Organic matter and soil microorganisms – Investigations from the micro- to the macro-scale

Dedicated to Univ. Prof. Dr. Dr. h.c. Winfried E. H. Blum on the occasion of his 60th birthday

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Organische Substanz und Bodenmikroorganismen – Untersuchungen auf unterschiedlichen Skalenebenen

Univ. Prof. Dipl.-Ing. Dr. Dr. h.c. mult. Winfried E. H. Blum zu seinem 60. Geburtstag gewidmet

1. Introduction

Soils are extremely heterogeneous entities (COLEMAN and CROSSLEY, 1996). The spatial variability of soil organic matter dynamics involving various soil microbial processes can be manifested at the microscale, plot scale, landscape- and regional scale (PARKIN, 1993). The variability of chemical and microbiological properties of agricultural and forest ecosystems is well known at the plot scale (BONMATI et al., 1991; VAUGHAN et al., 1994; BAHRI and BERNDTSSON, 1996; STORK and DILLY, 1998); much less information is available on the microscale. In soil ecology, hierarchical approaches have gained interest over the last two decades (ALLEN and STARR, 1982; O'NEILL et al., 1986). TISDALL and OADES (1982) described the hierarchical nature of processes responsible for structuring soils. In many soils, microorganisms and/or microbially mediated processes contribute to forming aggregate structure across five orders of magnitude, beginning at the level of clay particles (0.2 µm), through fungal and plant debris (2 µm and 20 µm), up to a 2-mm diameter soil crumb. A hierarchical approach to soil organisms that considers three different

Zusammenfassung

Dieser Artikel gibt einen Überblick über neuere Untersuchungen zur organischen Substanz und zu Bodenorganismen, die auf unterschiedlichen Skalenebenen durchgeführt wurden. Es wird der Frage nachgegangen, ob eine Übertragbarkeit von Ergebnissen, die auf kleinskaligem Niveau gewonnen wurden, auf eine höhere Ebene möglich ist. Pico- und nano-skalige Untersuchungen haben bisher zur Ermittlung der Struktur und der chemischen Zusammensetzung der organischen Substanz und der Mikroorganismen sowie deren Wechselwirkungen beigetragen. Da der Abbau der organischen Substanz im Boden sehr stark von der räumlichen Verteilung dieses Substrates abhängig ist, haben Untersuchungen auf der mikroskaligen Ebene in den letzten Jahren zum Verständnis des Mechanismus des Kohlenstoff- und des Stickstoffkreislaufes beigetragen. Mikroskalige Untersuchungen wurden häufig an unterschiedlichen Aggregatfraktionen oder spezifischen Mikrohabitaten durchgeführt. Auf der Plotebene wurde hauptsächlich der Einfluß der Bodenbewirtschaftung auf den Umsatz der organischen Substanz untersucht. Bei diesen Untersuchungen wurde u.a. die Quantität und Qualität der Ernterückstände und ihre zeitliche und räumliche Verteilung und das Verhältnis von ober- und unterirdischer Biomasse in Abhängigkeit vom Nährstoffangebot getestet. Die vorliegenden Daten werden in den letzten Jahren auch verstärkt zur Modellierung auf der regionalen Skala verwendet. Geostatistische Methoden konnten räumliche Zusammenhänge zwischen bodenbiochemischen Prozessen und Standortseigenschaften auf der Landschaftsebene ermitteln. Aus der Literaturübersicht kann der Schluß gezogen werden, daß Untersuchungen auf den einzelnen Skalenebenen nur spezifische Fragen beantworten können und daß ein vollständiges Verständnis eines Ökosystems nur durch die integrative Betrachtung von Untersuchungen aller Skalenebenen gewonnen werden kann.

Schlagworte: Organische Substanz, mikrobielle Biomasse, Bodenenzyme, Skalierung.

Summary

The aim of the present study is to provide an overview of recent investigations on soil organic matter and soil microorganisms at different hierarchical levels (pico-, nano-, micro-, macro- and regional-scale) and to elucidate whether results at any one level can be up-scaled to higher hierarchical levels. Pico- and nano-scale investigations are used to reveal the structure and chemical composition of organic substances and microorganisms as well as the interaction between biota and humic substances. Since the decomposition rate of residues in soils depends much on their location within the soil, studies on the micro-scale enable researchers to delineate the mechanisms driving C and N turnover. During the last decade, micro-scale investigations concentrated either on aggregates yielded by different physical separation procedures or on different microhabitats characterized by high turnover of organic material. Plot-scale investigations were mainly perfomed to understand the influence of soil management on soil organic matter turnover; the parameters considered were changes in the quantity and quality of plant residues entering the soil, their seasonal and spatial distribution, the ratio between above- and below-ground inputs, and changes in nutrient inputs. In addition, many plot-scale investigations of chemical and microbiological properties from the range of different soil ecosystems provide not only a useful database to explain potential changes within a single field or plot, but also a database with which to model processes on the regional scale. Landscape-scale analyses by geostatistical methods are now recognized as a useful tool for identifying and explaining spatial relationships between soil biochemical processes and site properties. In conclusion, investigations on each level of resolution may answer specific questions, but a complete understanding of a soil ecosystem requires an integrative view of investigations at all levels of resolution.

Key words: Organic substances, microbial biomass, soil enzymes, scaling.

levels is outlined in BEARE et al. (1995). In the aggregatusphere, bacteria, amoebae, and certain nematodes have varying degrees of success in gaining access to the prey biota of interest, whereas at a coarser level of resolution interactions between microbes and the fauna in the rhizosphere or at the soil litter-interface have been documented. At the next level of resolution, which extends from many centimeters to several meters across the landscape, the activities of macrofauna such as earthworms or burrowing beetles, influence microbial, chemical and physical processes of soils. The hierarchical view of COLEMAN et al. (1992) and COLEMAN and CROSSELY (1996) covers the range from a molecular level up to watersheds and beyond (Figure 1). The conceptual diagram clearly shows that investigations on each level of resolution may answer specific questions, but that a complete understanding of a soil ecosystem requires an integrative view of investigations at all levels of resolution.

Therefore, the aim of the present study is (1) to give an overview of recent investigations on soil organic matter and soil microorganisms at different hierarchical levels (pico-, nano-, micro-, macro- and regional-scale) and (2) to elucidate whether results at the single level can be up-scaled to higher hierarchical levels.

2. Pico- and nano-scale investigations

Pico- and nano-scale investigations are used to expose the structure and chemical composition of organic substances and microorganisms as well as the interactions between biota and humic substances. The study of soil microorganisms can identify the organisms, unravel their relationships, determine their numbers, and measure the rates of physiological processes (PAUL and CLARK, 1996). The last decade has been marked by a rapid increase in the number, reliability and sensitivity of techniques in soil microbiology. Beside process-oriented studies and physiological analyses (e.g. plate counts, enzymes, thymidine uptake), the estimation of signature molecules such as phospholipids, ergosterol, respiratory quinones and ATP content can improve our understanding of the biotic activity, biomass and structure of the microbial community (KATAYAMA and FUJIE, 2000). For example, the isoprenoid quinones present in the membranes of mitochondria or chloroplasts are essential components in electron transport systems of most soil microorganisms. Since many such microorganisms have only one major quinone and since the major quinone does not change with their physiological condition, this biomarker is a high potentially powerful indicator of microbial diversity.

Whereas signature molecules rapidly decomposed after the death of soil microorganisms, physical and chemical

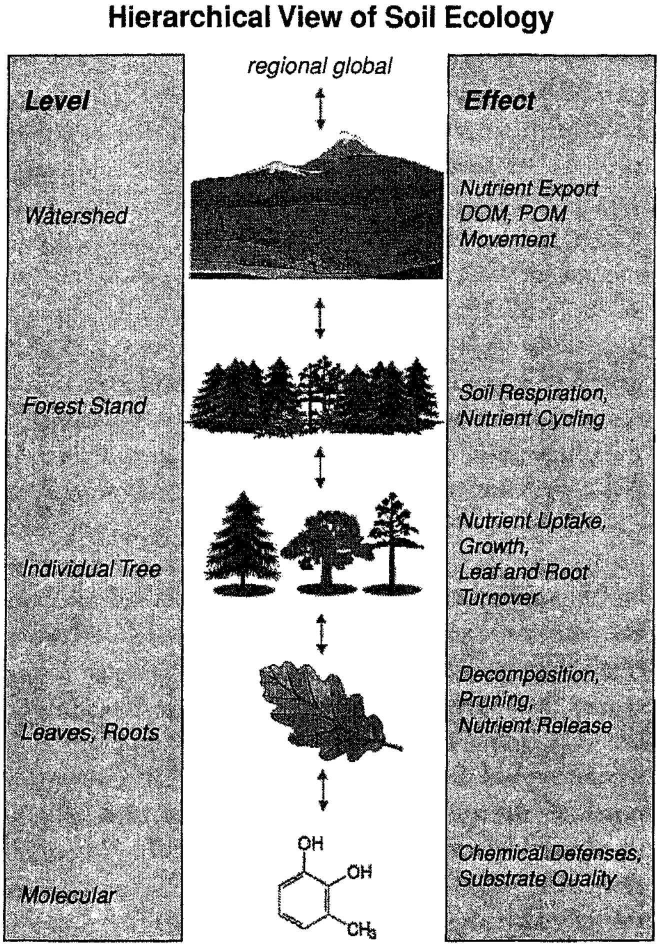

Figure 1: Hierarchical view of soil ecology ranging from a molecular level up to landscape level. Feedbacks from vegetation and soil interaction can reverberate back down to the molecular level, flows are potentially bidirectional (modified after COLEMAN and CROSSLEY, 1996)

Abbildung 1: Hierarchische Gliederung bodenökologischer Untersuchungen von der molekularen bis zur Landschaftsebene. Wechselwirkungen zwischen Pflanzen und Böden können sich auch auf die molekulare Ebene auswirken; Flüsse können in beide Richtungen gelesen werden (modifiziert nach COLEMAN und CROSSLEY, 1996)

protection of humic substances are responsible for the slow degradeability of soil organic matter according to the model of JENKINSON and RAYNER (1977). Physical protection will predominantely be discussed in the following chapter. Chemical protection of organic substances is apparently be a major factor for long turnover half-lives of hundreds to thousands of years, although recent studies show that physical protection of SOM may result in similarly low turnover rates as chemical stabilization (HAIDER, 1999). The degradeability of soil organic matter depends on its chemical composition. In the past considerable efforts were undertaken to characterize SOM and specifically humic substances by various analytical techniques. Especially ^{13}C-NMR (e.g. KROSSHAVN et al., 1992), pyrolysis-soft ionization mass spectrometry (e.g. SCHULTEN and SCHNITZER, 1992) and FT-IR spectroscopy (e.g. HABERHAUER and GERZABEK, 1999) and to some extent enzymatic methods (JAHNEL et al., 1993) have advanced our knowledge about the structural details of humics, their predominant functional groups and elemental composition. We know that humic substances contain mainly aromatic structures (phenolic structures, lignin monomers and dimers), aliphatics (n-alkanes, n-alkenes, fatty acids, n-alkyl monoesters, mono- and polysaccharides) and N-compounds (SCHULTEN and SCHNITZER, 1992) in different proportions governed by origin and history of SOM. Little information has been obtained to date on their conformational structures, although efforts were recently undertaken to investigate the macromolecular structure and the impact of environmental factors (CONTE and PICCOLO, 1999; MYNENI et al., 1999). Further insights can be envisaged by using molecular modelling techniques (e.g. SCHULTEN and SCHNITZER, 1997). These emerging methods and the different approaches for deriving 2- and 3-dimensional models are discussed in detail in GERZABEK et al. in this issue.

In the late 1980s, the idea of micelle-like structures of humic substances consisting of smaller hydrophobic molecules emerged (WERSHAW et al., 1986); this contrasted with the previous dominant theory of macromulecules derived from SOM breakdown and subsequent polymerization. Returning to degradeability, the hydrophobic character of the micelles might play an important role. Figure 2 shows the result of a laboratory soil incubation experiment conducted with a ^{13}C-labelled model substance (2-decanol). The addition of humic acids (HA) of different hydrophobicity, as characterized by ^{13}C NMR spectroscopy, resulted in varying mineralization of the ^{13}C-labelled substance. Increasing hydrophobicity – HA from compost was less hydrophobic than HA from lignite – retarded the mineralization of the model substance (PICCOLO et al., 1999). These findings might be important for controlling mineralization and for characterizing SOM in the future.

In conclusion, pico- and nano-scale investigations helped to clarify the structure and chemical composition of soil microorganisms and organic substances as well as the interactions between biota and humic substances. These results boost our understanding of chemical and biological processes and structures at larger scales. A very impressive example is the use of molecular techniques (phospholipid

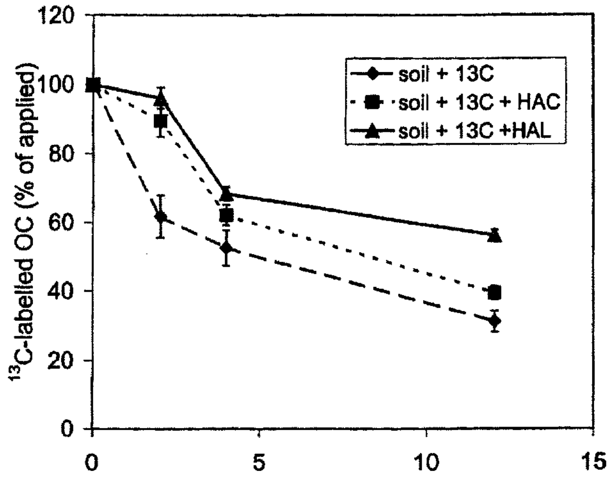

Figure 2: Mineralization of ^{13}C-labelled organic carbon (OC) as influenced by addition of humic acids from compost (HAC) and lignite (HAL). Mean values and standard deviations (n = 3). Redrawn from PICCOLO et al. (1999)

Abbildung 2: Der Effekt von Huminstoffen gewonnen aus Kompost (HAC) und Lignit (HAL) auf die Mineralisierung von ^{13}C-markierter organischer Substanz (OC). Mittelwerte und Standardabweichungen (n = 3). Umgezeichnet aus PICCOLO et al. (1999)

fatty acids and DGGE profiles) that explain the distribution of the soil microbial communities at the micro-scale (KANDELER et al., 2000).

3. Micro-scale investigations

The decomposition rate of residues in soils depends much on their location within the soil (LADD et al., 1996). Therefore, studies on the micro-scale enable researchers to delineate the mechanisms driving C and N turnover. During the last decade, micro-scale investigations concentrated either on aggregates yielded by different physical separation procedures or on different microhabitats charcterized by high turnover of organic material.

Physical separation of organo-mineral particles and micro-aggregates is typically based on two different concepts: i) separation according to particle density and ii) separation according to particle size. Density fractionation in heavy organic liquids yields light fractions, which are considered to be highly available, less composed material, whereas the heavy fractions are considered to be associated with mineral compounds, representing more processed and humified organic material (CHRISTENSEN, 1992). Size fractionation releases macro-organic matter and organic matter associated with mineral particles forming organo-mineral particles and micro-aggregates of contrasting structure, function and stability. Size fractions are typically gained by wet sieving and particle sedimentation or centrifugation, which accelerates sedimentation processes. Both concepts of particle separation are commonly based on physical disruption of soil aggregates using sonication (TISDALL and OADES, 1980; CHRISTENSEN, 1986; STEMMER et al., 1998) or mechanical treatments (GUPTA and GERMIDA, 1988; JOCTEUR MONROZIER et al., 1991, LENSI et al., 1995). Generally, soil organic matter quality ranges from hardly decomposed plant debris with a high C/N ratio and low density located in the coarse fractions to highly processed humic substances forming organo-mineral compounds with narrow C/N ratio and high density found in clay separates. The decrease in the C/N ratio with decreasing particle size and increasing particle density reflects the state of organic matter mineralization and humification. This pattern controls the soil microbial and enzymatic properties of organo-mineral particles and micro-aggregates isolated from mineral soils. Figure 3 illustrates a characteristic distribution pattern of soil organic matter, C/N ratio and microbial properties within particle size separates yield from a minimum tilled soil. Commonly, much of the soil microbial biomass is associated with the smaller sized fractions (fine silt and clay); the highly variable amount of microbial biomass within the coarse fractions strongly depends on the quantity and quality of the macroorganic matter located there (JOCTEUR MONROZIER et al., 1991; KANDELER et al., 1999a; STEMMER et al., 1999). Investigations on the structural diversity of the microbial community using the PLFA pattern and 16s rRNA gene fragments gave strong evidence that the microbial biomass within the clay fraction was mostly due to soil bacteria. In contrast, a high percentage of fungal-derived PLFA was found in the coarse sand fraction containing particulate organic matter (KANDELER et al., 2000). Enzyme activities of size fractions largely depend on the enzyme investigated and the fractionation procedure (LENSI et al., 1995; LADD et al., 1996). STEMMER et al. (1999) and KANDELER et al. (1999a) showed that the microbiological properties and enzyme activities of the coarse-sized fractions are strongly influenced by tillage practices; these fractions are therefore a valuable indicator for organic matter input and management changes. Note that, generally, isolated size or density separates do not reflect the complex interactions within an intact and highly structured soil aggregate where e.g. macroorganic matter is closely covered by clay-sized organo-mineral particles.

During the last decade, several attempts were made to investigate undisturbed microhabitats charcterized by high

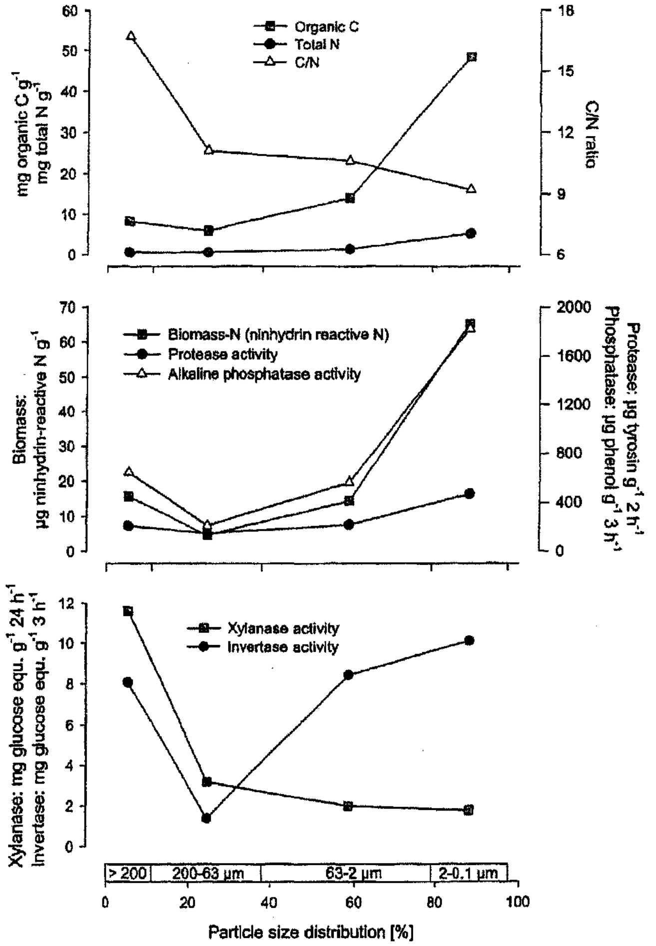

Figure 3: Distribution of soil organic matter and microbiological properties within particle size separates of a fine-sandy loamy Haplic Chernozem, which was applied to minimum soil tilling over a 7-year period

Abbildung 3: Verteilung der organischen Substanz und mikrobieller Eigenschaften auf die Korngrößenfraktionen eines seit 7 Jahren minimal bearbeiteten (Frässaat), feinsandig-lehmigen Tschernosems

turnover of organic material. These microhabitats may be a relatively small subset of total soil volume (COLEMAN and CROSSLEY, 1996). For example, the rhizo-, drilo-, und detritusphere are important microhabitats presenting "hot spots" of microbial activity (BEARE et al., 1995). In many cases, complex experimental designs and/or samples strategies were necessary to obtain microscale soil samples for chemical and microbiological analyses. TARAFDAR and JUNGK (1987), TARFDAR and MARSCHNER (1994) and GAHOONIA and NIELSEN (1991) used 0.1 – 0.2 mm slices of soil cores that were separated from the root mat by a 53 µm nylon mesh to investigate the nutrient uptake of plant and microbial processes in the rhizosphere. The abundance of rhizosphere microorganisms and their activities decreased within the range of several millimeters to levels found in the bulk soil. The expansion of the rhizosphere depended on the excretion of easily degradable organic substances by roots, mass flow and the diffusion of dissolved organic substances used as substrates by soil microorganisms. In addition, micro-scale slices (0.2 mm) gained by a freezing microtome were used to characterize gradients of dissolved organic matter and soil microbial processes at the soil-litter interface (KANDELER et al., 1999b). The scale of the soil-litter interface ranged from 1.1 – 1.3 mm, in which the gradients of protease, xylanase and invertase activities followed an exponential function ($y=c + \exp(b_0 + b_1x_1 + b_2x_2)$). The authors explained their results by the high local release of substrates driving C and N turnover within the 1–2 mm from the surface of the litter. The investigations on the drilosphere involved larger scales than those on the rhizo- and detritusphere. For example, TIUNOV and SCHEU (1999) showed that organic carbon and toal nitrogen increased in burrow walls of *Lumbricus terrestris* L. by factors of 1.8-3.5 and 1.3-2.2 at distances of 0-4 mm and 8-12 mm from earthworm burrows. The high specific respiration (qO_2) and the fast growth response to nutrient additions indicated that the microbial community in the burrow walls contained a larger fraction of metabolically active microorganisms, adapted to continuous resource additions by earthworm faeces and mucus. The authors concluded that these burrows are stable microhabitats which sustain a large and active microbial community. Therefore, the activity of soil microorganisms probably plays an important role.

In conclusion, micro-scale investigations over the past few years have improved our understanding of mechanisms driving C- and N-turnover. Since the areas of high activity in soil are heterogeneiously distributed within the soil matrix, hot spots of activity may be <10% of the total soil volume, but may represent >90% of the total biological activity in most soils worldwide (BEARE et al., 1995). Until now, the up-scaling of data from the micro- to the plot- or regional scale remains difficult because spatial distribution patterns at these scales are still incompletely known.

4. Plot-scale investigations

During the last decades, investigations at the plot scale were the dominant sampling strategy for soil chemical and biological studies. Accordingly, a representative number of soil samples were taken from the study site (arable land, grassland, forest) and combined to a bulk sample or treated as separate samples. Usually, random samples were combined from homogeneous, representative areas that were described by uniform soil type, soil texture and habitat characteristics (ÖHLINGER et al., 1993; ÖHLINGER, 1994, 1996). Whereas samples of agricultural soils were mainly taken from specific soil depths (e.g. 0–20 cm or 0–30 cm layers), samples of forest soils were taken from specific soil horizons (e.g. litter horizon, A horizon). Many plot-scale investigations elucidated organic matter turnover, the content of microbial biomass and microbial processes of different vegetation types and soil types. Table 1 summarizes acitivities of enzymes involved in carbon-, nitrogen-, phosphorous- and sulphur-cycling.

Table 1: The response of dehydrogenase and enzyme activities involved in carbon-, nitrogen-, phosphorus- and sulphur-cycling to the type of vegetation and soil (n.d. not determined)
Tabelle 1: Einfluß des Vegetations- und des Bodentyps auf die Dehydrogenaseaktivität und Enzymaktivitäten des Kohlenstoff-, Stickstoff-, Phosphor- und Schwefelkreislaufes

soil enzyme activity	range of activities	vegetation / soil type	reference
xylanase activity mg glucose g^{-1} 24 h^{-1}	13 – 24	spruce forest / n.d.	VON MERSI et al. (1992)
	0.28 – 8.0	beech forest / n.d.	ZECHMEISTER et al. (1991)
	3 – 17	agricultural land / n.d.	TABATABAI, BREMNER (1969)
	1.8 – 3.0	grassland / Orthic Luvisol	KANDELER, EDER (1993)
	0.68 – 1.02	agricultural land / Eutric Cambisol	KANDELER, MURER (1993)
	0.75 – 2.00	agricultural land / Haplic Chernozem	KANDELER et al. (1999c)
	0.38 – 1.15	crop rotation /Phaeocem, Lithosol,Cambisol	KANDELER et al. (1996)
	0.24 – 1.83	agricultural land / Haplic Luvisol, Entisol	STEMMER et al. (1999)
ß glucosidase µg p-nitrophenol g^{-1} h^{-1}	20 – 55	grassland / Pachic Arguistoll	AJWA et al. (1999)
	62 – 98	agricultural land / Argixeroll	BURKET, DICK (1998)
	36 – 160	forest / Haplohumult	BURKET, DICK (1998)
	70 – 130	crop rotation / Fluvisol	CURCI et al. (1997)
	130 – 310	crop rotation / Hapludalf	DENG, TABATABAI (1996)
	71 – 86	crop rotation / Pachic Ultic Argixerolls	MILLER, DICK (1995b)
protease activity µg tyrosine g^{-1} 2 h^{-1}	150 – 520	agricultural land / Haplic Chernozem	KANDELER et al. (1999c)
	224 – 514	pasture / Typic Dystrochrept	HAYNES, WILLIAMS (1999)
	315 – 468	crop rotation / Phaeocem, Lithosol,Cambisol	KANDELER et al. (1996)
	120 – 430	wheat seeds / loamy sand	BADALUCCO et al. (1996)
	198 – 288	crop rotation / Haplic Luvisol	FRIEDEL et al. (1996)
	304 – 624	agricultural land / Eutric Cambisol	KANDELER, MURER (1993)
arginine deaminase activity µg N g^{-1} h^{-1}	2.5 – 5.0	grassland / Pachic Arguistoll	AJWA et al. (1999)
	1.7 – 2.0	crop rotation/Phaeocem,Lithosol,Cambisol	KANDELER et al. (1996)
	4.0 – 11.0	forest / sandy soils	DILLY, MUNCH (1995)
	0.1 – 1.3	crop rotation / Fluventic Ustochrept	FRANZLUEBBERS et al. (1995)
arylsulfatase activity µg p-nitrophenol g^{-1} h^{-1}	30 – 50	grassland / Pachic Arguistoll	AJWA et al. (1999)
	115 – 340	agricultural land / Hapludoll	KLOSE et al. (1999)
	6.9 – 213	pasture / Typic Dystrochrept	HAYNES, WILLIAMS (1999)
	21 – 49	forest / Podzol	STADDON et al. (1998)
	1460 – 5912	forest / various soil types	GARCIA, HERNANDEZ (1997)
	50 – 350	crop rotation / Hapludalf	DENG, TABATABAI (1997)
	28 – 58	crop rotation/Phaeocem,Lithosol,Cambisol	KANDELER et al. (1996)
alkaline phosphatase µg p-nitrophenol g^{-1} h^{-1}	40 – 80	grassland / Pachic Arguistoll	AJWA et al. (1999)
	40 – 790	agricultural land / Aeric Vertic Epiaqualfs	KIM et al. (1998)
	100 – 500	crop rotation / Hapludalf	DENG, TABATABAI (1997)
	181 – 225	crop rotation / Ustochrept	CHANDER et al. (1997)
dehydrogenase µg TPF g^{-1} 24 h^{-1}	114 – 155	crop rotation / Haplumbreps, Hapludalfs	BEYER et al. (1999)
	2 – 9	n.m. / Palexeralf	MARZADONI et al. (1996)
	0.6 – 0.9	crop rotation / Fluvisol	CURCI et al. (1997)
	68 – 97	crop rotation / Ustochrept	CHANDER et al. (1997)
	148 – 207	crop rotation / Fluventic Xerochrept	PERUCCI et al. (1997)
	59 – 153	crop rotation /Phaeocem, Lithosol,Cambisol	KANDELER et al. (1996)

The plot-scale investigations of agricultural soils were mainly performed to understand the influence of soil management on soil organic matter turnover; the parameter included changes in the quantity and quality of plant residues entering the soil, their seasonal and spatial distribution, the ratio between above- and below-ground inputs, and changes in nutrient inputs (CHRISTENSEN, 1996). Using random samples of homogeneous, representative fields, most researchers have focused more on temporal variability than on the spatial heterogenity of different soil properties. Therefore, intensive studies of organic matter changes on different time scales (from one vegetation period to ca. one hundred years) were used to predict future changes and to improve soil protection. Since changes in SOM content are undetectable within 1 or 2 decades in temperate regions, datasets of long-term experiment are used to evaluate soil organic matter models (POWLSON et al., 1996). Figure 4 presents an example from the Ultuna long-term field experiment in Sweden (Uppsala). This experiment was established in 1956 on a clay loam (Eutric Cambisol) and is based on equal amounts of organic carbon (2000 kg C_{org} ha^{-1} yr^{-1}) applied through different organic amendments in comparison to reference plots. Therefore, changes in organic carbon contents in topsoil (0-20 cm) can be directly related to the stability of the organic matter applied. Figure 4 shows the development of C_{org} contents over the last 42 years. The bare fallow plot lost approximately one third of its initial C_{org} content due to mineralization of the initial soil C_{org}. Root and stubble input (cereals, rape and fodder beet) in the non-nitrogen treated plot yielded slightly higher C_{org} contents compared to bare fallow. Green manure, being the least stabilized organic material, kept the C_{org}-level nearly constant; materials less available for microbial degradation and mineralization, like animal manure and peat additions, increased this level considerably. Note that changes in humus contents are still ongoing and no equilibrium between C_{org} input and mineralization has been observed in the Ultuna experiment until now. Changes in microbial biomass and microbial processes are manifested over a shorter time scale (CHRISTENSEN, 1996). The recognition that soil microbial properties vary seasonally and that long-term variability exsists has improved our understanding of soil dynamics (DALAL et al., 1991; FRIEDEL et al., 1996; SALINAS-GARCIA et al., 1997). Therefore, in many cases these investigations helped to predict the expected changes in soil and their speed. For example, the response of xylanase activities in the top soil of a chernozem to reduced tillage intensity was detectable within the first year of the experiment, whereas significant treatment effects on microbial biomass and N-mineralization were observed after a 4-year period (KANDELER et al., 1999c). The authors explained the slow response of substrate-induced respiration to the altered tillage based on differences in biomass C turnover rates. Moreover, many plot-scale investigations showed that organic amendments (plant material, animal residues, sewage sludge) to soil are rapidly decomposed by microbial transformation, releasing essential nutrients such as N, P, and S. Stimulation of microbial biomass and enzyme activities in soil is usually greater in organically versus inorganically fertilized soils (GIANFREDA and BOLLAG, 1996). In addition, crop rotations which maintain plant root activity most of the year and have biennial legume-green manure incorporations, can improve soil chemical and microbiological properties within a relatively short time of 2 years (MILLER and DICK, 1995a, b).

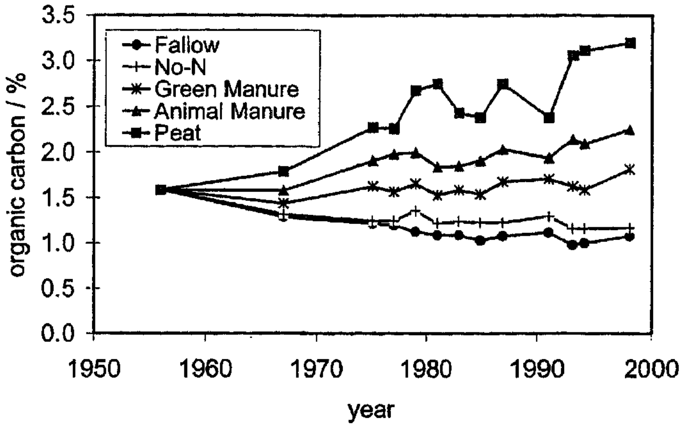

Figure 4: Development of organic carbon contents in topsoil samples (0-20cm) of the Ultuna long-term field experiment since its start in 1956. Fallow = bare fallow, No-N = plots without nitrogen fertilization; (redrawn from GERZABEK et al., 1997, amended with data from 1998)

Abbildung 4: Entwicklung der Gehalte an organischem Kohlenstoff im Oberboden (0-20 cm) des Ultuna Langzeitfeldversuches seit dessen Anlage im Jahr 1956. Fallow = Dauerschwarzbrache, No-N = Fruchtfolge ohne Stickstoffdüngung, Green Manure = Grasschnitt; Animal Manure = Stallmist; Peat = Torf; (umgezeichnet aus GERZABEK et al., 1997, ergänzt um Daten des Jahres 1998)

In conclusion, many plot-scale investigations of chemical and microbiological properties from the range of different soil ecosystems provide us with a useful database to explain potential changes within a single field or plot due to soil management and various other anthropogenic actions; at the same time, they also provide valuable data to model processes on the regional scale.

5. Regional-scale investigations

Basic information on spatial distribution patterns and spatial continuity of soil microbial populations and biochemical processes is necessary to describe soil microbiological properties at a regional scale (WIRTH, 1999). However, concepts and research activities at these scales are still sparse (PENNOCK et al., 1992; BERGSTROM et al., 1998; STORK and DILLY, 1998; WIRTH, 1999). To date, most reports have focused on soil quality and fertility (SMITH et al., 1993; CAHN et al., 1994) and only rarely on landscape-scale spatial distribution of soil biochemical processes (PENNOCK et al., 1992; HALVORSON et al., 1995; BERGSTROM et al., 1998). The distribution patterns of soil microbial properties on this scale can be traced back to several physical, chemical and biological dependencies. The characterization of these interactions is essential to achieve a better understanding of complex ecosystem processes (GOOVAERTS, 1998). For example, TSCHERKO (1999) reveal site factors (land use, soil type, contamination) that correlate with soil microbial properties and she identifies those microbial variables most sensitive to these factors. In order to determine the importance of factors which influence microbial biomass, N-mineralization and enzyme activities (xylanase, urease, phosphatase, arylsulfatase) on a more regional scale, 2500 values of microbial properties across different ecosystems derived from previous studies in Central Europe were pooled and analyzed for underlying trends by conventional statistical procedures combined with fuzzy operations (TSCHERKO, 1999). The data entities were collected from 10 different soil types over the last 10 years. The evaluation of the data (two-factorial ANOVA including organic carbon as a covariate) revealed a significant influence of land use (forest, grassland, arable land) and soil type on microbial properties (Table 2). Additionally, total organic carbon explained half of the variability of the enzyme activities (phosphatase, β-glucosidase, dehydrogenase, urease, xylanase) and one third of the heterogeneity of the microbial biomass (microbial C and N). In a multivariate, hierarchical view (cluster analysis with fuzzy numbers including all microbial variables), land use was the strongest factor, contamination the weakest factor governing the size of soil microbial properties at the ecosystem level. Soil type turned out to be an important site factor as it summarizes climatic, topographical and geological conditions, acidification, and vegetation influence (TSCHERKO, 1999). A multivariate approach of fuzzy set operations revealed microbial biomass and arylsulfatase activity to be sensitive indicators for contamination across different ecosystems.

In addition, a basis for further studies on the relationships between biochemical soil parameters and site properties can be obtained from the Soil Information System BORIS, which has been developed by the Federal Environment Agency in Vienna (SCHICHO-SCHREIER, 1994; SCHWARZ et al., 1994 bzw. 1999). BORIS holds more than 1.25 million records from more than 8500 sites from all Austrian provin-

Table 2 : Influence of soil type and land use on soil microbial processes. Given are the F-values of the factor variables (soil type, land use) and of the co-variate (total organic carbon TOC), level of significance (*** $P<0.001$, ** $P<0.01$, * $P<0.05$, ns not significant), total degrees of freedom (df) and explained variance (R^2) of two-factorial ANOVA

Tabelle 2: Einfluß von Bodentyp und Landnutzung auf bodenmikrobiologische Prozesse. F-Werte der Variablen (Bodentyp, Landnutzung) und der Co-Variate (Gesamtgehalt des organischen Kohlenstoffs), Niveau der Signifikanz (*** $P<0.001$, ** $P<0.01$, * $P<0.05$, ns nicht signifikant), Freiheitsgrade (df) und erklärte Varianz (R^2) der zwei-faktoriellen ANOVA

Microbial biomass/activity	Co-variate TOC	Land use	Soil type	FACTOR Interaction	df	R^2
Biomass N	84.9***	5.2**	72.3***	a)	7	0.67
SIR[b)]	1712.4***	574.0**	95.2**	55.2**	9	0.74
Alkal. Phosphatase	1872.5***	618.2***	164.5**	134.3***	7	0.76
Arylsulfatase	50.7***	15.2***	57.4**	a)	6	0.47
β-Glucosidase	1007.0***	2.8ns	3.2*	a)	4	0.87
Phosphatase	1351.0***	486.5***	0.1ns	a)	5	0.87
Dehydrogenase	582.1***	25.4***	63.8**	26.8***	6	0.69
N-Mineralisation	1004.0***	578.0***	72.5**	238.2**	11	0.70
Pot. Nitrification	208.8***	8.9**	108.6**	19.2***	7	0.45
Protease	4832.4***	138.9**	59.9**	13.1***	8	0.89
Urease	4203.6***	671.9***	32.6**	53.0**	11	0.83
Xylanase	2884.0***	82.6**	47.2**	19.8***	11	0.75

a) due to a singular matrix, higher-order interactions have been suppressed
b) SIR (Substrate-induced respiration)

cial soil surveys (except Salzburg and Vorarlberg), the Forest Soil Monitoring System, Austria-wide cesium data and more than 25 additional investigations. The records comprise site descriptions (more than 150 parameters), soil profile descriptions (more than 50 different parameters) and analytical measurements (more than 300 different parameters). All these parameters – among them more than 20 different biochemical soil parameters (ÖHLINGER, 1996) – have been included in the "Data Key Soil Science" (SCHWARZ et al., 1994), which forms the basis for the harmonization of different investigations. An expansion of soil parameters in the BORIS Soil Information System is possible at any time and will be included in the next edition of the "Data Key Soil Science".

Via BORIS INFO, a meta data information system (internet address: http://www.ubavie.gv.at/ under Umweltsituation/Boden/BORIS) each user can obtain the information on which parameters have been investigated and on where and how to access them. Submitting in a query for all parameters dealing with soil microorganisms reveals that 405 samples from 280 sites have been investigated. The location of these sites is shown on a map of Austria (Figure 5a). Until now, 19218 samples from 5782 sites have been analyzed for the organic matter content of soils (Figure 5b). Since the analytical procedure used in the different labs was not completely uniform for all analyses, only partially comparisons are possible. For 4074 sites it was possible to calculate the weighted mean of topsoils (0–20 cm) according different land use (Table 3). As expected the average organic matter contents of forests are higher than those of grassland, which

Table 3: The response of organic matter (%) in top soils (0 – 20 cm) to land use. Evaluation of the Soil Informationsystem BORIS of the Federal Environment Agency Vienna (The data were provided by the federal provinces of Burgenland, Lower Austria, Carinthia, Upper Austria, Styria, Tirol and the Federal Forest Research Center.)

Table 3: Organische Substanz (%) in Oberböden (0 – 20 cm) unterschiedlicher Landnutzung. Auswertung aus dem Bodeninformationssystem BORIS des Umweltbundesamtes Wien (Die Daten wurden von den Ämtern der Landesregierungen von Burgenland, Niederösterreich, Kärnten, Oberösterreich, Steiermark, Tirol und der Forstlichen Bundesversuchsanstalt aus den jeweiligen Bodenzustandsinventuren zur Verfügung gestellt.)

	Forest	Grassland	Arable Land	Others
Mean	10.90	6.88	2.81	5.17
Median	8.06	5.17	2.40	2.70
Min	1.21	1.35	0.40	0.80
Max	61.20	55.90	40.20	70.95
n	792	1168	2068	46

are higher than those of arable land. The spatial distribution is shown in Figure 6. In eastern Austria with its higher percentage of arable land, the values are lower than in the western provinces, where there is more grassland and forest.

Few studies examined in detail the relationships between soil physico-chemical properties and microbial indices at a larger scale. At the landscape scale significant correlations have been found between organic carbon and microbial biomass (WARDLE, 1992; STORK and DILLY, 1998), and enzyme activities (DUTZLER, 1977a, b; BERGSTROM et al., 1998; WIRTH, 1999). Based on this knowledge, models can be fitted to estimate and upscale soil microbial activity values at unsampled locations. Geostatistical models (WARRICK et al., 1986; GOOVAERTS, 1998) have been a tool to estimate spatial dependencies and to predict soil attribute values. Spatial dependencies of soil microbial biomass at the landscape scale ranged from 13 m (STORK and DILLY, 1998) to 28 m (WIRTH, 1999), the spatial dependency of soil basal respiration from 10 m (STORK and DILLY, 1998) to 61.2 m (WIRTH, 1999). BERGSTROM et al. (1998) determined spatial dependencies of phosphatase and arylsulfatase at ranges of 19 m and 16 m, respectively, and no spatial dependency of dehydrogenase, urease, glutaminase, and β-glucosidase.

Knowledge of the spatial dependency of soil microbial attributes helps to interpret their ecological meaning at the ecosystem level (BERGSTROM et al., 1998). Similarity of ranges between soil physico-chemical and microbial attributes points to ecological identity. Unfortunately, biochemical processes in the soil are dynamic, leading to variation in both space and time. Landscape-scale analyses by geostatistical methods are a useful tool for identifying and explaining spatial relationships between soil biochemical processes and site properties. However, further model improvements should focus on identifying and mapping time-space patterns using modern approaches like fuzzy classification and geostatistical interpolation.

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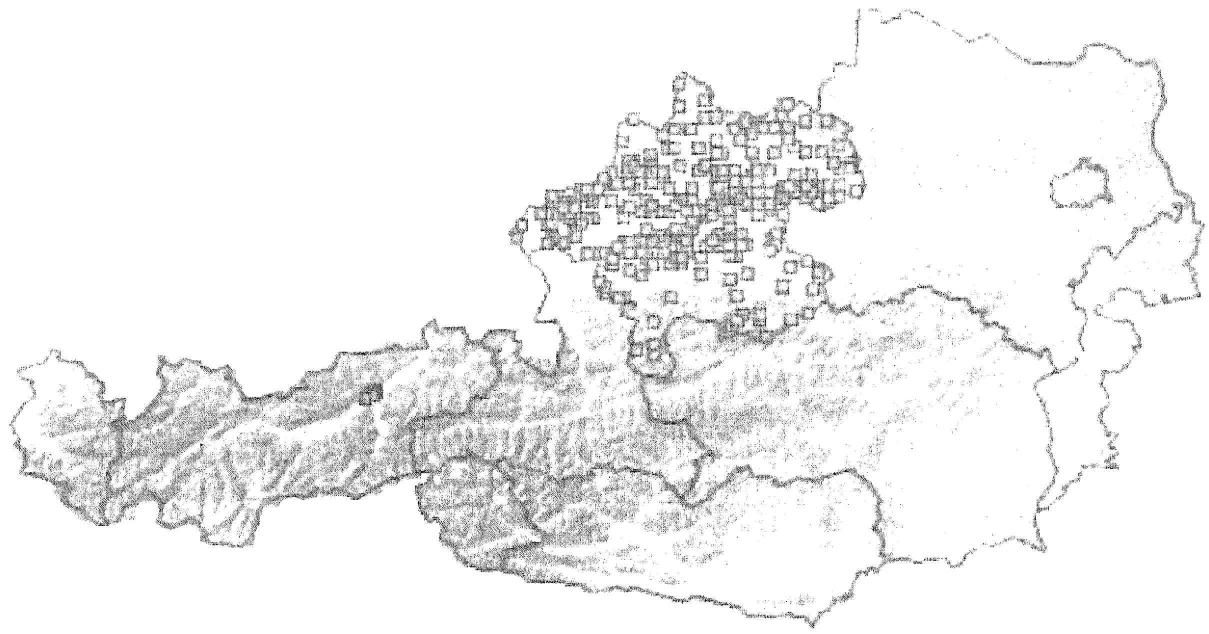

Figure 5a: Result of query to the Austria-wide Soil Information System BORIS: Representation of sites at which biochemical soil parameters have been investigated

Abbildung 5a: Abfrageergebnis aus dem österreichweiten Bodeninformationssystem BORIS: Darstellung der Standorte, an denen biologisch-chemische Parameter untersucht wurden

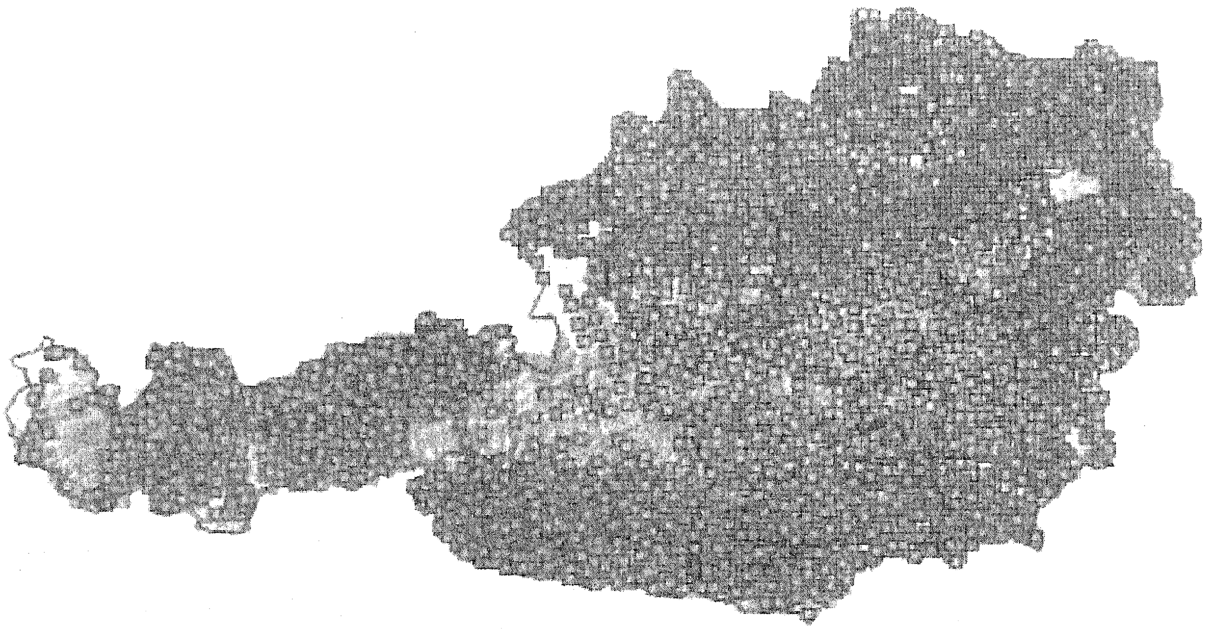

Figure 5b: Result of query to the Austria-wide Soil Information System BORIS: Representation of sites at which organic matter has been investigated

Abbildung 5b: Abfrageergebnis aus dem österreichweiten Bodeninformationssystem BORIS: Darstellung der Standorte, an denen organische Substanz untersucht wurde

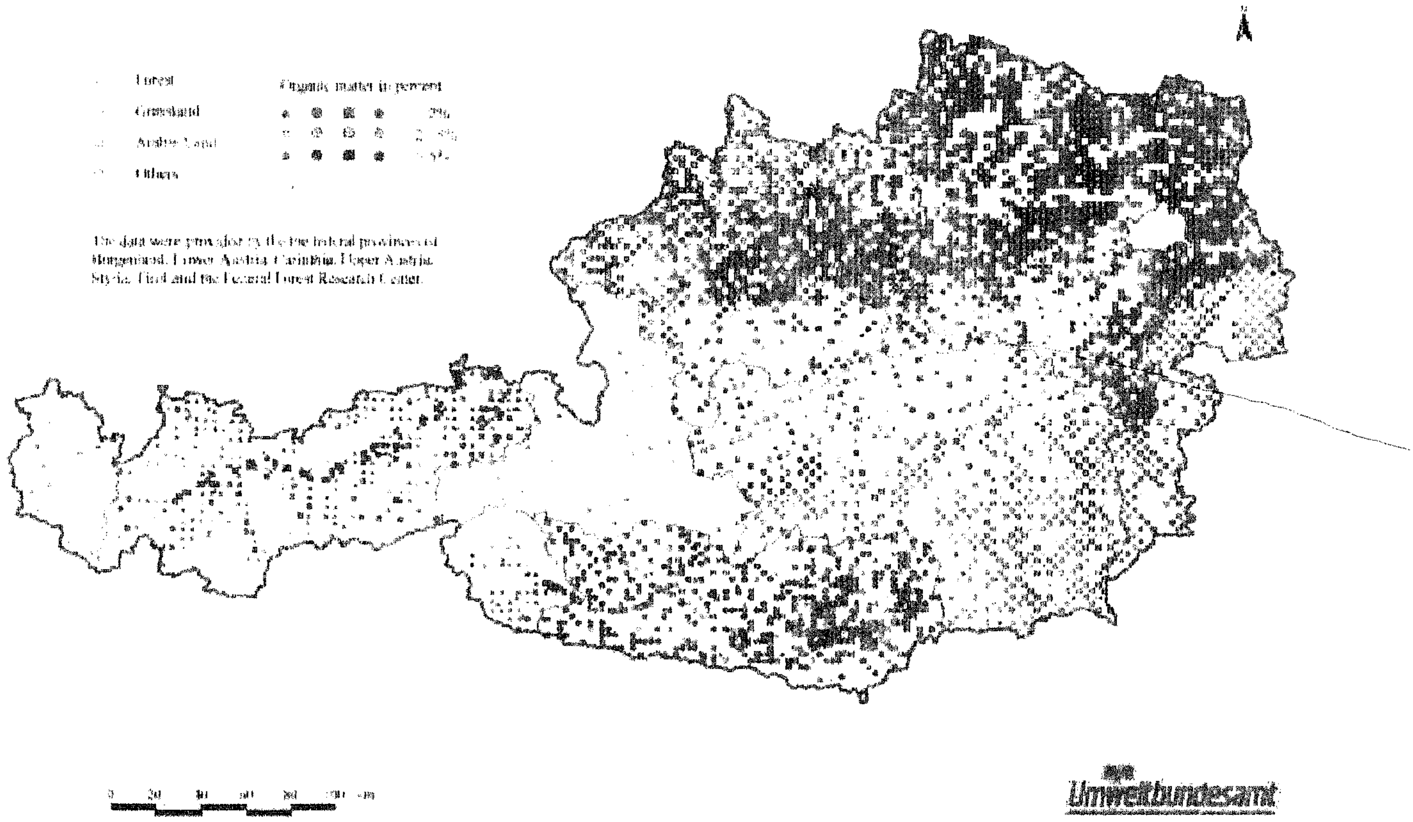

Figure 6: Distribution of organic matter in topsoils (0–20 cm) according to land use
Abbildung 6: Verteilung der organischen Substanz in Oberböden (0– 20 cm) nach Landnutzung

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