

# Storage of Soil Samples for a Retrospective Biological Assessment in the German Environmental Specimen Bank



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# Background

The German Environmental Specimen Bank is an important component of ecological environment observation in Germany. One objective is the long-term monitoring of soil quality in selected ecosystems to detect changes caused by organic and inorganic contaminants. So far emphasis has been laid on chemical analyses of the sampled soils: Different samples are routinely taken from selected ecosystems all over Germany. Immediately after sampling, the soil samples are sieved to < 2 mm and stored over liquid nitrogen. Frozen samples are prepared and homogenized for further storage. It is now planned to extend this resource to additionally include biological parameters. For this purpose, methods for the conservation and reactivation of soil samples are needed, which do not change the soil microflora and mesofauna or which cause only calculable changes. We performed preliminary investigations to compare potential effects of various soil conservation and reactivation methods on selected soil organisms.

# **Conservation and reactivation methods**

Conservation: (I) -18 °C, (II) -18 °C and transfer into liquid nitrogen after one week, (III) shock freezing (liquid nitrogen) Reactivation: (I) fast (one day at 20 °C), (II) slow (3 d 4 °C; 3 d 20 °C)

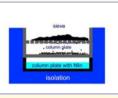
Measuring parameters:

Microflora: activities: respiration, nitrification, DMSO-reduction; structure: PLFA, T-RLFP

Nematodes: biocoenosis







#### Results

Assessment of the influence of different tests on the investigated biological soil parameters upon long-term storage (3 months)

Freezing and storage	-18	-18 °C		-18 °C/liquid nitrogen		liquid nitrogen	
Thawing	fast	slow	fast	slow	fast	s lo w	
Microbial activity							
Basal respiration	+	+	+	+	+/-	+	
SI-respiration	+	+/-	+/-	+/-	+/-	+/-	
Pot. NH₄*-oxidation	+/-	+/-	+/-	+/-	+/-	+/-	
DMSO-reduction	+/-	+/-	+	-	+	-	
Microbial biocoenosis							
PLFAs total	+/-	+/-	•	-	+/-	+/-	
PLFA Gram -: Gram +	-	-	-	-	-	-	
T-RFLP	-	+/-	-	+/-	-	+/-	
Nematode biocoenosis							
Total abundance	+/-	+/-	-	-	+/-	+/-	
Dominance ")	-	-	-	•	-	-	
Family occurrence *)	+	-	+	+/-	+	-	

\*) Due to insufficient data for long-term storage data on short-term storage were also considered. +: Comparable with control; +/-: soil-dependent effect; -: variation from control (as - is marked: microbial activity, PLFA: statistically significant and > 25 % variation from control; nematode biocoenosis > 25 % variation; T-RFLP: significant variations indicated)

#### Recommendations

- > Conservation: All tested procedures are applicable
- > Reactivation: Fast thawing
- Results on activities: Comparable to fresh soil
- Results on structure: Interpretation should be restricted to diversity (presence/absence of individual PLFAs, species)



# Conclusion

- ✓ The procedure commonly used for the conservation of soil samples is also suitable in the context of the analysis of biological parameters.
- Despite minor limitations concerning the interpretation of results, it is expected that trends of biological parameters can be pointed out using soil samples stored in the Environmental Specimen Bank.
- ✓ Further investigations concerning the weather conditions at the date of sampling revealed that the water content during storage (10 %, 50 %, 90 % of WHC<sub>max</sub>) had no significant effect on the results.