method, also applied by TOGASHI (1990). Marked sawyers were released again by either placing them on stems of two dead spruce trees or releasing them from hand at a central point of the trapping area (RP: 47° 46'34" N, 15° 02'29" E, 898 m above sea level).

Figure 7: Release of marked *Monochamus* spp. at the central release point in Hundsau, Dürrenstein Wilderness Area. Left: *M. sutor* male, right: two *M. sartor* females

By recapture of marked beetles, flight distances of *Monochamus* spp. between the central release point and the recapturing trap were measured. The influence of wind speed and wind direction was included in these analyses. The measurement station with the rotation anemometer (type 263 VR, Kroneis, Vienna) was situated in the centre of the study site near the release point. Data were kindly provided by E. BLACKWELL and V. WIMMER (Institute of Forest Entomology, Forest Pathology and Forest Protection, University of Natural Resources and Life Sciences, Vienna).

3.3.6 Bycatch analysis

During the trapping period (10 July to 20 August 2012), bycatches were killed immediately after trap emptying through ethyl acetate. They were analysed regarding species spectrum as well as flight activity. Particularly most frequently trapped insects, other cerambycids and potential carriers of PWN were focussed in examinations. However, only those bycatches with a size of at least 3 mm were quantitatively recorded, whereas smaller ones just give an overview of the species complex. Generally, all bycatches were determined to family, whereas, most often caught insects were classified to genus or species. Determination was done with the help of keys by: BENSE (1995), FREUDE, HARDE & LOHSE (1967 – 1979), REITTER (1911) and GRÜNE (1979) as well as online keys by LOMPE (2002 – 2013) and BENISCH (2007 – 2013).

For most recurrent bycatches, Kendall's tau correlation analyses with the weather factors and sawyer catch were computed analogous to *Monochamus*. Furthermore, lure preferences were examined with special focus on bark beetles, their natural enemies and species colonising the same substrate as *Monochamus* spp., due to the application of the unspecific attractants α-pinene and G2D in part. Statistical analysis was carried out in the same procedure as for *Monochamus*.

4 Results

4.1 *Monochamus* spp. emergence from logs

Monochamus sartor emergence period extended from 9 to 18 June 2012 (Figure 8). A total of seven beetles hatched from the logs D1-D7. Beetles found on control days 11 and 18 June possibly eclosed also on 9 or 10 June and 16 or 17 June respectively, since no control of logs occurred on these days. Eclosion began after a period of days with mean air temperatures above 22 °C. Warm temperatures from 7 to 9 June elicited hatching of the first four beetles, notwithstanding subsequent decreasing temperature values down to 15.5 °C at minimum. Moreover, from 16 to 18 June, mean temperatures reached 22.8 to 25.1 °C, ensuing in beetle eclosion. Despite temperatures above 22 °C from 19 to 22 June (22.9 to 26.8 °C) and from 28 to 30 June (22.7 to 27.1 °C), no adult emergence occurred after June.

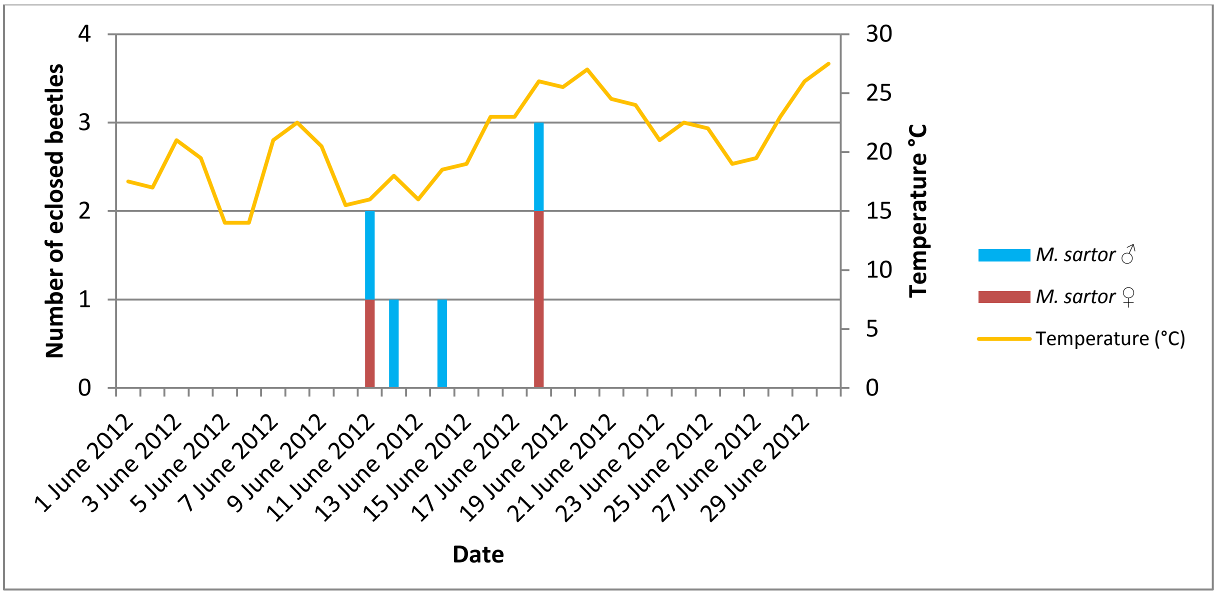

Figure 8: Eclosed *Monochamus sartor* from logs D1-D7, cut in Hundsau, Dürrenstein Wilderness Area and mean air temperature

Stem dissection in April 2013 showed that a total of ten *Monochamus* had remained in the wood (Table 9). Except one living larva in first prepupal stage (Figure 9), all of them died in different developmental stages. The larva was put back into its tunnel; stem parts were screwed together again and placed into a breeding cage in laboratory, whereupon a male *M. sartor* in good condition but with one incomplete antenna eclosed on 8 May 2013. Three of the four dead adult *M. sartor* were completely developed. However, one female lacked both front legs and one antenna. One leg and the antenna were found unsclerotisised in the tunnel. One specimen was first suspected to have escaped through the covering net by biting, since the stem and the net showed a circular borehole. But a fully developed, dead adult *M. sartor* male was found in the end of the tunnel near the borehole, when stem part D1 was dissected (Figure 9). Besides these dead specimens, some larval galleries were found completely empty.

Table 9: Successfully eclosed and dead *Monochamus sartor* adults as well as dead *Monochamus* spp. pupae and larvae remaining in the stems D1-D7, N = 17

	Eclosion 2012	Dead adults	Dead pupae	Dead larvae	Eclosion 2013
Monochamus spp.	0	0	1	4	0
M. sartor female	3	1	0	0	0
M. sartor male	4	3	0	0	1

Figure 9: Left: bitten hole into the covering net by one *Monochamus sartor* male found dead in stem D1, middle: living *Monochamus sartor* larva in first prepual stage from D5, right: dead braconid wasp recovered from D5

No *M. sutor* emerged from the logs or were found as dead adults in stems. The larvae and the pupa could not be determined to species. But most probably they belonged to *M. sartor*, since all other found beetles were determined as *M. sartor* and the tunnels of the remaining were similar in size and shape to those of *M. sartor*. Besides *Monochamus*, two dead, fungal infected larvae of other cerambycids as well as one dead parasitoid wasp (Braconidae, Figure 9) were found when dissecting the stems.

4.2 Trapping *Monochamus* spp.

4.2.1 Weather influence on flight activity

Trap catch in the study area Hundsau from 10 July to 20 August 2012 amounted to a total of 497 *Monochamus* spp. specimens (Figure 10). For both species, captures were female biased. Caught *M. sartor* had a sex ratio of approximately 2.6:1 and *M. sutor* 1.5:1.

Weather conditions influenced beetle flight, as became apparent in the amount of sawyer catches (Figure 11). High mean and maximum air temperature values increased beetle catch, whereas low air temperatures resulted in a low number of captured beetles. Precipitation also negatively affected *Monochamus* spp. trap catches.

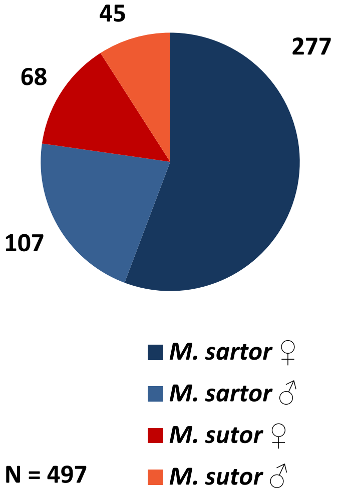

Figure 10: Total trap catch of *Monochamus* spp. in Hundsau, Dürrenstein Wilderness Area during the entire study period

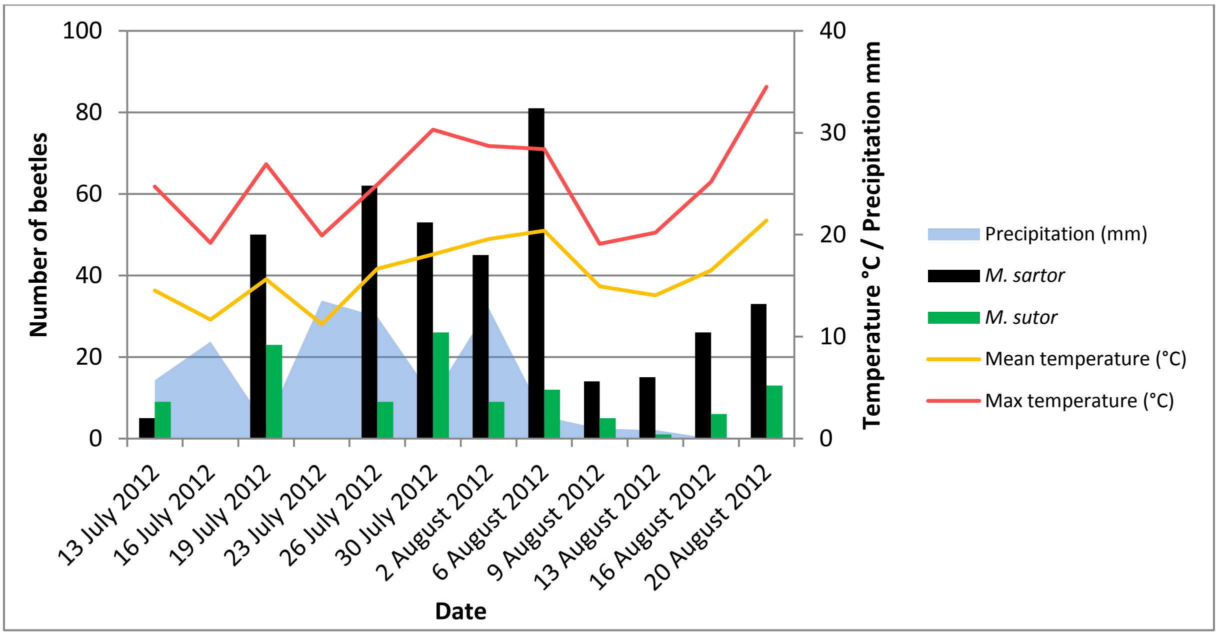

Figure 11: Catch of *Monochamus sartor* and *M. sutor* beetles in 3-4-day trapping periods, mean and maximum air temperature as well as precipitation in Hundsau, Dürrenstein Wilderness Area during the entire investigation period

A decrease of mean and maximum air temperature to 13.1 °C and 19.9 °C, respectively, resulted in no catch on 16 and 23 July; although flight was registered at warmer temperatures before. These temperatures, when no flight occurred, were highest daily values for the respective 3-4-day periods. Lowest temperatures with ascertained *Monochamus* spp. flight amounted to 15.0 °C mean and 19.1 °C maximum, which were also the reached peaks (7 to 9 August).

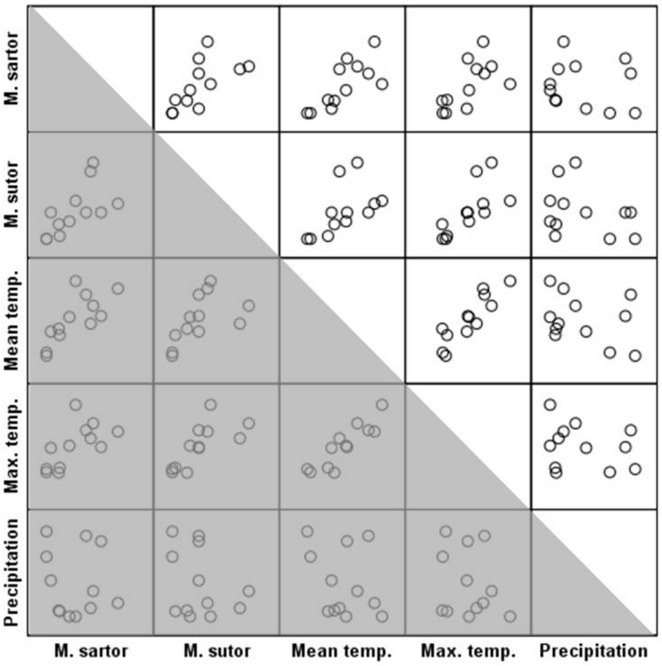

Figure 12: Correlation of total captured *Monochamus sartor* and *M. sutor* per 3-4-day period and the weather factors mean and maximum air temperature as well as precipitation in Hundsau, Dürrenstein Wilderness Area

M. sartor catch was significantly correlated with mean and maximum air temperature (Figure 12, Table 10). However, no significant correlation was computed for *M. sartor* catch and precipitation. For captured *M. sutor*, the correlations with mean and maximum air temperature were likewise significant. As for *M. sartor*, no significant interdependence was calculated for precipitation. Besides sawyer catch and weather factors, capturing numbers of both beetle species themselves were strongly significantly correlated (Table 10).

Table 10: Correlation analysis of *M. sartor* and *M. sutor* catch and weather conditions, n = 12

Correlation variables	**Kendall's tau**	**Significance (2-tailed)**
M. sartor & mean air temperature	0.626	0.005
M. sartor & max. air temperature	0.504	0.023
M. sartor & precipitation	-0.123	0.582
M. sutor & mean air temperature	0.657	0.004
M. sutor & max. air temperature	0.657	0.004
M. sutor & precipitation	-0.173	0.445
M. sartor & *M. sutor*	0.630	0.005

During a 5-hour trapping observation on 19 July, the direct connection between weather conditions and *Monochamus* spp. catch was evidenced (Figure 13). At the first trap control at 10:00 am on 19 July, a total of 36 *Monochamus* spp. specimens was counted. They possibly resulted from the entire trapping period from 16 to 19 July. Five hours later, traps were checked again and further 37 *Monochamus* spp. were found.

Within these five hours, air temperature rose from 20 °C to 25 °C. From 16 July, when no sawyers were found in the traps, temperatures increased to a maximum on 19 July. Decreasing temperatures and increasing precipitation after 19 July lead to no *Monochamus* spp. catch until 23 July.

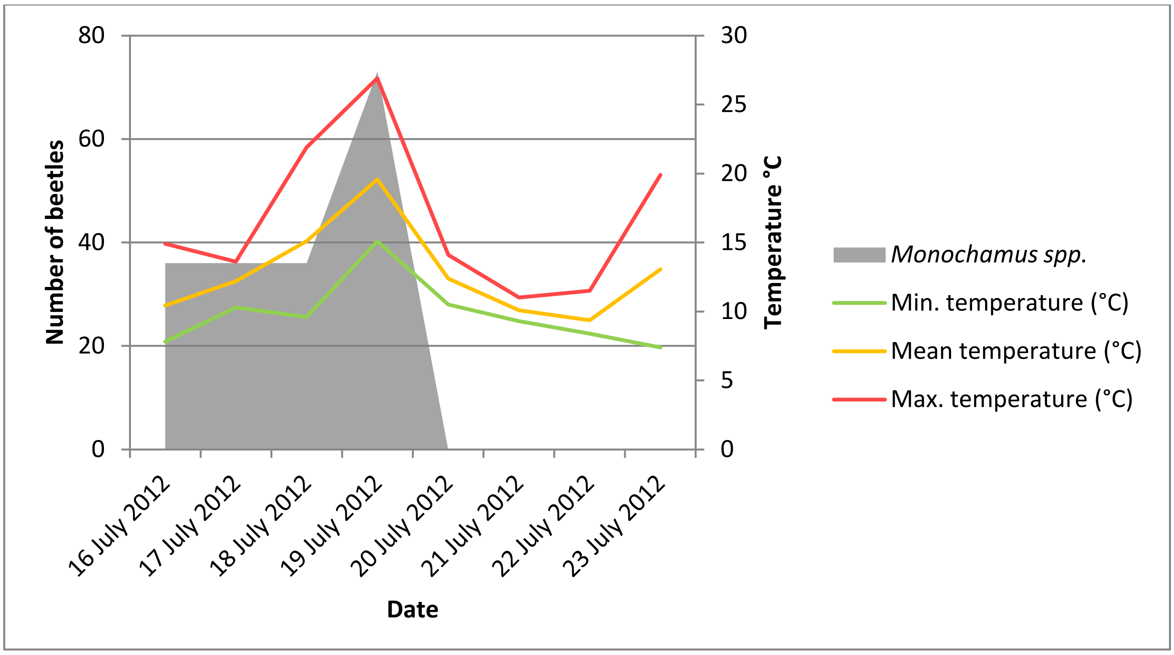

Figure 13: Catch of *Monochamus* spp., minimum, mean and maximum air temperature in Hundsau, Dürrenstein Wilderness Area from 16 to 23 July 2012; capture increase ascertained on 19 July 2012 was measured during a 5-hour trapping observation.

4.4.2 Behaviour of captured and released beetles

Table 11: Number of alive and dead *Monochamus sartor* and *M. sutor* females and males in receptacles during the entire trapping period in Hundsau, Dürrenstein Wilderness Area

Species	Sex	Alive	Dead
M. sartor	Female	222	55
	Male	91	16
M. sutor	Female	52	16
	Male	36	9

Sawyer behaviour was studied during emptying of collection buckets, transport to the release point and marking. High temperatures lead to activity and aggressiveness of the beetles; this was particularly apparent on 19 July before and between heavy thunderstorms. Fierceness was mainly expressed in trap buckets and little transport boxes by big strong *Monochamus* males against the smaller individuals. In worst case, this resulted in dead or injured females lacking extremities. Of all trapped *M. sartor*, a loss of 20 % of females and 15 % of males was noticed. Likewise more *M. sutor* females (24 %) than males (20 %) died; overall loss was proportionally higher compared to *M. sartor* (Table 11). Furthermore, not only beetles of the same genus, but also other Cerambycidae were attacked in buckets. Aggressive or frightened

Monochamus made squeaking, chirping sounds as deterrent measure through rubbing of the pronotum against the dorsal plate described by HELLRIGL (1971). Besides fighting, occasionally copulation in buckets, boxes or at the release point was noticed. This was favoured by suitable weather conditions when sawyers were quiet or moderately agile.

4.2.3 Lure attractiveness for *Monochamus sartor*

Mean amounts of caught female and male *M. sartor* (Figure 14) indicate the same tendency for the respective attractant during several trapping periods. However, male catch was always lower than female catch, throughout all periods (I – IV) for all treatments. For *M. sartor* females, the differences in lure preference were not significant (H-test: p = 0.653). However, for *M. sartor* males significant dissimilarities were established (H-test: p = 0.023) between G2D alone and G2D + α-pinene (U-test: p = 0.012). Catch of *M. sartor* males did not differ significantly between G2D + α-pinene and G2D + α-pinene + smoke (U-test: p = 0.032) as well as between G2D and G2D + α-pinene + smoke (U-test: p = 0.897), respectively (Figure 15).

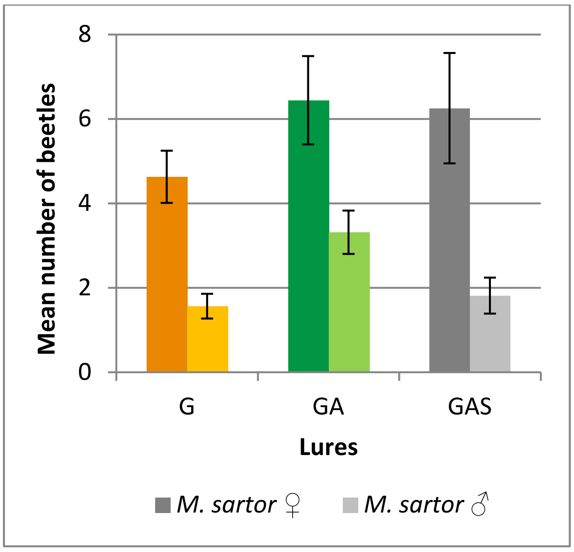

Figure 14: Mean numbers of captured *Monochamus sartor* females / males and standard error with all treatments: G (G2D), GA (G2D + α-pinene) and GAS (G2D + α-pinene + smoke volatiles), during the entire trapping period (40 days)

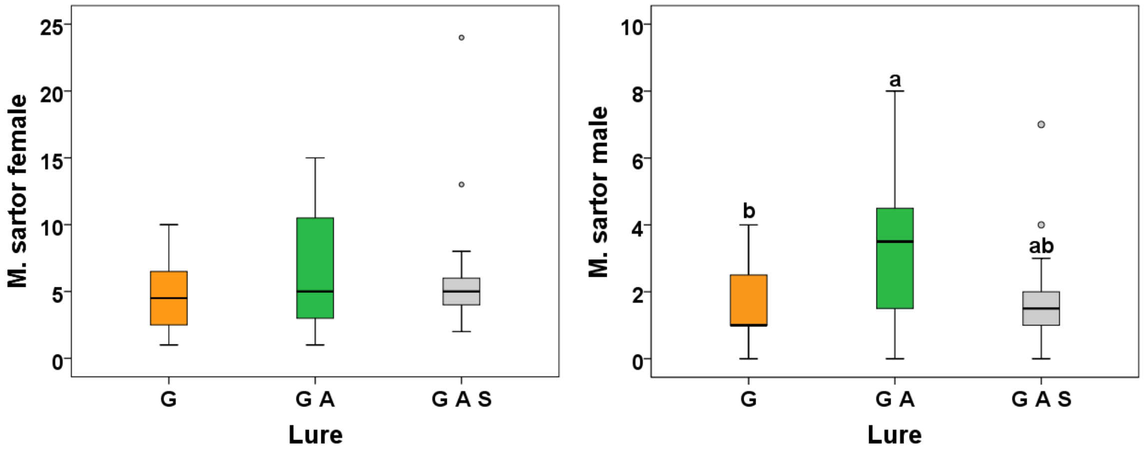

Figure 15: Number of captured *Monochamus sartor* females and males with lures G (G2D), GA (G2D + α-pinene), GAS (G2D + α-pinene + smoke volatiles), median as centre line in each box, 25th as bottom and 75th percentile as top of each box, outliers marked with dots. Significant differences marked with letters were established by Kruskal-Wallis H-test and followed up by pairwise Mann-Whitney U-test (corrected α=0.017).

Trapping periods

Regarding the trapping periods separately for the attractants (Figure 16), lowest catch with G2D + α-pinene and G2D + α-pinene + smoke odours was established in periods I and IV, whereas during the second (19 July to 30 July) and third (30 July to 9 August) period luring effect increased. However, capture with G2D alone was highest in the third and fourth trapping period.

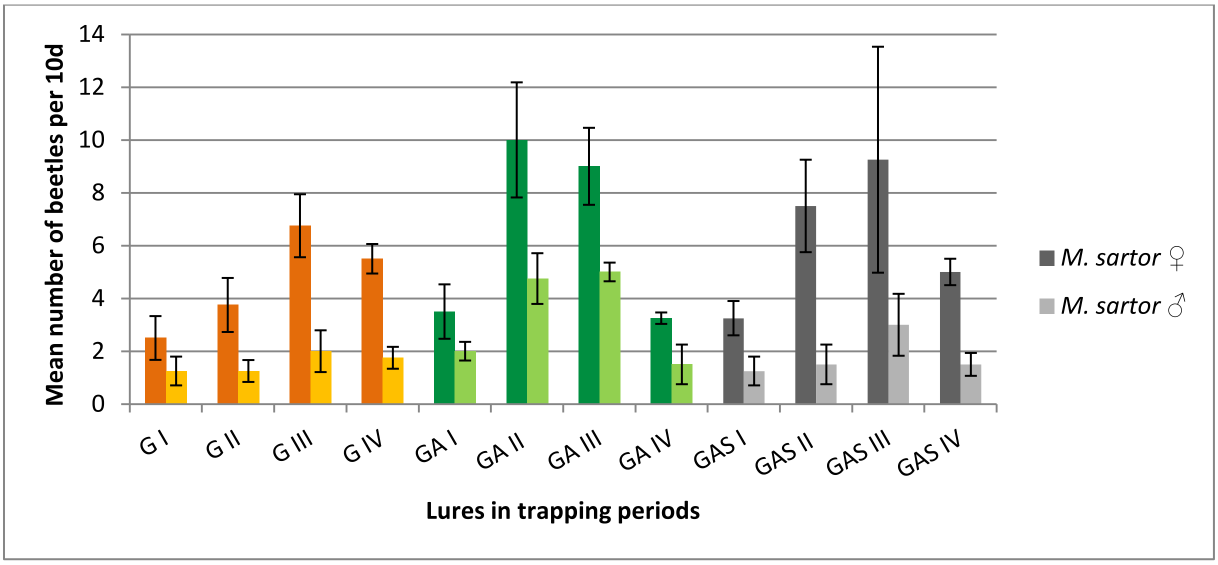

Figure 16: Mean numbers of captured *Monochamus sartor* females / males and standard error with all treatments: G (G2D), GA (G2D + α-pinene) and GAS (G2D + α-pinene + smoke volatiles), during 10-day trapping periods I to IV

Trap groups

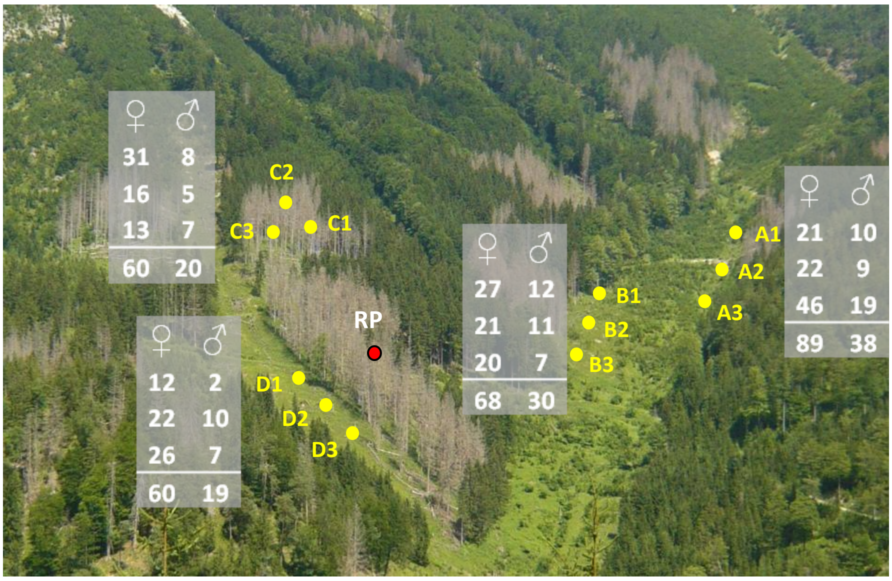

Figure 17: Sum of captured *Monochamus sartor* females and males per trap with all treatments of attractants during the entire trapping period

More *M. sartor* females than males were caught in all trap groups (Figure 17). Regarding absolute numbers of captured beetles, trap group A was preferred by *M. sartor* females and males above all other groups. Trap group B achieved the second-highest capturing result, whereas both C and D caught the lowest sum of females and males.

4.2.4 Lure attractiveness for *Monochamus sutor*

Total *M. sutor* catch was relatively low, compared with *M. sartor*. Only slight dissimilarities were established during the entire trapping period for all attractants (Figures 18, 19). Neither for females (H-test: p = 0.346) nor for males (p = 0.948), these differences in lure preference were significant. Male catch indicated a strong similarity of lure attractancy.

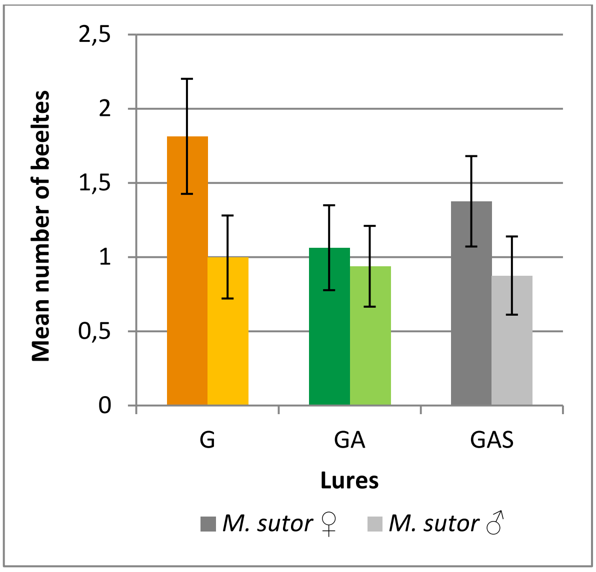

Figure 18: Mean numbers of captured *Monochamus sutor* females / males and standard error with all treatments: G (G2D), GA (G2D + α-pinene) and GAS (G2D + α-pinene + smoke volatiles), during the entire trapping period (40 days)

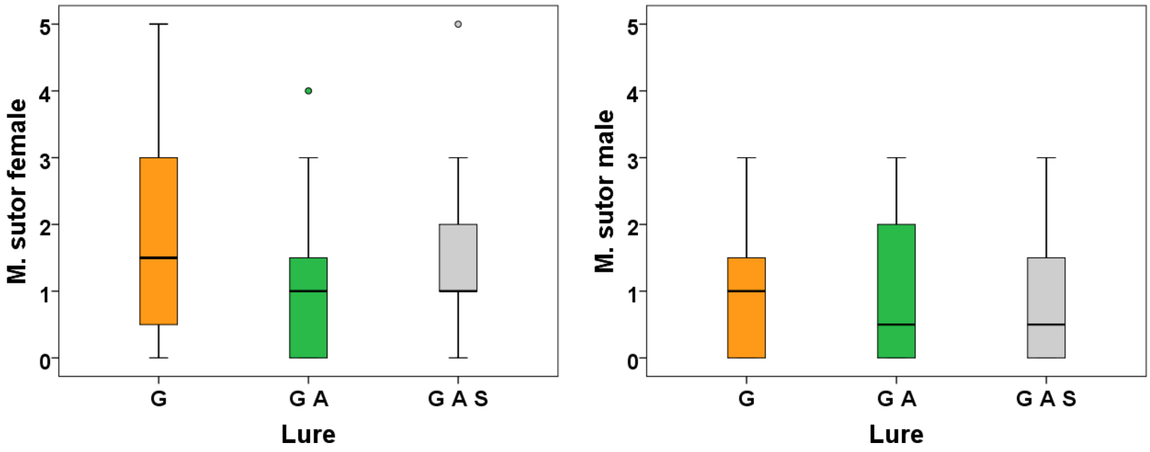

Figure 19: Number of captured *Monochamus sutor* females and males with lures G (G2D), GA (G2D + α-pinene), GAS (G2D + α-pinene + smoke volatiles), median as centre line in each box, 25th as bottom and 75th percentile as top of each box, outliers marked with dots

Trapping periods

Considering the trapping periods separately (Figure 20), females showed comparable temporal trends in preference of G2D and G2D + α-pinene. During period I, most females were caught with these two odour blends. However, when smoke volatiles were added, *M. sutor* female catch concentrated in period II (19 July to 30 July). No trend in lure preference or period was evident in male flight activity; capturing numbers did not strongly vary for the lure combinations throughout all periods.

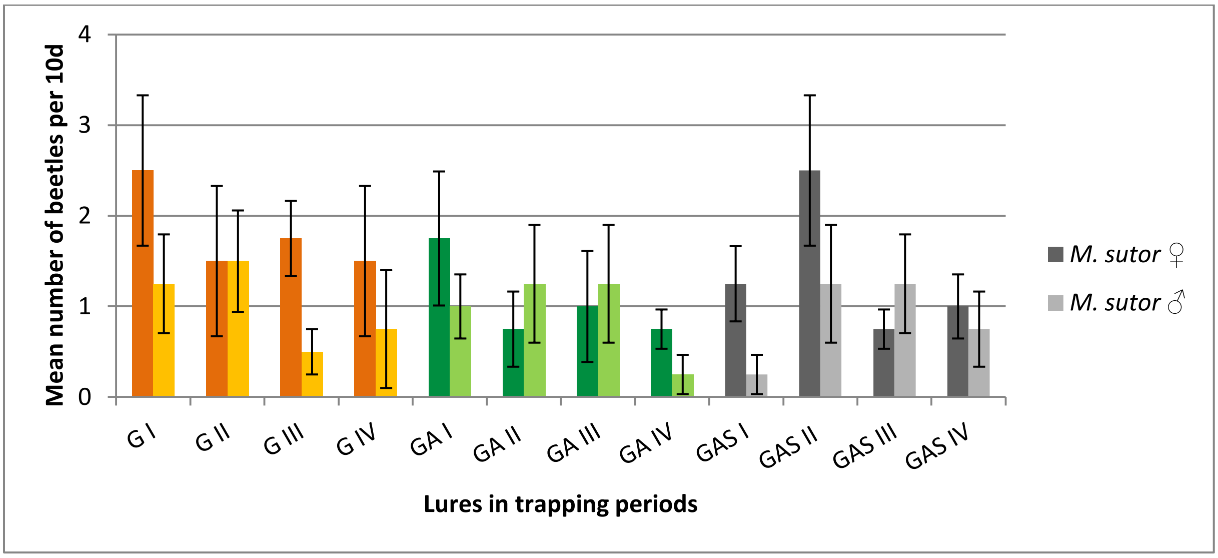

Figure 20: Mean numbers of captured *Monochamus sutor* females / males and standard error with all treatments: G2D, G2D + α-pinene, G2D + α-pinene + smoke volatiles, during 10-day trapping periods I to IV

Trap groups

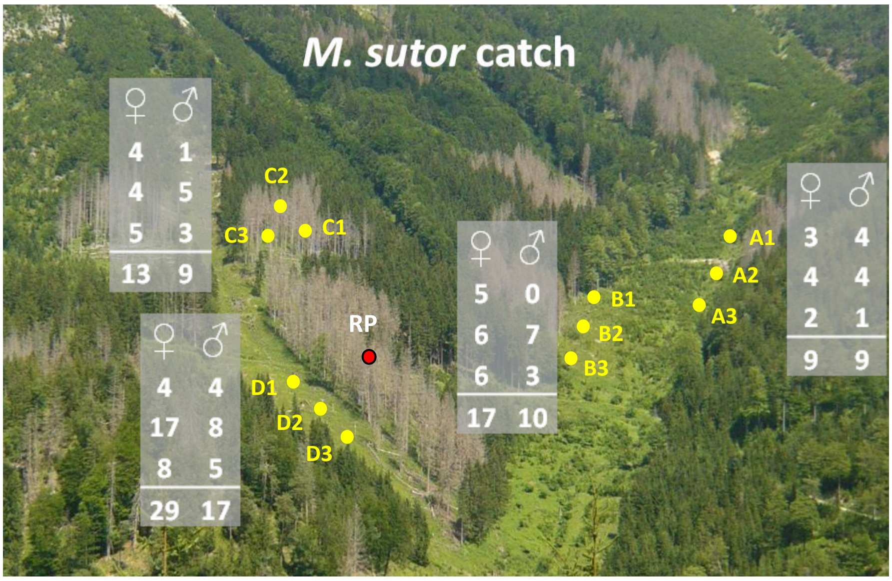

Figure 21: Sum of captured *Monochamus sutor* females and males per trap with all treatments of attractants during the entire trapping period

In contrast to the trapping results of *M. sartor*, trap group A was least preferred by *M. sutor* females (Figure 21). Few more female beetles were caught in group C and more in B. Considering males; an almost equal low amount was counted throughout the first three trap groups. However, group D captured most females and males. Especially trap D2 attained 17 females in sum, more than twice as many as any other group.

4.2.5 *Monochamus* spp. recapture

Table 12: Distances between traps and release point, recaptured *Monochamus*, time interval between release and recapture

Trap	Distance (m)	Recapture	Time (d)
A1	387	*M. sutor* ♂	4
A2	337	-	
A3	314	-	
B1	222	*M. sartor* ♂	7
B2	195	*M. sartor* ♂	7
B3	161	*M. sutor* ♀	14
C1	157	-	
C2	170	*M. sutor* ♂	4
C3	120	*M. sutor* ♀	4
D1	44	-	
D2	73	*M. sutor* ♂ *M. sartor* ♀	7 7
D3	131	*M. sartor* ♀	11

Four *Monochamus sartor* and five *M. sutor* were recaptured from 19 July to 20 August 2012 (Table 12). This resulted in 1.3 % and 5.8 %, respectively, of the total of 308 released *M. sartor* and 85 *M. sutor*. *Monochamus* spp. were recaptured on average seven days (maximum 14 days) after release. The time span did not differ between species.

Mean measured flight distances amounted to 155 m for *M. sartor* and 182 m for *M. sutor*. The maximum of 387 m to the most distant trap A1 was reached by one *M. sutor* male (Figures 22, 23). After release in the centre of the study site, the sawyers dispersed in all directions traversing vigorous and dead spruce stands as well as open areas, since all four trap groups were represented. The dominant wind direction during the recapturing period was east-northeast with a low velocity of 9 km/h on average, followed by north-northeast, north-northwest and west-northwest. However, no link between recapture and wind direction was established. No trap or trap group was particularly favoured. Amounts of recapture were too small for statistical analysis. Furthermore, no preferences of certain attractants were observed during the recapture experiment.

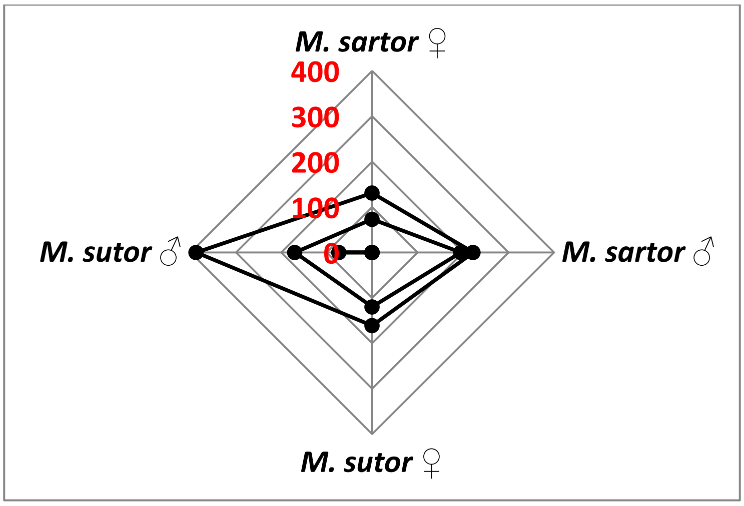

Figure 22: Minimum (centre) and maximum flight distances of released and recaptured *Monochamus sartor* and *M. sutor* females and males

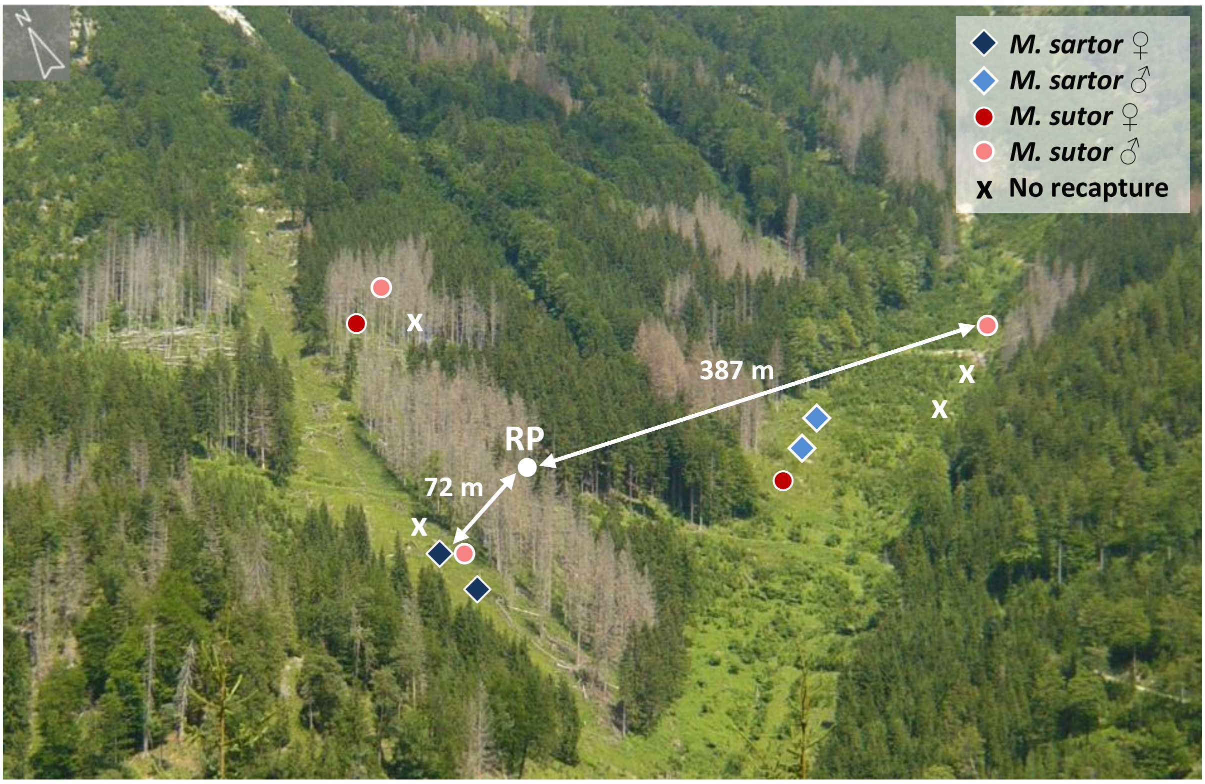

Figure 23: Recaptured *Monochamus sartor* and *M. sutor*, central release point (RP) and flight distance, symbols represent individual beetles.

4.3 Bycatches

4.3.1 Species spectrum

During the entire trapping period from 10 July to 20 August 2012, at least representatives of 25 coleopteran families were caught (Table 13). The captured Cerambycidae (N = 95) and Buprestidae (N = 24) were classified to species level (Figures 25, 26). Other Coleoptera that were quantified and determined to genus or species are listed in Table 14.

Table 13: Spectrum of captured beetle families

Family, subfamily
Cerambycidae
Curculionidae
Curculionidae, Scolytinae
Anthribidae
Rhizophagidae
Cleridae
Silphidae
Cisidae
Erotylidae
Mycetophagidae
Nitidulidae
Buprestidae
Coccinellidae
Endomychidae
Geotrupidae
Histeridae
Scarabaeidae
Scarabaeidae, Cetoniinae
Mordellidae
Elateridae
Staphylinidae
Cantharidae
Oedemeridae
Lycidae
Byturidae
Dascillidae
Lampyridae
other Coleoptera

Furthermore, bycatch consisted of the coleopteran families Staphylinidae, Rhizophagidae, Cleridae (particularly *Thanasimus formicarius*), Curculionidae (esp. Scolytinae), Mordellidae, Cisidae, Histeridae (esp. *Plegaderus vulneratus*) and Endomychidae (*Endomychus coccineus*). However, except Cleridae, Mordellidae (48) and Endomychidae, they were not quantified due to their size smaller than 3 mm.

Regarding Scolytinae, all three groups were represented: *Crypturgus* sp. (Scolytitae), *Ips typographus*, *Pityogenes chalcographus*, *Pityokteines vorontsovi* (Ipitae), *Polygraphus poligraphus* (Hylesinitae). Some bark beetles were damaged by predators, so that classification was impossible.

A total of 41 Coccinellidae was captured, among them two specimens of the invasive *Harmonia axyridis* in traps D1 and D2 on 30 July (Figure 24). These traps were situated in a distance of approximately 3.5 km from the next human settlement.

Figure 24: Captured *Harmonia axyridis*

Besides Coleoptera, representatives of ten other insect orders were caught (Figures 27, 28). The only xylobiont non-coleopteran bycatch were the siricids *Urocerus gigas* and *Sirex juvencus* (24 and four specimens).

Furthermore, representatives of three arthropod orders plus one gastropod were trapped. Pseudoscorpionida (17 specimens) constituted the most frequent other arthropod bycatch, besides Araneida (two) and Isopoda (one).

Table 14: Quantified and classified catch of Coleoptera

Family	**Genus / Species**	**Number**
Curculionidae	*Phyllobius argentatus*	1
Antrhribidae	*Anthribus albinus*	2
Cleridae	*Thanasimus formicarius* *Trichodes apiarius*	136 19
Silphidae	*Necrophorus vespilloides* *Oiceoptoma thoracicum*	4 1
Erotylidae	*Tritoma bipustulata*	1
Coccinellidae	*Coccinella septempunctata* *Adalia bipunctata* *Harmonia axyridis*	38 1 2
Endomychidae	*Endomychus coccineus*	20
Geotrupidae	*Geotrupes* sp.	7
Histeridae	*Platysoma* sp.	1
Scarabaeidae	*Trichius fasciatus*	6
Cantharidae	*Rhagonycha fulva* *Cantharis livida*	7 1
Oedemeridae	*Anogcodes fulvicollis* *Sparedrus testaceus*	3 1
Lycidae	*Dictyoptera aurora*	1
Byturidae	*Byturus* sp.	1
Dascillidae	*Dascillus cervinus*	1
Lampyridae	*Lamprohiza splendidula*	2

Coleoptera

Cerambycidae

Acanthocinus griseus
Acanthoderes clavipes
Arhopalus rusticus
Cyrtoclytus capra
Judolia cerambyciformis
Leptura rubra
Spondylis buprestoides
Rosalia alpina

Buprestidae

Buprestis haemorrhoidalis
Chrysobothris chrysostigma
Chrysobothris igniventris

Cleridae

Thanasimus formicarius
Trichodes apiarius

Scarabaeidae

Cetoniinae
Trichius fasciatus

Silphidae

Necrophorus vespilloides
Oiceoptoma thoracicum

Figure 25: Plate of selected non-target Coleoptera, captured in Dürrenstein Wilderness Area from 10 July to 20 August 2012

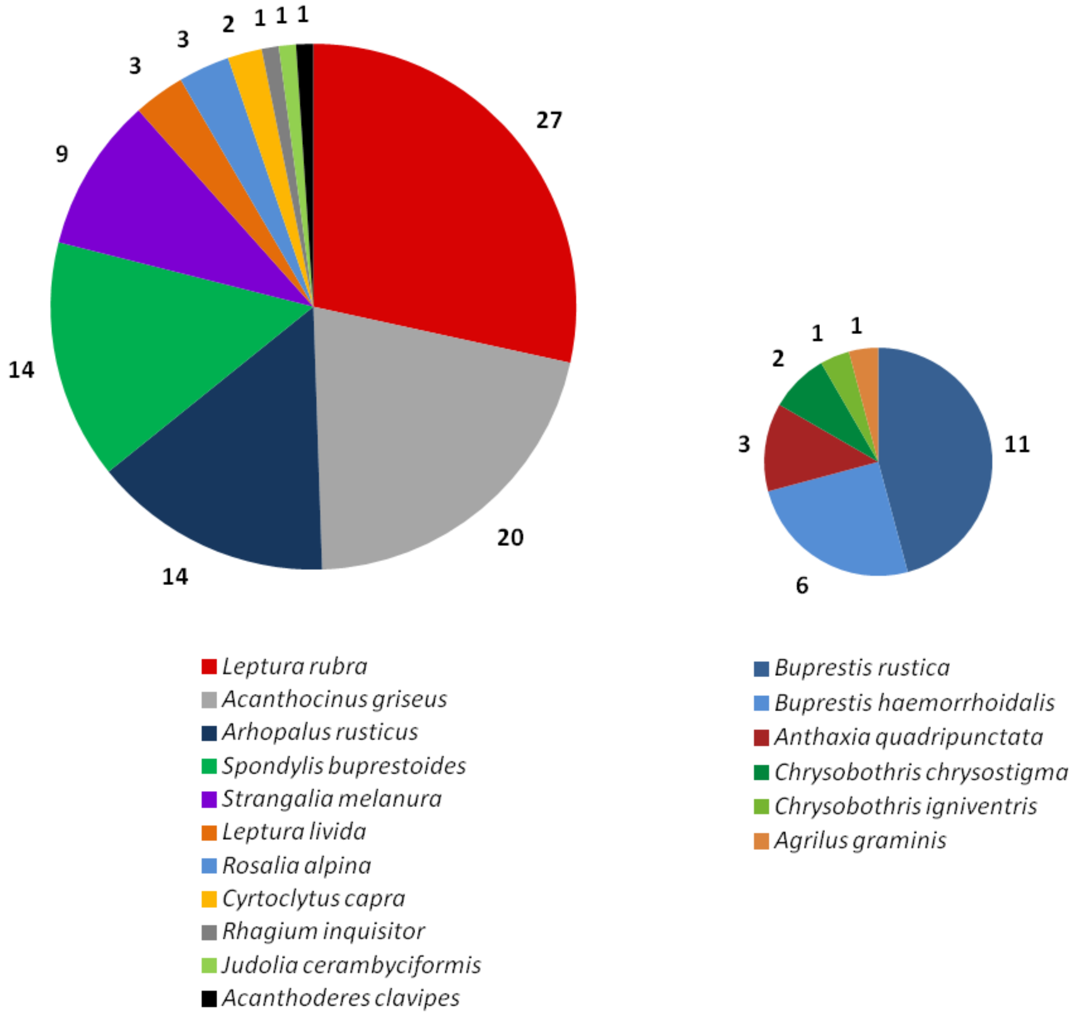

Figure 26: Bycatch of Cerambycidae (N = 95) and Buprestidae (N = 24) in Hundsau, Dürrenstein Wilderness Area from 10 July to 20 August 2012

Figure 27: Plate of selected non-target Hymenoptera and Lepidoptera, captured in Dürrenstein Wilderness Area from 10 July to 20 August 2012

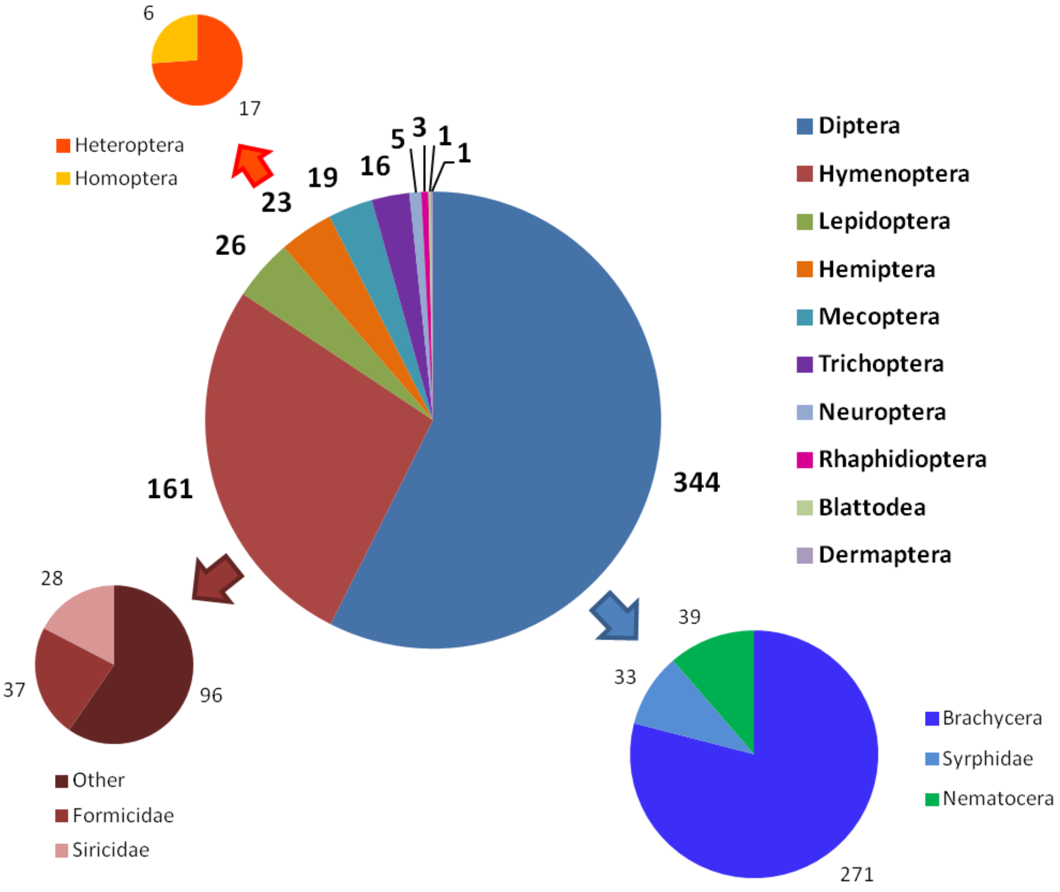

Figure 28: Bycatches of insect orders (central pie chart), suborders and families, except Coleoptera in Hundsau, Dürrenstein Wilderness Area from 10 July to 20 August 2012

4.3.2 Weather influence on flight activity of bycatches

Correlation analysis of the most frequently caught cerambycids *Acanthocinus griseus, Leptura rubra, Spondylis buprestoides* as well as the siricid *Urocerus gigas* and weather conditions resulted in no significant interdependence (Table 15). However, the capturing numbers of *Arhopalus rusticus* and the bark beetle predators *Thanasimus formicarius* and *Trichodes apiarius* were significantly correlated with mean and maximum air temperature (Table 15, Figure 30).

Table 15: Correlation analysis of bycatches and weather factors, n = 12

Correlation variables	**Kendall's tau**	**Significance (2-tailed)**
A. griseus & mean air temp.	0.268	0.251
A. griseus & max. air temp.	0.302	0.196
A. griseus & precipitation	0.152	0.517
A. rusticus & mean air temp.	0.520	0.034
A. rusticus & max. air temp.	0.520	0.034
A. rusticus & precipitation	0.056	0.820
L. rubra & mean air temp.	0.291	0.207
L. rubra & max. air temp.	0.226	0.326
L. rubra & precipitation	-0.244	0.292
S. buprestoides & mean air temp.	0.341	0.150
S. buprestoides & max. air temp.	0.376	0.114
S. buprestoides & precipitation	0.103	0.665
T. formicarius & mean air temp.	0.657	0.003
T. formicarius & max. air temp.	0.504	0.023
T. formicarius & precipitation	0.154	0.491
T. apiarius & mean air temp.	0.626	0.008
T. apiarius & max. air temp.	0.558	0.018
T. apiarius & precipitation	-0.085	0.719
U. gigas & mean air temp.	0.382	0.102
U. gigas & max. air temp.	0.448	0.055
U. gigas & precipitation	0.268	0.255

But graphs might indicate a trend towards weather influenced flight behaviour of all cerambycid bycatches and the siricid (Figure 29). Capturing periods from 14 to 16 July and from 20 to 23 July 2012 with low temperatures and high precipitation attained no or lowest catch throughout all considered non-target species. However, rising temperatures and almost no precipitation in the end of the study period were not linked to higher catch except for *T. formicarius*.

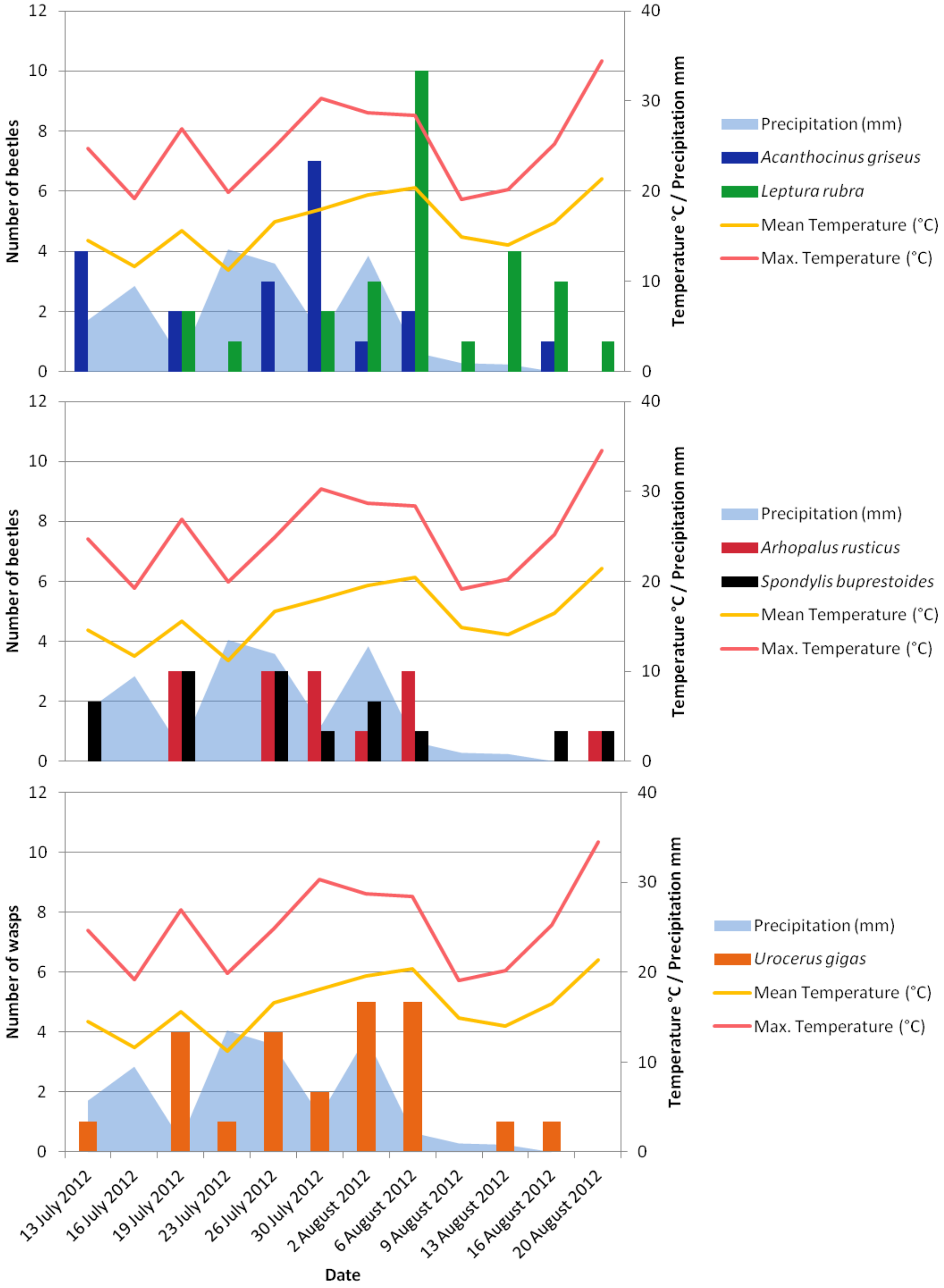

Figure 29: Catch of *Acanthocinus griseus*, *Leptura rubra*, *Arhopalus rusticus*, *Spondylis buprestoides* and *Urocerus gigas*, mean and maximum air temperature as well as precipitation in Hundsau, Dürrenstein Wilderness Area during the entire investigation period

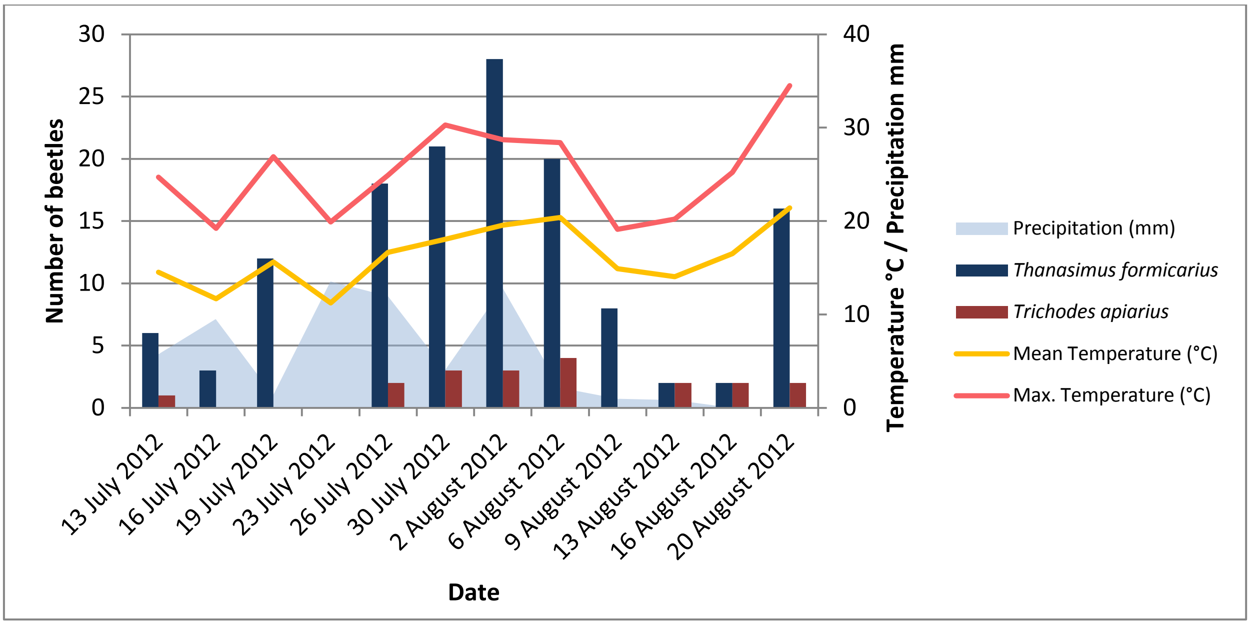

Figure 30: Catch of *Thanasiums formicarius* and *Trichodes apiarius*, mean and maximum air temperature as well as precipitation in Hundsau, Dürrenstein Wilderness Area during the entire investigation period

Furthermore, significant correlations were established between all cerambycid bycatches as well as *Thanasimus formicarius* and *Monochamus* spp. catch, whereas, *Leptura rubra* did not show any interdependence (Table 16). For capturing numbers of *Trichodes apiarius* and *Urocerus gigas*, only the correlations with *M. sartor* catch were significant.

Table 16: Correlation analysis of bycatches and *M. sartor*, *M. sutor* catch, n = 12

Correlation variables	**Kendall's tau**	**Significance (2-tailed)**
A. griseus & *M. sartor*	0.506	0.031
A. griseus & *M. sutor*	0.570	0.017
A. rusticus & *M. sartor*	0.823	0.001
A. rusticus & *M. sutor*	0.708	0.005
L. rubra & *M. sartor*	0.309	0.182
L. rubra & *M. sutor*	0.150	0.524
S. buprestoides & *M. sartor*	0.516	0.031
S. buprestoides & *M. sutor*	0.528	0.029

Correlation variables	**Kendall's tau**	**Significance (2-tailed)**
T. formicarius & M. sartor	0.585	0.009
T. formicarius & M. sutor	0.551	0.015
T. apiarius & M. sartor	0.630	0.008
T. apiarius & M. sutor	0.419	0.082
U. gigas & M. sartor	0.569	0.016
U. gigas & M. sutor	0.360	0.132

4.3.3 Lure attractiveness for bycatches

Most recurrent bycatches were analysed in regard to odour preference (Figures 31 – 33). Besides the xylobiont insects (Cerambycidae, Siricidae) and bark beetle predators (Cleridae), the seven-spot ladybird (*Coccinella septempunctata*) and *Endomychus coccineus* were investigated.

A similar trend towards highest attractancy of G2D + α-pinene was apparent for *Acanthocinus griseus* (H-test: p = 0.299) and *Arhopalus rusticus* (H-test: p = 0.049), but without significant differences in pairwise lure blend comparisons. *Leptura rubra* and *Spondylis buprestoides* were each lured in equal intensity by G2D + α-pinene and G2D + α-pinene + smoke volatiles and lowest by G2D. No *S. buprestoides* was caught with G2D alone, resulting in significant dissimilarities (H-test: p = 0.008). But pairwise attractant comparisons did not confirm this significance. G2D alone was more attractive for *L. rubra* compared to the other Cerambycidae. Attractancy did not differ significantly between the lure blends for *L. rubra* (H-test: p = 0.640).

Considering *Thanasimus formicarius* and *Trichodes apiarius,* they showed an inverse trend of lure preference. While *T. formicarius* was highest attracted by G2D + α-pinene, this lure was less preferred by *T. apiarius*. However, the differences were not significant for both (H-tests: p = 0.068, p = 0.452). Likewise, for *C. septempunctata* (H-test: p = 0.857) and *E. coccineus* (p = 0.525), no significant dissimilarities were established. The ladybird slightly preferred G2D + α-pinene. *E. coccineus* was equally lured by G2D and G2D + α-pinene; with addition of smoke volatiles attractancy marginally decreased.

The wood wasp *Urocerus gigas* showed a tendency, which was not comparable to the beetles, except *S. buprestoides*. No response to G2D alone, moderate reactions to G2D + α-pinene and highest orientation towards the full mixture G2D + α-pinene + smoke volatiles were established (H-test: $p < 0.001$). The difference was significant for comparison of G2D and G2D + α-pinene + smoke volatiles (U-test: $p = 0.001$).

Bycatch of Buprestidae was too low to analyse statistically; Table 17 gives an overview of capturing numbers with the different treatments.

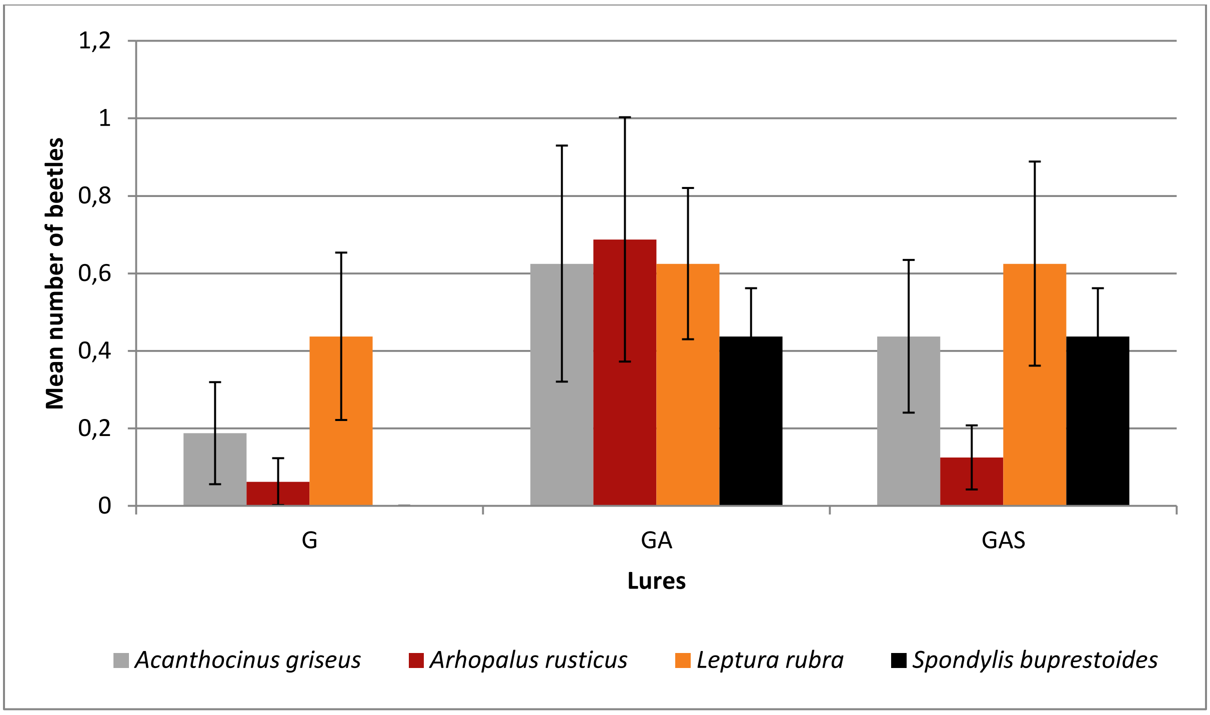

Figure 31: Mean numbers and standard error of captured *Acanthocinus griseus*, *Arhopalus rusticus*, *Leptura rubra* and *Spondylis buprestoides* for all treatments: G2D, G2D + α-pinene, G2D + α-pinene + smoke volatiles, during the entire trapping period (40 days)

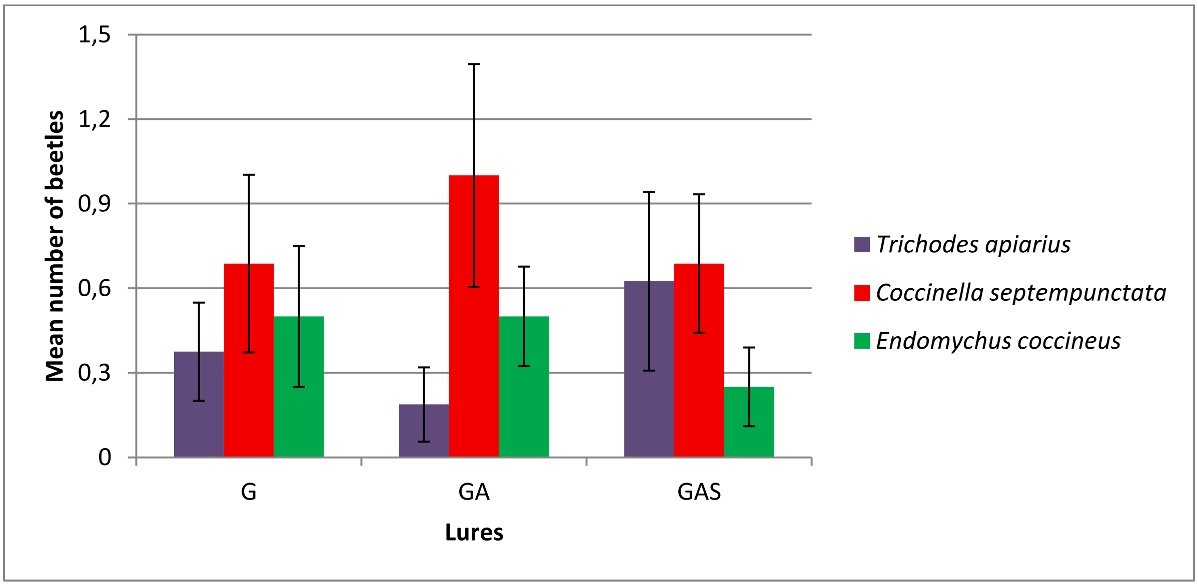

Figure 32: Mean numbers and standard error of captured *Trichodes apiarius*, *Coccinella septempunctata* and *Endomychus coccineus* for all treatments: G2D, G2D + α-pinene, G2D + α-pinene + smoke volatiles, during the entire trapping period (40 days)

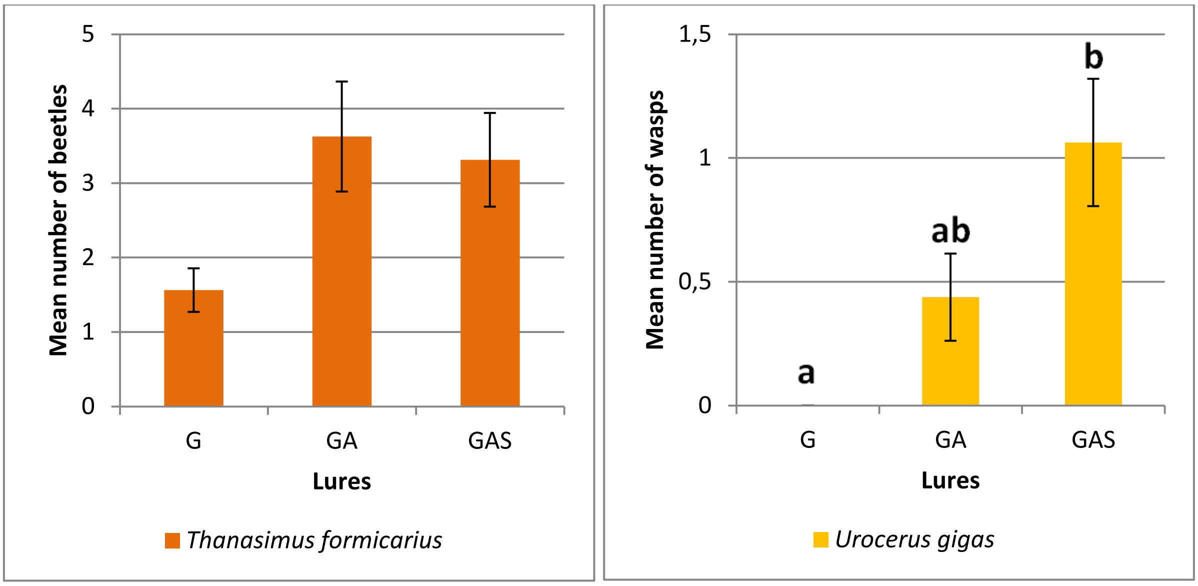

Figure 33: Mean numbers and standard error of captured *Thanasimus formicarius* and *Urocerus gigas* for all treatments: G2D, G2D + α-pinene, G2D + α-pinene + smoke volatiles, during the entire trapping period (40 days). Significant differences marked with letters were established by Kruskal-Wallis H-test and followed up by pairwise Mann-Whitney U-test (corrected α = 0.017).

Table 17: Number of captured Buprestidae with applied attractants G (G2D), GA (G2D + α-pinene), GAS (G2D + α-pinene + smoke volatiles) during the entire trapping period (40 days)

Species	**Lure**		
	G	GA	GAS
Buprestis rustica	3	3	5
Buprestis haemorrhoidalis	3	2	1
Chrysobothris chrysostigma	0	1	1
Chrysobothris igniventris	0	0	1
Anthaxia quadripunctata	0	3	0
Agrilus graminis	0	1	0

5 Discussion

Since previous research in Europe focussed on PWN main vector *M. galloprovincialis*, this study provides first insights into phenology and flight behaviour of the potential vectors *Monochamus sartor* and *M. sutor*.

Monochamus spp. emergence from logs

Direct weather influence on *Monochamus* spp. phenology was apparent during the beetle emergence experiment. *M. sartor* hatching followed mean temperatures above 22 °C. This connection was also observed by TANIWAKI et al. (2004), where peaks of *M. alternatus* eclosion were caused by high temperatures. Under homeothermic conditions, no peaks of eclosion were established. This revealed the stronger effect of weather conditions compared to the sawyer's innate rhythm. Likewise for the European *M. galloprovincialis*, a temperature related emergence was discerned in Portugal. Peaks of hatching occurred earlier in warmer years; the whole eclosion period was brought forward in course of the year (NAVES et al., 2008). Laboratory experiments showed a linear correlation of temperature from 15 °C to 30 °C and development rate during the last larval stage until emergence. Considering the number of hatched *M. galloprovincialis* adults, 23 °C were optimum. In principle, a thermal sum of 822 degree-days was calculated for 50 % adult eclosion (NAVES and DE SOUSA, 2009).

In our experiment, no further *M. sartor* emerged from the logs after 18 June 2012, in spite of mean air temperatures exceeding 22 °C in the end of June as well as in July and August. However, dead adults, larvae and one pupa were found remaining in the stem parts. In two of four cases, the pupal chamber was apparently created too deep in the wood, so that beetles were unable to exit the stem; distances of 4 cm or more were too far to reach the stem surface. Thus, beetles starved to death, as it was established for *M. galloprovincialis* as well (PHRAME, 2007). Naturally, the distances extend from 1.5 to 3 cm with 2.0 cm on average and beetles spend up to four days with boring out (HELLRIGL, 1971). One of the dead sawyers lacked extremities, indicating disruption of the pupal stage. One further adult *M. sartor* could not get through the harder wood of a branch while attempting to exit the stem. Cutting the tree in the natural habitat at the beginning of May 2012 during the critical time of final tunnel building as well as transport and placement of stem parts under changed environmental conditions were presumably linked to loss of orientation and interruption. Marking the upper and bottom side of stem parts and the directions in the natural habitat for correct placement during surveillance as well as more careful transport may help to limit disturbance of the sensitive larvae.

Cause of death of one *M. sartor* male, which successfully bored the exit hole but withdrew into the stem again, is unclear. Such an untypical behaviour suggests disturbance while leaving the stem. Also irritation by gnawning synthetic material is possible, since one hole bitten into the covering gauze net originated from this log. Starving in the stem, owing to unsuitable weather conditions for flight while finishing the exit hole, can be the cause of death. Dying of exhaustion by boring out from a pupal chamber, which was constructed too deep in the wood might be a possible other reason. Besides adults, one pupa and larvae died within the stems. Reasons might be various, such as natural death due to pathogens and parasitoids (HELLRIGL, 1971, NAVES et al., 2008) or development disruption by tree cutting and transport.

The living larva found in the log gives evidence of the possibility of a two-year life cycle of *Monochamus* spp., as it is described for *M. sartor* and *M. sutor* under unfavourable climatic and feeding conditions or late oviposition. In these cases, larval development cannot be completed within one year (HELLRIGL, 1971, BAKKE and KVAMME, 1992). It is important to note that the Dürrenstein logs had no bark when stored at BFW; bark feeding of the larva could therefore be excluded after May 2012. Apparently, it remained dormant as mature larva for one year. For both *M. sartor* and *M. sutor*, a larval diapause without feeding or activity during the entire vegetation period is known to occur; even when larval development was finished and larvae already entered the first prepupal stage in previous autumn (HELLRIGL, 1971).

Throughout the beetle emergence experiment, only *M. sartor* eclosed or remained within the logs. Therefore, it is likely that the larvae and the pupa also belong to this species. No *M. sutor* was found, although in the natural habitat where the tree was cut, *M. sutor* was also present. A reason might be the choice of stem parts with diameters above 20 cm for the study, known to be the breeding habitat of *M. sartor*, whereas *M. sutor* mainly colonises thinner tree parts (8 to 14 cm). This partition of the host tree is caused by interspecific competition, since in the Alps, *M. sartor* is strongly competitive in spruce (*Picea abies*), according to HELLRIGL (1971).

Trapping *Monochamus* spp.

The bigger amount of captured *M. sartor* compared to *M. sutor* in the trapping experiment in Hundsau, Dürrenstein Wilderness Area might indicate higher population densities of *M. sartor*. Throughout the study, no *M. saltuarius* was caught, which is a complementary species to *M. sartor* and *M. sutor* in higher elevations, likewise populating *Picea abies*. It is considered as most frequent *Monochamus* species in the South Tyrolean Alps (HELLRIGL, 2010). Surveys of ZÁBRANSKÝ (2001) proved the occurrence of *M. saltuarius* in the Wilderness Area. Due to morphological similarity to *M. sutor* (HELLRIGL, 1971, BENSE, 1995), it cannot be entirely excluded that individual *M. saltuarius* were captured but not recognised prior to release. However, all specimens that were brought to the laboratory were identified as *M. sutor* or *M. sartor*.

In our studies, black multiple funnel traps were applied. These have been successfully used for *M. galloprovincialis* and *M. sutor* in Spain (REPHRAME Periodic Report, 2012). Generally, traps with a black silhouette are appropriate for catch of wood-colonising Cerambycidae and Buprestidae, according to DE GROOT and NOTT (2001, 2003). Multi-funnel traps achieved lowest capturing results of *M. scutellatus*, *M. obtusus* and *M. clamator*. This was attributed to the funnel construction, which complicated landing of sawyers. Beetles rather fell down outside the traps (MCINTOSH et al., 2001). However, experiments by REPHRAME collaborators demonstrated that cross traps and the multi-funnel trap type used in this experiment are equally suitable. Teflon coating (as used in our experiment) was shown to be the more important factor (REPHRAME Periodic Report, 2012).

The employment of multiple funnel traps in Wilderness Area Dürrenstein led to a total of almost 500 captured *Monochamus* specimens. However, no comparative studies were conducted with other trap types there. To prevent insect escape from the dry collection cup (MOREWOOD et al., 2002), we used special deep buckets with a Teflon coating (ECONEX®, Spain), which were recommended by PAJARES et al. (2004).

In order to enhance their visual attractiveness, traps were installed on open space. Trap installation was arranged in randomised blocks in all REPHRAME studies. In Dürrenstein Wilderness Area, traps were hung on frames approximately 2 m above ground, as it was also done by MOREWOOD et al. (2002) and JURC et al. (2012). However, capturing results of *M. galloprovincialis* in Portugal from May to September were highest when traps were placed in the canopy. At ground level and on the trunk, only 24 % of the total amount were caught

(PHRAME, 2007). Height of trap installation should be species dependent. *Monochamus* spp. have different requirements on their breeding locations. *M. galloprovincialis* needs small wood diameters – the trunks of small or the canopies of large pine trees (HELLRIGL, 1971), which explains more frequent capturing there in Portugal. Likewise, *M. sutor* breeds in tree parts of same diameter classes in spruce. This might be another reason for low trapping amounts in our experiments, besides a possible supplanting by *M. sartor*. *M. saltuarius* prefers even smaller diameters as branches and tree tops. In contrast, *M. sartor* prefers large diameters (HELLRIGL, 1971), which should favour high catches with traps near the ground in Dürrenstein Wilderness Area in 2012. Moreover, time after adult emergence influences host seeking of *Monochamus* spp. Since *M. scutellatus* was stronger attracted to traps with a large, prominent silhouette during the last weeks of its flight period. This could be explained by aimed search of trunks as mating and oviposition sites, instead of green canopies for maturation feeding immediately after eclosion (DE GROOT and NOTT, 2003).

Weather influence

The 3-4-day control intervals in the trapping experiment in Dürrenstein Wilderness Area allowed studying weather influences on *Monochamus* spp. flight behaviour. Significant positive correlations of *M. sartor* and *M. sutor* catches and mean and maximum air temperature were established. High temperatures were directly linked to enhanced capture of both species, whereas cooler weather conditions reduced the sawyers' flight activity. Significant interdependence between *M. sartor* and *M. sutor* catch further showed the similarity in temperature dependent reaction of both species. In the study area, lowest daily mean temperature for registered flight was 15.0 °C, highest mean temperature without flight activity was 13.1 °C. This suggests a threshold temperature, sufficient for *M. sartor* and *M. sutor* flight, between these values. Since flight was observed at a maximum of 19.1 °C in one period and not observed at 19.9 °C in another period, maximum air temperature seemed not to be such a decisive factor for flight initiation of these sawyer species. However, additional influence of precipitation or global radiation on respective days or several days before may be assumed. Due to the short time span of observation, no definitive temperature threshold for flight activity can be derived. Nonetheless, our experiment gave first indications.

Similar weather dependent flight behaviour was established for the Asian *M. alternatus*, which requires at least 18 °C for activity (KOBAYASHI et al., 1984) and whose dispersion is favoured by rising maximum air temperature (TOGASHI, 1990). The European scolytine *Ips typographus* also starts flight activity and first tree infestation at certain air temperature thresholds and thermal sums (BAIER et al., 2007). Likewise, flight initiation of bark beetles *Ips pini* and *I. lecontei* in North America is strongly weather dependent. These Ipitae also stopped flight at cool temperatures, despite preceding flight beginning at warmer weather conditions. With increasing temperatures, the bark beetles showed enhanced flight activity again (GAYLORD et al., 2008), similarly to *Monochamus* spp. in Dürrenstein Wilderness Area. In our study, high amounts of precipitation at dates without sawyer catch might have additionally negatively influenced activity, although no significant interdependence between daily rainfall and *Monochamus* spp. capture was observed. Beetles remained calm when released on rainy days, while they mounted the stem or flied immediately under warm and sunny weather conditions. Studies of *M. alternatus* evidenced rainy weather strongly limiting walk and totally preventing flight (KOBAYASHI et al., 1984). European sawyers generally tend to increased flight activity on sunny, warm hours of the day, which was already ascertained by HELLRIGL (1971).

M. sartor and *M. sutor* flight was registered in Dürrenstein Wilderness Area in the morning as well as in the afternoon independently from trap catches or beetle release. Moreover, in a breeding experiment in cages at BFW Vienna, beetles of both species were watched feeding, mating and ovipositioning during the day. This conflicts with observations of mainly nocturnal *M. alternatus* in laboratory, showing walking, feeding and mating activities primarily from 8 pm to 5 am. Nonetheless, temperature was also considered as decisive factor here (NISHIMURA, 1973).

To our knowledge, no detailed studies of weather related flight behaviour of *Monochamus* spp. exist. In order to better examine direct weather effects on flight activity, daily trap controls are recommendable. Comparisons of beetle catch within a few hours on one day, as done during the 5-hour trapping observation to some degree, could also be appropriate. To determine the start of *Monochamus* spp. flight period in course of the year, trapping must commence in the end of May or beginning of June. Although, response to lures, which indicate the presence of mating partners or oviposition sites, might be low during the first weeks after emergence, when sawyers are in the phase of maturation feeding (HELLRIGL, 1971, MOREWOOD et al., 2002).

Trapped *Monochamus* showed increased activity, aggressiveness and faster movement in collection cups and transport boxes at high temperatures and particularly in thundery and oppressive air. Such behaviour was probably enhanced by too high beetle accumulation in small space, which consequently led to defence and elimination of competitors.

The results of beetle emergence control, varying capturing numbers in Dürrenstein Wilderness Area as well as the sawyers' activity in the receptacles indicate a key role of temperature in *Monochamus* phenology and behaviour.

***Monochamus* spp. response to attractants**

The main goal of the trapping experiment in Dürrenstein Wilderness Area in 2012 was testing the response of *M. sartor* and *M. sutor* to attractants. Traps were baited with all categories of lures known to influence the sawyers' host seeking and mate finding. Positive reactions of *M. galloprovincialis* and North American sawyers to bark beetle pheromones (ALLISON et al., 2003, PAJARES et al., 2004) suggested the inclusion of ipsenol and 2-methyl-3-buten-2-ol, which are constituents of the commercial lure G2D (Galloprotect 2D®, SEDQ, Spain), in our study. This bait further contains the *Monochamus* aggregation pheromone compound 2-undecyloxy-1-ethanol, which is present in *M. sutor* (REPHRAME Periodic Report, 2012), *M. galloprovincialis* and several North American and Asian species (PAJARES et al., 2010, TEALE et al., 2011, ALLISON et al., 2012, FIERKE et al., 2012). By means of Galloprotect Pack® (SEDQ, Spain) application, the host volatile α-pinene was added, in order to enhance attractance. Besides proven orientation of *M. galloprovincialis*, *M. scutellatus* and *M. mutator* towards this host odour (PEDDLE, 2000, DE GROOT and NOTT, 2003, IBEAS et al., 2006), behavioural studies evidenced sawyer response to fire injured trees (TRÄGÅRDH, 1929, HELLRIGL, 1971, RASKE, 1972, MEYER, 2002, SAINT-GERMAIN et al., 2004). Thus, the addition of smoke volatiles was hypothesised to further increase the luring effect.

For *M. sartor*, the same tendency of favouring G2D combined with α-pinene was obvious in both sexes, similar to *M. galloprovincialis* in Spain (PAJARES et al., 2010). However, only *M. sartor* males showed a significant difference between G2D and G2D + α-pinene. The addition of smoke volatiles did not further increase attractance for both sexes, but it was slightly higher than for G2D alone. Because of registered catch with G2D alone, we assume that *M. sartor* responds to the congeneric pheromone compound, not only to the scolytine substances. GC-MS analysis of volatiles released by one individual *M. sartor* male evidenced the emission of this compound, albeit only in lower quantities than *M. galloprovincialis* or *M. sutor*. The compound was not emitted by a *M. sartor* female of the same age (HALL, pers. comm., 2013). The analysed beetle was an offspring of the Dürrenstein Wilderness Area population from 2012.

In contrast to *M. sartor*, female *M. sutor* were mainly lured by G2D alone, second by G2D + α-pinene and less by the combination with smoke odours, as Spanish tests proved exactly in the same way (REPHRAME Periodic Report, 2012). Though, these were only trends and no significant dissimilarities were established in our experiment. Males did not show any intensified orientation towards a certain lure blend. Their catch was almost equally low for all attractants, despite reactions similar to females that were registered in Spain (REPHRAME Periodic Report, 2012). High variations in lure preference between the trapping periods were not observed for *M. sutor*. In general, both sexes of *M. sutor* were captured in quantities that were too low to be sufficiently representative. To deduce trustworthy and significant information on kairomone and pheromone influenced flight behaviour, catch amount has to be increased. This might be achieved by placing traps in the tree canopies, instead of ground level or by lengthening of the study period, as discussed above. Another reason for low trapping amount could be a general rare occurrence of *M. sutor* in Dürrenstein Wilderness Area.

Generally, more females than males of both species were caught throughout all treatments, although HELLRIGL (1971) assumes a natural sex ratio of 1:1 in all European *Monochamus* species according to mass breeding experiments. A female dominated imbalance was also observed in trapping experiments with α-pinene for *M. scutellatus* and *M. mutator* (PEDDLE, 2000). In our study, this was probably favoured by application of the aggregation pheromone compound in all lure combinations, which naturally attracts females stronger than males (PAJARES et al., 2010). Similar results were obtained by FIERKE et al. (2012), when more females than males of both *M. scutellatus* and *M. notatus* were caught with 2-undecyloxy-1-ethanol. In our third treatment, oviposition stimulating smoke volatiles (TRÄGÅRDH, 1929) might not be responsible for the higher amount of captured females, since host trees for mating and egg deposition are selected by males before female arrival (NAVES et al., 2008). As cerambycid females are generally more frequently present on future larval hosts than males, conclusions from capturing result to beetle eclosion sex ratio are impossible (FIERKE et al., 2012).

In experiments of ALLISON et al. (2003), including several host volatiles and bark beetle pheromones – but no sawyer pheromone, female and male *M. scutellatus* and *M. clamator* catch was almost equal or even slightly more males were captured. Further studies employing every category of attractants solely are required to determine that lure with the decisive effect on sex-specific catch. With that, also species-specific differences in lure preference could be demonstrated.

Since our study was the first trapping experiment on *M. sartor* and *M. sutor* in Dürrenstein Wilderness Area, several groups of attractants with presumed high luring effects according to other surveys (ALLISON et al., 2003, IBEAS et al., 2006) were combined to catch as many sawyers as possible. This strategy was successful. We evidenced that both species positively respond to the employed lures. This result was particularly important for *M. sartor*, whose chemical ecology has not been analysed before. On the basis of these findings, separate use of single attractants in a follow-up study is possible in order to analyse the effect of each on the sawyers' flight activity. Further research could also explain species dependent differences in lure preference and confirm or disprove the tendencies found in this study.

Tests of smoke volatiles independent from other host odours are required in particular, because currently, their attractancy for European *Monochamus* spp., except *M. galloprovincialis* (REPHRAME, Periodic Report, 2012), has not yet been investigated in detail. In our study, both *M. sartor* and *M. sutor* did not show any increased response. Also female *M. scutellatus* did not prefer burnt over unburnt logs for oviposition. Emission of certain luring odours by fire damaged trees stimulating *M. scutellatus* egg deposition is unclear. Thus, host finding of this species appears to be opportunistic and all kinds of stressed conifers are appropriate (BRETON et al., 2013). *M. sartor* and *M. sutor* might also follow such host seeking strategies, explaining no enhanced catch with smoke volatiles in Dürrenstein Wilderness Area in 2012. However, continued research is required to understand the exact behavioural patterns of *Monochamus* spp.

Furthermore, the attractiveness of the aggregation pheromone compound 2-undecyloxy-1-ethanol should be analysed separately to prove its importance for orientation of *M. sartor* and *M. sutor*, as it was already done for *M. galloprovincialis* (PAJARES et al., 2010).

Not only other combinations of the present lures, but also different release rates, causing species or sex dependent variations in reaction (IKEDA et al., 1980a, PEDDLE, 2000, IBEAS et al., 2006, PAJARES et al., 2010) should be considered. Moreover, the installation of a control trap group or one additional blank trap per baited trap block would determine the sole effect of visual attraction for *M. sartor* and *M. sutor*.

Recapture

In order to get an insight into dispersal flight behaviour of *M. sartor* and *M. sutor* in Dürrenstein Wilderness Area, flight distances, directions and time were measured during a recapture experiment. Recorded maximum flight distances of 387 m representing our maximum trap distance likely do not correspond to real maximum flight performance. It is possible that released *M. sartor* and *M. sutor* spread anywhere in the study area before being recaptured in one of our traps. So, determined flight distances do not necessarily reflect actual flight capacities of *M. sartor* and *M. sutor,* which might be probably as high as those of *M. scutellatus* or *M. galloprovincialis*. For the North American *M. scutellatus*, travelling distances of at least 10 km are reported (RASKE, 1972). But also the European *M. galloprovincialis* is known to fly up to 8.3 km and 3.6 km on average, examined during a Spanish mark-release-recapture survey. The most preferred trap, however, was situated in a distance of 2.8 km from the release point. Possibly, the arrangement of this trapping experiment, with a release point in an agricultural environment more than 2 km remote from coherent forest and nearest traps (GALLEGO et al., 2012), induced wider dispersion of beetles, that may be quite different from our study. Because in Dürrenstein Wilderness Area, sawyers found suitable breeding and feeding conditions as well as baited traps in direct surrounding of the release point. Our results of flight distance measurement were rather comparable with those of TOGASHI (1990), who recorded average dispersion distances of 10 to 20 m per week for *M. alternatus* during the first month after eclosion. Other studies of this species established that 75 % of recaptured beetles covered a distance of 100 m and few were caught 1 to 2.4 km away. *M. alternatus* dispersal behaviour between islands and disjunct forests is characterised by flight of 800 m, up to 3.3 km maximum (KOBAYASHI et al., 1984).

In our experiment, real required time to reach the recapturing trap was not measured, because it was unknown whether beetles stayed for some days on the release trees, as it was observed for *M. alternatus* remaining there from approximately 1.4 to 11.5 days on average (TOGASHI, 1990). Only some individuals were observed flying immediately, most climbed up the release trees while others stayed at the points where they were placed with apparent effects of weather conditions at the time of release. However, conditions for dispersal activity were favourable in Dürrenstein Wilderness Area because of high sawyer density per tree at the release site and sparse stand density (TOGASHI, 1990). Mean times of 8 days for *M. sartor* and 6.6 days for *M. sutor* were shorter than calculated means for *M. galloprovincialis* in Spain, which varied trap dependent from 8 to 33 days (GALLEGO et al., 2012). Also 48 days maximum duration of recapture in the Spanish experiment exceeded our measured 14 days for one *M. sutor*. Larger trap distances between 2.1 and 8.3 km to the release point, compared with about 0.04 to 0.39 km in Dürrenstein Wilderness Area, probably explain longer time intervals until *M. galloprovincialis* were recaptured.

In order to exactly determine flight time and distances of released beetles, equipping sawyers with senders might be one solution. However, sender stability and durability for several weeks are required, besides a detection radius up to some kilometers. Reflecting tags fastened on beetles and transceivers for location, as applied by WILLIAMS (2004) for *A. glabripennis*, are not appropriate for *Monochamus* spp., due to short range, functional periods and high fragility. Moreover, no distance measurement, beyond simple detection, was possible with this method.

During the Spanish study of *M. galloprovincialis* dispersal, flight directions of released beetles were mainly influenced by wind direction and wind speed. Most beetles flew against the dominating wind well-directed towards the source of volatiles (baited traps). This proved flight capacity and ability to localise pheromone and kairomone odours over large distances (GALLEGO et al., 2012). In Dürrenstein Wilderness Area, no wind correlated flight behaviour was observed. In spite of prevailing winds from east and north, all trap groups attained recapture and no group was especially preferred. The average wind speed of 11 km/h from main wind directions in our study was similar, but slightly higher than measured velocities in Spain.

Recapture percentage of both species was generally low with 1.3 % for *M. sartor* and 5.8 % for *M. sutor* of the total released beetles. These proportions were even lower than 7 % recaptured *M. alternatus* or at least 11 % *M. galloprovincialis* after release (KOBAYASHI et al., 1984, GALLEGO et al., 2012). Predation by birds or small mammals at the release point might be one reason for low percentage, since some marked elytra were found there the next release date three to four days later. These predators may have been attracted by high beetle density at the release site. Moreover, some of the beetles likely suffered from being in the trap collection cups for some time.

Bycatch

Throughout our trapping experiment, also non-target organisms were analysed in order to get an overview of the species spectrum. Furthermore, possible influence of used attractants and weather factors on flight activity was examined.

We confirmed the occurrence of the major part of captured longhorn beetle species in Dürrenstein Wildness Area, as they were already classified as present by ZÁBRANSKÝ (2001) during studies from 1998 to 1999. Moreover, our experiment proved the presence of *S. buprestoides* and *A. griseus*, previously classified as "possible" and "expected", respectively, but not found by ZÁBRANSKÝ. Besides the target species *M. sartor* and *M. sutor*, caught *R. alpina* and *C. capra* are listed in the European Red List of Saproxylic Beetles within category "Least Concern" (NIETO and ALEXANDER, 2010). The protected *R. alpina* seemed not to be frequent in Dürrenstein Wilderness Area according to ZÁBRANSKÝ (2001). Capture in our traps and bark beetle traps by BLACKWELL (pers. comm., 2012) as well as observations of several specimens in trapping season 2012, however, indicate a recurrent incidence of this species in the study area. Moreover, 2 *C. capra* were caught in Hundsau, only found once by ZÁBRANSKÝ (2001) and thus declared as rare.

Similar to Cerambycidae, we proved the presence of some Buprestidae. *C. igniventris* and *C. chrysostigma*, which were not recorded in studies of ZÁBRANSKÝ (2001) or only larval galleries indicated former occurrence in the Wilderness Area, respectively. Particularly the remarkable find of *C. igniventris* (determination confirmed by ZÁBRANSKÝ, 2013) was not expected for Dürrenstein Wildness Area, because this species usually does not populate mountainous habitats (ZÁBRANSKÝ, pers. comm., 2013).

Monochamus spp. associated species described by HELLRIGL (1971) were rarely caught in our study; some bark beetles were probably not recorded due to their small size. We captured *A. griseus*, *A. quadripunctata* as well as the scolytines *I. typographus*, *P. chalcographus* and *P. poligraphus*, in correspondence with HELLRIGL (1971). Moreover, the wood wasps *U. gigas* and *S. juvencus*, were trapped. *Sirex* spp. are known to be associated with *M. sartor*, often populating slightly damaged trees before *Monochamus* spp. (HELLRIGL, 1971).

Regarding coccinellid bycatch, capture of the invasive *H. axyridis* was of particular interest, since it was caught at 3.5 km distance from the next human settlement and approximately 8 km from the next village. The find demonstrates again the invading capacity of *H. axyridis*.

Our capturing result does not represent the complete xylobiont insect and beetle species spectrum of Hundsau, Dürrenstein Wilderness Area. Since with baited traps, only one investigation method was applied and trapping period was too short. According to ZÁBRANSKÝ (2001), continuous studies over several years are required to gain a detailed insight into the prevailing entomofauna of an area. This is inconsistent with conclusions of JURC et al. (2012), who recommended a 3-year monitoring with traps being sufficient for determination of the insect diversity.

Regarding weather influence on cerambycid, clerid and siricid bycatch, similarities to *M. sartor* and *M. sutor* were only established for *A. rusticus*, *T. formicarius* and *T. apiarius.* Bark beetle predators benefit from adaptation to flight behaviour of their prey, apparent in comparable temperature thresholds for flight initiation of *Enoclerus* spp. (Cleridae), *Temnochila chlorodia* (Trogositidae) and the scolytines *Ips* spp., *Dendroctonus* spp. in North America (GAYLORD et al., 2008). Rising air temperatures in the end of the investigation period did not increase catch of *L. rubra, A. griseus* and *U. gigas*, which was possibly conditioned by natural cease of flight or oviposition period and end of adult lifetime (BENSE, 1995, RICKINGER, 1998 - 2013). On the contrary, *S. buprestoides* and *A. rusticus* are active from June to September (BENSE, 1995), evident in captured individuals by the end of August. Generally, few clear connections of bycatch flight behaviour and weather factors or *M. sutor* catch might be caused by low capturing amounts. Differences between the life styles and habitat colonisation of *L. rubra* and sawyers or *A. griseus*, *A. rusticus* and *S. buprestoides* may be responsible for missing correlations (BENSE, 1995).

In our experiment, non-target species that colonise wood or nourish from captured objects might have been attracted actively by lures or prey (chemical cues).The employ of α-pinene possibly lured non-target species with any association to conifers. Direct attraction to certain applied baits was supposed for other Cerambycidae, Buprestidae, Siricidae, bark beetles and their predators such as Cleridae (*T. formicarius*, *T. apiarius*) and Rhizophagidae (*Rhizophagus* sp.) as well as carrion beetles (Silphidae: *N. vespilloides*, *O. thoracicum*). Bycatch of xylobiont insects was also registered in many trapping experiments for *Monochamus* spp. all over the world. In Canada, MCINTOSH et al. (2001), MOREWOOD et al. (2002) and DE GROOT and NOTT (2003) captured inter alia *Arhopalus* spp., *Buprestis* spp., *Chrysobothris* spp., *Sirex* spp. and *Urocerus* spp. – only to name those genera that were caught during our study as well. Due to similar habitat requirements, trap types representing a prominent vertical silhouette are appropriate for capturing large woodborers (MCINTOSH et al., 2001). Consequently, with employment of baited multiple funnel traps, catch of other wood-breeding insects cannot be excluded and is rather most probable. Studies from Slovenia proved the attractiveness of traps with α-pinene and several bark beetle pheromone components for *S. buprestoides*, *A. rusticus*, *R. inquisitor*, *L. rubra*, *S. melanura* and several other Cerambycidae and xylobiont insects, besides *Monochamus* spp. (JURC et al., 2012). In Italy, where also G2D and G2D + α-pinene were tested, longhorn beetle bycatch spectrum consisted of *A. rusticus*, *L. rubra*, *R. inquisitor* and *S. buprestoides* for instance (RASSATI et al., 2012). On the contrary, during *M. alternatus* trapping in Asia (2-undecyloxy-1-ethanol, α-pinene and ethanol differently combined), bycatch was very low, so that lure attractancy for non-target insects was excluded (TEALE et al., 2011). Thus, besides trap type, application of attractants plays a decisive role for bycatch complexity. In particular, the presence of the *Monochamus* spp. pheromone compound and scolytine pheromones was important. Another factor determining bycatch species spectrum is surely the habitat and host tree species. All studies named above were mainly conducted in pine forests, in small extent in fir or spruce dominated forests, and near sawmills. One-to-one comparisons to our experiment carried out in a mountainous spruce stand in a nature reserve are therefore not possible.

In Dürrenstein Wilderness Area, most frequently captured cerambycids as well as *T. formicarius* and *U. gigas* showed highest preference for lure combinations containing α-pinene and lowest for G2D alone. With use of the latter, even no *S. buprestoides* and *U. gigas* were caught at all, suggesting the role of α-pinene in host selection for these species. *A. griseus, T. formicarius* and especially *A. rusticus* capture decreased through employment of smoke odours. Decline of *A. rusticus* catch with smoke volatiles was not expected, due to reported orientation of congeneric *A. tristis* and *A. ferus* towards burnt bark and trees (SUCKLING et al., 2001, SANTOLAMAZZA-CARBONE et al., 2011). Generally, derivation of attractant preferences of captured non-target cerambycid species is difficult, since total trapping results amounted from 14 to 27 individuals per species. Attractiveness of applied lures for *R. alpina* can be excluded, because this species exclusively develops in deciduous trees (ZÁBRANSKÝ, 2001). Catch of three individuals was supposed accidental and caused by proximity of breeding trees to our traps.

Bark beetle predators were most probably strongly lured by scolytine pheromone components ipsenol and methyl-butenol, both components of G2D, obvious in catch registered throughout all treatments. Extremely high trapping amounts of *T. formicarius* were also observed during *M. galloprovincialis* monitoring in Brandenburg, Germany in 2012 (HIELSCHER, pers.

comm., 2013). Likewise, JURC et al. (2012) recorded clerid bycatch in Slovenia, dominated by *T. formicarius*.

Although buprestid bycatch was much too low (total of 24 specimens) for statistical analysis, attractiveness of smoke volatiles may be hypothesised for species associated with forest fire, such as *Chrysobothris chrysostigma* and *C. igniventris* (ZÁBRANSKÝ, pers. comm., 2013).

The captured Endomychidae (*E. coccineus*), Mycetophagidae, Cisidae and Erotylidae could have been either actively lured by traps and attractants, indicating weakened conifers, since all these families colonise dead wood and subsist on fungi (BENISCH, 2007 - 2013). The recurrently trapped *Epuraea* sp. (Nitidulidae), in concordance with JURC et al. (2012), and *Plegaderus vulneratus* (Histeridae) most likely orientated towards bark beetle pheromone compounds, because these species are known to feed on scolytine larvae in bark of dead coniferous trees (MÖLLER, 2009).

Throughout our experiment, bycatch amount was relatively high, especially regarding bark beetle predators. This is similar to observations for instance by JURC et al. (2012) with a proportion of 7 % and HIELSCHER (pers. comm., 2013). Hence, modifications of the attractant blends reducing bycatch are desirable. As concluded from other REPHRAME studies, α-pinene attracts unspecifically and should therefore be excluded from baits when bycatch of non-target organisms is of concern. Potentially replacing α-pinene by smoke volatiles may be another option that deserves further attention (REPHRAME Periodic Report, 2012).

Conclusions

Regarding the target species *M. sartor* and *M. sutor*, the trapping experiment in Dürrenstein Wilderness Area in 2012 evidenced the efficacy of the applied trapping method for monitoring of these potential PWN vector species. Capturing results proved the suitability of Teflon coated 12-unit multiple funnel traps with deep dry collector cups for live catch. Further, the appropriateness of employed lures Galloprotect 2D® alone or combined with α-pinene (Galloprotect Pack®) or both α-pinene plus smoke volatiles was established, especially for the females of both species. Moreover, the study demonstrated the attractiveness of traps and lures for non-target organisms. α-Pinene might have played a central role for conifer associated bycatch species.

M. sartor and *M. sutor* catch was highest at warm air temperatures, when beetles show high activity. Generally, warm temperatures can be considered as eliciting factor for adult emergence and flight. Trapping beetles from June to September would probably cover the entire adult flight period and consequently raise total trapping amount. In course of the recapture experiment, a suitable beetle marking technique was established and demonstrated a dispersal capacity of at least 400 m within two weeks. These measures allow targeted observation and will aid effective pest management of *M. sartor* and *M. sutor* in case of PWN introduction to Austria. Though, further studies are desirable to validate these findings or improve the methods.

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