

Introduction

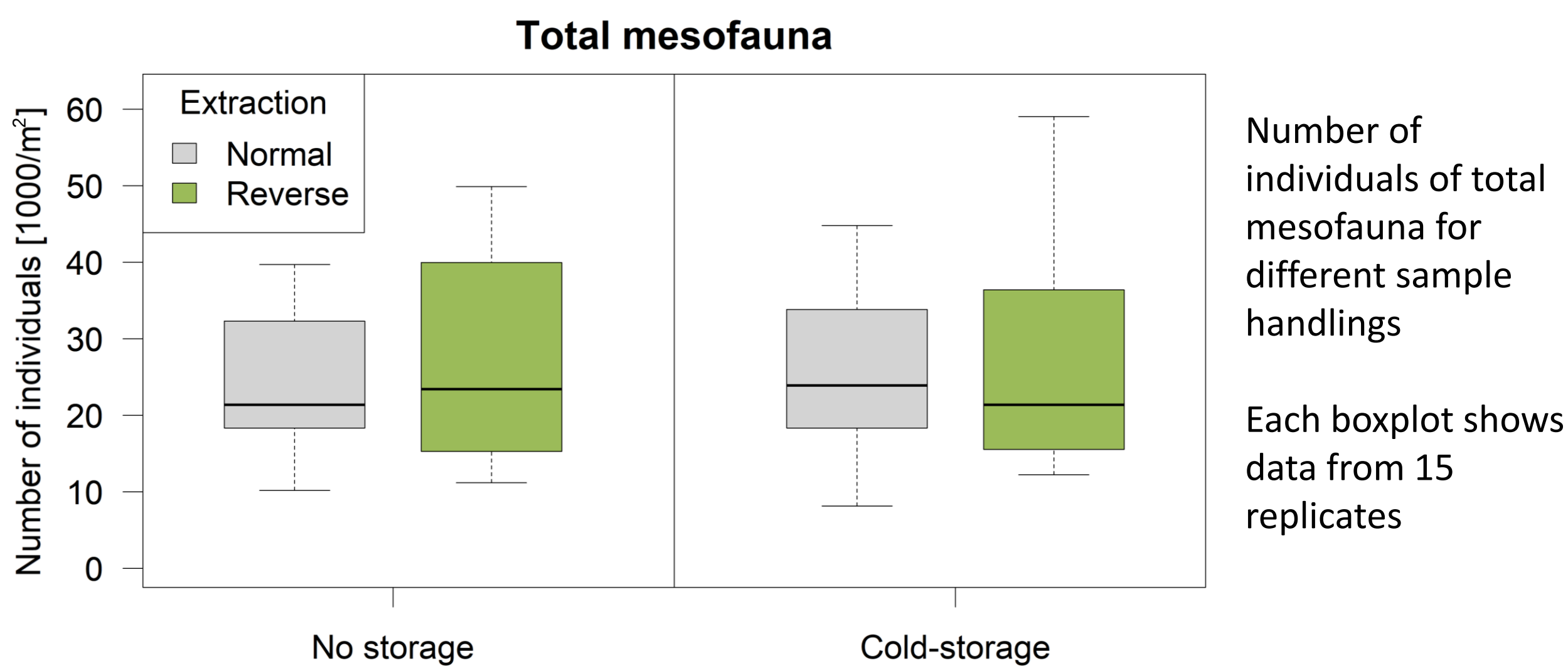


- Mesofauna field studies can be conducted as higher tier refinement option
- Such studies investigate effects on species composition and abundance at community/population level under realistic exposure conditions
- While planning mesofauna field studies, different conceptual and technical aspects need to be considered
- This poster focusses on
 - (a) the impact of technical aspects of sample handling (extraction & storage)
 - (b) characteristics of a suitable sampling design (number of replicate plots and soil cores taken per plot)

Methods and sample handling

- Collembola and Acari species were heat-gradient extracted (ISO 23611-2; Macfadyen, 1961) from soil cores from 15 grassland plots (4 soil cores per plot)
- The four soil cores represent four different combinations of sample handling and storage:
 - (i) normal orientation of the sample during extraction (same as in the field) & no storage
 - (ii) reverse orientation (upside down) & no storage
 - (iii) normal orientation & storage under cooled condition before extraction for 7 days at 5°C
 - (iv) reverse orientation & cold-storage

Using generalized linear mixed effect models, effects of sample handling and storage on the extraction efficacy were tested. **No difference between sample handlings was detected, except for one case.**



Variable	Poduromorpha	Entomobryoidea	Isotomoidea	Symphyleona	Total Collembola	Gamasina	Oribatida	Other Acari	Total Acari	Total Mesofauna
Individuals [1/m²]	3962	2664	10850	373	17849	1934	3156	3487	8585	26434
normal extract. - no storage	2677 #	not significant (no difference between sample handlings)								
normal extract. - cold-storage	4326									
reverse extract. - no storage	4734 #									
reverse extract. - cold-storage	3799									

significant difference ($\alpha > 0.05$) between marked sample handlings

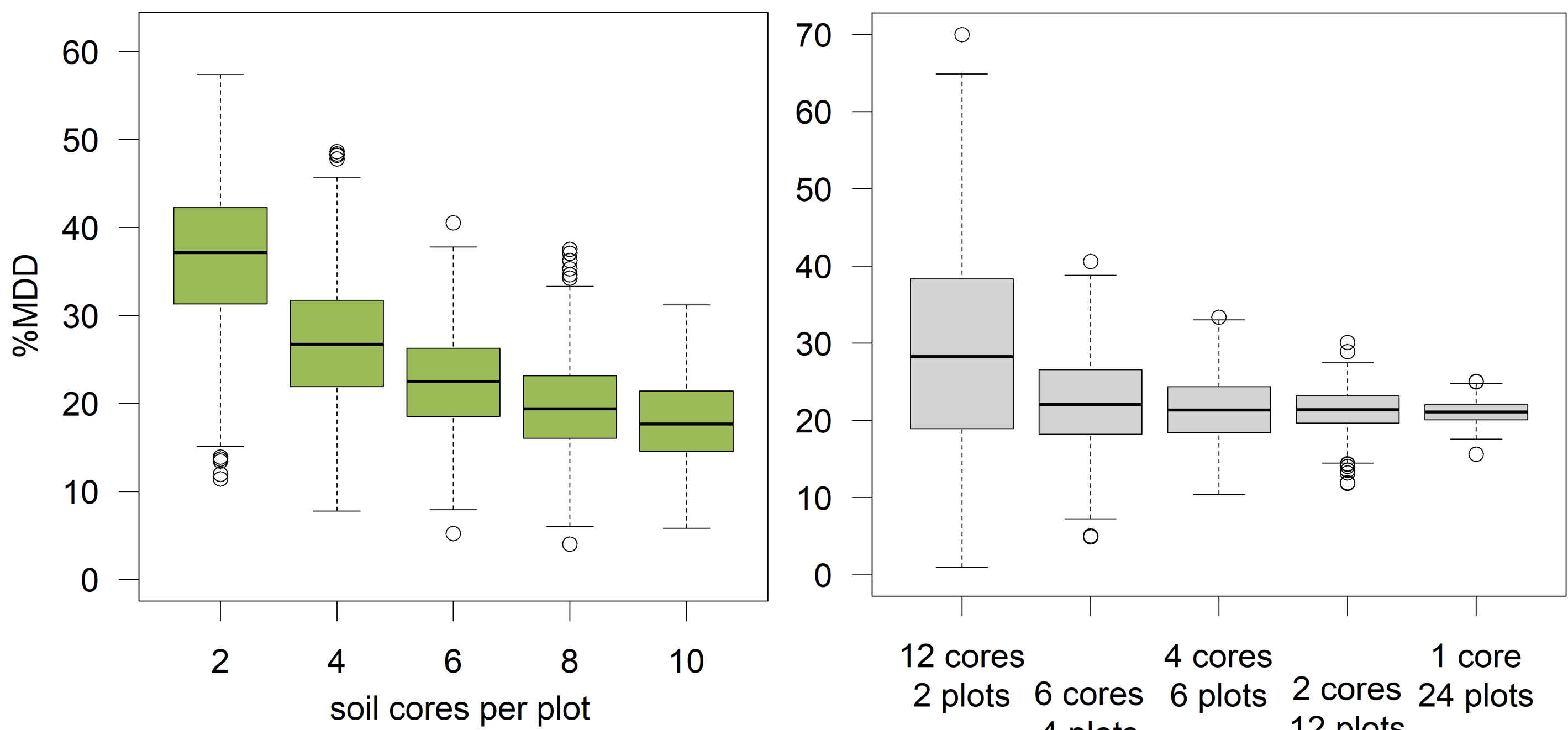
Study design and sample size

The large data set was used to simulate different realistic sampling designs to illustrate the relationship between number of replicates and minimum detectable difference (MDD; Brock et al. 2015) for this type of field study.

- Individual numbers of total mesofauna in all 60 soil cores were used and represent a characteristic variance
- Different study designs were resampled from the data, differing in number of soil cores & replicate plots (see figure below)
- For each setting 1000 virtual studies were sampled with the given number of plots for each control and treatment, respectively. Then the minimal difference between the means of a treatment and the control that could be detected by the conducted t-test, given as percentage of control mean, was calculated (shown as %MDD in Figure to the right)

→ Increasing number of soil cores taken per plot reduces the variance between plots

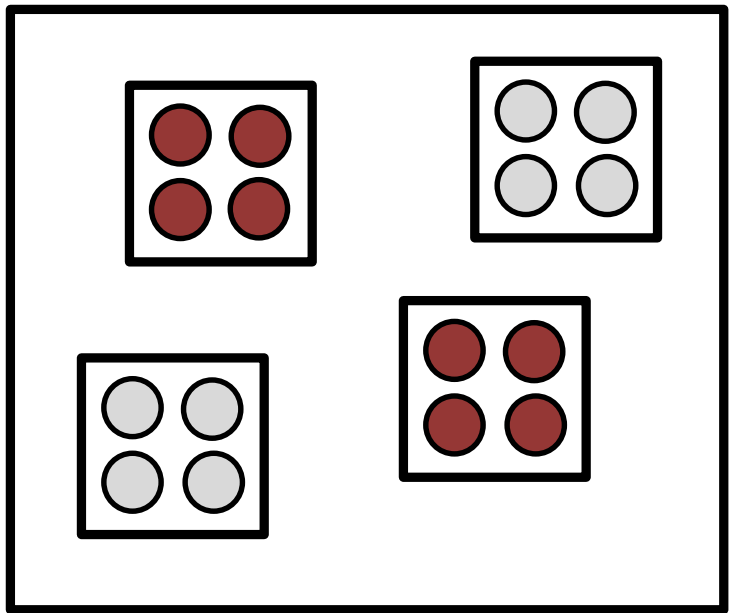
→ Increasing number of plots is more beneficial than increasing number of soil cores per plot



Variance of %MDD of 1000 samples for different combinations of number of replicate plots and number of soil cores: (left) 4 replicate plots for control and treatment, respectively, with differing number of soil cores per plot, and (right) 24 soil cores distributed on different number of control plots

Statistical analysis of mesofauna field studies:

- n replicate plots were sampled
- m soil cores were taken at each plot



1. Sum up number of individuals in all cores at each plot
2. Log-transform individual numbers per plot
3. Estimate difference between **control** and **treatment** plots by statistical test



Overall Conclusion



- It is crucial to apply a suitable sampling design (e.g. in terms of number of replicates) to ensure an appropriate explanatory power of the study
- Six soil cores taken on four replicate plots each, appear to be a good compromise of sample size and working effort. Four replicates are recommended in the current guidance for earthworms field studies (ISO 11268-3), which is often used for mesofauna field studies as well. A further increasing in sample size (either number of plots or soil cores) can improve the accuracy of results
- Orientation of soil cores during heat-gradient extraction of organisms (i.e. either normal or upside down) and their intermediate cold-storage (compared to direct extracted samples) had no effect on the extraction efficiency of the most important mesofauna groups