Flattening the curve: approaching complete sampling for diverse beetle communities

RYAN C. BURNER,1* TONE BIRKEMOE,1 JENS ÅSTRÖM2 and ANNE SVERDRUP-THYGESON1
1Faculty of Environmental Sciences and Natural Resource Management, Norwegian University of Life Sciences, Ås, Norway and 2Department of Terrestrial Ecology, Norwegian Institute for Nature Research (NINA), Trondheim, Norway

Abstract. 1. Insects are a hyper diverse and ecologically important group. Their high diversity, however, presents challenges in sampling methodology, because rare species are unreliably detected with low sampling effort. However, the relationship between effort and species detections, critical for effective monitoring and evaluation of population trends, is too seldom quantified.

2. We sampled forest beetles for 3 months in a 4-ha stand of mixed deciduous forest in southeastern Norway using 110 flight intercept (four types) and Malaise traps, the highest trap density (29 traps ha−1) that we have seen reported. We examined species accumulation curves to quantify the benefits of each additional trap, compared capture rates among several trap designs and trap emptying frequencies, and tested for spatial autocorrelation.

3. In total, we captured 566 beetle taxa (19 854 individuals) from 52 families, yet our species accumulation curve was only beginning to flatten. Trap types differed considerably in their effectiveness. Nevertheless, 20 of our most effective window traps detected 75% of all taxa in our dataset. We found no evidence of spatial correlation within the scale of the study (100 m radius), nor did trap-level forest covariates (5 m radius) explain much variation.

4. This implies that low-to-moderate sampling effort dramatically underestimates species richness, but that a limited number of effective traps can nonetheless achieve relatively thorough sampling for some applications. Immediate trap surroundings and spacing appeared unimportant. Insect ecologists should take particular care in selecting trap types, and be cautious comparing studies that employed different trap types.

Key words. Bayesian joint species distribution models (JSDMs), Coleoptera, flight intercept window traps, HMSC, Malaise traps, sampling effort, spatial autocorrelation, species accumulation curves.

Introduction

Insects are a hyper diverse and ecologically important group, providing a number of ecosystem services (Noriega et al., 2018). Their diversity, numbers, and sheer biomass make them a fascinating and important topic of study (Bar-On et al., 2018), but this same diversity also presents challenges as we try to understand the ecology of various taxonomic and functional groups and the roles they play in different environmental contexts. Ecological research on all but the most common species must typically rely on observational studies, often consisting of little more than records of the locations and habitats in which species have been detected (Ovaskainen et al., 2019).

Making inferences about ecological processes, species habitat requirements, and biotic interactions from distributional data is challenging, especially for hyper diverse groups of organisms. When sampling intensity is low, a large proportion of species remain undetected (‘false negatives’) at each site (Martikainen & Kouki, 2003). The power to make inferences from observational studies under
these conditions is typically low (Kellner & Swihart, 2014). This is a particular challenge because most communities contain a high proportion of rare species (Stork, 1988; Novotný & Basset, 2000; Cunningham & Lindenmayer, 2005).

Here, we focus on the challenges of sampling beetle communities, a notoriously species rich group (Stokland et al., 2012; Ulyshen, 2018), in forests. Sampling beetles at a single site typically involves one or more traps, placed randomly or targeting microhabitats of interest, and deployed for several weeks or months when the insects are most active. Relatively little information is available quantifying species’ detection rates and sampling adequacy, but there is evidence that typical sampling effort misses many species (Martikainen & Kouki, 2003; Martikainen & Kaila, 2004; Hedgren & Wessel, 2008).

Boreal forests in the Nordic countries are home to a diverse beetle fauna. Our sampling at over 500 sites (one to five traps per site) in Norwegian boreal forests over the past two decades has detected approximately 70 species (95% CI 15–160; 1450 total species) per site (Sverdrup-Thygeson & Ims, 2002; Birkemoe & Sverdrup-Thygeson, 2015; Sverdrup-Thygeson et al., 2017; Burner et al., 2021b), likely a small fraction of the true species richness. Sampling across the boreal zone that sample beetles using a handful of traps per site have often found only weak effects of environmental covariates at a variety of scales (Similä et al., 2002; Fossestøl & Sverdrup-Thygeson, 2009; Sverdrup-Thygeson et al., 2014a; Jacobsen et al., 2015, 2020). This is likely due in part to the complex (and perhaps random) processes that contribute to beetle community assembly and movement patterns and to the difficulty of choosing environmental variables that describe the most relevant features of habitats. Yet, to the extent that typical sampling protocols are detecting only a small proportion of the species inhabiting a given site, the difficulty of explaining and predicting beetle communities is likely also due to the noisy data resulting from large numbers of false negatives (Driscoll, 2010).

The proportion of species detected at a site increases with increasing sampling intensity, but limited budgets mean that allocating additional sampling effort per site reduces the number of sites that can be sampled, which can limit a study’s explanatory power. Sampling effort is also subject to diminishing returns, making it important to quantify the relationship between effort and detection to optimise sampling protocols (Driscoll, 2010). Sampling efficacy, however, is a function not only of effort but also of method. The effectiveness of different trap designs is known to differ (Siitonen, 1994; Hyvärinen et al., 2006; Missa et al., 2009; Burner et al., 2020), but researchers too seldom quantify these differences when comparing data collected using varying methods. There are also logistical choices to be made in distributing sampling effort – traps should only be deployed as long and emptied as frequently as is necessary, because both require time and effort that might otherwise be used to sample additional sites. The increasing use of DNA-based methods for species identification also places stricter restrictions on the amount of rainwater that can be tolerated in traps, pushing a demand for shorter intervals between trap emptying events (Aström et al., 2020). It is therefore important to quantify when and how often emptying should occur.

Additionally, traps at a site must be adequately spaced to provide independent samples, but spatial autocorrelation appears to vary by scale and among groups of beetles (Blanchet et al., 2013; Horak, 2013; Steinke et al., 2021). This is an important methodological consideration, but quantifying autocorrelation patterns can also help to understand the spatial scales at which environmental filtering may structure beetle communities, a persistent question that has received much attention (Bergman et al., 2012; Sverdrup-Thygeson et al., 2014b; Jacobsen et al., 2015). Researchers frequently try to explain insect capture rates using environmental variables collected at different spatial scales, and there is evidence that covariates from the immediate trap location (Missa et al., 2009; Sverdrup-Thygeson et al., 2014b; Burner et al., 2021b) as well as at the scale of several kilometres (Sverdrup-Thygeson & Lindenmayer, 2003; Sverdrup-Thygeson et al., 2014b; Rubene et al., 2017) can influence which species are captured at a site, although these remain open and important questions. But how important are small-scale differences in habitat within a single forest stand? If within-stand environmental covariates or autocorrelation influence species distributions, then getting a representative sample from a forest stand will be challenging.

Researchers typically test the relevance of environmental covariates at different spatial scales by comparing the explanatory effects of these covariates (Holland et al., 2004). However, the challenge with this approach, particularly for a group like forest beetles where most covariates typically have low explanatory power, is that weak effects might indicate poor covariate choice rather than inappropriate spatial scale. Examining spatial autocorrelation patterns, however, can provide clues to the relevance of various spatial scales (De Knegt et al., 2010), unconfounded by potentially flawed covariate choices. The presence of autocorrelation does not demonstrate conclusively that environmental variation is responsible (Václavík et al., 2012) – it could also be caused by, e.g., dispersal or other behavioural factors (depending on the scale) – but the absence of any autocorrelation at a given scale should provide evidence that it is not a relevant scale of environmental variation, assuming detection probabilities are consistent and high enough to detect adequate signal (Mc New & Handel, 2015). Spatial detection patterns are further complicated in taxa that, like beetles, have several life stages (larvae, flying adults) that differ in their habitat, behaviour, nutrition, and mobility (Harvey et al., 2011), with only adults entering the traps.

Research to better understand insect communities is thus predicated on having adequate sampling effort and effective sampling strategies. Understanding patterns of spatial autocorrelation, necessary in formulating sampling methods, can also be informed by the often frustrating search for meaningful covariates at different spatial scales (Sverdrup-Thygeson et al., 2014b). Yet, sampling-related questions are often addressed using inferences from large-scale studies with sparse sampling, rather than being rigorously tested. To determine how to optimise beetle sampling strategies, we conducted intensive high-density beetle sampling in a stand of mixed deciduous forest in southeastern Norway. We asked:

1. How much sampling is required to detect most species in a community?
2. How do several trap types, including those designed to divert rainwater, compare in their capture rates?
Is small-scale spatial autocorrelation an important consideration, and what does this reveal about the importance of forest covariates?

**Materials and methods**

**Study area**

We sampled beetles from a 3.8 ha stand of mixed deciduous forest in southeastern Norway (Fig. 1) (Burner et al., 2021a). The stand was dominated primarily by European beech (*Fagus sylvatica*), aspen (*Populus tremula*), birch (*Betula spp.*), linden (*Tilia spp.*), oaks (*Quercus spp.*), Scots pine (*Pinus sylvestris*), spruce (*Picea spp.*), and several other less numerous deciduous species. This stand is on university property and last had trees harvested 69 years ago, although in some parts trees have not been removed for 103 years (Veidahl et al., 2017). Many large diameter trees are found there, including many beeches >75 cm in diameter, and there is much fallen dead wood of various ages and sizes. The presence of many large diameter deciduous trees makes this a somewhat atypical site for Norwegian forests. The site is in a mixed agricultural and wooded landscape.

**Beetle sampling**

To compare the performance of several trap designs and trap emptying protocols, we deployed 110 traps within our study area from 26 May to 27 August 2020. These included 10 Malaise traps (Bugdorm, Taiwan), 20 IBL-2 flight intercept ‘window’ traps (CHEMIPAN, Warsaw, Poland) with a device used to divert rainwater (Burner et al., 2020), and 80 custom cross-pane flight intercept traps (Fig. 2). Sixty out of the 80 total cross-pane flight intercept traps were fitted with a polyvinyl chloride (PVC) elbow and mesh screen (1 mm² mesh size) in the downspout meant to prevent rainwater from entering the collection bottle where it dilutes preservation liquid, which could lead to degradation that affects both morphological and DNA metabarcoding identification techniques. The triangular IBL-2 trap has a capture surface of 3950 cm², and the cross-pane traps have two panes totalling 1600 cm².

Collection vials were covered with foil (to prevent UV damage to insects) and filled with 85% ethanol in the Malaise traps, and with a 70:30 mixture of propylene glycol and ethanol (95%) in the flight intercept traps. Malaise traps were set with the base touching the ground. Flight intercept traps were set with the base of the trap window roughly 1 m above the ground. Traps were hung between two trees and were 0.5 m to 2 m from these

![Figure 1. Map of beetle sampling locations. Inset shows location in southeastern Norway. Colour of points signifies trap and treatment type (Table 1). Created using 'ggmap' R-package (Kahle & Wickham, 2013).](image-url)
trees. The top funnel of each of the cross-pane flight intercept traps was also fitted with a 750 ml collection container, with an entry hole located in the centre of the top funnel. Although these top containers did capture some insects (e.g. Diptera spp.), they captured almost no beetles and we stored their contents separately and excluded them from our analyses for this reason.

We used six trap/treatment combinations (Table 1), which differed in the frequency at which they were emptied and refilled, and (for the cross-pane trap design) in whether they included the elbow and mesh screen meant to allow rainwater to escape rather than entering the beetle collection bottles. After the designated time interval, trap contents were collected and frozen and the preservative liquid was replaced. Beetles were then identified morphologically by an expert taxonomist (Sindre Ligaard, independent consultant) to species (where possible) or genus. Taxonomy follows the GBIF taxonomic backbone (GBIF Secretariat, 2021), which is based on the Catalogue of Life checklist (Bánki et al., 2021).

Traps were placed in 20 clusters in our study area (Fig. 1). Each cluster consisted of one trap each from our five flight intercept trap treatments (see Table 1). Additionally, each even-numbered cluster included a Malaise trap. Clusters were spaced between 10 and 50 m apart and were in areas open enough for unimpeded insect flight. Within each cluster, traps were placed roughly in a line or circle (depending on availability of sites for hanging), with 1–15 m between traps and varying the order of the trap types. Our randomly varied spacing within and among clusters meant that pairwise distances between traps were well distributed from 1 to 160 m (Fig. S1). Coordinates for each trap are in the Supporting Information Table S1.

To describe the habitat around each trap and cluster, we collected information on several environmental covariates (Table S1). Within a 5 m radius of each trap, we counted deciduous and coniferous trees (> = 10 cm diameter). We also visually estimated a relative sun exposure index for each trap on a

Table 1. Trap treatments for comparison of beetle trap effectiveness in mixed forest. Malaise and several types of flight intercept (F.I.) traps were emptied at different intervals and tested with and without a rainwater excluding device. Treatments beginning with ‘W’ signify flight intercept (‘window’) traps. For trap types see Fig. 2.

| Treatment | Trap type          | Rainwater diversion device? | Number of traps | Empty interval (weeks) |
|-----------|--------------------|-----------------------------|-----------------|------------------------|
| Ma        | Malaise            | -                           | 10              | 4                      |
| W-IBL     | IBL-2 F.I.         | Yes                         | 20              | 4                      |
| Wx1       | Cross-pane F.I.    | No                          | 20              | 4                      |
| Wx2       | Cross-pane F.I.    | Yes                         | 20              | 4                      |
| Wx3       | Cross-pane F.I.    | Yes                         | 20              | 8 + 4*                 |
| Wx4       | Cross-pane F.I.    | Yes                         | 20              | 12                     |

*Traps emptied after 8 weeks were deployed again for the final 4 weeks of the study.
scale of 1 (shaded) to 5 (sun exposed) by looking towards the sun through the forest canopy facing southward from each trap on a single sunny day between 1100 and 1300 h. Finally, we measured the total volume of standing and lying dead wood (≥ 10 cm diameter) within 5 m of each trap. Each of these covariates has been linked to beetle occurrences (Seibold et al., 2016; Vogel et al., 2020; Burnet et al., 2021b). We chose the scale of 5 m to examine trap-level small-scale variation because this minimised the overlap among adjacent traps. We also obtained estimated monthly treatments for our trapping period from the 1 km resolution ERAS-Land climate reanalysis (Copernicus Climate Change Service (C3S), 2017).

Analyses

To compare capture rates among different trap types, emptying frequencies, and trap periods, we generated species accumulation curves by sequentially adding traps in a random order (500 replicates) in R (R Core Team, 2021). We did this with all traps combined and also separated traps by treatment and season to compare. When comparing among trap treatments we merged capture records from all trap periods for each trap (i.e., for those traps emptied several times). When comparing capture rates among the three 4-week trap periods of the season, however, we compared only those traps emptied every 4 weeks and separated capture data by trap period. We estimated the total number of species present at our site using the iChao1 estimator, based on the total count of individuals captured of each species (Chiu et al., 2014).

To test for spatial autocorrelation in capture patterns, and to determine the extent to which small-scale habitat covariates could explain capture rates, we used the Bayesian joint species distribution model (JSDM) framework from the ‘Hmsc’ R-package (Tikhonov et al., 2020). We used four models (Table 2) to answer four related questions. To test for spatial autocorrelation in species richness and total beetle abundance at the scale of individual traps (pooled across trap period), we fit a model with a lognormal Poisson error distribution and log link function to each of the trap period datasets with species richness and total abundance as response variables. We included trap treatment as a categorical fixed effect, with treatments X2, X3, and X4 merged into a single category because these traps had very similar capture rates (see Results). We also included a spatially explicit random effect of site, which allowed us to test for spatial signal in the residuals for each trap site and estimate the spatial scale at which that signal was relevant, based on an exponential covariance function (Ovaskainen & Abrego, 2020). Any spatial signal thus detected is ‘residual’ in the sense that it includes only spatial autocorrelation in species distributions that is not explained by the fixed effect covariates.

Additionally, we fitted a similar model independently to each of 149 beetle species detected in 10 or more traps (species with enough locations to test for spatial signal). The response variable in this case was presence/absence of a given species by trap, fit using a probit distribution. Finally, we fitted a model to the presence/absence dataset of 253 species detected in five or more traps (species with enough locations to test for effects of covariates). This model was like the previous model except that it included our environmental covariates and a random effect of cluster rather than site (due to a lack of spatial signal in the previous model – see Results). All models were fitted with default non-informative priors and using initial values for the fixed effects estimated from a simple linear model. For each model, we used three Markov chain Monte Carlo (MCMC) chains, each run for 125 000 iterations, with the first 75 000 discarded as burn-in. One thousand samples were drawn from the remaining iterations at thinning intervals of 50. Model convergence was evaluated by examining effective sample size (all >1000) and scale reduction factors (all <1.1).

Results

We detected 558 beetle species (19 854 individuals) from 60 families, as well as eight taxa that could be identified only to genus (Table S2). These included 22 near threatened, four vulnerable, and four endangered species (160 individuals in total), according to the Norwegian red list classification (Henriksen & Hilmo, 2015). Most species (74%) were detected in fewer than 10 of our 110 traps, and 146 species (26%) were detected in only a single trap (Fig. S2). When considering total abundance, most species were represented by fewer than 10 individuals (66%), and we detected only a single individual of 136 species (24%). The iChao1 estimate predicted that actual species richness at our site was 714 (95% CI: 681–756). Of 30 red-listed species, 11 were detected in only a single trap, 9 in two traps, and the remaining 10 species were detected in 3 to 22 traps.

| Response variable | Distribution | Fixed effects | Random effects |
|-------------------|--------------|---------------|---------------|
| Species richness  | Lognormal Poisson | Trap type     | Site (spatial) |
| Total abundance   | Lognormal Poisson | Trap type     | Site (spatial) |
| Occupancy (149 spp.)* | Probit       | Trap type     | Site (spatial) |
| Occupancy (253 spp.)** | Probit       | Trap type, environmental covariates† | Cluster |

*Species distributions were fitted independently for each species detected in ≥ 10 traps (n = 149) to examine spatial autocorrelation in each species.
**Joint species distribution model included all species detected in ≥ 5 traps (n = 253) to test for effects of environmental covariates.
†Environmental covariates included sun exposure, number of deciduous trees (≥ 10 cm diameter), number of coniferous trees (≥ 10 cm diameter), and log-transformed volume of coarse woody debris (≥ 10 cm diameter), all within a 5 m radius of each trap.

© 2021 The Authors. Insect Conservation and Diversity published by John Wiley & Sons Ltd on behalf of Royal Entomological Society, Insect Conservation and Diversity, 15, 157–167
Traps differed considerably in the number of species and individuals that they caught (Fig. 3); IBL-2 flight intercept traps and Malaise traps had more individuals and species than the cross-pane traps, and cross-pane traps without the water diversion device (‘X1’) performed better than traps with the device (‘X2’, ‘X3’, and ‘X4’). We did not, however, observe evidence of different overall capture rates among the cross-pane traps with water diversion devices (‘X2’, ‘X3’, and ‘X4’), whether emptied at 4, 8 (+ 4), or 12-week intervals (P > 0.88). The water diversion devices did, however, reduce the amount of water entering capture bottles when traps were checked at 4-week intervals (P < 0.05; Fig. S3). We found no evidence that the number of species identified from a trap was related to the amount of liquid in that trap at the time of emptying (F_{1.177} = 0.82, P = 0.37).

Differences in overall patterns of species accumulation among the trap types mirrored the relative richness of species caught by each (Fig. 4). The 20 IBL-2 flight intercept traps captured 442 species, which was about 2.6 times more than the 145–200 species caught by 20 cross-pane traps (95% confidence interval). This difference in capture rates was roughly proportional to the difference in capture surface area (2.5x).

---

Figure 3. Species richness and total abundance of beetles captured per trap in different trap treatments in southeastern Norway. Different letter codes (A–D) show which traps differ according to a Tukey post hoc test (P < 0.001). Traps include cross-pane flight intercept traps (‘Wx1-4’), Malaise traps (‘Ma’), and IBL-2 (‘W-IBL’) flight intercept traps. Total richness and abundance are summed across the entire trapping period. Boxes show the 25th, 50th (median), and 75th percentiles, and whiskers extend to the furthest data point within 1.5 times the interquartile range from the edge of the box. Circles mark additional data points outside that range. For trap and treatment details and sample sizes, see Fig. 2 and Table 1.

Figure 4. Species accumulation curves for several trap types used to sample beetles in southeastern Norway. On the left, each of three trap designs is compared to total species accumulation of all traps for the entire season (a). On the right, cross-pane window traps emptied at different intervals, and with and without a rainwater exclusion device, are compared. Coloured regions represent 50% and 95% confidence intervals (CIs), based on 500 simulations. IBL-2 flight intercept traps captured the most species, followed by Malaise traps and smaller cross-pane ‘X’ flight intercept traps (a). Traps without the rainwater device caught more species than traps with the device, whatever the trap emptying frequency (b). For trap and treatment details, see Fig. 2 and Table 1.

© 2021 The Authors. Insect Conservation and Diversity published by John Wiley & Sons Ltd on behalf of Royal Entomological Society.
between the IBL-2 and cross-pane traps. Yet, a total of 80 cross-pane traps (the surface area equivalent of 32 IBL-2 traps) captured only 307 species. Among the different cross-pane trap treatments, 20 traps without the rainwater diversion device captured 220 species, whereas an equal number of traps with the device captured only 132–144 species (regardless of trap emptying frequency).

Traps emptied in late June captured 83% of the total species captured by all 4-week traps (Malaise, IBL-2, X1, and X2) across the entire season (Fig. 5). The July trapping period captured 60% of the total species (including new additions totalling 11% of all species). Although 47% of total species were detected in August, only 4% of total species were new additions to the species list from this month.

Our joint species distribution model showed that the small-scale (5 m radius) environmental covariates we included had little effect. This model explained 2%–76% (mean = 30%; Tjur $R^2$) of the variation in presence/absence of each species, but fewer than 10% of the 253 species in this model were correlated with one or more environmental covariate (95% Bayesian credible interval; Table S3). Instead, variance partitioning revealed that most (79%) of the model’s explanatory power was attributed to the trap type covariate, with 3%–6% each for the four environmental covariates and 6% for the random effect of trap cluster (Fig. S4).

Differences in capture rates among window trap types were consistent among the 249 beetle species included in this model. Compared to Wx1 traps (no water device), 59% of beetle species were captured less often in cross-pane traps with the device (Wx2/3/4), and no species were captured more often (95% support; Fig. S5). The larger IBL-2 traps captured 74% of species more often than the Wx1, and only 2% of species were captured less often. However, the comparison with Malaise traps was more mixed. Malaise traps performed better than Wx1 traps for 21% of beetle species but worse for 21% and did not differ from the Wx1 traps for the remaining 58% of species.

We found no evidence of spatial autocorrelation at the scale of this study; species detected in fewer than 10 and 15 clusters were on average captured in only 1.1 and 1.3 traps per cluster, respectively, which did not differ from the random expectation (Fig. S6). When we fitted a species distribution model separately for each of the 149 most common species, including trap type as a fixed effect and a spatially explicit random effect of site, there was no strong evidence for spatial autocorrelation in any species; in estimates of the relevant scale of autocorrelation, 95% credible intervals spanned most or all the range of between-trap distances and included zero (Fig. S7). The same was true for models of species richness and total beetle abundance.

**Discussion**

We sampled a stand of mixed deciduous forest in southeastern Norway with the highest level of sampling intensity (29 traps ha$^{-1}$) that we have seen reported. We detected at least 558 distinct species, 16% of all beetle species found in Norway and a high number for 3 months of sampling in such a small area (<4 ha). Indeed, even with such intensive sampling, our species accumulation curve appeared to be only beginning to flatten and true richness at our site was estimated to be 20%–34% higher using iChao1 (Chiu et al., 2014), and perhaps higher still, given that some subset of species will not be susceptible to trapping with our methods and timing. Our mixed deciduous forest study site likely provides more diverse habitat than much of the spruce-
dominated managed forest in Scandinavia, but nonetheless our findings indicate that typical beetle sampling only detects a small fraction of the species occurring in a site. This demonstrates that estimating species richness is difficult, even with intensive sampling (Wikars et al., 2005). Consequently, analyses that require reliable observation of species in the long tail of the abundance distribution will be very costly.

However, differences in common species, or in community metrics like richness and overall abundance, may be detectable through space and time with more feasible sampling efforts. Our most effective trap type captured most species using only 20 traps (Fig. 4). But, inferences for the rare species that we seldom observe will be difficult indeed (Martikainen & Kaila, 2004; Engen et al., 2008; Guillera-Arroita, 2017), including for those red-listed species that are difficult to observe (Martikainen & Kouki, 2003). Some apparently rare species in a given study may also simply represent ‘tourist’ species that are only passing through that particular habitat (Gaston et al., 1993).

Different trap types capture different numbers of species (as is expected), but species accumulation curves from these traps can nonetheless appear to flatten at different levels (Fig. 4); if we had only sampled beetles in this stand using the cross-pane flight intercept traps, we would have estimated that the total number of species present was probably less than 400. This implies that there is a filtering effect on the type of species that the various trap types capture, capping the potential observed species richness at different levels. Our JSDMs revealed that the IBL-2 flight intercept traps performed better than the cross-pane traps for most beetle species, but the effectiveness of Malaise traps clearly differs among species. A variety of methods are thus needed to sample beetles effectively. It is also important to note that our methods targeted flying adult beetles, but the inclusion of larval sampling, pitfall traps, beating and fogging would likely have produced additional species (Allison et al., 1993; Basset et al., 1997; Driscoll, 2010).

Because capture rates can vary considerably among trap types, even when the difference in design is as small as e.g. (in our case) the presence or absence of a rainwater diversion device (Burner et al., 2020), it is also important to use consistent methods among years and sites. Trap effectiveness can also vary with species traits (Burner et al., 2020), which also influence species distributions (Burner et al., 2021). These sources of variability mean that if methods must change during a study, or when results from several studies are going to be merged, it is important to have at least some data comparing sampling methods to understand the effects of these methods (Økland, 1996).

Studies that aim to understand any but the most common and easily trapped beetles are likely limited by incomplete sampling. This is a feature of many inventories of diverse organisms. Many locally rare species, which are the hardest to sample, are likely to be geographically rare as well (Burner et al., In Revision), meaning that neither intense sampling at a few sites nor sparser sampling in many sites is likely to be optimum for rare species (Martikainen & Kouki, 2003; Cunningham & Lindenmayer, 2005). This highlights the tradeoffs between sampling ‘width’ and ‘depth’, and the optimum for any given study will thus depend on both the objectives and the relative costs. If the species of interest have patchy and ephemeral distributions, one might sample more locations, and if the species is hard to detect, one might look more carefully at each location (Tyre et al., 2003). Care should be exercised when making conservation choices based on a comparison of species lists among forest types unless sampling has been intense.

Our findings are not entirely bad news for beetle ecologists, however. Trap emptying frequency does not appear to be very important (unless DNA identification methods will be used; Åström et al., 2020), reducing time and financial burdens of sampling. The lack of spatial autocorrelation within this stand suggests that environmental variation at the scale of tens of metres is also relatively unimportant. This means that selection and spacing of trap sites within a stand are relatively unimportant and unlikely to bias results. Regarding the timing of sampling, June was the most important month for beetles of the 3 months we studied (at least for our region, and under the climate conditions experienced in 2020). Later months add progressively fewer new species, meaning that shorter sampling seasons would have relatively little impact if the peak period was covered. However, we no doubt missed a minority of species that have their activity peaks earlier in spring or later in the autumn. Also, small-scale (5 m) environmental covariates appear to be relatively unimportant. It is possible, however, that we did not detect small-scale effects of covariates because our trap sites were relatively homogeneous. Sun exposure of trap sites (Missa et al., 2009; Seibold et al., 2016; Vindstad et al., 2020), nearby tree species (Sverdrup-Thygeson & Birkemoe, 2009; Burner et al., 2021b) and amount of dead wood (Jacobsen et al., 2015; Müller et al., 2020) have been shown to be important in beetle and other insect studies. Sun exposure in particular was relatively homogeneous among our traps, with 79% of sites scoring either two or three in our one to five index.

The primary limitation of our study is of course that we sampled only a single forest stand in a single season. This means that we cannot yet assess how generalizable our results are, nor can we address the importance of forest stand or landscape-level covariates or spatial autocorrelation above stand scale (Blanchet et al., 2013; Rubene et al., 2017). Nevertheless, our conclusions about autocorrelation and (especially) sampling intensity are likely to hold in many study systems.

We have demonstrated that within-stand covariates are unlikely to determine the distributions of beetles within a stand, perhaps because most species are relatively mobile and not limited by distance at that scale. The scale of our study, however, does not let us address the relative importance of stand and landscape level covariates in determining beetle communities. Nor could we address temporal autocorrelation given the duration of our study. An important next step in understanding beetle sampling would be to expand this intensive sampling effort to a larger number of diverse forest stands in a landscape context (Sverdrup-Thygeson et al., 2014b).

Based on our results, then, we suggest that typical levels of sampling effort result in many false negatives. Given the variation in trap effectiveness, a consistent mix of trap types might be advisable. Switching trap types among sites or years should
be avoided, and when combining data from multiple trap designs a comparison of trap catches under equal conditions should be made. Traps placed relatively closely together appear to provide independent samples from a beetle community, and variation in catches between micro-sites within a relatively homogenous area thus can likely often be attributed to chance.

Acknowledgements

This research was funded by the Norwegian Environment Directorate as part of an ‘Agreement on monitoring hollow oaks and insects in hollow oaks’. The Norwegian University of Life Sciences (NMBU) workshop designed and produced the cross-pane flight intercept traps. Thanks to Sindre Liggard for identifying the beetle species and to Lindsay Burner, Ruben Roos, and Ross Wetherbee for assistance in the field. High-performance computing resources were provided by Frederick H. Sheldon and Louisiana State University (LSU HPC).

Conflict of Interest

The authors have no conflict of interest to declare.

Author Contributions

R.B.: Conceptualization, Methodology, Data collection, Formal analysis, Writing – Original draft; T.B.: Conceptualization, Methodology, Data collection, Writing – review and editing; J.A.: Methodology, Writing – Review and editing; A.S.-T.: Conceptualization, Methodology, Data collection, Funding acquisition, Writing – Review and editing.

Data Availability Statement

Data are available on Zenodo: https://doi.org/10.5281/zenodo.5544556.

Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Pairwise distances among all traps for beetle sampling

Figure S2. Distribution of sites (traps) occupied (left) and total abundance (right) of 566 beetle taxa trapped in southeastern Norway. Four species with a total abundance >550 (679–1414) are excluded from the total abundance histogram.

Figure S3. Capture bottle fluid % fill at time of emptying flight intercept traps (boxplots). All bottles were originally filled to 30% with a mix of ethanol and propylene glycol, and additional liquid means that rainwater entered the traps. All traps were deployed for 12 weeks; text above each group of plots shows frequency of trap emptying. All treatments shown except ‘Wx1’ were equipped with a mesh screen device meant to divert rainwater from traps. Blue dots (scale at right) show cumulative precipitation at the trap site during the trapping period for each trap. Different letter codes (A-B) show results of Tukey pairwise significance test between traps for fluid fill values (p < 0.05). Precipitation estimates come from the ERA5-Land climate reanalysis (Copernicus Climate Change Service (C3S), 2017). For trap and treatments see Fig. 2 and Table 1.

Figure S4. Variance partitioning from HMSC Bayesian joint species distribution model (JSDM) with 253 beetle species captured in southeastern Norway. Each vertical bar represents one species. Trap design explained most (79.3%) of the variance in the model. Covariates included trap design, several environmental covariates (see legend and Table 2), and a random effect of trap cluster. Legend shows mean proportion of variance explained by each covariate.

Figure S5. Relationship between trap type (x-axis) and probability of capturing 253 beetle species (y-axis). Values represent the estimated regression coefficients (95% support) from an HMSC Bayesian joint species distribution model. These coefficients are relative to the intercept category, Wx1. Wx2/3/4 traps, which unlike Wx1 have a device meant to divert rainwater, performed less well for most species. The larger W-IBL traps performed much better for most species. Malaise traps, however, performed much better for some species but much worse for others. For more details on model and trap design see Table 2 and Fig. 2.

Figure S6. Comparison of number of traps and number of clusters occupied by 566 beetle taxa in southeastern Norway. Clusters of traps (ca. 20 m radius) consisted of 5–6 traps, with 2–5 m between adjacent traps and 5–25 m between adjacent clusters. Each point represents one beetle species. The grey ribbon represents a null model simulation (95% CI), in which traps were randomly assigned to clusters 1000 times. The blue dotted line represents a scenario where captures are maximally spatially overdispersed relative to the null model, meaning that a species on that line is captured in exactly one trap per cluster. All clusters were in a single 4-ha stand of mixed deciduous forest. Species present in fewer than 10 and 15 clusters were detected by 1.1 and 1.3 traps per cluster on average, respectively. Almost all points fall within the grey ribbon, indicating that beetles are no more or less clustered than expected by chance, and that spatial autocorrelation in captures is not present. For a complementary test of spatial autocorrelation, see Fig. S7.

Figure S7. Estimated scale of spatial autocorrelation in 149 beetle species, as well as in species richness and total abundance patterns, based on 110 traps placed in a forest stand in southeastern Norway. All 95% Bayesian credible intervals, estimated by a spatially explicit random effect of trap site in species distribution models, spanned nearly the full range of between-trap distances, similar to the model priors for this effect. This indicates a lack of spatial signal in the capture data. For a complementary test of autocorrelation, see Fig. S6.

Table S1. Locations and surroundings of 110 beetle traps deployed in twenty clusters in southeastern Norway. Each covariate was measured in a 5 m radius around the trap.

Table S2. List of beetle taxa detected in flight intercept and Malaise traps in southeastern Norway. Beetles were identified
morphologically by an expert taxonomist (Sindre Ligaard, independent consultant). Taxonomy follows the GBIF taxonomic backbone (GBIF Secretariat, 2021), which is based on the Catalogue of Life checklist (Bánki et al., 2021).

Table S3. Number of beetle species responding positively and negatively (95% support) to four environmental covariates in joint species distribution models. Out of 253 total species. Covariates were measured within a 5 m radius of each trap. For model description see methods and Table 2.

References

Allison, A., Samuelson, G.A. & Miller, S.E. (1993) Patterns of beetle species diversity in New Guinea rain forest as revealed by canopy fogging: preliminary findings. Selbyana, 14, 16–20.

Áström, J., Birkemoe, T., Dahlø, S., Davey, M., Ekrem, T., Endrestøl, A., Fossay, F., Nystad Handberg, Ø., Hansen, O., Magnusen, K., Majanen, M.A.M., Navrud, S., Staverløkk, A., Sverdrup-Thygeson, A. & Ødegaard, F. (2020) Forslag til nasjonal insektovervakning - Erfaringer fra et pilotforsøk samt en nyt-kostnadsanalyse. NINA Rapport 1725. Norsk institutt for naturforskning, Trondheim.

Bánki, O., Roskov, Y., Vandepitte, L., DeWalt, R.E., Remsen, D., Schalk, P., Orrell, T., Keping, M., Miller, J., Aalbu, R., Adlard, R., Adriaensens, E., Aedo, C., Aescht, E., Akkari, N., Alonso-Zarazaga, M.A., Alvarez, B., Alvarez, F. & Anderson, G. (2021) Catalogue of life checklist (Version 2021-09-21). Catalogue of Life. https://doi.org/10.48580/d4sv.

Bar-On, Y.M., Phillips, R. & Milo, R. (2018) The biomass distribution on Earth. Proceedings of the National Academy of Sciences, 115, 6506–6511.

Basset, Y., Springate, N.D., Aberlenc, H.P. & Delvare, G. (1997) A Workshop on acidic and neutral rainwater: a critical review. Atmospheric Environment, 31, 135–141.

Birkemoe, T. & Sverdrup-Thygeson, A. (2021a) Near-natural forests harbor richer saproxylic oak beetles. Forest Ecology and Management, 466, 118124.

Burner, R.C., Birkemoe, T. & Sverdrup-Thygeson, A. (2021b) Examining the relative importance of forest and landscape scale factors on beetle diversity in boreal forest. Forest Ecology and Management, 487, 119023.

Burner, R.C., Stephan, J.G., Drag, L., Muller, J., Ovaskainen, O., Potter, M., Siitonen, J., Skarpaas, O., Doerfler, I., Gossner, M.M., Schall, P., Weisser, W.W. & Sverdrup-Thygeson, A. (2021) Traits mediate environmental responses and species associations of forest beetles in ways that differ among bioclimatic regions. Journal of Biogeography. https://doi.org/10.1111/jbi.14272

Burner, R.C., Stephan, J., Birkemoe, T., Wetherbee, R., Muller, J., Siitonen, J., Snall, T., Skarpaas, O., Potter, M., Doerfler, I., Gossner, M.M., Schall, P., Weisser, W. & Sverdrup-Thygeson, A. & (In Revision) Functional structure of European forest beetle communities is enhanced by rare species. Biological Conservation.

Chiu, C.-H., Wang, Y.-T., Walther, B.A. & Chao, A. (2014) An improved nonparametric lower bound of species richness via a modified good-turing frequency formula. Biometrics, 70, 671–682.

Copernicus Climate Change Service (C3S) (2017) ERA5: Fifth generation of ECMWF atmospheric reanalyses of the global climate. Copernicus Climate Change Service Data Store (CDS).

Cunningham, R.B. & Lindemayer, D.B. (2005) Modeling count data of rare species: some statistical issues. Ecology, 86, 1135–1142.

De Knecht, H.J., Van Langevelde, F., Coughenour, M.B., Skidmore, A.K., De Boer, W.F., Heitkönig, I.M.A., Knox, N.M., Slotow, R., Van Der Waal, C. & Prins, H.H.T. (2010) Spatial autocorrelation and the scaling of species–environment relationships. Ecology, 91, 2455–2465.

Driscoll, D.A. (2010) Few beetle species can be detected with 95% confidence using pitfall traps. Austral Ecology, 35, 13–23.

Engen, S., Sæther, B.E., Sverdrup-Thygeson, A., Grøtan, V. & Ødegaard, F. (2008) Assessment of species diversity from species abundance distributions at different localities. Oikos, 117, 738–748.

Fossetti, K.O. & Sverdrup-Thygeson, A. (2009) Saproxylic beetles in high stumps and residual downed wood on clear-cuts and in forest edges. Scandinavian Journal of Forest Research, 24, 403–416.

Gaston, K.J., Blackburn, T.M., Hammond, P.M. & Stork, N.E. (1993) Relationship between abundance and body size: where do tourists fit? Ecological Entomology, 18, 310–314.

GBIF Secretariat (2021) GBIF Backbone Taxonomy. Checklist dataset. https://doi.org/10.15468/39omei.

Guilleria-Arroita, G. (2017) Modelling of species distributions, range dynamics and communities under imperfect detection: advances, challenges and opportunities. Ecography, 40, 281–295.

Harvey, D.J., Hawes, C.J., Gange, A.C., Finch, P., Chesmore, D. & Farr, I. (2011) Development of non-invasive monitoring methods for larvae and adults of the stag beetle, Lucanus cervus. Insect Conservation and Diversity, 4, 4–14.

Hedgren, O. & Weslien, J. (2008) Detecting rare species with random or subjective sampling: a case study of red-listed saproxylic beetles in boreal Sweden. Conservation Biology, 22, 212–215.

Henriksen, S. & Helgum, O. (2015) The 2015 Norwegian Red List for Species. Norwegian Biodiversity Information Centre, Norway.

Holland, J.D., Bert, D.G. & Fahrig, L. (2004) Determining the spatial scale of species’ response to habitat. BioScience, 54, 227.

Horak, J. (2013) Effect of site level environmental variables, spatial autocorrelation and sampling intensity on arthropod communities in an ancient temperate lowland woodland area. PLoS One, 8, e81541.

Hyvärinen, E., Kouki, J. & Martikainen, P. (2006) A comparison of three trapping methods used to survey forest-dwelling Coleoptera. European Journal of Entomology, 103, 397–407.

Jacobsen, R.M., Burner, R.C., Olsen, S.L., Skarpaas, O. & Sverdrup-Thygeson, A. (2020) Near-natural forests harbor richer saproxylic beetle communities than those in intensively managed forests. Forest Ecology and Management, 466, 118124.

Jacobsen, R.M., Sverdrup-Thygeson, A. & Birkemoe, T. (2015) Scale-specific responses of saproxylic beetles: combining dead wood surveys with data from satellite imagery. Journal of Insect Conservation, 19, 1053–1062.

Kahle, D. & Wickham, H. (2013) ggmap: spatial visualization with ggplot2. The R Journal, 5, 144–161.

© 2021 The Authors. Insect Conservation and Diversity published by John Wiley & Sons Ltd on behalf of Royal Entomological Society., Insect Conservation and Diversity, 15, 157–167
