Formation of biogeochemical interfaces in soils as controlled by mineral and organic components

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Abstract
The formation of soil interfaces is controlled by the type of particle surfaces present and the assemblage of organic matter and mineral particles. The formation of interfaces was studied with artificial soils which were produced in a long-term biogeochemical laboratory incubation experiment (3 and 6 months). The experiment used clay minerals, iron oxides and charcoal as major model components controlling the formation of interfaces because they exhibit high surface area and microporosity. Soil interface characteristics are analyzed in relation to microbial community structure and function that developed after 6 months of incubation. Already after 6 months of incubation the artificial soils exhibited different properties in relation to their composition. Major effects are observed for artificial soils formed in the presence of montmorillonite, ferrihydrite and charcoal.

Key Words
Biogeochemical interface, soil formation, clay, iron oxides, char, microbial community structure and function, DSC

Introduction
The biogeochemical interfaces of soils are a dynamic and hierarchically organized system of various organic and inorganic constituents and organisms, the spatial structure of which defines a large, complex and heterogeneous interface (Totsche *et al*. 2010). Major solid phase constituents of soils are rock fragments, minerals like quartz, carbonates, clay minerals and the (hydr)oxides of iron, manganese, and aluminium. Carbonaceous materials including charred organic carbon of biogenic origin (Biochar) and, of course, the soil organic matter (SOM) are a second major ingredient of soils. These components are of different provenience, complexity and molecular size. Their arrangement during pedogenesis results in the spatial and temporal development of complex biogeochemical interfaces in soils. The natural diversity in soil structural components leads to isolated areas of enhanced biogeochemical activity, popularly known as hot spots, and at the same time zones of lower biogeochemical activity in soils. The existence and importance of such a differentiation of active and inactive spots in soils, i.e. a patchy distribution of soil properties, are recognized in the ecological community, but a solid understanding of the underlying mechanisms that produce this differentiation is still lacking. We hypothesize that the formation of soil interfaces is controlled by the type of particle surface(s) present and the assemblage of organic matter and mineral particles. We consider clay minerals, iron oxides and charcoal as major components controlling the formation of interfaces because they exhibit high surface area and microporosity. The objective of our work is to characterize the heterogeneous architecture of soil interfaces as it develops depending on the type of particle surface present and to link it with the structure and function of the microbial communities. The formation of interfaces is studied in batch incubation experiments with inoculated artificial soils consisting of model compounds, thus artificial soil materials comprised of model compounds with increasing complexity of interfaces were created.

Methods
Artificial soil
The components used for the artificial soil incubation experiment were quartz sand, clay minerals (illite and montmorillonite), iron oxide (ferrihydrite), aluminium hydroxide (boehmite) and charcoal (Table 1). A microbial inoculum for this experiment was obtained from the water extractable community of a Eutric Cambisol from Ultuna, Sweden. Organic matter obtained from sterilized manure was used as C and nutrient

source during incubation. The soils were incubated at a water content of 60% of the water holding capacity. Soil solution consisted of 0.01 M $CaCl_2$. Samples were obtained after 3 days (t=0), 3 months (t=1) and 6 months (t=2) of incubation.

C and N content
Carbon and nitrogen content were determined after 3 and 6 months of incubation.

Table 1. Composition of the artificial soils

	A	B	C	D	E	F	G	H
Manure	X	X	X	X	X	X	X	X
Quartz	X	X	X	X	X	X	X	X
Montmorillonite	X			X	X			
Illite		X		X		X	X	X
Ferrihydrite			X			X		X
Boehmite							X	
Charcoal					X			X

Differential scanning calorimetry
Six months incubated artificial soil samples were analyzed in a ramp of -50°C to 550 °C with 10 K min^{-1} heating and synthetic air as purge gas (50 mL min^{-1}). DSC measured the energy transformation in the samples occurring during heating. In this experiment, the combustion process was monitored. This analysis gives insight into the development of organic matter quality from the manure in the soils under the influence of various minerals and charcoal with respect to thermal stability.

Microbial community and function
Microbial communities were followed by DGGE of 16S rRNA gene fragments amplified from directly extracted DNA. Samples were taken at Day 1, 9, 31 and 90. Microbial community structure was analyzed by fingerprinting of PCR products of the 16S rRNA genes from DNA extracted from soil. As an example for microbial community function, degradation of wheat litter was investigated in a batch incubation experiment using the different artificial soils that had been incubated for 3 months. As an indicator abundance pattern of the alkane monooxygenase, gene (*alkB*) which catalyzes the degradation of plant waxes and other alkane containing substances was chosen.

Results and Discussion
C and N stabilization
C contents in soils (figure 1 left) E and H are higher, due to the presence of charcoal. Already after 6 months of incubation, the different artificial soil materials show a large difference in C stabilization, although the amount of C substrate added as manure was similar for all soils (figure 1 right). C losses are specifically low in the presence of oxides ferrihydrite (soil F) or boehmite (soil G). Losses are high from both soils containing charcoal (E and H), indicating that the charcoal has a stimulating effect on C mineralization or is itself mineralized already in the beginning of the incubation. Nitrogen was lost from the soil at a relatively constant ratio to carbon loss with a ratio of C loss/N loss between 8 and 12.

Differential scanning calorimetry
The DSC thermograms show –besides a weak endotherm below 100°C indicating water evaporation- pronounced exotherms in the temperature region I (200°C-400°C) and region II (400-550°C) (figure 2). This may in a first attempt be interpreted as combustion of two types of organic matter differing in thermal stability. The two most striking observations are: (i) In the exotherms of region II distinctly structured peaks occur only for the samples A and D containing montmorillonite, (ii) the exotherm in region II differs significantly between the samples containing charcoal and the samples without charcoal.

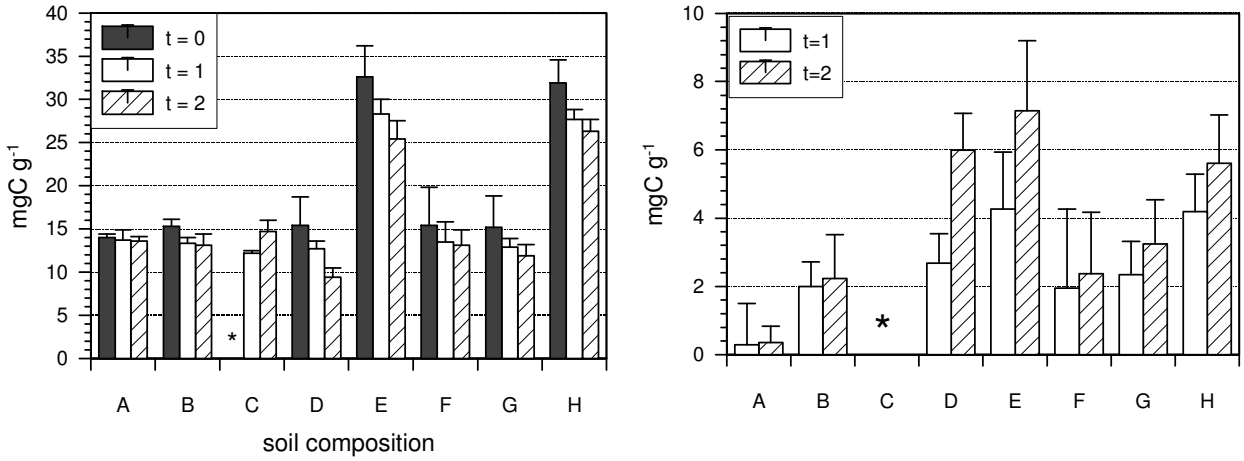

Figure 1. Carbon concentration of the artificial soils (left) after 3 days (t=0), 3 (t=1) and 6 (t=2) months of incubation, and amount of carbon lost from the soils calculated from the carbon concentration at t=0 minus the carbon concentration at t=1 and t=2 respectively (right). Results and standard deviations were calculated from the three replicates for each soil. * not determined.

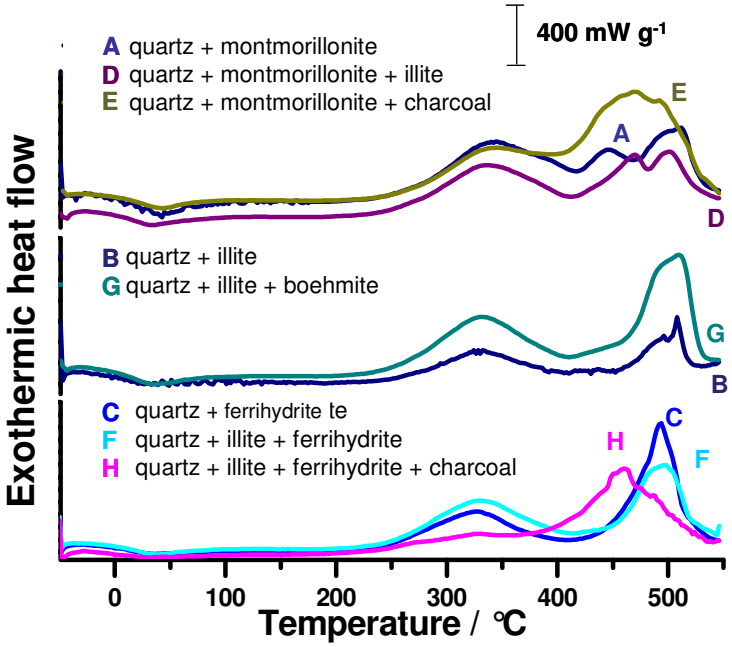

Figure 2. Exothermic heat flow from selected artificial soils measured by differential scanning calorimetry.

Microbial community

Soil-like bacterial communities' patterns could already be observed 9 days after inoculation. Charcoal was found to strongly influence the reassembly of bacterial communities. UPGMA clusters analysis of communities' profiles showed that separate clusters were always formed for those samples with charcoal. Effects of montmorillonite, illite and iron oxides on microbial communities could clearly be observed until day 90. Bacterial communities of samples with montmorillonite differed from those of the corresponding samples with illite. Separated clusters for samples with iron oxides were also formed in betaproteobacterial and actinobacterial communities, but no clear difference was observed between ferrihydrite and boehmite.

Alkane monooxygenase

Already three months after the incubation of the different soil mixtures clear differences in the abundance of the *alkB* gene during litter degradation are visible in the different artificial soil mixtures at different interfaces (figure 3). Mainly those artificial soils that contain montmorillonite have a tendency to lower alkB numbers at the soil-litter interface. The obtained numbers are already comparable to values that can be found in fully developed soils.

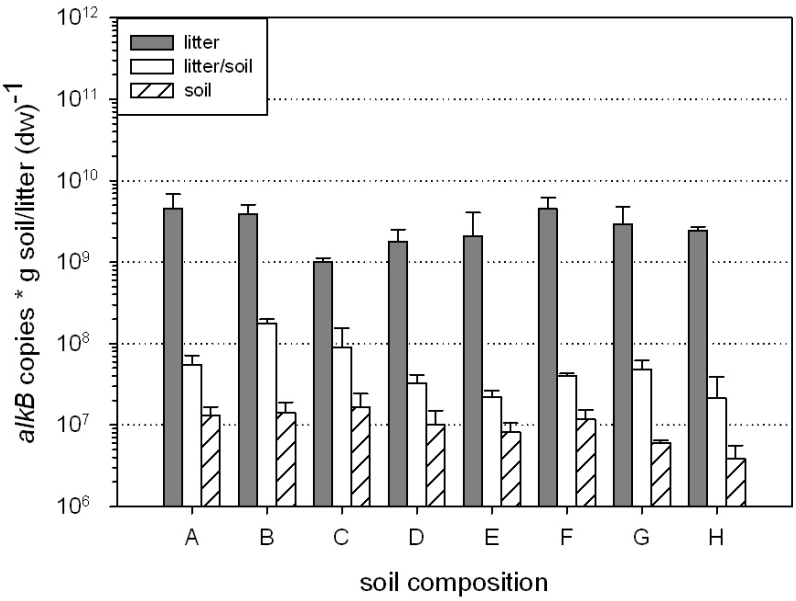

Figure 3. Abundance of the alkane monooxygenase gene (*alkB*) during degradation of wheat litter at different interfaces (litter, litter/soil, soil) after two weeks of incubation in the different artificial soils at t=1. The bars indicate standard errors (n=3).

Conclusions

Already after 6 months of incubation, the different artificial soil materials show a large difference in C stabilization, although the amount of C substrate added as manure was similar for all soils. At the current stage of research, a contribution of charcoal to OM stability cannot be excluded but has to be shown in further analyses. The preliminary results suggest, however, that the minerals present during formation of biogeochemical interfaces, determine stabilization and quality of organic matter.

The presence of charcoal and mineral composition had a clear effect on the development of the microbial community after only 3 months of incubation. Furthermore, clear differences in the microbial community function, expressed in the abundance of the *alkB* gene, which is involved in the degradation of plant waxes during litter degradation are visible in the different artificial soils at different interfaces. The presence of an expandable clay mineral (montmorillonite) and of ferrihydrite had clear effects on the formation and properties of the biogeochemical interface in the artificial soils, as well as the presence of charcoal. These differences in the biogeochemical interface formation also lead to different sorptive properties of the interface for phenanthrene (Pronk *et al.* 2010).

References

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