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Optimal sample and plot size for inventory of field and ground layer vegetation in a mature *Myrtillus*-type boreal spruce forest

Jyrki Jalonen, Ilkka Vanha-Majamaa & Tiina Tonteri

Jalonen, J., Vanha-Majamaa, I. & Tonteri, T., Finnish Forest Research Institute, P.O. Box 18, FIN-01301 Vantaa, Finland

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Field and ground layer vegetation in a mature Norway spruce (*Picea abies* L.) forest of *Myrtillus*-type in Kuorevesi, southern Finland, was sampled on 31 plots of sizes 0.5, 1 and 2 m^2 to find optimal balance among sampling effort, number of observed species, and accuracy of mean species-cover estimates. Rarefaction and bootstrapping were used in the data analysis. According to the results, the addition of species is slow yet continuous after the first few plots. The expected number of species in samples taken with the different plot sizes decreases in the following order: $2 > 1 > 0.5$ m^2. Results implied that plant species have different optimum plot sizes with respect to the accuracy of the mean cover estimates. However, representative registering of species present needs a larger sample and plot size than a relatively accurate estimation of the mean cover of the individual species. Consequently, representative registering of species sets the minimum limits for the number and size of the sample plots.

Key words: bootstrapping, rarefaction, understorey vegetation

INTRODUCTION

There are neither standard sample and plot sizes for inventory of field and ground layer vegetation in any vegetation community, nor a standard method to establish these entities (Du Rietz 1921, Braun-Blanquet 1964, Küchler & Zonneveld 1988, Barkman 1989). Some sample- and plot-size suggestions for boreal vegetation communities have been given by Økland (1990). However, optimal sample and plot size for discrete vegetation communities should be determined independently with respect to the objectives of the study. It frequently appears that sample and plot sizes are not based on information about the vegetation community under study, nor have they been chosen by analytical means.

Generally, the number of detected species increases and the variance of the mean cover estimates decreases with increasing sample size (Arrhenius 1921, Braun-Blanquet 1964, McCune & Lesica 1992, Condit *et al.* 1996). This suggests that as many plots as possible should be used. Variance of the mean cover estimates should be minimized to maximize accuracy in parameter estimations such as mean species-percentage cover

(Kenkel & Podani 1991). To achieve this, sample-plot size should be larger than the mean cluster size of the species studied (Kenkel *et al*. 1989). Therefore, sample-plot size should also be as large as possible, given the constraints of time and costs (Kenkel & Podani 1991).

Practical considerations usually limit the number of the sample plots in a study, but the effects of sample size limitation can be counterbalanced by increasing plot size. There is, however, a plot size beyond which the gains in the number of recorded species and smaller variance of mean species-cover estimates are offset by substantial increases in sampling effort (Podani 1984). Furthermore, the accuracy of visual estimations of percentage cover which is one of the most common methods to assess species abundance (Küchler & Zonneveld 1988) decreases with increasing plot size (Sykes *et al*. 1983, Jukola-Sulonen & Salemaa 1985, McCune & Lesica 1992). But against this, less species are detected with decreasing plot size (McCune & Lesica 1992). Forest-floor vegetation, due to numerous microsites, is particularly heterogeneous with many scarce species (McCune & Lesica 1992, Frisvoll & Prestø 1997). A large sample and/or plot size may be needed to detect most species in such communities. Vegetation sampling is, therefore, a compromise among the number of recorded species, accuracy of species percentage cover estimates, and the number and size of sample plots, i.e. sampling effort.

The aim of this study was to find the optimal balance among the number and size of sample plots, the number of detected species and the accuracy of mean species-cover estimates for the purpose of inventorying field and ground layer vegetation in mature *Myrtillus*-type spruce forests in the southern boreal vegetation zone.

MATERIAL AND METHODS

Study area and sampling

The study area (0.25 ha) was located in the municipality of Kuorevesi in southern Finland (61°53´N, 24°40´E) and represented a typical mature *Myrtillus*-type spruce forest (*sensu* Cajander 1926) of the southern boreal vegetation zone (Ahti *et al.* 1968). It was surrounded by several hectares of forest with similar composition and age (i.e. 140 years). Timber volume was 398 m^3 ha^{-1}, of which 86% was Norway spruce (*Picea abies* L.), 9% Scots pine (*Pinus sylvestris* L.), 4% silver birch (*Betula pendula* Roth), 1% pubescent birch (*Betula pubescens* Ehrh.), 0.06% European aspen (*Populus tremula* L.), and 0.04% European mountain ash (*Sorbus aucuparia* L.). The number of trees per hectare ($d_{1.3} \geq 10$ cm) was 879, of which 89% was Norway spruce, 6% Scots pine, 2% silver birch, 1% pubescent birch, 0.1% European aspen and 1% European mountain ash. The total volume of logs and snags was 15.3 m^3 ha^{-1}.

Thirty-two plot locations were randomized to the study area. To prevent the distribution of the plot locations from becoming too inequitable, the study area was subdivided into four equal-sized units with eight random plot locations in each. At each location, nested circular plots with an areal extent of 0.5, 1 and 2 m^2 were inventoried. Mistakenly, the 0.5 and 1 m^2 plots were not inventoried at one location, thus the location was rejected from the analysis. The total number of plots per each plot size is therefore 31.

Vascular plants, including tree saplings up to 1.8 m in height, bryophytes, hepatics and lichens were studied. Species abundance was estimated with the visual species-percentage cover method using a 0.1–100% scale with 0.1% increments up to 1%, and with 1% increments from there on. In addition, the entire study area was examined to record species not included in the quantitative samples.

Species coverage was assessed by three persons. The estimates were calibrated through simultaneous assessment of coverages, and comparison of the results until obtained species-percentage cover estimates were similar.

The nomenclature follows Hämet-Ahti *et al.* (1998) for vascular plants and Koponen *et al.* (1977) for bryophytes.

Data analysis

The expected numbers of species in samples of one to 31 plots were calculated using the rarefaction method (Sanders 1968, Hurlbert 1971, Simberloff 1972, Heck *et al.* 1975, Kouki & Haila 1985, Krebs 1989). To do this, one hundred random samples of sizes from one to 31 were drawn from the empirical sample of 31 plots. For each sample size, the expected number of species was calculated according to the rarefaction equation. The equation is given in Heck *et al.* (1975), Kouki and Haila (1985) and Krebs (1989); for example *see* also Smith *et al.* (1985). It should be noted that sample size cannot be extrapolated beyond the number of plots in the empirical sample, since samples are drawn without replacement. Theoretically, rarefaction assumes random spatial distribution of individuals and species, otherwise the expected number of species may be overestimated. Rarefaction is therefore best used within a single biotope (Fager 1972, Kouki & Haila 1985), as in our study.

Bootstrapping (Diaconis & Efron 1983, Efron & Tibshirani 1993) was used to analyze how the different plot sizes, and different sample sizes, in our case from 1 to 31 plots, affected the variance of the mean species-cover estimates. Bootstrapping is a resampling technique which uses the empirical sample to randomly generate samples of de-

sired size. The bootstrapped samples can be used in various statistical analyses.

Bootstrapping was done separately for each plant species. From the empirical sample of percentage cover estimates from the 31 plots, a random set of estimates was drawn one thousand times. The randomly drawn sets are the bootstrap samples. Bootstrap samples were drawn with replacement. Since the size of the bootstrap sample equals the number of sample plots, the bootstrap sample size was from one to 31, as in the rarefaction calculation. A mean species-cover estimate was calculated for each bootstrap sample. Variance of the mean species-cover estimates, calculated using the 1 000 bootstrap samples, represents the accuracy of the mean species-cover estimate. Bootstrapping with sample sizes from one to 31 were done independently for each of the three plot sizes.

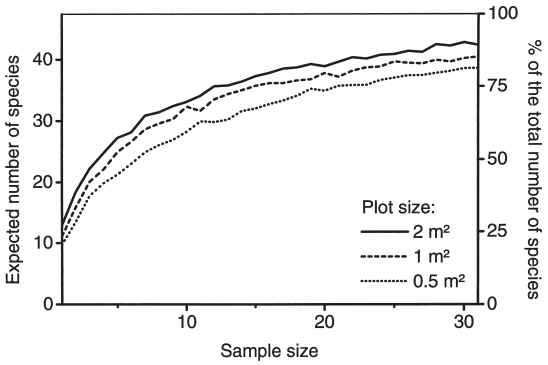

Fig. 1. Rarefaction values for the expected number of species in samples of one to 31 plots.

RESULTS

Expected number of species

After the first few plots, the expected number of species increased relatively slowly but continuously up to 31 sample plots, i.e., the species accumulation curves did not become asymptotic. The 2-m^2 plot had constantly the highest expected number of species (Fig. 1). Results of the 1-m^2 plot fell between those of the 0.5- and 2-m^2 plots.

Bootstrap variance

The results for four vascular plant species and four bryophyte species which typically inhabit Myrtillus-type spruce forests are shown to illustrate different patterns regarding how the variance of the mean species-percentage cover estimates of the bootstrap samples (bootstrap variance), responded to different plot and sample sizes (Fig. 2). The species chosen were *Deschampsia flexuosa* (L.) Trin., *Trientalis europaea* L., *Vaccinium myrtillus* L., *V. vitis-idaea* L., *Dicranum majus* Turn., *Pleurozium schreberi* (Bridd.) Mitt., *Polytrichum commune* Hedw., and *Sphagnum girgensohnii* Russow.

The bootstrapping results were similar to those of the rarefaction, i.e., with increasing sample size the bootstrap variance decreased quickly at first, but after the first few plots, the decrease was relatively slow (Fig. 2). The plot size having the lowest bootstrap variance varied among species. For *S. girgensohnii* and *T. europaea*, the 0.5-m^2 plots had the lowest variance, but the lowest variance for *D. majus* and *P. schreberi* was associated with the 2-m^2 plots. For *V. vitis-idaea*, the 1-m^2 plot had the lowest bootstrap variance. For *D. flexuosa*, *V. myrtillus* and *P. commune*, the plot size did not seem to affect the bootstrap variance at all.

DISCUSSION AND CONCLUSIONS

After the first few plots, an increase in the number of new species encountered is only gained through a considerable increase in sampling effort (Fig. 1). The reason why the accumulated new species curve did not become asymptotic seems to have been because of encounters with microsites which contain uncommon species. It might be possible to capture most microsites, and thus species present, in the study area by considerably increasing the sample size. But it depends on the study aims whether having a few more species in the quantitative sample is worth the extra sampling effort. A supplementary survey of microsites to obtain qualitative species data may be a better choice in many cases.

More species were detected using 2-m^2 plots than with 1-m^2 and 0.5-m^2 plots. However, 1-m^2 plots represent a good compromise. Although the 1-m^2 plots had a lower expected number of species and in many cases higher bootstrap variance than the 2-m^2 plots, smaller plots are somewhat faster to enumerate in practice, and cover estimates are more accurate (McCune & Lesica 1992).

The magnitude of the bootstrap variance is dependent on the mean abundance and spatial distribution of the studied species. The reason that

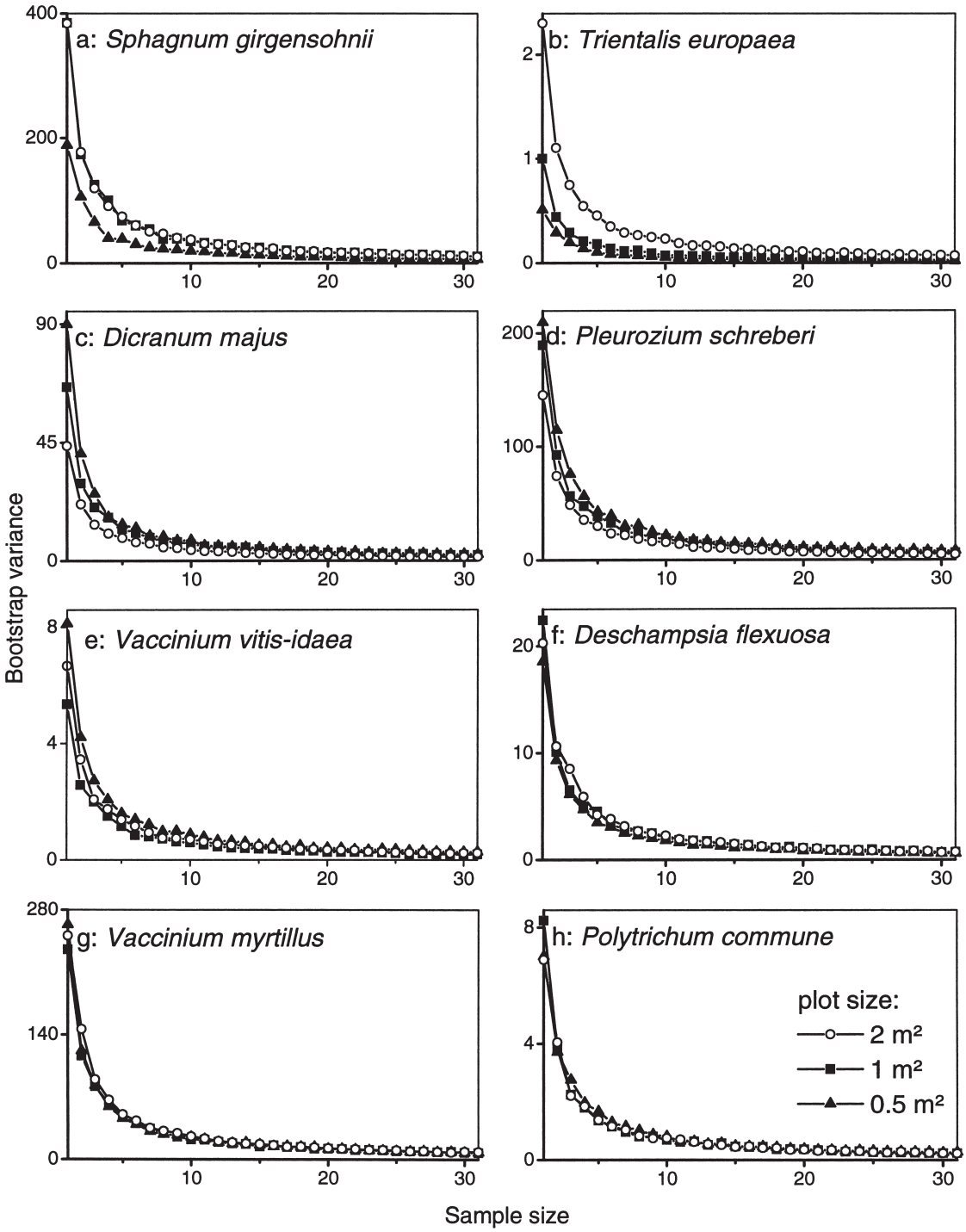

Fig. 2. Variance of the mean species percentage cover estimates of 1 000 bootstrap samples (bootstrap variance) of one to 31 plots.

plot size did not influence the bootstrap variance of *Deschampsia flexuosa*, *Polytrichum commune* and *Vaccinium myrtillus* is probably due to the great frequency of these species in our material (Fig. 2). *Sphagnum girgensohnii* and *Trientalis europaea* had the lowest bootstrap variance as-

sociated with the 0.5-m^2 plot, which may seem inconsistent, but if a species is relatively uncommon, an increasing number of zero observations decreases the variance of the mean cover estimates. Indeed, the frequency of *Trientalis europaea* was 70% higher with the 2-m^2 plots than with the 0.5-m^2 plots, while the overall frequency of this species remained low. In multivariate studies, when a sample is used to obtain estimates of more than one species, as the case usually is in vegetation studies, it is possible for each species to have a different spatial distribution, and thus a different optimal plot size (Kenkel & Podani 1991). Plot size affects sampling variance most profoundly in heterogeneous vegetation communities, where smaller plots are more likely to detect small-scale vegetation patches (Clapham 1932).

With all analyzed species, the bootstrap variance decreased to a relatively low level at a smaller number of sample plots than seems to be needed for representative registering of species present in the studied community. In other words, the aim to "optimally maximize" the number of registered species predetermines the minimum limits for sample and plots sizes, not the aim to "optimally minimize" the variance of the mean cover estimates.

Consideration of plot shape may be of importance in communities with a strongly clustered spatial pattern of species. More species are encountered using rectangular plots than circular or square ones of the same area in such communities (Clapham 1932, Bormann 1953, Podani 1984, Condit *et al*. 1996). However, in repeated inventories, circular plots are easier to place identically each time. They also have less edge effect than square plots and thus less sampling error (Goldsmith & Harrison 1976).

Sampling always includes some error. Misidentification and/or overlooking of species is a potential source of sampling error (Lepš & Hadincova 1992). Despite careful investigation, species may have remained undetected on our study area, which was large enough to contain numerous microsites. Variation between different observers' cover estimates is often high if coverages are either very low or very high (Tonteri 1990, Lepš & Hadincova 1992). If several persons are involved in the vegetation sampling, there should be a calibration exercise to increase consistency of the cover estimates. The optimal sample and plot size should be determined independently for discrete plant communities. A plant community with a different spatial pattern will probably need a different sample and/or plot size for optimal balance among sampling effort, accuracy of cover estimates, and the number of detected species.

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