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### **COSUST Special Issue: Wageningen Soil Meetings Session 3.1**

- Short review articles (3500 words) in which recent developments on the topic are presented, emphasizing the most important aspects.
- Short annotations to papers that the authors consider to be most interesting from all those published on the topic over the previous year (2010-11).

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### **Soil biodiversity, biological indicators and soil ecosystem services - an overview of European approaches**

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#### **Abstract (max 200 words)**

Soil biota are essential for many soil processes and functions, yet there is an increasing pressure on soil biodiversity and soil degradation remains a pertinent issue. Therefore, the sustainable management of soils requires soil monitoring, including biological indicators able to relate land use and management to soil functioning and ecosystem services. Since the 1990's, biological soil parameters have been assessed in an increasing number of field trials and monitoring

programmes across Europe. The development and effective use of meaningful and widely applicable bio-indicators however, continues to be a challenging tasks. This paper aims to provide an overview of current knowledge in relation to soil biodiversity characterization and assessment. Examples of European monitoring approaches and soil biodiversity indicators are presented, and the value of soil biodiversity databases for developing a better understanding of the relations between soil management and ecosystem functions and services is discussed. We conclude that integration of monitoring approaches and data sets, together with state-of-the art ecological expertise, offers good opportunities for advancing ecological theory as well as application of such knowledge by decision makers.

## **Introduction**

The Convention on Biological Diversity (CBD; URL: <http://www.cbd.int/>) and the Millennium Ecosystem Assessment [1] have underlined the relationships between, biodiversity loss and a decline in the capacity of ecosystems to support human well-being. Being the legally binding international agreement for the conservation and sustainable use of biological diversity, the CBD has stimulated a demand for a wide range of indicators suited to monitor trends in the state of biodiversity and natural resources [2]. Soils are a natural resource that must be secured for future generations. Although soils are, in theory, a renewable resource, rates of soil formation or restoration are too slow to cope with current rates of soil degradation. Soils also host an enormous biodiversity, both in terms of abundance, number of species and functions of organisms. These organisms and their interactions are fundamental to many soil processes and ecosystem functions, including organic matter decomposition, nutrient cycling, soil structure formation, pest regulation and bioremediation of contaminants. In aggregated form these processes and functions relate to ecosystem services that are of direct benefit to humans, such as food production, climate regulation, or provision of clean water [3] (Figure 1). Although biodiversity that is 'hidden' belowground has long received little attention compared to aboveground biodiversity, this attitude has clearly started to change. Loss of biodiversity in soils due to the expansion, intensification and mechanization of agriculture has been identified as a major problem across Europe. Related pressures include soil erosion, organic matter decline, compaction, contamination, salinization and climate change [4,5].

Different EU policies are in some way contributing to the protection of soils (e.g. regulation on water quality, pesticide use, waste management or nature protection), but aims and actions are scattered ([http://ec.europa.eu/environment/soil/index\\_en.htm](http://ec.europa.eu/environment/soil/index_en.htm)). The adoption of the EU Soil Thematic Strategy in 2006 was a first step towards a coordinated approach to ensure the protection of all soils in Europe [6]. Further integration of soil biodiversity conservation into EU agricultural and/or environmental legislation, however, is hampered because the level of knowledge has been considered insufficient to recommend policy. A better understanding of soil organisms, their distributions, interactions and functions in soils and how they translate into ecosystem services is

therefore essential to guide further action [4]. A necessary first step to achieve the conservation of soil biodiversity is a better knowledge on its spatial and temporal distribution and how this relates to soil management and habitat quality [4]. A crucial second step is to better understand and communicate the implications for ecosystem functioning, such that soil (biodiversity) conservation is taken into account in decision making. In the face of those needs, monitoring and assessment of biological soil parameters has been initiated in several countries, from small-scale field experiments up to national scale monitoring programmes. As a parallel activity, there has been much work on the development of biological soil indicators that relate soil management with soil functioning. This is very much a task in progress, but advancements in this field are accelerating.

This paper combines a literature review on soil biodiversity and biological soil indicators, with examples of monitoring programmes across Europe. We thereby aimed at addressing the following objectives:

1. To provide a brief overview of current knowledge and developments related to soil biodiversity (characterization) including associated functions
2. To discuss the development and monitoring of biological indicators that link soil management or habitat quality to soil biodiversity and ecosystem functions, based on European experiences.
3. To discuss needs and opportunities for data integration, data mining and communication of knowledge among stakeholders, in order to support the sustainable management of soil biodiversity and soil ecosystem services.

>> Fig 1.

## **Soil biodiversity**

Soil biota comprise the organisms that spend all, or part of, their life cycles belowground - ranging from the myriad of invisible microbes, such as bacteria, fungi and protozoa, to the macro-fauna, e.g. earthworms, ants and termites (<http://www.fao.org/ag/AGL/agll/soilbiod/>). Larger animals such as moles and voles are also considered soil fauna, but are rarely considered in soil biodiversity assessments due to their small numbers. Although, strictly speaking, plant roots belong to the soil biota their role is beyond the scope of this review. For a general introduction to all the different groups of soil biota and their functions we refer to the European Atlas of Soil Biodiversity [7].

One of the most complete definitions of soil biodiversity is derived from the CBD definition of biodiversity: 'Soil biodiversity comprises 'the variation in soil life, from genes to communities, and the ecological complexes of which they are part, i.e. from soil microhabitats to landscapes' [3]. It should be emphasized that such variation

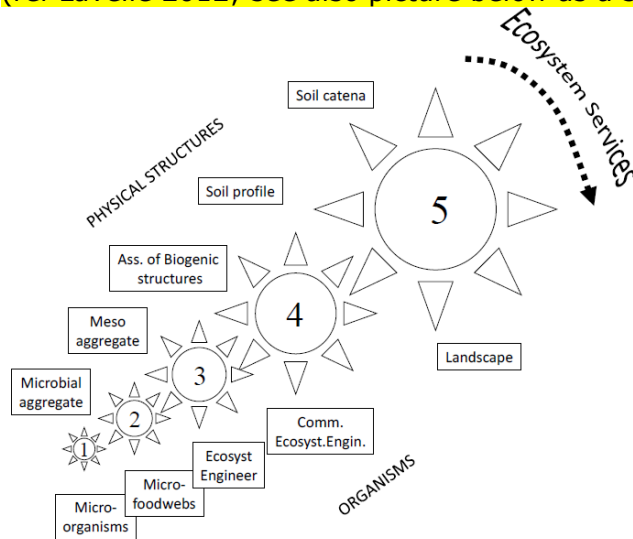
can be described in terms of three primary, and interrelated, attributes of biodiversity: composition, structure and function [8]. In the context of this paper, we then consider soil biodiversity as the quantity, variety and structure of all forms of life in soils, as well as related functions [9]. Taxonomic identification of soil organisms can be problematic because a vast amount of soil organisms has not yet been identified and many microbes are not culturable under lab conditions. However, relations between soil biodiversity and ecosystem functions tend to depend more on structural and functional diversity than on taxonomic parameters per se [10,11]. This phenomenon is partly explained by the high level of functional redundancy within species-rich soil communities [12,13]. As an exception several species (groups) have been identified for their unique role in specialized soil processes. These so-called "keystone species" have a disproportionate effect on certain soil functions [14]. Examples are fungal species that are capable of decomposing certain recalcitrant organic compounds [15], symbiotic micro-organisms involved in atmospheric N fixation or P uptake by plants [16] or bioturbators like earthworm species (see textbox).

Considering that the complex of biotic interactions, in conjunction with the abiotic environment, determine soil processes and functions, a more comprehensive characterization of soil biodiversity would be useful. Like ecosystems in general, soils are hierarchical systems with internal processes operating at each level of organization and interacting across levels. Hierarchy theory suggests that higher levels facilitate or constrain the behaviour of lower levels. Hierarchical relations between habitat characteristics, soil organisms and their interactions have been described by Lavelle (1997), while implications for ecosystem functions are discussed in Kibblewhite (2008) [17] and Lavelle (2012) [18]. For instance, the soil macrofauna (predominantly termites, earthworms, or ants) contain ecological groups that have the ability to dig in the soil profile, create burrows, nests and galleries while mixing, ingesting and/or excreting organo-mineral soil material. As they can modify the soil habitat in terms of physical structure and availability of resources to other soil organisms, those soil animals have been characterized as 'ecosystem engineers' [19]. The soil ecosystem engineers thus possess a strong effect on the distribution and activities of smaller soil organisms.

Accordingly, Kibblewhite et al. (2008) [17] and Turbé et al (2010) [3] classified soil organisms into broad functional assemblages that act at different spatio-temporal scales and can be related to different ecosystem functions (Fig 1). A distinction is made between 'decomposers' and 'nutrient transformers' (grouped as 'chemical engineers' by Turbe et al (2010)[3]); 'biocontrollers' (or 'biological regulators' [3]), i.e. small invertebrates, such as nematodes, springtails and mites, which act as herbivores or predate on other invertebrates or micro-organisms; and the 'ecosystem engineers' (Figure 1). For completeness, however, it should be noted that this broad classification does provide a generalization as multiple functions can be performed by the different functional assemblages and overlap in functions occurs across all levels (e.g. microbes can contribute to soil aggregate formation [20,21]). In line with the hierarchical framework of Lavelle (1997) it should be stressed that

the assemblages of organisms that form the functional groups do not operate in isolation but imply a high degree of interconnectedness between soil functions. This implies that an intervention that affects one function will inevitably alter the other functions [17,18]. This is also reflected in Fig XX.

(ref Lavelle 2012; see also picture below as a summary of the idea)



## Biological soil indicators

The concept of indicators is widely used in environmental monitoring, mainly in relation to anthropogenically induced disturbances and environmental change. In general terms, indicators have been defined as measurable surrogates for environmental end points that are in itself too complex to assess. Such indicators, either biological, physical or chemical, should be able to give information about the state and trends as well as the seriousness of the situation, and are assumed to be of value to environmental management goals and decision making ([3,8] [2]). The soil community provides a large number of potentially interesting bioindicators for environmental monitoring in response to a range of stresses or disturbances [17,22-25]. However, the diversity of applications and types of bioindicators can cause some confusion regarding the type of information they provide. According to Gerhardt [26], we define biological indicators as (groups of) organisms whose reactions (in terms of presence/absence, abundance, morphology, physiology or behaviour) give information on the condition of a habitat or ecosystem. They are useful in situations where the indicated factor is difficult to measure, or where the environmental factor is easy to measure but difficult to interpret in terms of ecological significance. Based on their application, Gerhardt distinguished between *environmental* bioindicators, which diagnose the state of the environment, and *ecological* bioindicators, whose response to environmental stresses is representative for the community as a whole.

These types of soil bioindicators have been applied in environmental risk assessment and monitoring of responses to land use (e.g. [27]), agricultural management (e.g. [28-33]) or soil contamination (e.g.[23,25]). The parameters being measured comprise different soil organisms selected for their sensitivity to soil management or environmental conditions and/or their relevance for important ecological functions (e.g. organic matter decomposition, N mineralization or soil structure formation) [25]. Recently, there has been an increasing interest in developing bio-indicators as proxies for soil functions or ecosystem services, or soil quality or soil health in general (refs). In fact, in the light of current information needs, the full spectrum from habitat characteristics to biotic responses and ecological functions should be considered in environmental assessments.

The hierarchical organization of the soil community and ecosystems in general suggest that soil biodiversity be monitored at multiple levels of organization (organism, population, community, ecosystem) and at multiple spatial scales (e.g. from plot to farm to landscape) [8,18]. Different organisms from the three broad functional groups or hierarchical levels described in the previous section have frequently been used as biological soil indicators. One examples from each group is briefly discussed here, including the advancement of molecular techniques and references for further reading (see textboxes). Other organism groups that have commonly been measured in monitoring programmes are micro-arthropods such as collembola (springtails), acari (mites) [8,31,34,35] and other mesofauna, e.g. enchytraeids (potworms) [31].

#### >> Textboxes

Criteria for the selection of indicators that adequately characterise soil biodiversity and soil functions and are suitable for monitoring purposes have been summarized by Ritz et al (2009) [36] and Turbé et al (2010) [3] (Table 2). Because it is highly unlikely that a single indicator will comply with all these criteria, in practice focus is on the development of sets of complementary indicators, including both biotic and abiotic parameters. Nevertheless, despite the fact that a multitude of indicators estimating some aspect of soil biodiversity exist no reference set of standardized indicators is available yet. Moreover, at this stage, relations between the biological parameters being measured and ecosystem functions have largely been based on assumptions, or at best expert knowledge, rather than empirical testing. This basically reflects an insufficient comprehensive understanding of soil communities and their biotic interactions to be able to predict how losses in soil biodiversity affect multiple soil functions. Other difficulties include the wide variety of objectives, stakeholders and environmental conditions to be addressed. These issues, as well as promising avenues for progress in indicator development and application, are discussed in the remainder of this paper.

#### >> Table 2.

## Examples of European approaches

Since the late 1980's, biological parameters have been assessed in an increasing number of studies, ranging from long-term agricultural field trials (e.g. [8,18,28,29,37-40]) to regional or national monitoring programmes (e.g. [27,31,35,36]). Currently there are over 15 European countries that have collected soil biological parameters as part of a large scale monitoring programme. Some examples are provided in Table 1. Ideally this would provide the foundation for integrated assessments of soil biodiversity across a wide range of situations in Europe. However, the information has been collected for different objectives and using a wide range of methods, and few indicators have consistently been used in national-scale monitoring [31,36]. Recent attempts to develop standardized indicator sets that comply with the criteria listed in table 2 are briefly reviewed here.

>>Table 1.

Frameworks for selecting bio-indicators for national-scale soil monitoring have been devised in, for example, France [25], the Netherlands [31,38] and the UK [36]. These frameworks had in common that a wide array of candidate indicators was assembled and tested for their suitability to be used in systematic measurement of soil biodiversity. Selected indicators had to comply with requirements such as (i) pertinence to predefined soil functions, including agricultural production, environmental interactions and habitat support and (ii) applicability to the range of ecosystems under consideration, (iii) ability to discriminate between soil types and (iv) technical, practical and financial criteria [36]. Linking of organism groups to soil functions generally involved literature survey and expert judgement [31,36]. Ritz et al (2009) [36] and Rutgers et al (2012) [32] used a systematic approach of stakeholder consultation to take into account a diversity of end-user requirements and priorities. It was concluded, however, that further work is needed to confirm the sensitivity of the indicators, their ability to discriminate between soil-land use combinations and their ecological interpretation [36]. One example of such work is the ongoing (2006-2012) French national BioIndicator programme [25]. Using homogeneous procedures and protocols, 47 biological parameters were assessed in a large number of sites differing in land use, agricultural management, contamination type and pollution levels. Those included microorganisms, fauna and flora at the community level (e.g. abundance, biomass, species and functional composition and ecological traits) as well as the organism level (e.g. gene expression). Their potential to be used as a bioindicator for national scale monitoring was validated based on their sensitivity to different environmental conditions and disturbances, and their accessibility and applicability by experts and non-specialist stakeholders.

In parallel with national initiatives, European research projects have been initiated to promote the standardisation of biological soil indicators, mainly through Framework Programmes (FP). An overview of those projects is given in Turbé et al (2010) [3].



Among those, the FP6 project ENVASSO (Environmental Assessment of Soil Monitoring; [9]) was the first attempt to develop a harmonised system for soil biodiversity monitoring across Europe. Standardized bio-indicator sets were defined and organized into different priority levels [9]. 'Level I' indicators included groups of organisms, corresponding with the abovementioned functional classification of Kibblewhite et al (2009) [17], as well as ecological functions: 1) abundance, biomass and species diversity of earthworms (or enchytraeids if no earthworms are present, e.g. in soils with low pH); 2) abundance and species diversity of collembola; and 3) microbial respiration. Depending on local objectives and available resources, the key indicators could be complemented with 'level II' or 'level III' indicators [9]. Procedures and protocols, based on ISO standards [41-43], were tested in pilot sites in France, Ireland, Portugal and Hungary to assess the efficiency and sensitivity of the indicators across a range of land-use categories at a European scale [9].

Comparison of data between consecutive samplings over multiple years indicated that species composition tends to be relatively stable, but abundances and biomasses were more variable, depending for example on weather conditions and crop rotations [9,25,31]. In order to interpret the results, there is a strong need to define baselines and reference and thresholds values for certain combinations of land use, soil type and climate. Such references do not yet exist at a European scale [9], although density ranges for different groups of organisms have been published for a selection of soil and land use types in the Netherlands [31] and France [44]. Among the objectives of the ongoing FP7 project Ecofinders are the standardisation of methodologies for the assessment of biological soil indicators, and characterisation of normal operating ranges for soil biodiversity according to climatic zones, soil types and land uses across Europe [45]. The increasing availability of ISO standards [41-43] for sampling procedures and analyses is an important step towards homogenization of operational procedures, but further work is still required in this field [9,46].

Another important challenge for biological soil indicators is to capture the variety of spatio-temporal scales over which environmental changes occur [3]. Depending on life history traits and dispersal characteristics, certain groups of soil organisms can respond slowly to changes in land-use or agricultural management [29]. Those observations emphasize the need for sampling designs with wide spatiotemporal coverage [9,17,29]. Long-term field experiments remain important to enhance our understanding of biotic responses with time after certain changes in management or land use occur, as well as underlying mechanisms [17,28,29,40,47,48].

### **Linking biological soil indicators and ecosystem services for decision support**

A major challenge in sustainable soil management is to support multiple ecosystem functions and services (Figure 1) and weighing of trade-offs in terms of societal needs. Until now, interpretation of biological soil indicators in terms of ecological

implications has largely been based on expert judgments [32,49]. A more robust and quantitative approach relies on empirical testing and development of models that incorporate scale issues and trade-offs. The decision support function of biological soil indicators also implies that they must facilitate communication with end users such as policy makers, land managers (including farmers) and technicians. Datasets derived from soil biodiversity assessment and large scale monitoring provide potentially important sources of information. Promising developments and approaches on those two issues are briefly reviewed here. One promising avenue for developing a predictive understanding of the linkages between habitat characteristics or disturbance and different ecosystem functions and services is based on ecological traits, or the morphological, physiological, behavioural or life-history attributes that determine the response of organisms to (changing) environmental conditions and can be linked to effects on ecosystem functions [21,49,50]. Important advantages of this approach include possible generalizations across eco-regions, independent from taxonomy [21,25,51]. Information on trait attribute values of soil organisms being accumulated in databases can be connected with the occurrence of species as an indicator, as was done for earthworm species by Peres et al (2012) [25]. For example, the size of organisms strongly determines their spatial aggregation patterns and dispersal distances, as well as their lifetimes and sensitivity to habitat disturbances with consequences for multiple, interconnected soil functions [3,19,52]. Mulder et al (2011) [53] showed how mining of databases of abiotic and biotic soil variables derived from the Dutch monitoring programme 'DSQN' (Table 2) can contribute to our understanding of the relations between soil characteristics, the (trait) structure of the soil community and ecosystem functioning. Three ecological concepts were explored and rendered promising results: allometry, i.e. the science of size-abundance relationships amongst organisms in soil food webs; stoichiometry, i.e. the biotic relationships in terms of chemical compositions (e.g. nutrient-to-carbon ratios) of plants and soil organisms; and the association of structural and functional aspects of food webs. The observed influence of ecological stoichiometry as a dominant independent predictor provides opportunities to develop a mechanistic model of invertebrate responses under different management regimes. Detritus-based food web modeling has successfully been used for quantification of nutrient and carbon flows based on soil biodiversity assessments [54]. Although organic matter decomposition is not only a key ecosystem function in its own right, but also the main source of energy driving other ecosystem functions (e.g. soil structure maintenance), such models have not yet considered this interconnectedness. Incorporating the interconnectedness of soil ecosystem functions provided by different functional assemblages in the soil community provides scope for predicting (interactions between) multiple functions and services (Kibblewhite, Brussaard, Lavelle).

Finally, in order to allow for informed decision making in soil management issues and implementation of the benefits of soil ecosystem services into policy, quantification and weighing of ecosystem services is essential. For quantification indicators should be fitted to so called 'utility' functions which transform the specific units of the indicator to a uniform scale for ecosystem service performance (EEA 2011). This is

not straightforward because ecosystem services act on different spatial and temporal scales. [19; 20] have shown how pragmatic choices enable quick quantification of soil quality through the performance of ecosystem services, based on monitoring of biotic indicators as well as abiotic soil properties, and can already be implemented in practical systems. Stakeholder involvement is central to the identification and prioritization of important ecosystem services by different end users [32,36]. Another strength of databases derived from soil biodiversity monitoring programmes is the development habitat-response relationships and communication tools to be used in stakeholder processes and for awareness raising [25]. When spatially presented, derived fundamental models have been used to demonstrate that different options in land-use planning and soil management resulted in highly different impacts on the biodiversity of soil organisms, including differences in functional attributes [53,55].

## **Synthesis and conclusions**

The sustainable management of soils across Europe requires monitoring of biological soil indicators that can be linked to soil ecosystem functions and services and translated into decision support tools. Development of such indicators, or sets of indicators, is not straightforward and represents an active field of research. Based on hierarchical theory, different functional assemblages of soil organisms can be distinguished and their interactions should be reflected in soil biodiversity assessments and indicator selection. No single indicator is universally applicable and different indicators are needed for different functions. So in practice focus is on the development of sets of complementary indicators that need to be validated across a wide range of environmental conditions using standardised methods to produce accurate and consistent results. Several European initiatives contributing to the selection of indicator sets and standardization of methods have been discussed here. However, despite considerable progress, major scientific and practical issues remain to be addressed.

LIST THOSE>>>spatiotemporal coverage, links with functions is of the not empirically tested, different objectives of , importance to connect monitoring and more indepth studies (LT trials) for validation and hypothesis testing. + In order to interpret the results, there is a strong need to define baselines and reference and thresholds values for certain combinations of land use, soil type and climate.

Integration and data mining of datasets resulting from soil biodiversity assessments and monitoring programmes offers unique opportunities to develop ecological concepts and models that predict effects of soil management on soil biodiversity and ecosystem services. Promising avenues include approaches based on the analysis of ecological traits. Studying the extent to which driving forces behind the partitioning of energy in the soil food web [49], influence multitrophic interactions and multiple soil functions is a fruitful area for future research. Finally, the knowledge thus generated should be applied in decision making, which requires simple and clear

communication to decision-makers. Databases on biological soil indicators have been applied already to rural and societal questions and for the development of tools for stakeholder processes and awareness raising.

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## Tables

Table 1. Examples of European national or regional soil biodiversity monitoring networks, their geographical coverage and types of indicators measured (source: Turbé et al 2010 [3] and unpublished data from the ECOFINDERS project).

Table 2. Seven criteria for the selection of biological soil indicators. Source: Turbé et al (2009)[3].

### Figure captions

Fig 1. The relationships between functional assemblages of soil organisms, aggregate ecosystem functions and ecosystem services. Ecosystem services represent aggregations of functional outputs of biological processes that are of direct benefit to the society (source: Kibblewhite et al 2008 [11])

### Textboxes

Examples from different broad functional groups that have frequently been used as biological soil indicators. a) Earthworms (picture: R.G. de Goede); b) nematodes. The picture represents a bacteria feeding nematode (source: Jeffery et al 2010 [8]); c) microorganisms. The picture on the left side represents bacterial cells (green) growing on the surface of a fungal hypha. The picture on the right shows a range of soil organisms grown in culture in the laboratory. Each different shaped and coloured colony represents a different 'species' of microorganism (source for both pictures: Jeffrey et al 2010 [8]).

### **Textboxes**

**a) Earthworms** live at the soil surface in the litter layer or in galleries and chambers that they dig in the soil. These invertebrates belong to the broad functional group of ecosystem engineers. They usually have limited abilities to digest litter and soil organic matter and rely on microbial digestion capabilities, by feeding directly on them and via their internal rumen systems [18]. By producing structures in the soil, in the form of burrows and excrements, they strongly modify the architecture of the soil and the habitat



for other soil organisms, including plant roots. Earthworms can play a particularly large role in litter transformation and incorporation of litter into the mineral soil, soil structure formation and the soil water balance, both in agricultural and (semi-)natural ecosystems [56]. Earthworms are often used as soil bioindicators contaminated soils, because of their sensitivity to soil contamination (heavy metals and organic contaminants) [25]. They also respond strongly to agricultural practices (e.g. tillage, crop rotations, pesticides application, organic matter inputs) [25,27,29,30]. Species ( $\pm 100$  in France) are classified into three ecological groups (anecics, endogeics and epigeics) that provide different functions and show different sensitivity to soil disturbances or chemical contamination [25,29,30,40]. Epigeic earthworms live at the soil surface and feed on plant litter. They are most sensitive to exposure to soil tillage, pesticides and contaminants. They

are important for litter transformation and contribute to nutrient cycling. Anecics create permanent vertical or sub-vertical burrows and deposit their excrements at the soil surface. They are very important for organic matter incorporation into the mineral soil, but are sensitive to ploughing. Endogeics feed on mineral soil enriched in soil organic matter, and therefore benefit from organic matter incorporation through tillage or the activities of epigeics or anecic earthworms [52]. They create networks of non-permanent, horizontal to sub-horizontal burrows, and deposit their excrements in the soil. Anecic and endogeic earthworms play a key role in the formation and maintenance of soil structure, thereby enhancing water infiltration and remediation of soil pollutants, and controlling soil erosion [27,28].

Total abundance or biomass of earthworms are commonly used as bioindicators, nevertheless the functional group diversity may be a better indicator of habitat quality and soil ecosystem functions [9,25,54]. An important advantage of earthworms as indicators is that taxonomic identification is relatively easy because of their body size and relatively low species richness. Earthworms can be observed with the naked eye and are commonly known, and are therefore suitable for communication purposes with stakeholders. On the other hand, their spatial variability in the field can be high, which makes representative sampling a laborious task.

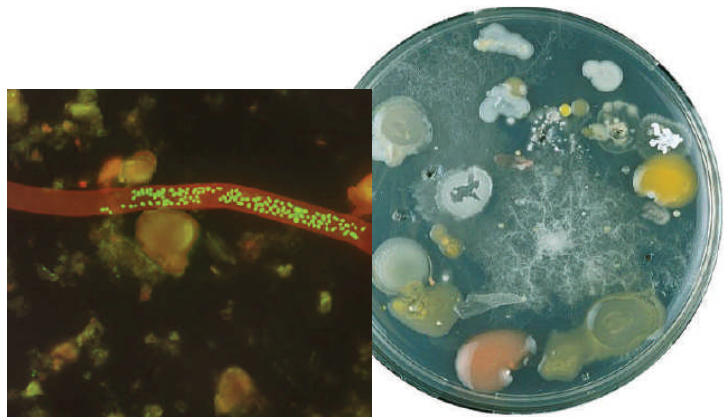
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**b) Nematodes** are biological regulators and represent one of most numerous and speciose groups. Nematodes in soils are trophically diverse and include economically important plant parasites. They show a high and diverse sensitivity to pollutants and because of their trophic diversity nematode assemblages do not only reflect their own fate, but also the condition of the bacterial, fungal and protozoan communities. These characteristics make them potentially interesting bio-indicators for soil health and soil disturbances [57]. Although nematodes can easily be sampled and extracted from soil, their identification is time consuming and requires expert knowledge. Previous studies demonstrate that the small subunit ribosomal DNA (SSU rDNA) gene harbours enough phylogenetic signal to distinguish between nematode families, genera and often species [58,59]. A robust & affordable quantitative PCR-based nematode detection tool for agricultural and scientific purposes, and comparable tools for the assessment of the ecological condition of soils are being developed [60]. Briefly this works as follows: after nematodes extraction from soil the nematode community is lysed, and after DNA purification the lysate is used to quantitatively characterize nematode assemblages. The difference in DNA contents of various life stages is limited, and different distributions of the life stages barely interfere with quantitative community analyses. Verification in recent field studies suggests that Q-PCR based analysis of nematode assemblages is a reliable alternative for microscopic analysis. The availability of an affordable and user-friendly tool might facilitate and stimulate the use of this ecological informative group of soil inhabitants.



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**c) Microorganisms:** Chemical engineers are soil organisms that decompose organic matter and





transform nutrients. Soil microorganisms dominate this functional group [3,17]. They indicate environmental changes by modifications in i) quantity/biomass, ii) structure and/or iii) activity [35,36,61]. Until now the impact of microbial biomass versus community structure on the active contribution of microbes to ecosystem processes is uncertain [35,62,63]. In some cases community structure was important for the maintenance of microbial function, like C and N cycling [64], but in others microbial biomass [65]. Functional redundant microorganisms exist, but their occurrence depends largely on function and environment considered [13,14,34]. Disconnections between factors driving microbial community structure and those driving its function further complicate indicator selection [65]. To comprehensively assess soil microbial diversity according to the definition of Bispo et al., 2009 [9] it is recommended to include indicators of each parameter group. However, the number of studies and monitoring networks using indicators of all three groups is limited (Table 1). Different methodological approaches are used to describe and quantify microbial diversity on the genotype, phenotype or metabolic level (method overview see [43]). To achieve progress in the area of microbial indicators it is of special importance to work on the definition and identification of microbial functional groups and their reaction to environmental changes [34]. The analysis of proteins expressed by microbial communities (metaproteome) is one emerging and promising tool to track new functional genes and metabolic pathways of soil microbes in the future [42]. Beside molecular based approaches new conceptual models are needed to link microbial diversity with ecosystem processes. The development of concepts describing the relationship between the stoichiometry of microbial biomass (e.g. the C, N and P status of soil microorganisms) and nutrient cycling is promising [41].

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