Bench sheet · EnGeL-FAUNA-MAR-MORPH

Sampling, MacFadyen extraction and sorting of soil microarthropods · version 1.1.1 (2026-11-20). Full protocol: EnGeL-FAUNA-MAR-MORPH. Tick each step when done; write deviations in Notes.

Date	
Operator	
Plate ID / samples	
Ethanol 96% lot	

Before starting

☐	No.	Step
☐	•	Define sampling points according to the sampling design of the study.
☐	•	Register the samples in the sample master data file to obtain EnGeL labels (type of sampling `Macf`).

1. Collecting the soil samples

☐	No.	Step
☐	1	At each sampling point, use a split corer to collect a soil sample.
☐	2	Open the corer. Optionally, take a photo of the soil core profile to complement the site characterisation.
☐	3	Separate the sample inside the corer into the litter/organic layer (including the humus horizon; variable thickness is expected) and the mineral soil.
☐	4	Discard the mineral soil in the split corer (mineral soil is collected with an additional PVC tube).
☐	5	Record the depth of the litter layer.
☐	6	Place the litter sample in a labelled plastic box (Annex 1).
☐	7	For the undisturbed mineral soil, remove the litter layer to expose the mineral soil and take a core to a depth of 5 cm with a PVC tube (5 cm diameter × 5 cm height, c. 100 cm^3).
☐	8	Drive the PVC tube gently into the soil using a wooden or rubber block and a mallet, to avoid compacting the soil.
☐	9	Remove the PVC tube from the soil with a spade placed underneath it. Remove excess soil around the tube with a knife.
☐	10	Close both ends with plastic caps, seal them with paper tape to preserve the soil structure, and place the tube in a labelled plastic bag.
!		**CRITICAL** Avoid disturbing the core structure and compacting the soil, which could kill the animals. The time between sampling and extraction should not exceed 5 days, to minimise effects of confinement and shifts in the micro-populations. Keep samples in an aerated space at room temperature until shipping.
☐	11	For shipping, pack the samples in a styrofoam box.

2. Extracting microarthropods

☐	No.	Step
☐	12	In the laboratory, set up the MacFadyen extraction system.
☐	13	Place each sample core upside down in a canister with a plastic or metal net (2 mm mesh) at the bottom.
☐	14	Connect the canister to a funnel attached to a collecting flask containing 25 mL of 96% ethanol.
☐	15	Build up a temperature difference of about 35–40 °C between the upper and lower parts: heat the upper part to 45–50 °C while cooling the lower part to a maximum of 10 °C (field temperature). Circulate heated air in the canister area and cooled air in the collecting area.
☐	16	Increase the temperature of the upper part gradually, starting at about 5 °C above field temperature during the first 2–3 days, to prevent sample desiccation and animal death.
☐	17	Then intensify the gradient (upper part 45–50 °C) for the remaining days. The total extraction time is 7 days.

3. Sorting and preserving

☐	No.	Step
☐	18	After extraction, label the animal samples for identification and prepare them for storage until sorting.
☐	19	Place each sample in a Petri dish under the stereomicroscope.
☐	20	Sort the animals into major groups and morphotypes according to life-form traits (pigmentation, antenna length, presence of ocelli, furca and scales/long hairs). Handle animals with a pipette or a fine brush.
☐	21	Transfer each group/morphotype to a plastic vial with 96% ethanol and store at room temperature (ideally in the fridge if there is space) for molecular analyses.

After finishing

☐	No.	Step
☐	•	Clean the bench with 70% ethanol.
☐	•	Empty the small bench bin into the right waste bin.
☐	•	Wash your hands.

☐	No.	Step
☐	•	Turn off equipment and lights.
☐	•	Lock doors, fridge and freezer.

Notes / deviations
