

Dry heat extractors: microarthropod extraction v1.1

Extraction protocol using Tullgren/Berlese extractors with heating from above is presented here. Please, adapt high-gradient extraction accordingly. Extraction is done for **at least 7 days through a 2 mm mesh directly into ~96% ethanol** which consequently serves as the fixing solution; no intermediate fixing agent is used. Consumable materials:

☐ Vials to store microarthropods (15-20 ml tubes are recommended; it should be
possible to close them tight to avoid evaporation), 1 per sample.
☐ Ethanol (~96%), 20-25 ml per sample.
☐ Printed ethanol-resistant labels, 1-2 per sample (inside and/or outside).
Instructions:

- 1. Place the funnels without sieves in the funnel holders.
- 2. Place or attach vials filled with 96% ethanol under the funnels (with labels).
- 3. Put soil and litter on the plastic shield on the table, <u>gently</u> break into several large pieces and discard visible earthworms and large macrofauna (> 1 cm).
- 4. Put a sieve on another plastic shield; put 2 layers of 2 mm mesh into the sieve.
- 5. Put litter and soil from the first plastic shield on top of the mesh.
- 6. Move the sieve with litter and soil on the first plastic shield and add soil that fell on the second plastic shield in the sieve.
- 7. Gently place the sieve with soil and litter on top of a funnel. Repeat for all samples.
- 8. Switch on the light to start the extraction.
- 9. Run the extraction until the sample is completely dry, but a minimum of 7 days.
 - a. The temperature should never exceed 50°C on the surface of the samples.
 - b. After 3 days check if ethanol is evaporated and add more, if needed.
- 10. When extraction is finished, remove the sieves from the funnels.
- 11. Remove the vials, fill them with 96% ethanol ($> \frac{2}{3}$) and close (with labels).
- 12. Store all vials in a freezer (c. -20°C).
- 13. Discard the soil and litter.

Important tips:

- Animals must be able to escape down: Do not overload the funnels. There should be
 a maximum of 3-4 cm soil thickness on the sieve and empty spaces around the
 sample, allowing animals to escape down. If the funnels are small, use several
 funnels to extract one sample. Soil monoliths can be gently broken down, but do not
 destroy soil aggregates to avoid locking/flatting animals.
- Extraction conditions should not be too harsh (e.g. too hot), because slow animals may dry out before being able to escape. Control T with a termometer.
- Ensure appropriate extraction conditions: a ventilated room with low humidity.
- <u>Sample cleanness is very important:</u> Avoid any dirt (soil) falling in the vials. Avoid vibration, do not shake the extractors. If you check the substrate with your hands, do it above the plastic shield table, not above the extractor.
- Before finishing the extraction, meticulously check the sample moisture: take 2
 largest samples out of the extractor and check for dryness by hand on a table. The
 soil should be perfectly and completely dry.