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Short Guidelines for Unique Methods

Educational Network on Soil and Plant Ecology and Management

Content

Soil Respiration

Teacher Dr. Tomáš Picek, Dr. Zuzana Urbanová

Contact University of South Bohemia, Department of Ecosystem Biology, Faculty of Science, Branišovská 1760, České Budějovice 370 05, Czech Republic

Methods

Soil Respiration *in situ*

- Direct measurement of soil respiration in the field using EGM4 with soil chamber
- Soil temperature measurement
- Soil moisture measurement

Potential soil respiration measurement under laboratory conditions

- Field sampling of surface soil layer in different types of wetlands
- Soil preparation in the laboratory

Incubation of soil samples under aerobic and anaerobic conditions

- Establishment of aerobic and anaerobic conditions in incubation flasks
- CO_2 and CH_4 production of soils measured by gas chromatograph

Calculation

- Respiration rate and rate of methane production in various soils and under variable conditions
- Estimation of active microbial biomass

Key words wetland soil, microbial respiration, aerobic, anaerobic, methane production

Learning objectives To show, how soil quality and aeration conditions affect microbial activities in soil and decomposition processes

Description of Methods

Soil respiration in situ

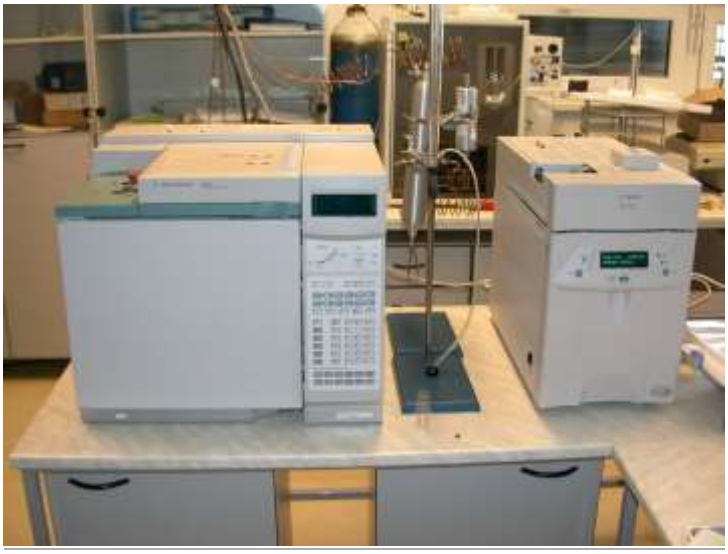
- Soil respiration is directly measured in the field using EGM4 (infrared gas analyzer) equipped with soil respiration chamber.
- Few sampling plots (at least 6) are selected at each site, vegetation is removed and plastic collars are inserted into the soil.
- Second day, the measurement is done using EGM4. The rate of soil respiration is expressed as g CO_2 m^{-2} h^{-1} and it is calculated automatically by the instrument (known chamber volume and area of soil covered by the chamber)
- Together with soil respiration the soil temperature in three depths (5cm, 10 cm, 20 cm) and soil moisture are usually measured as important variables and used for modelling of soil respiration on the study site

Soil respiration measurement in the field using EGM4 with soil respiration chamber.

Plastic collar inserted into the soil for soil respiration measurement.

Potential soil respiration measured under laboratory conditions

- Soil for laboratory respiration measurement is sampled at the same sites as used for *in situ* measurements. Surface layer is sampled (0-20 cm) at least in three replicates at each site
- In the laboratory, soil is homogenized by sieving through a 2 mm mesh
- A portion of each sample is dried to constant weight and weighed to calculate the dry matter content (later needed for calculation)
- 10 g of soil are weighed into glass bottles (100ml) and sealed airtight with rubber stoppers
- Two sets of each sample are incubated at 15°C for 2 days (one week), one in aerobic conditions (ambient air in headspace), the other one in anaerobic conditions (the headspace of flasks flushed with helium)
- The headspace of flasks is sampled by syringe (0,2 ml) and concentration of CO_2 and CH_4 in the headspace of the incubation flasks are measured in 12 hours intervals using gas chromatograph equipped with temperature conductivity detector for CO_2 analyzes and with flame ionization detector for CH_4 analyzes



Gas chromatographs (left) and incubation box with glass bottles with soil samples (right).

- Rates of CO_2 and CH_4 production are then calculated as an average for the whole incubation period

Calculation:

1. *Volume of CO_2 in the headspace of incubation flask*

$$G = c_{CO2} \cdot V_G / 1000 \qquad [\mu l\ CO_2]$$

C_{CO2} concentration of CO_2 measured by gas chromatograph (ppm)

V_G volume of the headspace of incubation flask (ml)

2. *Volume of CO_2 dissolved in soil solution*

$$L = \frac{0{,}928 \cdot G \cdot V_L}{V_G} \qquad [\mu l\ CO_2]$$

0,928 – solubility coefficient of CO2 in water at 20°C

V_L volume of soil solution in incubation flask V_L = (1 – dry weight) . wet weight of sample (ml)

3. *Total volume of CO_2 produced per 1 g of soil*

$$T = \frac{(G + L)}{\text{(weight of dry soil)}} \quad [\mu l\ CO_2\ g^{-1}]$$

4. Respiration rate

$$Y = \frac{0{,}536\ .\ (T \text{ end of incubation}) - T \text{ start of incubation})}{\text{Length of incubation}} \quad [\mu g\ C\text{-}CO_2\ g^{-1}\ day^{-1}]$$

- coeficient 0,536 for recalculation from μl CO_2 to μg C-CO_2

5. Estimation of active microbial biomass

$$Cmic = 433\ .\ \log_{10} Y + 59{,}2 \quad [\mu g\ C\ g^{-1}]$$

Y (μl CO_2 g^{-1} h^{-1})

For calculation of methane production rate the same formulas are used, only solubility coefficient is different for CH_4.

Recommended literature

Brady, N. C.: The nature and properties of soils. MacMillan Publ. Comp., New York, 2002.
Reddy, K.R., DeLaune R.D. Biogeochemistry of Wetlands, 2008.

Soil Zoology

Teacher	Apl. Prof. Dr. Manfred Wanner
Contact	Brandenburg University of Technology Cottbus - Senftenberg, Faculty of Environment and Natural Sciences, Dept. Ecology wanner@b-tu.de
Methods	Field sampling methods and laboratory procedures in soil zoology •Estimation of the overall biological activity in the field •Litterbags •Minicontainers •Bait lamina strips •Field sampling of animals •Soil samples – abundance of endogeic animals •Pitfall traps – activity-density of (larger) epigeic animals •Eclector traps – abundance of (hidden) insect larvae •Exhaustor – collecting individual arthropods from surfaces •Chemical extraction – collecting earthworms •Laboratory methods •Dynamic extraction of soil samples – Gradient methods •Berlese-Tullgren extraction •MacFadyen and Kempson extraction •Baermann funnel •Direct examination of a soil sample by hand sorting •Direct examination of a soil sample under the microscope
Key words	Soil animals, Field Sampling, Extraction Methods
Learning objectives	

Description of Methods

Estimation of the overall biological activity in the field

Litterbags

• Litterbags are used for litter decomposition studies
• Decomposition of leaf litter is measured by weight loss of bagged leaf litter
• Different mesh sizes of the bags may be used to exclude macroarthropods, micro-arthropods, or microfauna
Target: overall organismic activity, incl. microorganisms & fungi

Minicontainers are highly standardized small litterbags, arranged in rows and inserted vertically or horizontally into the soil.

The bait-lamina test is an easy and fast method to investigate the `feeding activity' of soil organisms, a quantity reflecting complex decomposition processes.
The test consists of vertically inserting 16-hole-bearing plastic strips filled with a plant material preparation (as bait) into the soil.

Field sampling of animals

Pitfall traps
• A useful, inexpensive, and rapid method for assessing communities of macroarthropods
• But: pitfall traps have only limited usefulness for assessing population sizes
• Trapping success depends on behaviour (activity) of the epigeic arthropods!
• Activity-density, NOT abundance is measured!
• Statistical analysis: only nonparametric tests are allowed!

Pitfall traps consist of cans or jars, (yoghurt pots are often used), set flush with the soil surface, and containing some chemical as preservatives (e.g. a mixture of water, alcohol, and acetic acid).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Eclector traps
• A capture method which is reputed for producing specimens of species seldom captured otherwise:

Diptera larvae living in the soil can be counted indirectly by catching the hatching adults, which react phototrophic and fly towards the opening of the tent-like eclector trap

• Eclectors do not destroy the soil structure!

Target organisms: predominantly hatched diptera


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Exhaustors

a simple “vacuum cleaner” (or suction pipe) used by mouth to catch mobile arthropods

…do not confuse the suction tube with the other…

Active extraction of earthworms: Formalin/Mustard - Method

• Water suspensions of chemical repellents
• Application with a watering can several times on a defined area
• Worms in the upper soil try to escape to the soil surface


©Elmer

Active extraction of earthworms: Octet – Method (Electricity)

• Based on the law of electrodynamics (condenser, electric field)
• 8 electrodes in circular configuration, changing electric fields
• Worms are exposed to electricity and try to escape to the soil surface

Soil samples by a soil coring device

Effective core diameter: usually 50 mm for soil samples

• Soil samples can be used for soil analyses (physical, chemical, biological)

• With respect to collect (endogeic) soil animals, soil samples can be used for extraction procedures (Berlese funnel extraction or flotation, see below)
• Most quantitative samples of microarthropods (e.g., Collembola or mites) are taken from soil cores taken by 0-5 and 5-10 cm in depth. Such a soil core, especially that of the upper layer, will contain several hundred soil animals!

Target organisms: Predominantly endogeic micro- and mesofauna


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Laboratory methods – dynamic extraction of soil samples – gradient methods

<u>Berlese (- Tullgren) extraction</u>
Extraction principle:
the soil environment is
- dark
- moist
- cool

Illuminating by a light bulb will establish a gradient of light/darkness; dryness/moisture and heat/cool.
Highly recommended for soil microarthropods which will follow the gradient actively and finally fall into the tube filled with a preservative chemical.

Target organisms: predominantly active endogeic soil mesofauna (microarthropods as mites and springtails etc.)


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MacFadyen-extraction

• For a variety of medium-sized soil arthropds (mites, springtails….)
• Heat and moisture gradient
• Improved technical equipment (water cooling, infrared light, computer regulation)

Target organism: predominantly active endogeic soil meso- and macrofauna


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Kempson-extraction

• For relatively large soil arthropods (beetles, spiders, isopods….)
• Heat and moisture gradient
• Improved technical equipment (water cooling, infrared light, computer regulation)

Target organism: Predominantly active endogeic soil (meso-) and macrofauna (arthropods)


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Bearmann Funnel

• A “wet” gradient method
• Extraction of minute, water-dependent animals, especially nematodes, enchytraeids, and tardigrades.
• A thin soil sample is placed on a sieve in a funnel filled with water and exposed to light and heat – similar to the “dry” Berlese-extraction
Target organisms: predominantly active endogeic soil microfauna (nematodes, tardigrades, enchytraeids, rotifers)

Hand-Sorting of soil animals

• Pros
  - If skilled staff, precise and complete results
  - Living & dead organisms collected
  - direct method
  - no sophisticated equipment necessary
• Cons
  - only experienced staff
  - time consuming!
  - manpower costs!
  - hand-sorting of animals have to be done immediately after soil sampling

Target organisms: all organisms within a soil sample

Recommended literature

•Coleman, D.C.; Crossley, D.A.Jr.; Hendrix, P.F.: Fundamentals of Soil Ecology. 2nd ed.; Elsevier, Amsterdam (2004)
•Dunger, W.; Fiedler, H.J.; Methoden der Bodenbiologie, Gustav Fischer, Jena (1997)
•Dunger, W. Tiere im Boden. Die Neue Brehm Bücherei, Ziemsen Verlag, Wittenberg-Lutherstadt (1983) – reprint now available!

Allelopathy

Teacher Pr. Catherine Fernandez & Virginie Baldy

Contact Institut Méditerranéen de Biodiversité et d'Écologie marine et continentale, Marseille, France
catherine.fernandez@imbe.fr; virginie.baldy@imbe.fr

Methods Highlighting allelopathic potential with fast laboratory bioassays
Methodological aspects for more realistic results
- Choice of the target species
- Role of soil microorganisms
- Allelopathy of volatile compounds
- Allelopathy in the field

Key words Allelopathy; Secondary metabolites

Learning objectives

Description of Methods

Highlighting allelopathic potential with fast laboratory bioassays

The method described herebelow allow the assessment of allelopathic potential in a short-term experiment. It is useful for screening of allelopathic potential of various species/organ/ages but some refinements to this method can lead to more realistic conclusions

Experimental design

Water-soluble compounds are probably the most involved in allelopathic interactions, thus water extracts of the donor species are prepared to test for their allelopathic effect on a target species. *Lactuca sativa* is commonly used as target species in such bioessays for its rapid germination and growth and supposed sensitivity to allelochemicals.

- Donor species leaves, roots or litter extracts are prepared by soaking 20 g (dry weight) in 200 ml of distilled water (10 % dry weight). Extracts are prepared at room temperature and left in darkness for 24 hours. Diluted solutions (5 and 2.5%) are prepared from the mother solution.
- Trials are carried out using glass Petri dishes
- 25 seeds of target species are placed on two sheets of Whatman® n°4 filter papers,
- Petri dishes are watered with 2 ml of distilled water (Control) or 2 ml of maceration of each extract.
- 3 to 5 replicates (Petri dishes) per treatments are realised

Measures on the target species

- **Germination**: Seed germination in each petri dish is counted every day. *Lactuca sativa* germinates very quickly, most seeds will germinate the first two days. Two parameters can then be calculated:
    - Germination speed is calculated for each petri dish using the Kotowski's velocity factor:
      Cv = 100 * (ΣNi / ΣNiTi), with Ni = number of germinated seeds at Ti and Ti = number of days from the beginning of the experiment.
    - Final germination rate is calculated as [number of germinated seeds/number total of seeds]*100
- **Growth**: After 5-7 days, the length of the hypocotyle and radicle of all *Lactuca* seedlings can be measured

Statistical analyses

- Germination percentage can be analysed by a Chi2 test
- Germination velocity and length of the hypocotyle, radicle and total length of seedlings are analysed by ANOVAs

<u>Methodological aspects for more realistic results</u>

Choice of the target species

Choose several target species co-occurring with the donor species. Allelopathic response are often species-specific

Role of soil microoganisms

Soil microorganisms can alleviate or increase allelopathic effects. Allelochemicals can also be adsorbed on soil particles. Using natural soil instead of filter paper in the Petri dishes give more realistic results.
The role of soil microorganisms can be investigated by comparing the allelopathic effect in petri dishes with sterilised soil.

Allelopathy of volatile compounds

Emission of volatile compounds can also have an allelopathic effect on target species. This can be tested by placing the petri dishes with the target species seeds in sealed pots. In half of the pots, leaves (or roots) of the donor species are placed in tea bags hung in the pot. In the other half, empty tea bags are placed.
The rest of the bioassay remains the same.

Allelopathy in the field

Evidencing allelopathy in the field is difficult due to the difficulty to distinguish between allelopathy and resource competition. Several methods have however been proposed.

- Comparing bioassay results with field patterns

To reach a convincing evidence of the occurrence of allelopathic interactions in natural ecosystems, laboratory bioassays should be compared with field patterns.

A field inventory of the plant species present in the presence / absence of the donor plant species can be performed first. We expect that plants that are found only in the absence of the donor species are sensitive to its allelochemicals (high inhibition in bioassays) while plants that can be found in its presence are insensitive or stimulated.

It is also possible to introduce the target species in the presence/absence of the donor species in the field and monitor its germination and growth to compare with laboratory results.

- Use of activated carbon

Activated carbon is known to adsorb and remove allelochemicals, thus reducing or cancelling allelopathic effects. Introduction of activated carbon in the field can be a solution to examine if allelopathy is occurring. However, methodological problems in link with modification of soil properties have been recorded on calcareous soils.

- Greenhouse realistic design

Greenhous experiment with natural soil and leachates directly collected from the donor species (instead of prepared extracts) have been proposed (Viard-Crétat et al. 2009 Ann. Bot.).

Recommended literature

- Inderjit, Callaway RM (2003) Experimental designs for the study of allelopathy. Plant Soil 256:1–11.
- Zeng R.S., Mallik A. U., Luo S.M. (2008). Allelopathy in Sustainable Agriculture and Forestry. Springer, New York, USA
- Reigosa M., Pedrol N., Gonzalez L. (2006). Allelopathy: a physiological process with ecological implications. Springer, Dordrecht, Netherlands

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Soil Physics

Teacher	Prof. Endla Reintam
Contact	**Estonian University of Life Sciences, Institute of Agricultural and Environmental Sciences, Department of Soil Science and Agrochemistry, Kreutzwaldi Str. 1, 51014 Tartu, Estonia, endla.reintam@emu.ee**
Methods	Field sampling methods and laboratory procedures in soil physics: Bulk density Particle density Porosity Water holding capacity, water content Soil structure and its stability
Key words	soil bulk density, water content, porosity, structure, structural stability
Learning objectives	estimation of soil physical quality depending on management and land use

Description of Methods

Sampling

Equipments: steel cylinders, hammer, knife, spade, plastic boxes, waterproof permanent marker
Push cylinder in the soil, do not compact soil inside of cylinder;
Use knife or spade to take cylinder out, so that cylinder remains fulfilled with the soil;
Cut off excess soil and place cylinder into plastic box for further analyses.


Bulk density

Bulk density is soil mass divided by unit volume. In its natural state, a soil's volume includes solids and pores, therefore, a sample must be taken without compaction or crumbling to correctly determine bulk density (Dm).

$$D_m(g/cm^3) = \frac{p}{V} \quad , \qquad (1)$$

Dm – soil bulk density (g m^{-3})

p – Oven dry soil weight (g),

V – volume of cylinder (cm3); $V=\pi r^2 h$ where $\pi=3.14$, r – radius of the cylinder in cm, h – height of the cylinder in cm.

To measure the bulk density:

1) Take the undisturbed soil sample with the cylinder;
2) Clean the cylinder from outside (no extra soil outside the cylinder);
3) Put the cylinder with the soil to the oven and let it to dry at 105°C for 12 hours;
4) Weight the dry soil with the cylinder, write the result;
5) Empty the cylinder, clean it (dry) and weight the cylinder, write the result;
6) Now you can calculate the oven dry soil weight by removing from total weight (cylinder + soil) the weight of empty cylinder;
7) Measure the cylinder and find it volume;
8) Calculate the soil bulk density according to the equation above;
9) If you need to know the water content of the sample, you need to weight the soil with the cylinder before putting the sample to the oven. Use the equations below.

Porosity:

Porosity is that portion of the soil volume occupied by pore spaces. This property does not have to be measured directly since it can be calculated using values determined for bulk density and particle density. Finding the ratio of bulk density to particle density and multiplying by 100 calculates the percent solid space, so subtracting it from 100 gives the % of soil volume that is pore space.

Particle density (De) is the volumetric mass of the solid soil. It differs from bulk density because the volume used does not include pore spaces.

1) Fill 100 ml with distilled water; weight the flask with the water.

2) Weight ca 20 g oved dry soil; put it in to the flask (emptied from water).
3) Include to the soil ca 25 ml distilled water, shake and boil slowly ca 10 min to remove air from the soil.
4) After cooling, fulfil the flask with the distilled water and weight again.

De (g/cm^3)= p/v; (2)
where v=n+p–m,
p – oven-dry soil weight (g),
n – flask weight with distilled water (g),
m – flask weight with distilled water and soil (after boiling) (g),
v – volume of soil taken for analysis (cm^3).

Particle density represents the average density of all the minerals composing the soil. For most soils, this value is very near 2.65 g cm^{-3} because quartz has a density of 2.65 g cm^{-3} and quartz is usually the dominant mineral. Particle density varies little between minerals and has little practical significance except in the calculation of pore space.
Particle density can be calculated as follow:
Humus content under 10%:

$$D_e = 2.67 - 0.03x \ , \quad (3)$$

Humus content over 10%:

$$D_e = 2.50 - 0.0163x \ , \quad (4)$$

Histosols:

$$D_e = 1.38 + 0.008x_0 \ , \quad (5)$$

where De particle density (g cm^{-3}); x – soil humus content (%) and x_0 – decomposition level of the peat.

Total porosity can be found:

$$P_{tot}(\%) = \frac{D_e - D_m}{D_e} \times 100 \ ; \quad (6)$$

where, P_{tot} – total porosity (%)
De – density of solid particles (g cm^{-3})
Dm – bulk density (g cm^{-3})

Water content at the measurement, maximum water holding capacity, field capacity

1. Take undisturbed soil sample with the cylinder out of the box, clean cylinder from outside and weight the sample.
2. Cover the bottom of the sample with filter paper and tight with rubber band;

3. Put the sample to the bowl and fulfil the bowl with the water so that only the bottom of the cylinder will be in the water;
4. Cover the cylinders with plastic and leave for 24 hours;
5. Take fulfilled samples off from bowl, set for a 10 min on the moist filter paper, weight the sample;
6. Put the sample on the sandbox (60 hPa suction; pF1.8), leave there for 2 weeks;

7. Take samples from sandbox and weight.
8. Dry the soil sample to a constant weight (24 hours in a hot-air oven at 105 degrees C).
2. Weigh the sample, clean the cylinder. Record the weight of the empty cylinder and calculate the volume of the cylinder.

Calculate the bulk density, porosity and water content of sample (at sampling, maximum, pF1.8).

Calculations of water content

1) Water content on the field:

$$V_f(\%) = \frac{W_f - W_d}{W_d} \times 100 \times D_m, \qquad (7)$$

where, V_f – water content at field (vol%),
W_f – weight of wet (as it was on the field) soil (g)
W_d – weight of oven dry soil (g)
D_m – bulk density (g cm^{-3})
(You will get gravimetric water content if you will not multiply the result with Dm)

2) Maximum water holding capacity:

$$V_{\max}(\%) = \frac{W_w - W_d}{W_d} \times 100 \times D_m, \quad (8)$$

where, V_{max} – maximum water content after saturation (vol%),
W_w – weight of wet soil after saturation and short stay on filter paper (g)
W_d – weight of oven dry soil (g)
D_m – bulk density (g cm^{-3})

3) Water content at field capacity (pF1.8):

$$V_{fc}(\%) = \frac{W_{fc} - W_d}{W_d} \times 100 \times D_m, \quad (9)$$

where, V_{fc} – maximum water content after sandbox at pF1.8 (vol%),
W_{fc} – weight of wet soil after sandbox (g)
W_d – weight of oven dry soil (g)
D_m – bulk density (g cm^{-3})

4) Aeration porosity at field capacity:

$$Paer\ (\%) = P_{tot} - V_{fc}, \quad (10)$$

where,
P_{tot} – total porosity (%)
V_{fc} – soil water content at pF1.8 (%)

Soil structure

Good structure is the basis of good soil quality. It regulates soil aeration and gaseous exchange rates, the movement and storage of water, soil temperature, root penetration and development, nutrient cycling and resistance to structural degradation and erosion. It also promotes seed germination and emergence, crop yields and grain quality. Good structure also increases the window of opportunity to cultivate at the right time and minimises tillage costs in terms of tractor hours, horsepower requirements and the number of passes required to prepare the seedbed.

Assessment:

- Remove first the 0 – 5cm topsoil that contains dense and compacted root system without disturbing soil.
- Remove a 20cm cube of topsoil with a spade.
- Drop the soil sample a maximum of three times from a height of one metre (waist height) onto the firm base in the plastic box. If large clods break away after the first or second drop, drop them individually again once or twice. If a clod shatters into small units after the first or second drop, it does not need dropping again. Do not drop any piece of soil more than three times.
- Part each clod by hand along any exposed fracture planes or fissures.
- Transfer the soil onto the large plastic bag.
- Move the coarsest parts to one end and the finest to the other end. This provides a measure of the aggregate-size distribution. Compare the resulting distribution of aggregates with the three photographs in Figure.

		
Good condition: Good distribution of finer aggregates with no significant clodding.	Moderate condition: Soil contains significant proportions of both coarse firm clods and friable, fine aggregates.	Poor condition: Soil dominated by extremely coarse, very firm clods with very few finer aggregates.

Aggregate stability – slaking test:

Slaking is the breakdown of large, air-dry soil aggregates (>2-5 mm) into smaller sized microaggregates (<0.25 mm) when they are suddenly immersed in water. Slaking indicates the stability of soil aggregates and resistance to erosion, and suggests how well soil can maintain its structure to provide water and air for plants and soil biota when it is rapidly wetted. High soil stability suggests that organic matter is present in the soil to help bind soil particles and microaggregates into larger, stable aggregates. Slaking results in detached soil particles, reduced infiltration and plant available water, and increased runoff and erosion and causes surfake sealing.

Assessment:

Select 3 air-dry aggregates, 4–6 cm diameter. Place soil fragments in the mesh of 1 cm diameter. Observe the soil fragment for 5–10 minutes. Refer to the stability class table below to determine the quality.

		
Good condition: No change, water is clean	Moderate condition: Aggregate breaks down but some ones remain intact on the top	Poor condition: Aggregate breaks down completely into sand grains

Recommended literature

Soils: Visual Soil Assessment (VSA). DEFRA.
http://adlib.everysite.co.uk/adlib/defra/content.aspx?id=000HK277ZX.0HDECKKQLJIF9JD
Youtube – soil structural stability:
http://soilquality.org/indicators/slaking.html

https://www.youtube.com/watch?v=GOos10UyRwY
https://www.youtube.com/watch?v=MOZi33vVsOA
Herrick J.E., W.G. Whitford, A.G. de Soyza, J.W. Van Zee, K.M. Havstad, C.A. Seybold, and M. Walton. 2001. Field soil aggregate stability kit for soil quality and rangeland health evaluations. Catena 44:27-35.

Oxygen Measurement in Soil Samples

Teacher	Prof. Dr. Marian Kazda
Contact	**Universität Ulm, Institut für Systematische Botanik und Ökologie** **Albert-Einstein-Allee 11** **D-89081 Ulm** **marian.kazda@uni-ulm.de**
Methods	Oxygen Measurement in Soil Samples
Key words	oxygen, optical measurement

The optical measurement system (Presens Inc.) allows also an assessment of oxygen saturation in soils. First tests were done in wetlands by Mainiero and Kazda (2005) and a similar approach was applied also in the Summer School "Soil&Water" within the framework of the EduSaPMan network.

The samples collected in standard 100 cm^3 metal cylinders can be taken from different parts of the soil profile and analysed for regular soil parameters. For the assessment of oxygen dynamics, these cylinders were put in plastic cups and flooded until the complete soil was saturated. Optical oxygen sensor was inserted into the soil sample through the plastic lid and changes in oxygen saturation were assessed by the "FIBOX LCD" (Presens Inc., Regensburg, Germany). Periodical measurements allows to detect changes over time and to relate their dynamics to other soil properties.

Fig. 1: Experimental set-up showing the FIBOX LCD device and polymer-based optical oxygen sensors inserted into soil samples.

References

Mainiero, R. and M. Kazda 2005: Effects of Carex rostrata on soil oxygen in relation to soil moisture. Plant and Soil 270: 311-320.

Experimental climate change – precipitation manipulation (*in situ*)

Teacher Dr. Ilja M. REITER, biologist specialized in ecophysiology, research engineer CNRS

Contact **Research federation ECCOREV FR3098, CNRS, Europole Mediterranéene de l'Arbois, Bât CEREGE, BP 80, 13545 Aix-en-Provence**
+33 623485078

Methods Experimental climate change – precipitation manipulation (*in situ*)

Key words
global change, climate change, temperature, precipitation, CO_2, ozone

Experimental method Rain manipulation

- static exclusion – gutters and fixed covers
- dynamic exclusion – roofs, covers
- controlled irrigation – sprinklers, chambers

Learning objectives

Precipitation (rain, snow, hail, dew) and atmospheric humidity are climatic variables that are potentially subjected to change. The expected change strongly depends on the latitude, the annual patterns vary widely. Change for precipitation may be in quantity, frequency, intensity, seasonality, and type. Many biological components of the ecosystem are subjected to changes by altered precipitation/ humidity, i.e. (phyllosphere, plants, fungi, soil fauna).

The design of an experimental device needs to be tuned to the research question, the financial means, and manpower, in particular for the case of ecosystem research, which usually is strongly multidisciplinary as to identify key processes.

A wide range of rain exclusion systems exist worldwide being either static (fixed gutters or covers) or dynamic ((re)movable covers). Further, precipitation may be controlled in chambers or field sites by irrigation (sprinklers, drip-irrigation, flooding); for chambers, defined water quantities usually are applied by drip-irrigation or similar. The most common aim is to alter water availability to plants and animals in the ecosystem. Therefore, it may be necessary that the soil outside the exclusion plot are physically separated from the soil of the exclusion plot itself by means of barriers to limit lateral diffusion or transport of soil and surface water.

The design of a rain exclusion system may alter the environment (aboveground and belowground) beyond the precipitation, through shading, thermal reflection, and change in the boundary layer, soil-intrusive installations or other. The location of the device above or below the canopy (phyllosphere, undergrowth) and grid of the spatial pattern e.g. gutters (distribution/ impact on soil fauna) can play an important role for the experimental outcome.

Description of Methods

Static rain exclusion systems

Static rain exclusion systems can well reduce a defined fraction of precipitation, as e.g. predicted as the mean expected reduction in climate-change modelling exercises. Many rain exclusion systems are based on rain gutters or similar devices, with the proximity of these devices defining the excluded fraction, and which are 'permanently' installed either above or below a canopy. Disturbance of the ecosystem, installation and maintenance costs often are low, as long-life, off-the-shelf equipment can be bought and installed without heavy machinery. This specifically is the case for low canopies, such as agronomical fields, grass(land), and low forests (plantings, regeneration).

Installations may increase the disturbance of the ecosystem for two reasons. Whereas lateral flow of soil and surface water into the exclusion plot seems to be rather negligible for shallow soils such as Karstic Mediterranean sites, it is of major importance to implement lateral barriers or to have a sufficiently large buffer zone around the core plot, in deep soils like in tropical ecosystems, especially when rainfall is high. Further, above-canopy installations may demand strong foundations and supporting structures, that are put in place with heavy machinery, often needing access lanes or similar, all of which are bound to have unwanted impact on the experimental core plots.

As permanent installations are not only changing the precipitation quantity of a plot, but also the distribution of the precipitation, the light and temperature environment, the boundary layer, it is necessary to have similar, but non-excluding structures (e.g. rain-gutters turned upside down) for the control plots. Care must be taken that the non-excluding structures are installed in a way that don't favor unwanted redistribution of water (e.g. water running along these structures to 'collect' at another spot.

Dynamic rain exclusion systems

Dynamic rain exclusion systems are ideal to run experiments where a temporal, seasonal, or frequency variation is required, e.g. a stronger/longer summer drought period or total exclusion period, reduction of rain days/events/ rain types (e.g. thunder storms). These types of installations are far more complex and less autonomous than static systems, as they need a refined protocol to run the experiment, mechanic parts to move the cover or roof, a rain detection device or 'continuous' surveillance to decide to exclude or not, and electricity.

As some

Equipment and installation costs are higher, and the provision of electricity can be a major challenge for remote sites. The complexity makes these systems more prone to experimental errors and malfunctioning, need for maintenance and manpower demand, thus needing more financial support than static systems, and biasing the approach to run long-term experiments. Some of the ecosystem processes are highly sensitive to precipitation, especially the ones related to microorganisms like litter decomposition and processes in the phyllosphere. Here, total exclusion of rain events is a crucial criteria for the exclusion protocol. This is currently still difficult to fully automatize. A somewhat comfortable solution are network-capable roof/cover-controlling devices having internet access and man-machine interfaces for a manual control. The control relies heavily on meteorological predictions or on-site presence, which renders the closing and opening of covers even more difficult

for remote sites. However, unique results may be acquired with this method, in particular for question on microbiology and micro-meso fauna.

Irrigation systems

Northern latitudes are bound to have higher annual rainfall in the 'near' future, other regions will have changes in the seasonal rainfall pattern, more thunderstorms or other. Whereas for some parts of the growing season this may be of an advantage, an excess of water in the soil can lead to anaerobic conditions, which are unfavorable to root systems and have strong effects on aboveground physiology.

To provide 'natural' and homogeneous precipitation (rain) to experimental plots is still an experimental challenge, as e.g. varying rain intensity, or drop size over a certain range demands dedicated equipment. Water quality (ions) is an issue also as it's different from tap water. When irrigation is applied to the ground level (drip irrigation, flooding) or to the above-canopy level (sprinklers), e.g. the phyllosphere is not affected in the same manner. Further, the distribution pattern of the irrigation is of importance as 'one-sided' wetting of the rooting system, may still provoke strong signaling from the non/less-watered rooting system to the rest of the plant.

To consider,

with these methods we try to alter water availability to plant, microbes, and fauna. It is therefore important to evaluate, how the experimental treatment is 'seen' by the organism or the investigated process of interest. Water stock and loss can be low or high and thus humid conditions more or less persistent (e.g. in soils, litter or the phyllosphere). This needs to be taken into account when designing the protocol for the experimental treatments. Further, the treatments can change the relation between the water scenarios and the physiological processes that are linked, as due to changes in the overall resource state (e.g. energy) of the organism. The treatments add to the *in natura* variability of environmental conditions, which is one of the reasons why long-term experiments are important to understand changes in ecosystem functioning.

Recommended reading

Beier *et al.* (2012) Precipitation manipulation experiments − challenges and recommendations for the future. Ecology Letters 15:899-911

climmani.org - ClimMani Climate Change Manipulation Experiments in Terrestrial Ecosystems: Networking and Outreach

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