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Fakulteten för landskapsplanering, trädgårds- och jordbruksvetenskap

Pheromone-based monitoring of *Elater ferrugineus* as an indicator for species-rich hollow oak stands

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Abstract

The rare beetle *Elater ferrugineus* was sampled at 47 sites in the county of Östergötland, Sweden by means of pheromone-baited traps to assess its value as an indicator species for hollow oak stands rich in rare saproxylic beetle species. In addition *Osmoderma eremita* was sampled with the same method. These data were then compared against species survey data collected in the same sites by pitfall and window-trapping. Weighted odds ratio models revealed that both species co-occur with many red listed saproxylic beetle species, with *E. ferrugineus* being a somewhat better indicator for the rarest beetle species. Regression models showed that the conservation value of a site (measured as red list points or number of red listed species) increased with the number of specimens of *E. ferrugineus* and *O. eremita* caught. Accuracy of sampling by means of pheromone trapping turned out to be radically different for the two model species. *E. ferrugineus* traps put out during July needed to be up for only 6 days in order to gain full accuracy, whereas *O. eremita* traps needed to be out from early July to mid August in order to obtain full accuracy with one trap per site. By using *E. ferrugineus* or preferably both species as indicator species, effort spent in the field, and thereby the costs for saproxylic biodiversity sampling, could be greatly reduced compared with conventional methods, and the accuracy of classification of valuable habitat significantly improved.

Keywords: *Elater ferrugineus*, *Osmoderma eremita*, saproxylic, indicator, *Quercus* sp., pheromone, conservation

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Introduction

All over the world species are pushed towards and too often beyond the edge of extinction by human destruction and fragmentation of habitats (Pimm & Raven 2000, Chisholm et al. 2011). Many of the species that are driven to extinction are insect species. The hidden number of insect extinctions is probably high due to the fact that many insect species are yet to be described, or their populations cannot be monitored to the extent where we can properly assess their threat status (Myers et al. 2000). Identifying and protecting key areas or habitat types that have a high number of potentially threatened species is therefore essential in conservation biology (Myers et al. 2000). However, complete surveys are expensive, time-consuming and often prevented by the lack of taxonomic experts. Therefore the use of indicator species instead of complete surveys to locate sites with high species richness is common in conservation biology in order to save money and time (Margules & Usher 1981, Simberloff 1998). Many indicator species used are vertebrates, whereas insects are less frequently used as indicators except in aquatic environments where insects have been used for long time to determine the status of rivers or lakes (Bolger et al. 1991). But the use of invertebrates as terrestrial monitoring species has increased, especially among butterflies (Lepidoptera) (Margules & Usher 1981, Rosenberg et al. 1986, Ehrlich 1992, Oliver & Beattie 1993, Sparrow et al. 1994, Nilsson et al. 1995, Myers et al. 2000). Monitoring for specific insect indicator species is nevertheless often difficult and time consuming, regardless whether performed by means of trapping or active search. There is considerable risk of overlooking individual indicator species at a site, due to low precision of the methods typically used (Jansson 2009). However, pheromone-based trapping systems have been used in pest management for decades for efficient monitoring of a wide range of pest species. These systems are usually highly species specific, which should make them appropriate for conservation purposes in order to make surveys for indicator species more effective, but until recently these systems have been exclusively used in pest management (Bengtsson et al. 2006, Yasuda et al. 2010). The first pheromone identified for monitoring a rare and threatened insect species was the male-produced sex pheromone for *Osmoderma eremita* (Larsson et al. 2003). Using pheromones that attract indicator species, which in turn indicate sites with high species richness, is a promising way of monitoring sites with high conservation value. *O. eremita* is a species associated with hollow trees, and in Sweden it is mainly found in hollow oak (*Quercus* sp.) stands (Ranius & Nilsson 1997, Ranius et al. 2005). These hollow oak sites generally have high species richness (Nilsson et al. 1995, Økland et al. 1996). But hollow oak areas in Sweden have suffered from severe decline and fragmentation in the last 200 years, mainly due to the change in ownership of the oaks. Ownership of the oaks was transferred from the crown to local land owners in the beginning of 19th century, which resulted in many oaks being cut down (Eliasson 1999, Eliasson & Nilsson 2002). The fragmentation of the hollow oak stands in the landscape had implications for the flora and fauna connected to them (Eliasson & Nilsson 2002). Since many of the beetle species associated with hollow trees are assumed to have low dispersal rates, they are dependent on the continuity of hollow trees within a relatively narrow range (~2 km proposed by Nilsson and Baranowski 1994) (Nilsson & Baranowski 1994, Jansson 2006). Their low dispersal rates, in combination with the high

degree of habitat fragmentation, have caused many of the species connected to these habitats to be threatened (Gärdenfors 2010).

In 1994 Nilsson and Baranowski found that many click beetles species exclusively live in sites with long hollow tree continuity. However, they also claimed that these beetles would generally not be practically useful as indicators since they are believed to be under-sampled – meaning that even if they are present at a site, the chance of detecting them is low (Nilsson & Baranowski 1994). Among these beetles is the rare rust red click beetle (*Elater ferrugineus*), which was until 1993 only known from 25 sites in Sweden, and which is frequently associated with *O. eremita* (Nilsson & Baranowski 1994).

In 2002 it was discovered that the females of *E. ferrugineus* were attracted to the pheromone of *O. eremita*, which made it possible to monitor the presence of this rare species classified as Vulnerable in the Swedish red list of 2010 (Svensson et al. 2004, Gärdenfors 2010, Ljungberg et al. 2010). In 2007 the female sex pheromone of *E. ferrugineus* was identified, further expanding the possibility to monitor the abundance of this rare species (Tolasch et al. 2007, Svensson et al. 2011b). Holt (1993) stated that habitat interactions at different trophic levels work over different spatial scales, and species higher in a trophic web should be more affected by fragmentation than species lower in the trophic web. Since *E. ferrugineus* larvae are generalist predators, but nevertheless partly dependent on relatively few large prey species including *O. eremita*, then it is reasonable that it is more sensitive to habitat fragmentation than *O. eremita* (Holt et al. 1999, Holt 2002). Recent studies have shown that *E. ferrugineus* exhibits higher population fluctuations (Larsson & Svensson 2011) and depends on hollow tree resources at larger spatial scales (Ranius et al. 2011) than *O. eremita*. Therefore, *E. ferrugineus* could be a very sensitive and cost-effective indicator of the biological effects of landscape fragmentation, given the present potential for monitoring with a highly attractive pheromone.

O. eremita is protected by law in many European countries and is classified as Near Threatened according to the red list of 2010 in Sweden (Anonymous 1992, Gärdenfors 2010, Ljungberg et al. 2010), and had been found at approximately 130 sites around Sweden until 2005 (Ranius et al. 2005). It is exclusively dependent on the wood mould in hollow trees (Ranius & Nilsson 1997, Ranius et al. 2005). Several studies have shown that *O. eremita* is a species that indicated hollow oak stands with a high conservation value (Jansson et al. 2009, Ranius 2002). Pheromone-baited traps have been used in the field to estimate population sizes of this rare species; when comparing with previous studies carried out during several years with un-baited pitfall traps in the same region, the results showed that population sizes previously had been underestimated (Larsson & Svensson 2009). The possibility of calculating species abundance at a site with a relatively low work effort strengthens the potential for pheromone based systems as a tool for monitoring species abundance, which is necessary to be able to calculate the extinction risk of a species (Schultz & Hammond 2003, Larsson & Svensson 2011).

The main aim of this project was to evaluate the usefulness of pheromone-trapped *E. ferrugineus* as an indicator species for hollow tree stands with a rich saproxylic insect fauna.

This was studied by means of pheromone trapping of *E. ferrugineus* in sites previously surveyed during 1994-2010 by Nicklas Jansson (see e.g. Jansson 2009, Jansson et al 2009). The question addressed in the present study was to determine whether pheromone trap catch of *E. ferrugineus* is a good predictor for the presence of individual rare and threatened species, as well as for sites rich in rare species. Species matching with the red list of 2010 was used to test the hypothesis that *E. ferrugineus* is more abundant at sites with high richness of red listed saproxylic beetle species associated with hollow oak stands. The presence of *E. ferrugineus*, and its indicator potential, was compared with that of *O. eremita*, which has already been shown to have high indicator potential for saproxylic beetles (Ranius 2002, Jansson et al. 2009). Methods used in this project were matched against both the data on *E. ferrugineus* and *O. eremita* sampled by Jansson (2009) and data reported into Artportalen (Species Gateway), which is a site where anyone may report records of any Swedish species. In this way I have both determined the accuracy and usefulness of *E. ferrugineus* as an indicator species for hollow oak stands with very high conservation value, and the efficiency of the two pheromones in detecting the respective species compared to other methods.

Material & methods

Background of trapping methodology and datasets used in this study

Pitfall- and window-trapping

Jansson's surveys 1994-2010 were carried out with trapping in individual trees with pitfall traps and window traps (one of each in every individual oak sampled). Data obtained from Jansson (2009 *and pers. comm*) that has been used in the present study was obtained from four oaks per site, which yields approximately 40% of the species according to Jansson (2009) (see Figure 1). Only saproxylic beetle species associated with hollow oaks were used.

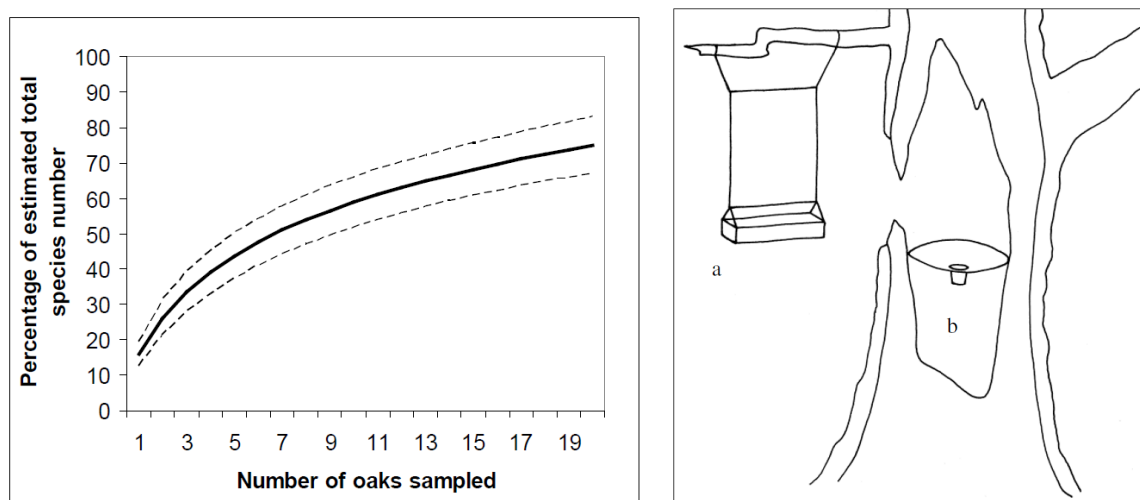


Figure 1. Rarefaction graph (left) showing the cumulative percentage of beetle species detected as a function of the number of oaks sampled. Right panel shows a hollow tree with a window trap (a) and pitfall trap (b). Figure panels obtained from Jansson 2009 with permission from Niklas Jansson.

Sites for trap placement

Data from Jansson's surveys in Östergötland, Sweden 1994-2010 were used to obtain 47 sites for the project. Since the aim of the project was to use the species diversity of whole sites, rather than the diversity of individual trees, some limitations were set to define what constituted a site. Firstly, the extent of individual sites was delineated by the area around the trap when the trees were less than 500 m apart from one tree to another. Secondly, the sites should not be closer than 500 m from each other, in order to avoid conflicts between traps since the sex pheromone of *E. ferrugineus* has been proven to be effective at large distances (Larsson et. al. *unpubl.*). The target position for each trap was the centroid of the four trees sampled per site in the 1994-2010 survey. All traps were mapped into ArcGIS (ESRI ArcMap 9.3.1, Redlands, California, U.S.A.) in order to check for error in placement.

Site naming

Since some of the sites have been used in earlier studies with different naming, sites with alternative naming in other studies are therefore listed here in order to reduce the name confusion. Names within parenthesis are names previously used for the same site in other studies; Brokind (Skolhagen), Fornhagen (Kalvhagen), Ravnäs (Rafnäs, Ravsnäs), Runstorp (Rundstorp), Skaggebo (Storängen).



Figure 2. Top left: marking scheme and marked *O. eremita* with individual nr. 52, top right: trap hanging from branch, down left: *E. ferrugineus*, down right: trap with *E. ferrugineus* beetles in a trap at Runstorp, background picture *Succisa pratensis* in front of oak trees (*Quercus robur*) at Norsholm. Photographs: K. Andersson

Artportalen (Species Gateway) records

Artportalen is a Swedish site where records of various animals and plants may be reported; in this case the records for *E. ferrugineus* and *O. eremita* have been assessed. Since the aim of using the Artportalen dataset has been to compare the different datasets and the methods used, some of the reports in Artportalen have been excluded. These are either data reported from Jansson's survey (Jansson 2009, Jansson et al 2009) that have been included in the dataset used in the present study, or data obtained by means of pheromone baited traps. Reports included in the present study are not older than 30 years; sites have been obtained with a precision of approximately 1 km radius around the trap position. Complications with using data of this type is that it may be of varying quality since both professional entomologists as well as laypeople may add records to this site, yielding a mix of results from surveys and spontaneous observations with very different methods. As an example of methods included in the Artportalen dataset are findings of frass (*O. eremita*) and pieces of insects such as elytra or pronotums. Another substantial problem is the inherent reporting bias, where absences of individual species at any particular site are seldom reported. Therefore, almost only presence data may be obtained from this site. Artportalen site visited 19/10-2011.

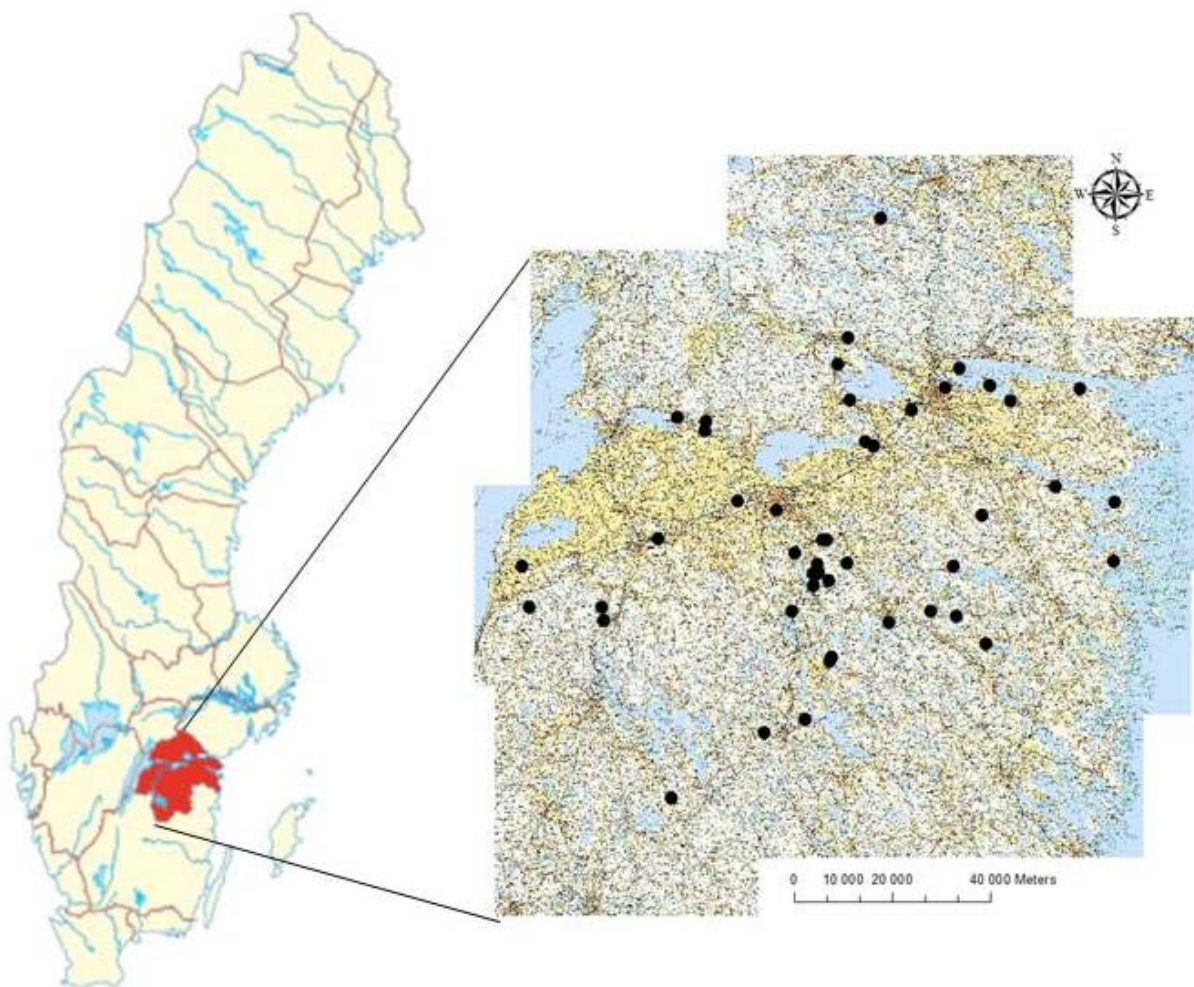


Figure 3. Maps of the county of Östergötland, Sweden with the sites sampled marked out. Maps from © Lantmäteriet, I2011/0032.

Model species

Elater ferrugineus, (Elateridae), Linnaeus 1758

The red click beetle *Elater ferrugineus* (synonym: *Ludius ferrugineus*) is Sweden's second largest click beetle (imago size about 17-24 mm, the largest click beetle to be found in hollow trees, see Figure 2) and lives in deciduous hollow trees such as oak (*Quercus* sp.) and beech (*Fagus sylvatica*) (Palm 1954, 1959, Nilsson & Baranowski 1994). The life cycle of *E. ferrugineus* is between 4-6 years probably depending on the amount of food available (Palm 1954, 1959). Larvae of *E. ferrugineus* are mainly predators and feed on other insect larvae such as larvae of *O. eremita* but given the chance it will also be cannibalistic (Nilsson & Baranowski 1994, Palm 1954, 1959). *E. ferrugineus* was until 1993 only known from 25 sites and is often thought to be an under-sampled species in surveys (Nilsson & Baranowski 1994, Jansson 2009).

Osmoderma eremita, (Cetoniidae), Scopoli 1763

The hermit beetle *Osmoderma eremita* (imago size 25-35 mm) is a saproxylic beetle associated with hollow trees (see Figure 2). The most important factor of the tree is the size and direction alignment of the hollow opening and the amount of wood mould inside (Ranius & Nilsson 1997). In Sweden it has only been found in deciduous hollow trees, where the larvae feed on the mycelia and chew the trunk from the inside and thereby enlarging the hollow (Ranius & Nilsson 1997, Ranius 2002, 2005). The normal life cycle of *O. eremita* in Sweden is about 3 years; imagos emerge in late June and the season is usually over before the end of August (Ranius & Nilsson 1997). *O. eremita* is known from 270 sites in Sweden but recorded in only 130 of these after 1990 in spite of extensive surveys. It is therefore highly plausible that the rate of extinction from sites is higher than the rate of colonization (Ranius 2005).

Pheromone trapping

Pheromone trapping can be a highly species-specific method, depending on the pheromone used and species aimed for. In the present project pheromones for *E. ferrugineus* and *O. eremita* have been used. The female-produced sex pheromone for *E. ferrugineus* attracts only males, but the pheromone for *O. eremita* has been shown to attract mostly females of *O. eremita* and females of *E. ferrugineus*, but in much lower numbers than the sex pheromone of *E. ferrugineus* (Larsson et al. 2003, Svensson et al. 2004, Svensson et al. 2011a).

All field trapping in the present study was performed with LS cross-vein funnel traps of the type according to Figure 2 and 4, (Svensson & Larsson 2008). Each trap was baited with both the sex pheromone for *E. ferrugineus* (7-methyloctyl (Z)-4-decenoate; Tolasch et al. 2007, Svensson et al. 2011b) and *O. eremita* (*R*)-(+)- γ -decalactone; Larsson et al. 2003). The sex pheromone of *E. ferrugineus* (>98% purity) was obtained from Erik Hedenström's research group at Mid Sweden University (Svensson et al. 2011b). Baits were

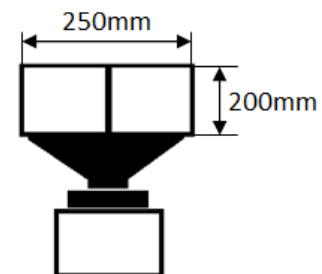


Figure 4. Principal scheme of LS cross-vein funnel trap, with measures of the cross-vein.

made from 200 μ L PCR tubes loaded with 2 μ L aliquots of neat compound, and pierced with an insect pin size 3 just below the lid to release the pheromone (Tolasch et al. 2007). The sex pheromone of *O. eremita* was obtained as a racemic mixture from Sigma-Aldrich, as the racemate can be substituted for the pure (*R*)-enantiomer (Svensson & Larsson 2008). Baits were made from 4 mL glass vials loaded with 600 μ L of neat pheromone, with a cotton dental roll (Celluron, Paul Hartmann, S.A., France) inserted as a wick. One trap per site, containing one bait for each of the two model species, was placed at approximately 2.5 m height from the ground on a branch facing north from the stem, in order to avoid sun exposure of the trap.

Traps were emptied every third day during the first three weeks and thereafter the traps that caught beetles were henceforth emptied at this interval, whereas the traps that did not catch beetles were checked less frequently in order to ease the work load of the fieldworkers. Beetles were marked with a queen bee marking pen (Uni paint marker PX-21, Mitsubishi Pencil co., LTD.) on the elytra with a unique combination of marks for each individual (see Figure 2 for marking scheme). Caught beetles were released in the field after marking. The release procedure for *E. ferrugineus* was to take the beetles approximately ten meters away from the trap-tree and throw them up into the air, which will force them into flight. Release of *O. eremita* was somewhat different since these beetles are not as prone to fly as *E. ferrugineus*. *O. eremita* was put on a tree at least 15 m from the trap-tree. In cases where the beetles flew from one trap to another these beetles were recorded as new to the trap where they ended up. Measurements of the movements of beetles between traps were calculated in ArcGIS (ESRI ArcMap 9.3.1, Redlands, California, U.S.A.) as the shortest possible geodesic route for the beetle (as the crow flies).

Trapping periods used in statistical analyses

The trapping periods used in statistical analyses (Period A/B) were first chosen as starting point for the traps to see if it would be possible to explain the variance in rare species only by trapping for 6 days in the beginning of July (Period A; 5 July-12 July). Second period (Period B; 23 July-30 July) was in late July in order to see the change between late and early July, respectively. Only new individuals caught within the respective interval were used; recaptures within the same period were not counted. For the statistics on this part, the sample size for Period A and whole season was (n-1) since one site was not activated until mid July (because permission was given late by landowner).

Red list points for individual sites

Red list score 2010 was calculated according to the CSPI (conservation priority index) (Jansson et. al. 2009). The red list points are calculated as a score for each site, where each species found in the site is given a certain score depending on its threat category (NL – Not Listed: 0p; LC–Least Concern: 0p; NT – Near Threatened: 1p; VU – Vulnerable: 3p; EN – Endangered: 5p) and these scores are summed per site.

Odds ratios and statistical analyses

Odds ratios are related to the probability of finding species x at a site with species y. If the odds are significant, and the odds <1 it means that species x is less frequent in sites with species y than in sites without species y. If it is =1 it means that the distribution is random,

and if it is >1 it means that species x is more frequent when species y is present. Odds ratios and weighted odds ratios are commonly used in other disciplines such as medicine but are less used in environmental sciences and ecology (Rita & Komonen 2008, Morris & Gardner 1988, Bland & Altman 2000). Odds ratios is a tool to measure the size of the deviation and its direction between proportions; as argued by Rita & Komonen (2008) this way of treating data should have more attention in ecology since it is a statistical sound way of treating proportions.

Odds ratios (OR) may be calculated according to Morris & Gardner (1988).

$$OR = \frac{(A * B)}{(C * D)}$$

Where: A= no. of sites with both species x (from Jansson data) and *E. ferrugineus* or *O. eremita* (from the present study results) present
 B= no. of sites with neither of the species present
 C= no. of sites with *E. ferrugineus* / *O. eremita* present and species x absent
 D= no. of sites with *E. ferrugineus* / *O. eremita* absent and species x present

An approximation of the confidence interval is then given by (Morris & Gardner 1988, Bland & Altman 2000):

$$\text{Standard error: } SE(\ln(OR)) = \sqrt{\frac{1}{A} + \frac{1}{B} + \frac{1}{C} + \frac{1}{D}}$$

The 95% confidence interval is then given by:
 $e^{(\ln(OR) \pm N_{(1-\alpha/2)} * SE(\ln(OR)))}$
 ($N_{1-\alpha/2}$) is 1,96, normal distribution with a two tailed 95% CI.

Odds ratios were calculated in Comprehensive meta-analysis V2™, Englewood, New Jersey, U.S.A.. This program calculates the exact confidence intervals and was used for all odds related calculations in this thesis. The categories used when species were grouped according to the red list categories in the statistical analysis were; not listed (NL), least concern (LC), near threatened (NT) and Vulnerable and Endangered as one (VU+EN).

Generalized linear model analysis (GLZ)

Generalized Linear Models (GLZ) have been increasingly used over the past 20 years in biology, e.g. soil science, fishery, marine biology and conservation (Schneider 2009, 1992, Lane 2002, Venables & Dichmont 2004, Hardin & Hilde 2007, Whittingham et al 2006). Many authors emphasize the benefits of using the GLZ instead of general linear models, which may be seen as a special case of GLZ (Schneider 2009, Hardin & Hilde 2007, benefits e.g. Lane 2002, Venables & Dichmont 2004, Whittingham et al 2006). The high flexibility of the GLZ with possibilities to handle data that are not normally distributed was the main reason to why I chose to do a GLZ in the analysis. By using GLZ in all analyses it made all models comparable with each other. The choice of normal distribution and log-link was based upon the goodness of fit from runs with other distributions (e.g. binomial, Poisson). In order to choose the best describing model, the best subset model with the lowest AIC (Akaike Information Criterion) was chosen when modeling. GLZ analysis was performed with normal

distribution and log-link (link function for the response variable, -chosen in the analysis program), in order to test the relationships between variables, *E. ferrugineus/O. eremita* catches (over given time period) vs. species richness, number of red listed species, red list point, number of NT species and number of EN/VU species. Catch data (predictor) were $\log(x+1)$ transformed in order to lower the variance that affected the residuals. Statistics were carried out in Statistica (StatSoft, Inc. (2011). STATISTICA (data analysis software system), version 10. www.statsoft.com), Tulsa, Oklahoma, U.S.A..

Graphing of accuracy

Accuracy gives a measure on how accurate a method is in detecting a certain species, and is dependent on the correct detections on sites of presence/absence as well as false detection of either presence/absence on each site. In this study I have used accuracy as a means of detecting how quickly a dataset reaches agreement with the whole season dataset; no other data than the pheromone trapping performed in this study have been used.

Accuracy is calculated by:

$$\text{Accuracy} = \frac{(a) + (b)}{(a + b + c + d)}$$

Where: a=number of sites correctly detected as sites with species x present
b=number of sites correctly detected as sites with species x absent
c= number of sites falsely detected as sites with species x present
d= number of sites falsely detected as sites with species x absent

Cumulative accuracy-graphs started at different times, gives an estimate for how long time a trap needs to be active in order to get 100 % accuracy at a site. Three-day intervals were used for traps that caught beetles, but at a few occasions the interval was up to five days, in which case trapped beetles (if any) were noted as captured on the last day. For *O. eremita* the principle was the same, except for a few more odd visits due to the low catch frequency at some of the sites presumed to lack *O. eremita*, with the longest being 17 days and ending with a capture.

Nestedness analysis

In order to show if the saproxylic beetle fauna in old hollow oaks displayed ordered extinction patterns among the surveyed species and sites, a nestedness analysis was performed.

Nestedness analyses show whether species in a community of organisms disappear in an ordered way when affected by destruction/ fragmentation of the habitat, with the most sensitive species disappearing first and the most resistant species last (Figure 5). The potential indicator value of individual species is closely related to the overall degree of nestedness among the species assemblages at different sites. In a highly nested system, the presence of a species at a site indicates with high probability the presence of other species less sensitive to extinction. By contrast, if species disappear in a more stochastic fashion, the potential indicator value of a species is lower, as it provides less information about the presence of other species.

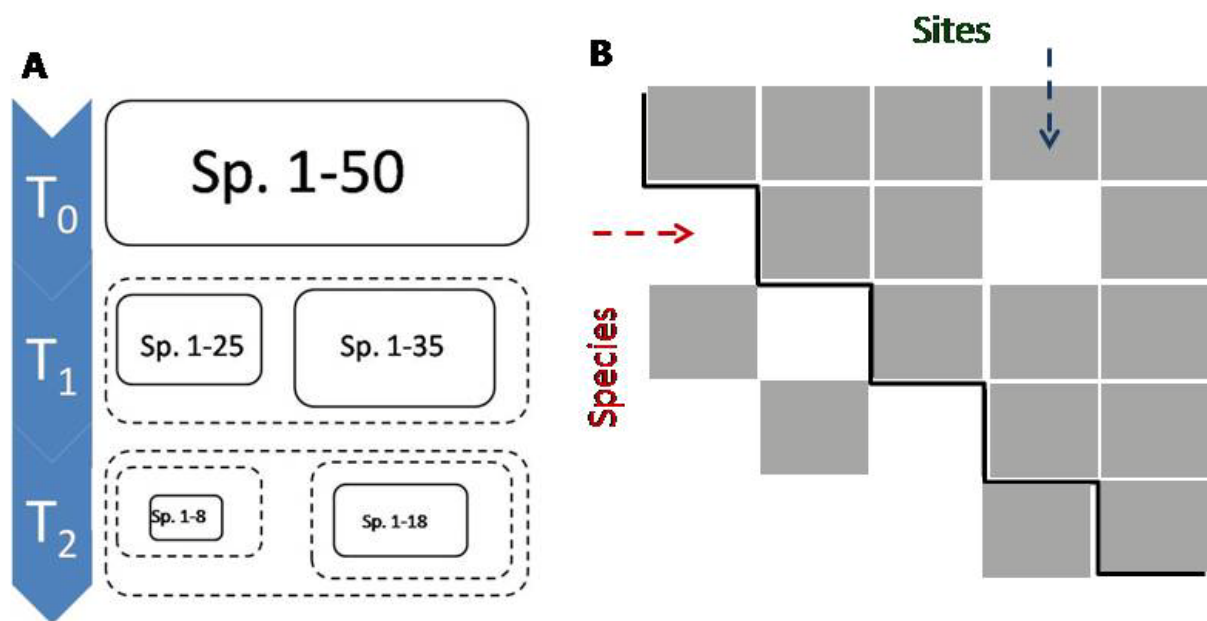


Figure 5. Graphical representations of principal nested extinction patterns.

A) At the beginning (T_0) before habitat fragmentation/ destruction all species 1-50 are able to exist within a single habitat. But as time goes and the habitat gets more and more fragmented and individual patches get smaller, the more species vanish from each patch. A good visualization to think of is found in Atmar & Patterson (1993) where the authors describes it as an island that becomes an archipelago due to raise of the sea level.

B) If species differ in their sensitivity to the effects of habitat destruction and fragmentation, they will disappear in a non-random order, with the most sensitive species disappearing first. This will result in a nested pattern of extinctions at different sites, when species are ordered according to an extinction hierarchy. This is illustrated in the figure as a matrix of presence and absence of individual species at different sites. Sites are represented as columns in the matrix, and individual species are represented as rows, with a grey square indicating presence of a species at a site, and a white square indicating an absence. If the process would be entirely deterministic, species would be ordered in a perfectly nested extinction hierarchy, with all presences to the right and above the line and all absences on the other side. In reality, extinctions are often stochastic to some degree, resulting in less than perfect order in the matrix. The degree of nestedness is usually represented as a temperature value representing the degree of entropy or randomness within the system. A temperature of 0 degrees represents a perfectly nested matrix, whereas a temperature of 100 degrees represents a totally random pattern of presence and absence.

The nestedness analysis will give a temperature which indicates if the species occur in a specific pattern in a hierarchical order of disappearance; a temperature of 0 degrees means that the species assemblage is perfectly ordered, whereas a temperature of 100 degrees means that species are totally disordered.

Nestedness temperature was calculated in the Nestedness Calculator by Atmar and Patterson (1993, 1995). Data from Jansson (2009) was used. Only presence/absence per site and species were used. 1000 iterations of randomized matrices with the same fill level as the one created by the dataset were performed in order to obtain the chance of generating a matching matrix by chance.

Results

In total 985 catches of 693 individuals of *E. ferrugineus* and 99 of 95 individuals for *O. eremita* were recorded during the summer 2011 in the present study. The maximum number of catches for one site (= one trap) was 143 catches of 91 individuals of *E. ferrugineus* at Runstorp (Figure 6). For the different periods the catches for *E. ferrugineus* were in period A: 246 individuals (18/18 active traps catching), period B: 138 individuals (19/19 traps catching). For *O. eremita* catches in each period were: period A: 17 individuals (12/27 traps catching) and for period B: 22 individuals (14/28 traps catching).

Percent re-captures of total catch for *E. ferrugineus* was 29.6 % (ranging between 5.3-50 %) and for *O. eremita* 4.0 % (ranging between 0-50 %) (Figure 6). The probability of re-capturing *E. ferrugineus* was higher than re-capturing *O. eremita*. Since the pheromone for *O. eremita* works as a kairomone and also attracts females of *E. ferrugineus*, beetles caught in traps that were emptied by me were sex determined in order to see the proportion of females in the traps. Seven of the *E. ferrugineus* in these traps were females and the total number of beetles caught in these traps was 182 (i.e. 3.85 % females, none of which were recaptured). Movements between traps differed between the two species with many more and longer movements detected for *E. ferrugineus*. Forty-two *E. ferrugineus* beetles moved between two different traps, ranging in distance from 500 m-9 900 m. One *O. eremita* moved between two different traps in the present study; the distance between these two traps was 500 m.

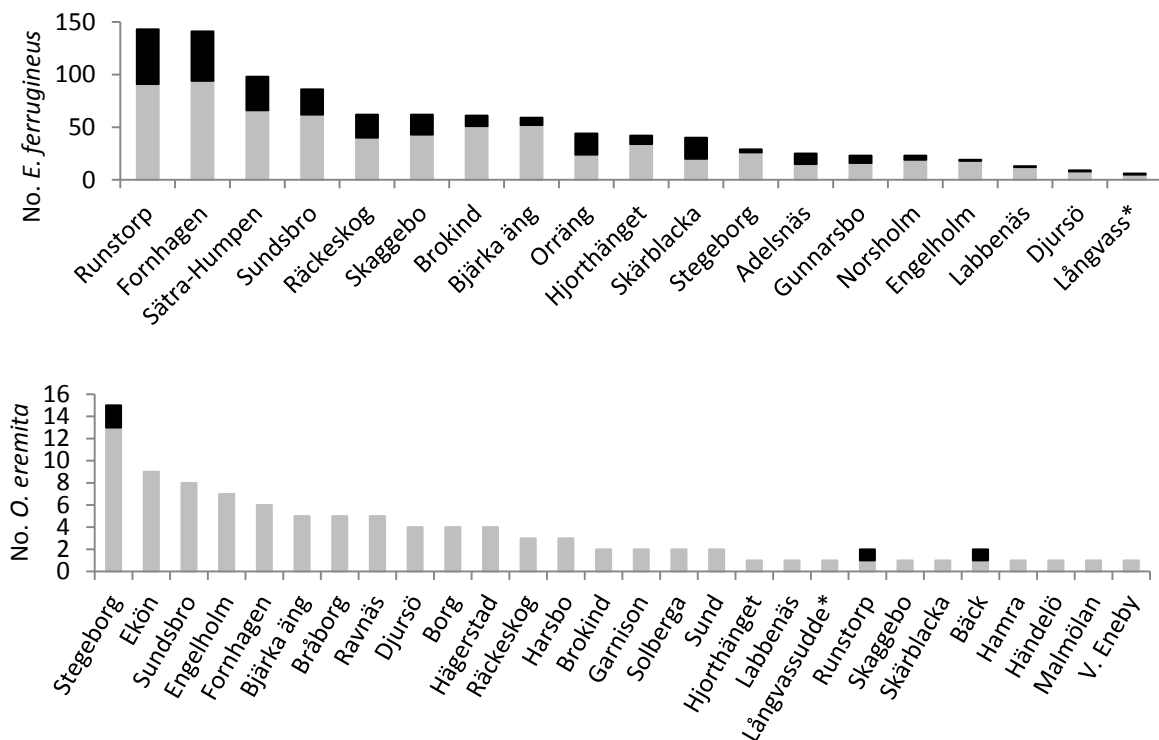


Figure 6. Graphs showing *E. ferrugineus* (upper) and *O. eremita* (lower) catches in pheromone traps. The number of unique individuals and re-captures are indicated with grey and black, respectively. *trapping at Långvassudde started 13 days after the others.

Comparing the results from Jansson's survey with the present study results revealed that *E. ferrugineus* was found at 19 sites in the present study; the five sites where *E. ferrugineus* was detected in the Jansson (2009) dataset were all included among the sites found in the present study. For *O. eremita* the results were more similar between the two surveys: 31 sites were found in the 1994-2010 surveys and 28 in 2011. The pheromone trapping was unable to detect *O. eremita* on five of its sites in Jansson's material from 1994-2010 surveys, but also detected this species at two sites where it was lacking in the data from 1994-2010 (Table 1 and Figure 7). Overall, the results for *E. ferrugineus* exhibit low convergence between different data sets, and demonstrate the added benefits of the pheromone in detecting its presence. In contrast, the three data sets for *O. eremita* display relatively high convergence, indicating a more balanced contribution from each data set. No entirely novel locality for *O. eremita* was detected in the present study, suggesting that few occupied sites remain undetected (Figure 7).

Table 1. The table shows presence/ absence of *Elater ferrugineus* and *Osmoderma eremita* on each site. x means that the species was only found in Janssons (2009) surveys, O means that it was only found during the 2011 survey, and ⊗ denotes that it was found in both surveys.

	<u>Elater ferrugineus</u>	<u>Osmoderma eremita</u>		<u>Elater ferrugineus</u>	<u>Osmoderma eremita</u>
Adelsnäs	○	x	Mauritzberg	-	-
Bjärka äng	⊗	⊗	Norrby sjön	-	-
Borensberg	-	-	Norsholm	○	x
Borg	-	⊗	Orräng	○	x
Brokind	⊗	⊗	Prästbola	-	-
Bråborg	-	⊗	Ravnäs	-	⊗
Bäck	-	○	Ruda	-	-
Börsjö	-	-	Runstorp	○	⊗
Disevid	-	-	Räckeskog	○	⊗
Djursö	⊗	⊗	Skaggebo	○	○
Ekön	-	⊗	Skärblacka	○	⊗
Engelholm	○	⊗	Solberga	-	⊗
Fornhagen	○	⊗	St.Löpgöl	-	-
Garnison	-	⊗	Stegeborg	⊗	⊗
Gunnarsbo	○	x	Sund	-	⊗
Hamra	-	⊗	Sundsbro	⊗	⊗
Harsbo	-	⊗	Sundsnäs	-	-
Hjorthänget	○	⊗	Svaltorp	-	-
Hägerstad	-	⊗	Sätra-Humpen	○	x
Händelö	-	⊗	Tisenhult	-	-
Labbenäs	○	⊗	Tolefors	-	-
Lagnebrunna	-	-	V Eneby	-	⊗
Långvassudde	○	⊗	Östantor	-	-
Malmölan	-	⊗			

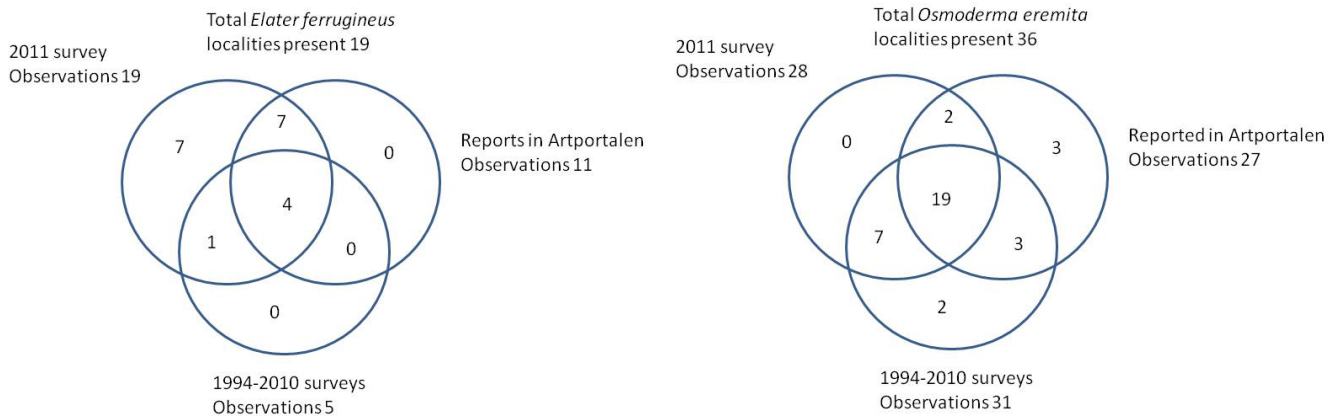


Figure 7. Venn diagrams comparing three different data sets containing occupied sites for the two model species *E. ferrugineus* and *O. eremita*. The two surveys used in this project (pheromone trapping in July-August 2011 and Jansson's pitfall/window trapping surveys 1994-2010), and the records from Artportalen (adjusted so that records included in Jansson (2009) are removed). Each circle represents a specific data set. When circles overlap it shows that these surveys all detected the presence of the species on the same site and vice versa. The upper left fields in the two diagrams show that the pheromone trapping in 2011 detected 7 previously unrecorded localities for *E. ferrugineus*, whereas no novel localities were detected for *O. eremita*. Data for Artportalen obtained 19/10-2011 from www.artportalen.se

Temporal catch pattern

Catches of *E. ferrugineus* were skewed towards the early part of the field season, with the highest number of new individuals being captured during the first week. A few monitoring traps placed at known localities with high density of *E. ferrugineus* prior to the field season ensured that the true beginning of the flight season was not missed, however. Catches remained relatively high during most of the field season, with most traps catching new individuals until the first week of August, after which only a few stray individuals appeared (Figure 8). In contrast, *O. eremita* catches exhibited no distinct peaks over the season, other than perhaps a brief initial series of captures and a subsequent skew in the catch towards the latter half of the season. The catch pattern of *O. eremita* suggested that the field season of this species was somewhat longer than that of *E. ferrugineus*. A suggested dip in the catches of both species after the first 10 days coincided with a brief spell of unusually cold, rainy weather (Anonymous 2011).

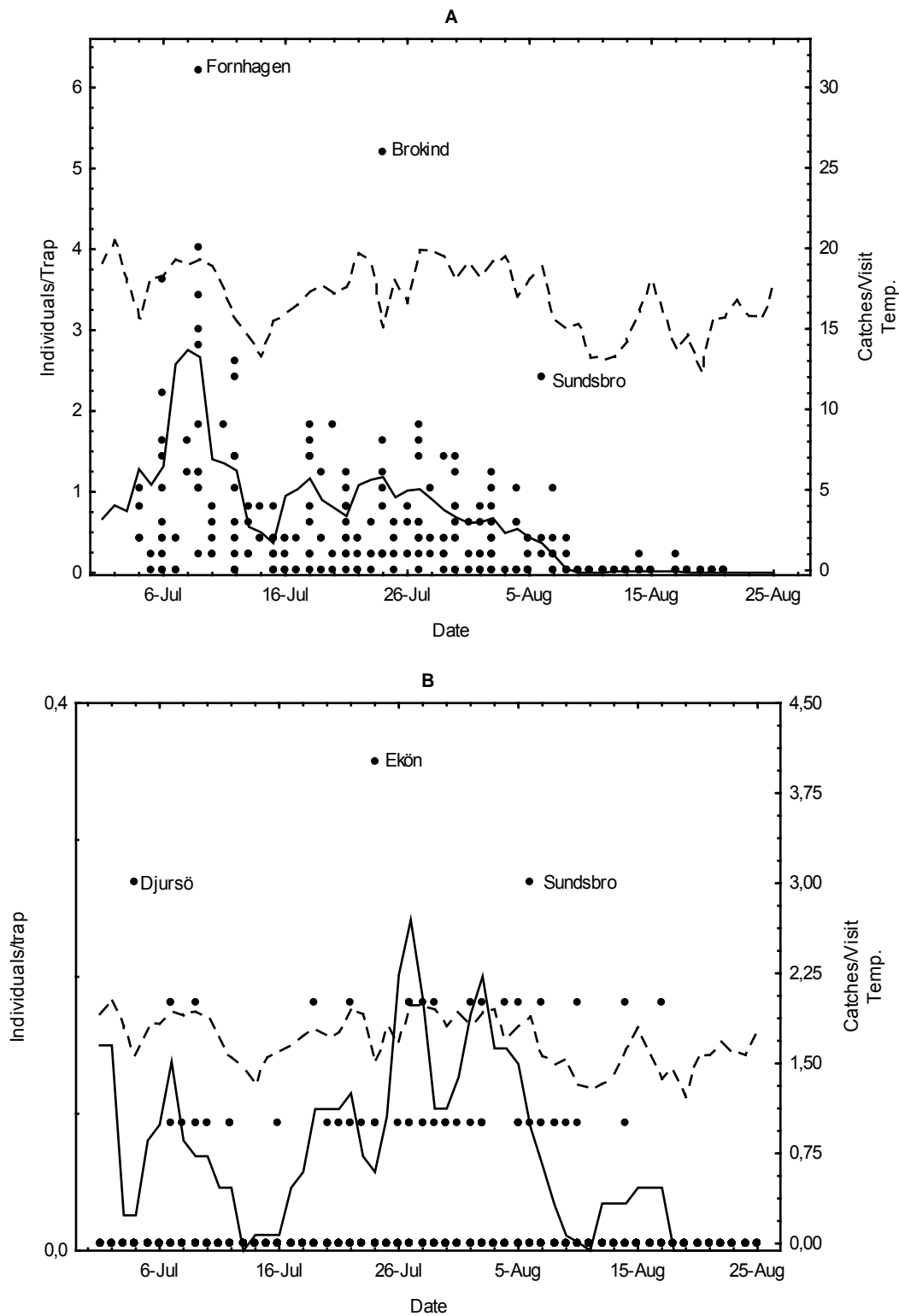


Figure 8. The temporal pattern of catches of new individuals per trap (dots, right y-axis) of (A) *E. ferrugineus* and (B) *O. eremita*. Solid lines denote average catches per day (left y-axis), dashed line is the mean day temperature at Malmslätt meteorological station (obtained from SMHI.se) (right y-axis, in the lower graph (B) the temperature is divided by 10). Note the differences in catch number and lack of distinct peaks for *O. eremita*. Note also that the individual points showing the highest catches in the two species all come from different sites in each species, suggesting that individual sites may exhibit peak densities at somewhat different times of the field season.

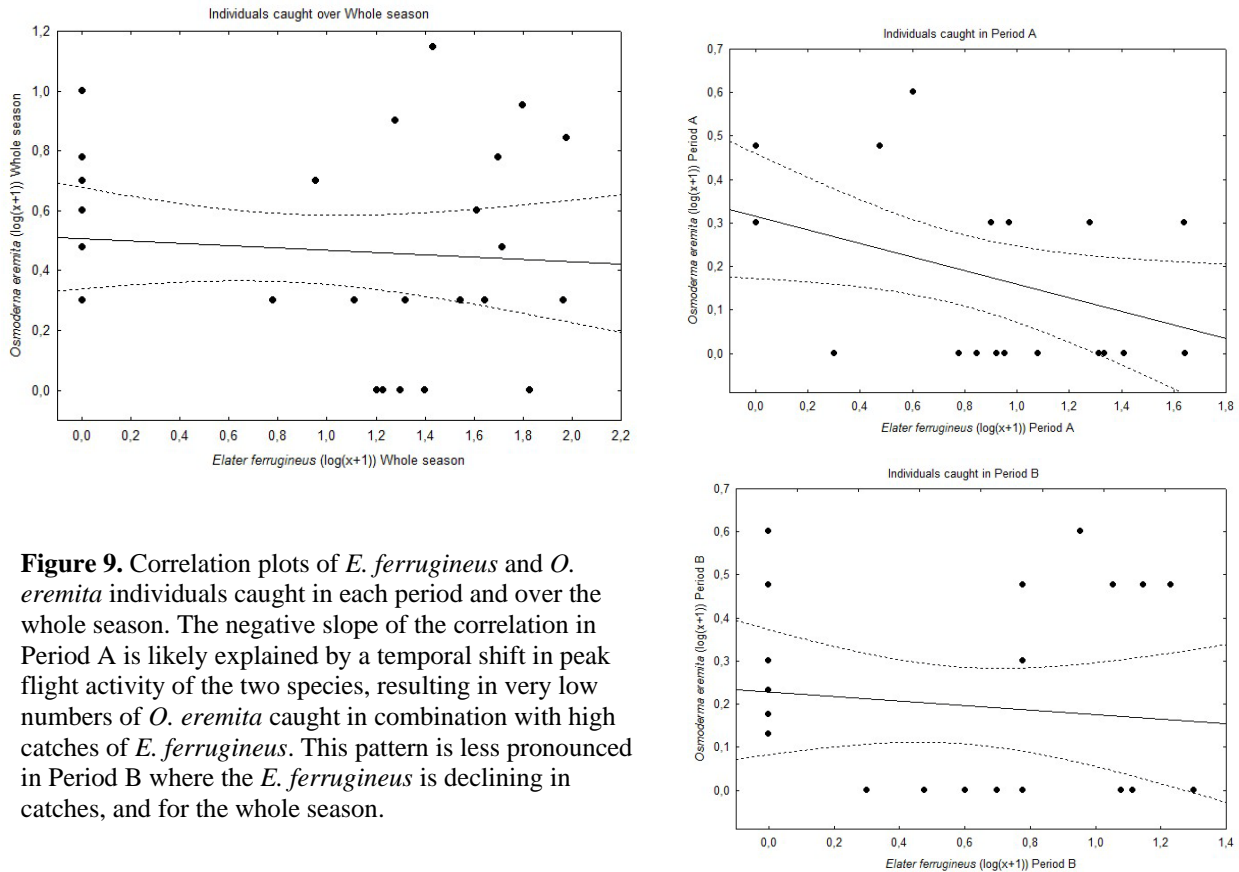


Figure 9. Correlation plots of *E. ferrugineus* and *O. eremita* individuals caught in each period and over the whole season. The negative slope of the correlation in Period A is likely explained by a temporal shift in peak flight activity of the two species, resulting in very low numbers of *O. eremita* caught in combination with high catches of *E. ferrugineus*. This pattern is less pronounced in Period B where the *E. ferrugineus* is declining in catches, and for the whole season.

GLZ (Generalized Linear models)

Correlation tests were carried out for variables used as predictors in the GLZ (period A & B and whole season). Comparisons between the *E. ferrugineus* and *O. eremita* abundance as measured by trap catch showed no strong correlations (r -values; period A: -0.43 ($n=23$), period B: -0.11 ($n=27$) and whole season: -0.095 ($n=33$) (see also Figure 9). The trend towards negative correlations, although not significant, is likely explained by the differences in peak flight activities by the two species, which resulted in high catches of *E. ferrugineus* in combination with low catches of *O. eremita*. Since the pattern is strongest in the sub-period A it may be explained by the differences in initial peak patterns of the two species with *E. ferrugineus* displaying a distinct peak at the beginning of the season where *O. eremita* lacks a peak (see Figure 8).

Abundance of *E. ferrugineus* and *O. eremita* were strong predictors for several variables that were used as indicators for the conservation value of a site. Figure 10 shows some examples of these relations, with *E. ferrugineus* and *O. eremita* individuals captured over the whole season plotted against four different indicators of conservation value (No. of species, No. of Red listed species, Red list points, and No. of Near Threatened species).

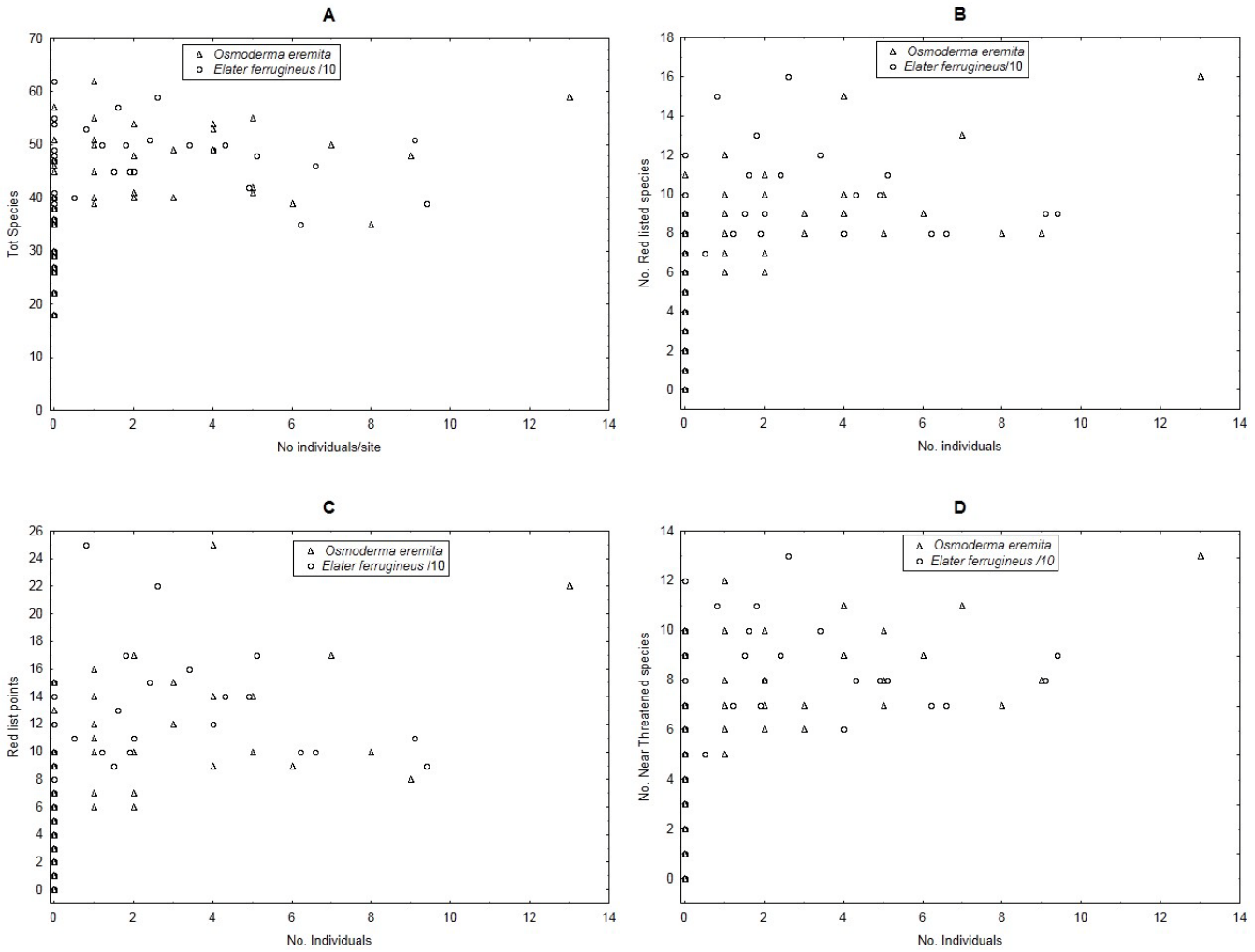


Figure 10. Scatterplots of the number of individuals caught over the whole season for both *E. ferrugineus* and *O. eremita* against several parameters used to indicate conservation values for individual sites; note that the catches of *E. ferrugineus* are divided by 10 in order to fit in the same plot as *O. eremita*. The graphs show the catch of *E. ferrugineus* and *O. eremita* plotted against (A) Total No. of species; (B) No. of Red listed species; (C) Red list points; (D) No. of Near Threatened species.

In each case, there was a rather abrupt threshold for the presence of *E. ferrugineus* or *O. eremita* at different sites. No individuals of either species were trapped at the sites displaying the lowest values for different conservation indicator variables.

In the GLZ analysis, *E. ferrugineus* abundance was the best predictor in four of the pairs of response and predictor variables: Red list point, VU+EN, and total species number vs. Period B, and VU+EN vs. Period A. In all other pairs the best describing model was a combination of the abundances of *E. ferrugineus* and *O. eremita*. When the catches over the whole season were used as predictors, the model always included the abundances of both *E. ferrugineus* and *O. eremita*. See Table 2 for details.

Table 2. GLZ (normal distribution log-link) results, continuous predictors in columns. Model 1: *E. ferrugineus* abundance (period x); model 2: *O. eremita* abundance (period x), (model 2 was tested but excluded since it was not the best explaining model in any case); model 3: Interactions of *E. ferrugineus* and *O. eremita* with intercept included in model.

Response	Predictor	Model	AIC	p-value	Intercept (CI)	Elater (CI)	Osmoderma (CI)
Tot. sp.	Period A	3	340.7	0.047	3.69 (3.61-3.78)	0.095 ((-0.0013)-0.20)	0.28 (-0.06)-0.62)
Tot. sp.	Period B	1	345.5	0.017	3.69 (3.61-3.78)	0.15 (0.018-0.29)	
Tot. sp.	Whole se.	3	334.4	0.002	3.64 (3.55-3.73)	0.069 ((-0.0072)-0.15)	0.22 (0.051-0.39)
R.l. sp.	Period A	3	237.5	0.00040	1.81 (1.63-1.98)	0.27 (0.089-0.46)	0.83 (0.27-1.39)
R.l. sp.	Period B	3	242.3	0.00048	1.82 (1.65-1.99)	0.39 (0.16-0.63)	0.42 ((-0.099)-0.94)
R.l. sp.	Whole se.	3	222.8	<0.0001	1.67 (1.50-1.85)	0.20 (0.08-0.33)	0.57 (0.31-0.84)
NT cat.	Period A	3	222.8	0.0015	1.74 (1.57-1.90)	0.21 (0.033-0.39)	0.80 (0.26-1.33)
NT cat.	Period B	3	228.3	0.0027	1.74 (1.58-1.90)	0.29 (0.062-0.52)	0.48 ((-0.028)-.98)
NT cat.	Whole se.	3	211.2	<0.0001	1.62 (1.45-1.79)	0.14 (0.014-0.26)	0.57 (0.30-0.83)
VU/EN cat.	Period A	1	126.4	0.0028	-0.69 (-1.28-(-0.11))	0.65 (0.14-1.16)	
VU/EN cat.	Period B	1	126.2	0.00039	-0.71 (-1.33-(-0.1))	1.03 (0.38-1.68)	
VU/EN cat.	Whole se.	3	116.8	<0.0001	-0.99 ((-1.65)-(-0.32))	0.64 (0.24-1.05)	0.080 ((-1.16)-1.32)
R.l.point	Period A	3	276.4	0.00064	1.96 (1.73-2.19)	0.34 (0.11-0.57)	0.91 (0.22-1.6)
R.l.point	Period B	1	280.4	0.00018	1.98 (1.76-2.19)	0.51 (0.22-0.80)	
R.l.point	Whole se.	3	261.3	<0.0001	1.80 (1.57-2.03)	0.28 (0.12-0.43)	0.61 (0.29-0.93)

Abbreviations used: tot. sp. = total number of species at a site; R.l. sp.: Red listed species found at a site; NT cat.: number of species categorized as Near Threatened at a site; VU/EN cat.: number of Vulnerable/Endangered species found at a site; R.l. point: Red list point

Weighted odds-ratios

Odds of finding any random species at a site with either *E. ferrugineus* or *O. eremita* compared to a site without either of these species were significantly higher ($OR_{E. ferrugineus}$: 1.53; $p < 0.0001$ and $OR_{O. eremita}$: 1.74; $p < 0.0001$). This indicates that sites with either *E. ferrugineus* or *O. eremita* are more species-rich since the chance of finding any random species included in this project was higher at sites with *E. ferrugineus* or *O. eremita*. Data for individual species are presented in Figure 11.

When categorizing individual species according to the red list of 2010 (see method part for categories), *E. ferrugineus* had a stronger connection to the most rare species (VU/EN) in this study (odds of finding a VU/EN species at a site with *E. ferrugineus* compared to a site without *E. ferrugineus*: 5.48; $p < 0.0001$). The corresponding odds ratio for *O. eremita* was not significant. Odds of finding species that are listed as NT in the red list of 2010 were greater for both *E. ferrugineus* and *O. eremita* ($OR_{E. fer.}$: 2.16; $OR_{O. ere.}$: 2.68; p -val.both < 0.0001). Beetles from the LC category were also significantly more frequently appearing when *E. ferrugineus* or *O. eremita* were present ($OR_{E. fer.}$: 1.53, p :0.047 and $OR_{O. ere.}$: 1.92, p : < 0.0001).

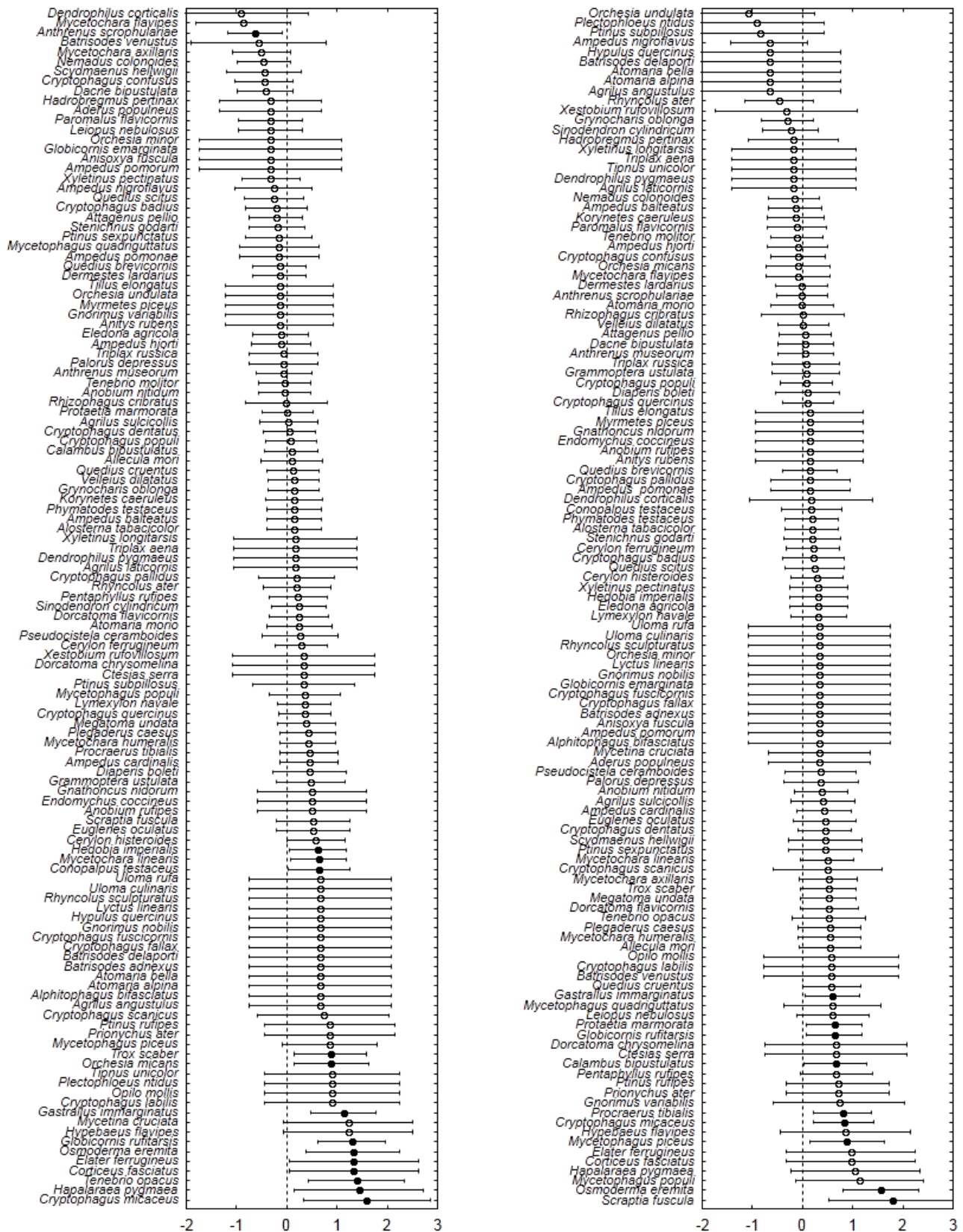


Figure 11. Graphs of logarithms of weighted odds ratio on presence/absence data on *E. ferrugineus* (left) and *O. eremita* (right) from 2011 vs. species presence/absence data from Jansson's (2009) surveys. Circles denote the odds ratio; filled circles indicate that the odds ratio is significant. 13 species occurred significantly (95% CI) more often when *E. ferrugineus* was present and one significantly more often when *E. ferrugineus* was absent. Two of the species that occurred more often were classified as VU and two as NT according to the Red list of 2010 (*E. ferrugineus* (VU) not included). Eight species occurred more often when *O. eremita* was present. Three of these were classified as NT according to the Red list 2010 (*O. eremita* (NT) not included). Only 47 traps were used in the present study, and many species were found at only few sites, resulting in rather large confidence intervals; nevertheless several significant co-occurrence patterns emerged.

Table 3. Weighted odds ratio for species that have a significantly higher probability of being found at a site with *E. ferrugineus* present (see fig. 11).

Species	OR	CI	Z-value	p-value	Red list category
<i>Conopalpus testaceus</i>	4.36	1.08-17.63	2.07	0.039	LC
<i>Corticeus fasciatus</i>	21.62	1.12-418.6	2.03	0.042	VU
<i>Cryptophagus micaceus</i>	39.00	2.15-708.8	2.48	0.0013	LC
<i>Elater ferrugineus</i> *	21.62	1.12-418.6	2.03	0.042	VU
<i>Gastrallus immarginatus</i>	13.33	3.03-58.62	3.43	0.00061	LC
<i>Globicornis rufitarsis</i>	19.56	4.24-90.15	3.81	0.00014	NT
<i>Hapalaraea pymaea</i>	27.44	1.44-523.5	2.2	0.028	LC
<i>Hedobia imperialis</i>	4.14	1.1-15.52	2.11	0.035	-
<i>Mycetochara linearis</i>	4.33	1.21-15.44	2.26	0.024	-
<i>Orchesia micans</i>	7.58	1.37-42.09	2.32	0.021	-
<i>Osmoderma eremita</i>	20.77	2.43-177.6	2.77	0.0056	NT
<i>Tenebrio opacus</i>	24.3	2.72-217.1	2.86	0.0043	VU
<i>Trox scaber</i>	7.37	1.43-38.08	2.38	0.017	-

*note that this is the species used as predictor in this case, but here the species is included based only on data from Jansson (2009).

Table 4. Weighted odds ratio for species that have a significantly higher probability of being found at a site with *O. eremita* present (see fig. 11).

Species	OR	CI	Z-value	p-value	Red list category
<i>Osmoderma eremita</i> *	36.40	6.24-212.4	3.99	<0.0001	NT
<i>Scryptia fuscula</i>	63.00	3.36-1180	2.77	0.0056	LC
<i>Procaerus tibialis</i>	6.33	1.68-23.88	2.72	0.0065	NT
<i>Cryptophagus micaceus</i>	6.67	1.66-26.76	2.68	0.0075	LC
<i>Mycetophagus piceus</i>	7.58	1.37-42.09	2.32	0.021	LC
<i>Globicornis rufitarsis</i>	4.33	1.21-15.44	2.26	0.024	NT
<i>Protaetia marmorata</i>	4.33	1.21-15.44	2.26	0.024	LC
<i>Gastrallus immarginatus</i>	3.90	1.13-13.45	2.15	0.031	LC
<i>Calambus bipustulatus</i>	4.62	1.10-19.50	2.08	0.037	NT

*note that this is the species used as predictor in this case, but here the species is included based only on data from Jansson (2009).

Thirteen species occurred significantly more often on sites with *E. ferrugineus* than without (Table 3), and one: *Anthrenus scrophulariae* (Dermestidae) occurred less frequently when *E. ferrugineus* was present (p-value: 0.022; z-value: -2.30; CI: 0.067-0.81). Four of the species that occurred more often with *E. ferrugineus*, excluding itself, are listed as least concern (LC), two as near threatened (NT) and two as vulnerable (VU) according to the Red list of 2010. Among the species that more often occurred at sites with *O. eremita*, excluding itself (Table 4), five are listed as least concern (LC) and three are listed as near threatened (NT) according to the Red list of 2010.

Accuracy

The accuracy for catching *E. ferrugineus* and *O. eremita* varied over the season 2011 (Figure 12). Traps that were set out from the beginning of July until late July reached 100 % accuracy within 6-9 days for *E. ferrugineus*. In contrast, traps for detection of *O. eremita* needed to be out from the beginning of July until early August in order to reach 100 %, whereas traps that were set out after 14th of July never obtained 100 % accuracy for *O. eremita*. For *O. eremita* the full accuracy with regards to all known sites would never be reached, since at five sites that were found 1994-2010 and another three from the Artportalen the pheromone traps failed to detect specimens. The true accuracy achieved for *O. eremita* compared to all known sites in this investigation therefore only corresponds to 78 %. Accuracy for *E. ferrugineus* was not affected by adding data from the surveys 1994-2010, as the latter included no additional sites.

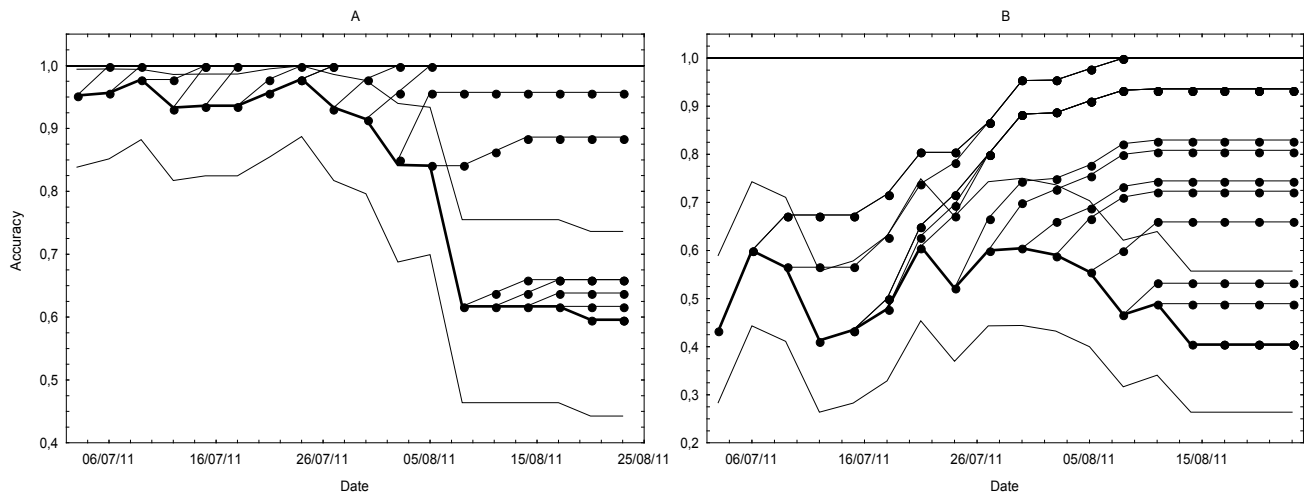


Figure 12. Accuracy graphs showing the overall agreement with the complete dataset of 2011: total number of sites with presence of *E. ferrugineus* (A) and *O. eremita* (B), based on pheromone trapping at different times during the field season.—Bold line denotes how the accuracy at each single visit (corresponding to three days of trapping) changes over the field season. The dotted lines around the bold line denote 95% CI. Thinner lines emerging from the bold line show the cumulative change in accuracy over time if trapping would have been started at that date and continued until end of the season. For example: if the trap is up on the 26th of July it will take only two visits before *E. ferrugineus* has reached an accuracy of 100%, whereas the accuracy for *O. eremita* will increase from 55% to about 80% accuracy over the next five visits and thereafter level out, as no catches were noted at new sites.

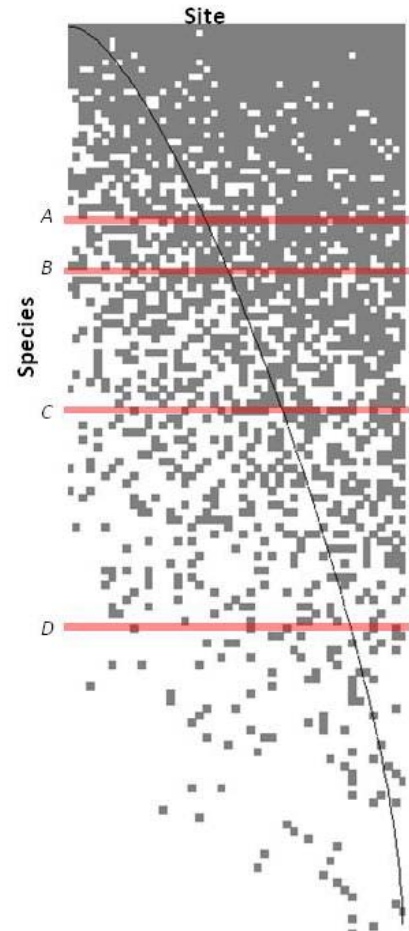
Nestedness analysis

The nestedness analysis showed that the saproxylic beetles had a nested pattern of occurrence among the sites that were used in this study, based on Jansson (2009). In the nestedness analysis I also included additional presence/absence data for *E. ferrugineus* and *O. eremita* from the present study as separate entries in the nestedness matrix, in order to determine the impact of detection method on the position of these two species in the nestedness hierarchy. The observed matrix (Figure 13) was filled to 34.5 % and had a temperature of 29.83 °, compared to the random matrices that had an average temperature of 77.98 ° (st. dev:1.7 °), which was significantly higher than the observed matrix ($p < 0.001$). Adding data from 2011 greatly affected the position of *E. ferrugineus* in the overall hierarchy, whereas that of *O. eremita* remained relatively unchanged. It is worth noting that *E. ferrugineus* appears to be perfectly nested within *O. eremita*, i.e. was found only at sites where *O. eremita* was present.

Discussion

This study showed that both model species were good indicators of the conservation value of individual localities. *O. eremita* has previously been shown to be a significant indicator for species richness of saproxylic beetles (Ranius 2002, Jansson et al. 2009). This study is the first to include both species, showing that *Elater ferrugineus* was a somewhat better indicator species for sites with very high conservation value. *E. ferrugineus* was shown to be more abundant in sites where many other rare saproxylic beetle species were found, and statistical models showed that the rarest saproxylic beetle species (EN/VU) were more frequently observed at sites occupied by *E. ferrugineus*. When looking at the co-occurrences of the two beetle species it is seen that *E. ferrugineus* is in no place present where *O. eremita* is absent (when adding the observations from Jansson, 2009). This is also apparent in the odds ratio diagrams, where *O. eremita* is significantly more often present on sites with *E. ferrugineus*

Figure 13. The presence of saproxylic beetles among sites found in the study of Jansson (2009), represented as a nestedness matrix calculated by a nestedness calculator (Atmar and Patterson 1993, 1995). Each row in the matrix represents one species and each column represents one site. A grey box means that the species is present on that site, and a white box means that the species is absent. Data from pheromone trapping have been added in order to give an illustration on their relationships with each other. **A:** *O. eremita*, Jansson (2009); **B:** *O. eremita*, present study; **C:** *E. ferrugineus*, present study; **D:** *E. ferrugineus*, Jansson 2009. In a perfectly nested community all filled boxes would be on the right side of the line. As the matrix is filled to less than 50%, the line has a concave appearance corresponding to the proportion of all squares that could potentially be filled. See also Figure 5.



but not the other way around (see Figure 11). This could indicate that *E. ferrugineus* is a beetle species with higher demands on the site than *O. eremita*, which in turn is believed to have relatively high site preferences (Ranius et al. 2005). Both the OR and GLZ models indicate that more of the rarest categories of beetle species appear more frequently where *E. ferrugineus* is present, but they are not significantly more common where *O. eremita* is present. Two species that are listed as VU and two NT occur significantly more often at sites where *E. ferrugineus* is found than sites without; for *O. eremita* four NT species were more frequently occurring in sites with *O. eremita*. This implies that sites where *E. ferrugineus* is found contain more rare species than those with *O. eremita*. However, it is important to note that a high number of species occur more often (but not significantly) with both *E. ferrugineus* and *O. eremita*, most easily seen in the odds ratios (Figure 11).

In the retrospective analysis of the different methods used in the present study, it is apparent that both the GLZ and OR analyses reveal the same pattern with *E. ferrugineus* being a somewhat better indicator for the most threatened saproxylic species. However, the necessary trapping effort was quite different between the two species; when using presence of *E. ferrugineus* much fewer trapping days were needed than for *O. eremita* in order to obtain a sufficient result. But it is also important to have in mind that the effort of using either of these, or preferably both is much lower than using more traditional methods such as pitfall and window-traps when performing surveys. The accuracy of *O. eremita* could presumably be increased by using more traps in the field in order to cover a larger area, since the added effort of emptying more pheromone-baited traps at a site is negligible.

The pattern with the more rare species co-occurring to a higher extent with *E. ferrugineus* indicates a nestedness pattern where *E. ferrugineus*, together with an array of other species, disappears earlier than *O. eremita* when moving toward less favorable conditions. It has been shown that *O. eremita* is dependent on the number of hollow oaks at a local scale, and that

comparatively few trees may sustain a local population of *O. eremita* (Ranius 2000). Since the two beetle species studied in the present investigation are believed to belong to different trophic levels, their sensitivity to fragmentation may be different (Holt 1993, 2002, Holt et al. 1999). In other studies where insects from different trophic levels have been studied in connection with the level of fragmentation (e.g. Davies et al. 2000) it has been shown that predatory species that decline have a higher risk of going extinct than species from lower trophic levels. The connection between spatial scale and trophic level is discussed by Holt (1993, 2002) where the author states that species higher in a food web have higher spatial demands in order to be able to persist in a fragmented landscape. Holt (1993) argues that if the area of the habitat is small and patchy the extinction risk of the predator is higher than for the prey. Species at higher trophic levels generally have smaller population sizes, making them more sensitive to stochastic fluctuations (see also Larsson and Svensson 2011). Patchier habitat distributions in the landscape could also make it difficult for the predator to track the prey, even if the amount of habitat would be enough to sustain a predator population (Holt 1993, Ranius 2000, Begon et al. 2006). The difficulties for the predator to track the prey may be further increased if the prey population decreases to a very low abundance (Begon et al. 2006). The area of suitable habitat at a larger spatial scale may therefore be more important for *E. ferrugineus* than for *O. eremita*. Bergman et al. (2012) showed that *O. eremita* is dependent on the oak density on a local scale (105 m). Ranius and colleagues (2011) predicted that *E. ferrugineus* is dependent on suitable trees on a much larger spatial scale than what has been reported for *O. eremita*.

For the concept of indicator species to be valid, species must not go extinct from individual sites in an entirely stochastic manner, which would make the presence or absence of individual species independent of each other. Ideally, species connected to a specific habitat should respond with different sensitivity to the same underlying factors leading to extinctions, causing a predictable hierarchy of presence and absence (See Figure 5 for a schematic representation of the concept). The nestedness analysis based on Jansson's (2009) data demonstrated that the saproxylic beetle species did indeed show a nested pattern of presence and absence, indicating a basically sound rationale for employing indicator species for conservation purposes among this guild of insects. The temperature of the nestedness matrix was 29.83 °, however, which could be seen as quite high for sites with many species tightly connected to the habitat, and therefore subject to quite uniform challenges to their persistence (Atmar & Patterson 1993, Eliasson 1999, Eliasson & Nilsson 2002). However, the relatively high temperature of the observed matrix is most likely heavily affected by the sampling method, by which approximately 60 % of the species remain undetected at each site (Jansson 2009). This underlying stochasticity is bound to raise the estimated temperature of the matrix, and a higher precision in the sampling of the species at each site would thus be needed for a relevant estimate of the true nestedness temperature. The nestedness pattern of saproxylic beetles shown here is otherwise consistent with the nestedness patterns demonstrated for butterflies in the Bornean rainforest after forest fragmentation in order to make room for oil-palm plantations, where some species seemed to disappear at certain levels of forest fragmentation in all fragments while others seemed to be more resistant (Benedick et al 2006). This pattern may resemble the fragmentation of broad leaf deciduous forests with hollow trees

in Sweden, characterized by gradual fragmentation of a previously widespread habitat with long continuity.

The saproxylic survey data set from Jansson (2009) represents a massive effort, and is very unusual in its scope and depth, combined with its stringent methodology. Nevertheless, it does not by itself suffice to determine relative extinction hierarchies and indicator status of saproxylic species. Because of the overall degree of uncertainty associated with the presence of individual species, the relative position of any species in the hierarchy could depend more on its probability of detection than its underlying dynamics of extinction and colonization. This is exemplified quite well by *E. ferrugineus* itself, which constituted a grossly under-sampled species with only 26 % detection of its present number of localities. For any array of saproxylic species that can be sampled with high accuracy, however, the Jansson (2009) data set constitutes an excellent framework for statistical evaluation of their utility as indicators. When including data from the present study, the presence of both *O. eremita* and *E. ferrugineus* among the investigated sites probably is quite near their true pattern of occupancy. For *O. eremita*, three more or less independent data sets display high convergence with regards to the identity of occupied sites, suggesting that few sites remain to be found. For *E. ferrugineus*, the documented capture probability of the sex pheromone for monitoring virtually guarantees detection of a resident population. The pattern of rarer species correlating with *E. ferrugineus* more often than *O. eremita* strongly suggests that *E. ferrugineus* is an indicator for sites that either are less fragmented or are less affected by extinction debts and should therefore have special attention (Benedick et al. 2006, extinction debt reviewed by Kuussaari et al. 2009). *E. ferrugineus* is also perfectly nested within *O. eremita* among the surveyed sites, since none of the sites that showed presence of *E. ferrugineus* lacked *O. eremita* (when data from Jansson (2009) was included), but 14 sites with *O. eremita* lacked *E. ferrugineus*. This pattern, and the considerable distance remaining between the two species in the overall nestedness hierarchy, further strengthens a nested community structure in the saproxylic beetle fauna in hollow oak areas. It provides further evidence that *E. ferrugineus* is dependent on the density of oaks on a larger spatial scale and thus disappears earlier in a fragmented landscape than *O. eremita* (see Figures 5, 11, 13). This could partly reflect that *E. ferrugineus* is directly affected by the number of trees with *O. eremita* – which is probably one of its most important prey species – but also that the former species is more sensitive to general processes of habitat dynamics affecting all species associated with hollow trees.

Further studies in order to estimate the dispersal capacity of *E. ferrugineus* males and females could shed light on whether *E. ferrugineus* is a species that has sex-linked differences in the ability of dispersal; something that was proposed for *O. eremita* by Dubois et al (2010). They showed under laboratory conditions that *O. eremita* are capable of flying up to 2 361 m (Dubois et. al 2010), which is much longer than most of the distances reported from field conditions; 180 m recorded by Ranius & Hedin (2001); 700 m recorded by Dubois & Vignon (2008); 500 m recorded by Svensson et al. (2011a); longest recorded flight in the present study for *O. eremita* was also 500 m. However, 1500 m was reported from Italy by Chiari (2011) which indicate that *O. eremita* has the potential to make longer dispersals, even though dispersals over 500 m are seldom reported. Larsson & Svensson (2011) reported strong

effects of temperature on dispersal propensity, which may explain the longer flights noted from Italian populations. Assessing the dispersal ability of these beetles is necessary in order to evaluate and make predictions for dispersal success into restored hollow oak areas. Pheromone-based systems would constitute a unique way of studying dispersal of beetles over a large spatial scale with mark-recapture procedures. Knowledge of the species dispersal capacities is essential for calculating the connectivity which is necessary for estimating metapopulation persistence over time (reviewed in Hanski 1998, Hanski et. al 1996).

Surveys by means of pheromone-baited funnel traps proved to be an accurate method for detection of both *E. ferrugineus* and *Osmoderma eremita*. When comparing *E. ferrugineus* data collected this year with the data collected by Jansson (2009) it could be seen that *E. ferrugineus* distribution in the county of Östergötland, Sweden had been underestimated. When comparing data from the present study with what was reported into the Species Gateway it was seen that these data collected during the summer 2011 increased the known sites with *E. ferrugineus* in the county of Östergötland with seven new sites (see Figure 7). The increase in the number of known sites for *E. ferrugineus* confirms that this species has been under-sampled in earlier surveys with other methods, which was proposed by Jansson (2009) and Nilsson and Baranowski (1994). Adding the present study's data to the nestedness graph it can be seen that *E. ferrugineus* changes position quite substantially where *O. eremita* remains almost at the same position, which further illustrates that *E. ferrugineus* is a species sensitive to sample method relative to *O. eremita*.

In terms of pheromone based monitoring, *O. eremita* was less abundant in the traps than *E. ferrugineus* – this is something that has been repeatedly shown in sampling from previous years (Larsson & Svensson 2009, 2011, Svensson et al. 2011a, Larsson et al *unpubl.*). The recapture rates and recapture distances, which were considerably higher for *E. ferrugineus* than *O. eremita*, indicate that differences in trap catch reflect fundamental differences in sampling range between the *O. eremita* pheromone trapping system and that of *E. ferrugineus* (Schlyter 1992, Östrand & Anderbrant 2003). This could be due to several different factors, including differences in attraction radius of the pheromone traps, and that *O. eremita* appears to be more sedentary than *E. ferrugineus* (Ranius & Hedin 2001, Larsson & Svensson 2009, 2011). In order to be able to split these concepts apart more studies are needed; it is difficult to disentangle these terms from studies conducted without proper control traps and direct behavioural observations (Schlyter 1992, Östrand et al. 2000, Byers 2009). Release-recapture experiments with pine sawflies *Neodiprion sertifer* have shown that the sampling range (= the greatest potential distances between release points and capture points) can be considerable greater than the active range of the traps (= the distances at which insects are able to perceive the pheromone and perform directed flights towards the traps) (Östrand et al. 2000, 2001, Östrand & Anderbrant 2003). Movements of the beetles are an indication of the more active flight habits of *E. ferrugineus* males (the longest movements of *E. ferrugineus* in the present study was 9 900 m where the longest movement for *O. eremita* was about 500 m). *O. eremita* has been shown to be highly sedentary with only 15 % of the adults dispersing from its natal tree. Swedish *O. eremita* spend most of their adult lives inside tree hollows, inaccessible to pheromone traps, and the longest recorded movements are approximately 500 m, consistent

with results from the present study (Ranius & Hedin 2001, Larsson & Svensson 2009, 2011, Svensson et al. 2011a). Dispersal flights for *E. ferrugineus* males in the present study are consistent with studies from previous years, where male individuals made movements over 10 000 m, (Larsson et al *unpubl.*).

Since the *E. ferrugineus* males are capable of moving great distances, there is a risk that a site may catch a beetle that is not from a local population, which may lead to misinterpretation of catches. The beetle may have flown in from an adjacent site where there is a resident population of *E. ferrugineus*. In the present study a number of beetles did fly in from adjacent populations (as shown by mark-recapture), but the total catch in every individual trap in the present study is likely too high to consist of only migrants that have flown in from adjacent areas. Only two traps caught less than 10 individuals of *E. ferrugineus* over the whole season. One of these sites (Långvassudde, five individuals) was up later than the others and thereby missed the peak of beetles due to lack of clearance to the land from the land owner (see Figures 6, 8). The other trap (Djursö, eight individuals) was placed very close to the water margin (Baltic Sea), where prevailing winds may have negatively affected the catch.

It could be important that each site where catches are recorded is evaluated so that no site is recorded as having a resident population of *E. ferrugineus* when it is a “tourist” (long flyer) that has been caught. To avoid misinterpretation, sites where the total catch is ambiguous could be subjected to extended trap effort. For *O. eremita* the risk of catching long-distance dispersers is negligible, as these beetles move very short distances (Svensson et al. 2011a).

The use of only one baited trap per site during a whole season proved to be sufficient to indicate presence of *O. eremita* in most cases. When evaluating the results from previous studies, *O. eremita* had been found at 31 sites compared with the 28 sites found with the present pheromone traps; five previous sites were not found 2011, however two new sites of *O. eremita* were detected in this study when compared to the data collected by Jansson (2009). This implies that *O. eremita* probably is generally over-sampled relative to other species when using pitfall and window traps, where the pitfall traps placed inside the hollow trees usually have a high probability of catching *O. eremita*, probably due to its sedentary ecology (Ranius 2001, Ranius & Nilsson 1997). Traps were placed according to a fixed centroid position 2011, rather than based on field cues such as large hollow trees, which would probably be applied if it were to be used as a method to survey sites with high conservation value.

When using trapping methods more adapted to field surveys (shorter periods than whole season), full accuracy was reached for *E. ferrugineus* within 6-9 days after activation as long as the traps were set out in July. For *O. eremita* the accuracy drastically decreased and could only reach 100 %, relative to the maximum number of detected sites in this study, if the traps were placed early in July and left out until mid August. This probably has much to do with the way that the traps were placed in field and that every site only had one trap. When using this method in the field my suggestion is to space out 3-5 traps and set one trap in the center of the area and the others more strategically chosen.

In order to be able to rank individual sites based upon the results from pheromone trapping more indicator species would be desirable. One suggestion could be the clearwing *Synanthedon vespiformis*, for which the pheromone is already determined. This species is considered as a pest species in most of Europe, this because it bores into the wood of stone fruit trees and other deciduous trees. The pheromone for this species has been identified in order to control this pest species in fruit orchards (Levi-Zada et al. 2011). But in Sweden *Synanthedon vespiformis* is believed to have a narrower host range with oak (*Quercus robur*) as its primary host tree and it is considered as vulnerable (VU) in the red list of 2010 (Gärdenfors 2010), which could make this pheromone applicable for conservation purposes in Sweden. Pheromones for further potential indicator species are lacking, but many of these species likely use sex or aggregation pheromones; the list could therefore be extended further. By using multiple traps baited with different pheromones, it would be possible to provide more fine-grained ranking of the sites according to the presence of more species besides *E. ferrugineus* and *O. eremita*, which are associated with the most valuable sites. The use of pheromone-baited traps in conservation would considerably increase accuracy while simultaneously reducing the costs of surveying large areas in order to find sites of interest for conservation methods. If the time assigned to field work is limited, this method would constitute a very good tool for decision makers. Especially since only one trap per site in one week is enough to obtain satisfying results.

Conclusions

The present study was carried out in some of the finest hollow oak areas to be found in Sweden, and the results from this study needs to be interpreted with that in mind. This study shows that both *Elater ferrugineus* and *Osmoderma eremita*, when using pheromone baited trapping, are highly useful as indicator species for rare species associated with deciduous hollow tree areas in Sweden. Many of the rarest species associated with hollow oak stands were present more often when either *E. ferrugineus* or *O. eremita* was present, with the former having a more tight connection with the most vulnerable species according to the red list of 2010 (Gärdenfors 2010). When evaluating the conservation value of individual localities, the sites housing *E. ferrugineus* should receive the highest priority.

Implications for management

Surveys by means of pheromone-baited funnel traps would constitute a very useful tool for conservation purposes both in order to locate sites with high species richness as well as monitoring dispersals and populations of rare and threatened species. Use of pheromone baited traps makes it possible to perform large surveys with relative low effort, as the high efficiency and species specificity makes interpretation of the catches fast and easy to carry out in the field.

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