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Different approaches to evaluating soil quality using biochemical properties

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Abstract

Soil biochemical properties are indicators of soil quality, but there is still no consensus as to how they should be used. We review the trends in their use over the last decade. Generally, biochemical properties related to the biocycles of the elements (C, N, P and S) are used to diagnose soil quality. These properties include both general biochemical parameters (i.e. microbial biomass C, dehydrogenase activity and N mineralization potential) and specific biochemical parameters (i.e. the activity of hydrolytic enzymes, such as phosphatase, urease and β-glucosidase). Biochemical properties can be used both individually, as simple indices, or in combination using complex equations derived from mathematical combinations or the application of statistical programs. The results described in the literature for both are contradictory and question the validity of the use of biochemical properties as quality indicators. Complex expressions, in which different properties are combined, are thought to be highly suitable for estimating soil quality, although their use is limited to the area and situation in which they have been described. Generally, the greatest problems posed by the use of biochemical properties as soil quality indicators include the lack of reference values, the contradictory behaviour shown by these properties when a soil is degraded, and the regional variations in expression levels. Most of these problems are derived from the scarce information available on the biochemical properties of soil. For this reason, obtaining soil quality indicators of general use will require a coordinated effort from the international scientific community to standardise the analytical methods and to compile databases of biochemical properties from soils under diverse geographic conditions and with different uses and management.
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1. Introduction

Over the last 10 years, soil quality has been one of the topics of greatest interest in soil science, so much so that a database such as the Soil Science CAB Abstracts® Database (CAB International Publishing) supplies more than 1500 publications that use the term 'soil quality' as a key word. This interest has been focused on defining the concept of soil quality and on searching for reliable ways for evaluating this quality. With regard to the definition of soil quality, in recent years the key involvement of the soil in crop production and in water and atmospheric purification has been recognised, thus emphasizing the role of the soil both for production and for environmental quality. This has led to a profusion of definitions of soil quality, but that of Karlen et al. (1997): "soil quality is the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation", is widely accepted. Similarly, attempts have been made to separate the concept of soil quality from that of soil health. The limits of the two concepts are not particularly clear, but it is currently accepted that the term quality refers to the aptitude of the soil for carrying out a specific function, while health refers to its overall condition (Doran and Safley, 1997; Doran, 2002).

In spite of the large number of scientific papers that choose soil quality as a keyword, less than 20% deal with

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the estimation and quantification of the level of soil quality. This lack of concern for soil quality evaluation may well be a reflection of its difficulty, since according to Nortcliff (2002), the search for quantitative indexes for soil quality is difficult. This is especially so when one considers that many changes take place over the long term, and thus a change in soil quality can only be perceived when all the effects are combined over a period of time. Authors often concentrate on finding out which soil properties best reflect the change in soil quality. Even though soil quality is affected by a large number of properties, the majority choose a small group of edaphic properties (basically biological and biochemical properties) to assess soil quality. Unfortunately, there is a notable absence of suitable and agreed evaluation procedures by which these properties are adequately measured. As a consequence of this lack of 'consensus', this review summarises the many attempts that have been made at the quantitative evaluation of soil quality. Its objectives are:

(a) To discuss which soils represent the maximum or optimal level of quality against which the measured quality of a given soil should be compared.
(b) To highlight which biochemical properties are usually employed in the evaluation of soil quality and why these are selected.
(c) To summarize the different ways in which these properties are used in order to evaluate soil quality and the problems that their use entails.
(d) To make suggestions about the future needs in the evaluation of soil quality using biochemical indicators.

2. The highest quality soils

Due to the lack of agreement on the definition of soil quality, there is currently no general consensus regarding the soils that should be considered of maximum quality. The different approaches to the latter point can be summarized in two options. The first considers that a maximum quality soil is the soil in equilibrium with all the components of the environment, i.e. a climax soil developed under climax vegetation. The second option considers that the maximum quality reference soils are soils capable of maintaining high productivity and of causing the minimum of environmental distortion.

The idea of using climax soils as the highest quality soils is not a new one; it was suggested by Fedoroff (1987) for the evaluation of soil degradation, and is based on the fact that soils that develop freely reach an equilibrium amongst their properties that leads to long-term stability in natural ecosystems. The use of climax soils as higher quality soils also makes it possible to have a reference to use in each geographic area, in comparison with the use of a single 'ideal soil'. Although the use of an ideal soil as a sole reference has been advocated by various authors (Arshad and Coen, 1992; Doran and Parkin, 1994; Sinclair et al., 1996), this idea has been criticized by others, since this 'ideal soil' will not always match maximum standards of quality (Sojka and Upchurch, 1999). In spite of this, climax soils have not been widely used as the best quality soils. Nonetheless, Rassmussen et al. (1989) used them for comparing different soil management systems, Cook and Hendershot (1996) in the establishment of admissible levels of lead pollution, Leirós et al. (1999) to evaluate the level of soil degradation from different causes, and Sánchez-Marañón et al. (2002) to study the effects of change in soil use on the quality of Mediterranean mountainous ecosystems. The reason for the infrequent use of climax soils could be that in the majority of the developed world these soils have practically disappeared. Furthermore, even though samples of climax soils are conserved in museums and research centres, often it is not possible to estimate the value of those properties selected as being quality indicators as storage may have altered these soil properties.

The other option for establishing maximum quality reference soils is to use soils capable of maintaining high productivity and of causing the minimum of environmental distortion; i.e. soils that maintain sustainable production, high levels of environmental quality, and human and animal health (Doran and Parkin, 1994; Jackson, 2002). The use of this option is justified by the fact that the productive function of the soil is still seen as being the most important one when determining its quality. From a temperate climate and a cropping system outlook there is the belief that the best quality soils would be those with characteristics similar to the Mollisols of central USA, independently of the geographic location in which the evaluation of soil quality is being carried out. The fact that the productive function of the soil has been considered as the most important factor has meant that most of the publications over the last decade have dealt with the estimation of soil quality of agricultural soils and their modification as a consequence of the soil management (e.g. ploughed as opposed to unploughed, organic fertilization as opposed to inorganic). A smaller number of publications evaluate the quality of degraded soils during the process of recovery or deal with the loss of soil quality by contaminating agents such as heavy metals, toxic wastes and pesticides.

3. Biochemical soil properties as quality indicators: why and which?

The selection of indicators that enable the quantification of soil quality is also important. In the correct functioning of a soil an immense number of physical, chemical and biochemical properties are involved. However, due to the impossibility of considering all these properties, it is necessary to make a selection. The selected indicators must satisfy a series of requisites: (a) sensitivity to the presence of the greatest possible number of degrading agents; (b) consistency in the direction of the change

undergone in response to a given contaminant; and (c) ability to reflect the different levels of degradation (Elliott, 1994). As Nortcliff (2002) points out, the evaluation of soil quality will have little or no value if the indicators are not selected rigorously. These must supply reference material to be able to measure trends and patterns, relate soil quality to other components of the system, and where trigger or precautionary thresholds are set, the quality may be assessed against these.

With regard to the selection of properties for use as indicators, Doran and Parkin (1996) consider a 'minimum data set' for use in soil quality evaluation, which includes physical (texture, rooting depth, infiltration rate, bulk density, water retention capacity), chemical (pH, total C, electrical conductivity, nutrient level) and biological (C and N microbial biomass, potentially mineralizable N, soil respiration) properties, although many of the properties included in this dataset, e.g. texture, do not satisfy the requirements given by Elliot (1994).

In general, the physical and physico-chemical parameters are of little use as they alter only when the soil undergo a really drastic change (Filip, 2002). On the contrary, biological and biochemical parameters are sensitive to the slight modifications that the soil can undergo in the presence of any degrading agent (Klein et al., 1985; Nannipieri et al., 1990; Yakovchenko et al., 1996). Hence, whenever the total sustainability of soil natural functions and its different uses has to be evaluated, key indicators must include biological and biochemical parameters. Considering the wide number of biological and biochemical properties involved in the functioning of the soil, Visser and Parkinson (1992) point out the need to consider different levels of study which entails the use of specific groups of properties. One level is that of the *biotic community*, which implies the use of properties that are related to the structure of the microbial population and classically used to verify the composition and distribution of different functional groups of microorganisms of soil. A second level involves *population studies*, which considers the dynamics of specific organisms or communities of organisms (biological indicators). A third level, *ecosystem level*, considers the use of properties that are related to the cycles of bio-elements (C, N, P and S), especially when related to the transformation of the organic matter in the soil; in other words, properties related to the size, diversity and activity of microbial biomass as well as to the activity of the soil hydrolytic enzymes.

The use of the biotic community option is generally restricted to the sphere of microbiology and used to verify the composition and distribution of different functional groups of microorganisms in the soil. Currently, the structure of microbial populations is usually determined with molecular techniques such as fatty acid profiles and DNA characterization, instead of the classical enumeration and identification techniques (Nannipieri et al., 2002), but these molecular techniques need costly equipment and specialized staff and thus their usefulness as a routine diagnostic tool is limited. However, with the development of diagnostic 'kits' this will become less of a problem. The use of biological indicators poses the problem of knowing which indicator responds to a specific soil treatment or contaminant. Therefore, in spite of being one of the better options, its use is severely limited by the fact that it is not always possible to know which contaminant has affected the soils. Thus, it is not surprising that the majority of biochemical soil properties used as soil quality indicators are biochemical properties that reflect either the activity of microbial processes (*general biochemical parameters*—Nannipieri et al., 1995) or that of hydrolytic soil enzymes (*specific biochemical parameters*). Moreover, these properties are capable of reflecting the actual flux of organic matter and essential elements in soil, and their transfer between individual pools, as well as between soils and other parts of the global environment. In general, ecologically related biological parameters should characterize soil as a dynamic part of the biosphere.

4. The different ways of using biochemical properties as indicators of soil quality

A consideration of the papers published from 1990 to 2003 having jointly as keywords: soil quality, soil biochemical properties and soil enzymes, shows that there are three approaches regarding the use of both general and specific biochemical parameters to estimate soil quality: (1) the use of individual properties; (2) the use of simple indexes; or (3) the use of complex indexes derived from combinations of different properties or deduced on the basis of statistical procedures. The aim of the following sections is to discuss the advantages and disadvantages of these approaches.

4.1. Individual properties

The use of a single biochemical property is a common approach used to estimate soil quality. This option has been employed when the concept of quality referred exclusively to the productive capacity (Moureaux, 1957; Lajudie and Pochon, 1956) or attempts were made to find the relationship existing between the production and value of a property (Sojka and Upchurch, 1999). Cultivation practices generally decrease the organic matter content of soils, and attempts have been made to establish a relationship between productivity and soil organic matter content. For estimation of soil quality using individual properties, 40% of the consulted published papers used a general biochemical parameter such as microbial biomass C, dehydrogenase activity, soil respiration, nitrogen mineralization capacity, FDA hydrolysing capacity or ATP content, whilst the remaining 60% considered a specific biochemical parameter such as urease or phosphatase activities.

Amongst the general parameters, the microbial biomass C is considered to be the most reliable (41% of authors) followed by dehydrogenase activity (28%) and N mineralization capacity (16%). Phosphatase (28%), β-glucosidase (16%) and urease (11%) activities are the most frequently used among the specific biochemical parameters and, appropriately, represent the P, C and N cycles. Microbial biomass C and N mineralization capacity have primarily been used to estimate changes in soil quality due to management and use, whilst dehydrogenase activity (as a general measure of viable microorganisms) has also been employed in soils affected by heavy metals and pesticides, as well as for the diagnosis of the degree of recovery of degraded soils.

Microbial biomass represents the living component of the organic matter of soil, excluding animals and plant roots. Although microbial biomass usually makes up less than 5% of soil organic matter (Dalal, 1998), it carries out many critical functions in the soil ecosystem, among which the following could be pointed out: it is both a source and sink for nutrients, it participates in the C, N, P and S transformations, it plays an active role in the degradation of xenobiotic organic compounds and in the immobilization of heavy metals, it participates in the formation of soil structure, etc. (Nannipieri et al., 2002).

Considering the key roles of microbial biomass in the proper functioning of the soil, its widespread use as a soil quality indicator is not surprising. Nevertheless, the results obtained by diverse authors show contradictory results. The microbial biomass C increased with the intensity of grazing in meadow soils (Holt, 1997; Banerjee et al., 2000) and with the cereal-pasture rotation (Dalal, 1998), decreased when the soil is cultivated (Caldwell et al., 1999; Kandeler et al., 1999b), and the effect of zero-tillage was unclear (Dalal, 1998). Furthermore, microbial biomass C increased in soils supplied with organic fertilizers (Dalal, 1998), showed an erratic behaviour for inorganic N fertilization (Singh and Singh, 1993; Ladd et al., 1994) and did not react coherently to the presence of herbicides (Voos and Groffman, 1997). Moreover, microbial biomass could not be considered as a good indicator of heavy metal toxicity in soils (Dalal, 1998). In laboratory experiments it has been observed that biomass decreases with the addition of Cd or Cu (Fig. 1); nevertheless, in field experiments Kandeler et al. (1996) observed that the microbial biomass increases when mixtures of metals are added to the soil, although other authors have observed decreases in microbial biomass in fields supplied with heavy metal polluted sludges (Chander and Brookes, 1993; Valsecchi et al., 1995; Filip, 2002). Taking these problems into account, we believe that the use of microbial biomass C as a reliable indicator is highly questionable.

The dehydrogenase activity of the soil is considered to be an indicator of the microbial redox system and of the oxidative activities of the soil (Trevors, 1984). Like microbial biomass C, this property has been used mainly to assess the influence of management on soil quality and

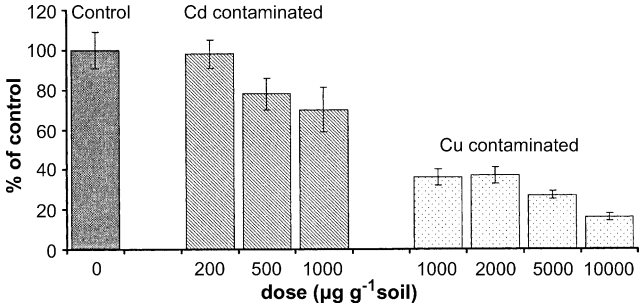


Fig. 1. Microbial biomass C in soils contaminated with different doses of cadmium and copper (three replicates for each treatment), as a percentage of the value of the microbial biomass C in uncontaminated soil (1200 mg kg^{-1}) used as a control (unpublished data from the authors).

contradictory results have also been obtained. For example, ploughing can both increase and decrease dehydrogenase activity (Bergstrom et al., 1998, 2000) whilst the addition of organic fertilizers, landfill effluents and industrial waste generally increases dehydrogenase activity (Bardgett et al., 1995; Pascual et al., 1999; Langer and Gunther, 2001). In general, this activity is not affected by the presence of heavy metals, unless in very high doses (Kandeler et al., 1996; Filip, 2002). Dehydrogenase activity has also been used to evaluate the degree of recovery of degraded soils, being considered as a good indicator, even in soils that have been contaminated by petroleum spillage (Margesin et al., 2000).

The nitrogen mineralization capacity refers to the capability of soils to transform organic nitrogen compounds into ammonium/nitrate under optimum moisture and temperature conditions over a given period of time. This determination has been used to assess the influence of soil management on soil quality. Significant seasonal variations limit its value as a soil quality indicator (Tscherko and Kandeler, 1999). It increases with light grazing (Banerjee et al., 2000) and with conventional ploughing (Kandeler et al., 1999b), although in the latter case the increase was explained by the mixing of the organic residues with the surface soil. Pankhurst et al. (1995) criticized the use of nitrogen mineralization capacity as a soil quality indicator because it is only scarcely affected by soil management, being only modified by additions of massive quantities of cattle slurry to the soil.

Among the hydrolases, acid phosphomonoesterase activity has been the most frequently used for estimating changes in soil quality due to either management or the presence of contaminants. It is a good index of the quality and quantity of organic matter in the soil (Jordan et al., 1995; Mullen et al., 1998; Bergstrom et al., 2000) and can be very high in arable soils as long as the levels of organic matter in the soil are maintained (Dick et al., 1994). For this reason, if the organic matter content of degraded soils increases during recovery, the level of this enzyme activity also rises (Gil-Sotres et al., 1992; García et al., 1997; Vance and Entry, 2000). Additionally, in soils affected by forest fires, phosphomonoesterase activity is clearly influenced by

fire intensity (Staddon et al., 1998) and increases after the fire in parallel with the soil recovery (Saá et al., 1993). Different studies have shown that this enzymatic activity increases as a consequence of organic fertilisation (Pascual et al., 1999; Chakrabarti et al., 2000) and decreases when phosphate fertilizers are used (Clarholm, 1993; Olander and Vitousek, 2000). Phosphomonoesterase activity is significantly decreased by the presence of lead (Marzadori et al., 1996) and other heavy metals (Kandeler et al., 1996), and the presence of pesticides in the soils only decreases it temporarily (Schäffer, 1993).

With regard to the enzymes involved in the carbon cycle, β-glucosidase has been the most widely used in the evaluation of soil quality in soils subjected to different management procedures. β-glucosidase activity was significantly lower in arable soils than in woodland and meadow soils (Bandick and Dick, 1999; Saviozzi et al., 2001). These findings could lead to the conclusion that β-glucosidase is a good indicator of soil quality, as it is negatively affected by some agricultural practices. However, other agricultural practices, such as organic fertilization, can increase the activity of this enzyme (Bandick and Dick, 1999; Pascual et al., 1999) thus reducing its value as an indicator of soil quality.

Urease, the enzyme that catalyses the hydrolysis of urea, has also been widely used in the evaluation of changes in soil quality due to soil management. Its activity increases due to organic fertilisation (Pascual et al., 1999; Chakrabarti et al., 2000) and after the addition of cattle slurry to the soil (Kandeler and Eder, 1993) and decreases as a consequence of ploughing (Saviozzi et al., 2001). Considering that both practices are normally carried out simultaneously in agricultural soils, it is clear that the use of urease as a soil quality indicator is of limited value.

The use of individual properties as soil quality indicators leaves a number of problems unsolved, confirming that which has been already indicated by Skujins (1978), Nannipieri et al. (1990) and Nannipieri (1994). These authors (and many others) consider that is conceptually wrong to assume that a single enzymatic activity, which reflects the enzymes catalysing a specific reaction in soil, could be taken as an index of more complex functions such as total microbial activity, soil fertility or soil quality, which all depend on many reactions and properties. But even in the case of a certain property having the capacity to reflect overall soil behaviour, in order for it to be considered a good indicator of soil quality it must fulfil another set of requirements since, in accordance with Dalal (1998), a good indicator should: (a) measure one or more functions of the soil; (b) be sufficiently sensitive to reflect changes due to disturbance, restoration or management; (c) provide reference, critical or threshold values; (d) be easily interpretable; and (e) be easy and inexpensive to obtain. None of the aforementioned biochemical properties satisfies all of these requisites, especially with regard to the existence of reference values. The first problem which severely limits individual biochemical properties as soil quality indicators is that even in high quality soils, such as climax soils, they show a high degree of variability due to climate, season, geographical location and pedogenetic factors (Table 1). It is also very difficult to compare the values obtained in one experiment with those from another, because they are generally obtained with different experimental protocols (especially in the case of enzymatic activities), either because there are no standardized methods or due to the soil samples having been subjected to different pretreatments (sample collection, sample storage, etc.) prior to their analysis. In order to mitigate the absence of reference values some authors use the percentages of the distribution of values (Kandeler et al., 1996) as an appropriate instrument for soil quality evaluation. However, this requires a large set of compiled data, and generally this is not the case.

These aspects, along with the erratic behaviour of biochemical properties, their inability to discriminate between the effect of the pollutant and any degradation that the soil may have undergone prior to its contamination, and the fact that, generally, they are not capable of discriminating contaminants according to the extent of degradation that they cause, have lead Trasar-Cepeda et al. (2000) to consider that individual enzymatic activities (and biochemical properties in general) have serious limitations as soil quality indicators.

Table 1
Variability of biochemical properties in soils from Galicia (N.W. Spain): range and seasonal coefficient of variation (seasonal CV)

	Range, seasonal CV (%)	
Biomass C (mg kg^{-1})	250–1468	25–28
Dehydrogenase activity (μmol INTF g^{-1} h^{-1})	0.10–0.34	22–46
N mineralisation capacity (mg kg^{-1})	1.4–70.0	28–72
Phosphomonoesterase activity (μmol PNP g^{-1} h^{-1})	2.33–15.76	23–46
β-glucosidase activity (μmol PNP g^{-1} h^{-1})	0.67–4.58	36–54
Urease activity (μmol NH_3 g^{-1} h^{-1})	3.17–49.78	40–48

PNP, *p*-nitrophenylphosphate; INTF, iodonitrotetrazolium formazan. Range refers to the variation of some biochemical properties in Ah horizons ($n=40$) of soils under climax vegetation (Atlantic oakwood) aiming to show the geographical and climatic variation of the individual properties. Seasonal CV shows the minimum and maximum values of the coefficient of variation of the individual properties (measured monthly over 1 year) for three soils located in areas of contrasting climate.

4.2. Simple indexes

Dalal (1998) proposed that the limitations shown by individual biochemical properties could be avoided by using simple indexes, i.e. relations between two biochemical properties. The ratio between two measured parameters represents a simple combination of two different measurements into a single criterion (Filip, 2002). However, and in spite of their simplicity, these indexes have not been generally adopted for evaluating soil quality. Among the most widely used simple indexes are the metabolic quotient (qCO_2), the death rate quotient (qD) and the ratio between biochemical properties and the total C and N soil content (specific activities if the biochemical property used is an enzymatic activity, Barriuso et al., 1988) or the C-biomass content (Kandeler and Eder, 1993; Landi et al., 2000).

qCO_2 is perhaps the most popular of these simple indexes. It represents the quantity of substrate that is mineralised per unit of microbial biomass carbon and per unit of time, and was initially used by Anderson and Domsch (1985). In general, in a distorted ecosystem the qCO_2 value increases in relation to a stable ecosystem (Dalal, 1998; Haynes, 1999). Nevertheless, the significance of this increase is unclear since it may be due to a drop in efficiency of substrate utilisation by the microflora, to a response of the microflora to adverse conditions (monoculture, acidification, high heavy metal concentrations, etc.), to the predominance of zymogene flora (r-strategists) over the autochthonous flora (K-strategists), or to the alteration of the bacteria/fungi ratio since they have different carbon use strategies (Dilly and Munch, 1998). The use of the qCO_2 has been also criticised because it can be insensitive to disturbance and ecosystem development, confounding the effects of disturbance with those of stress (Wardle and Ghani, 1995). In relation with soil quality evaluation, qCO_2 has been considered a good index of heavy-metal induced stress in soil (Brookes, 1995; Filip, 2002) as high qCO_2 values were found in long-term contaminated soils (Brookes et al., 1986). Moreover, several authors have indicated that highly productive soils have values of qCO_2 similar to contaminated soils (Valsecchi et al., 1995; Sparling, 1997). In addition, the values that have been suggested as being indicative of a stable soil ecosystem do not seem to have universal significance (Fig. 2). Therefore, this parameter may be of little value for soil quality evaluation, confirming that which was indicated by Dalal (1998). The same observations can be applied to the death rate parameter quotient qD, the C-loss quotient based on microbial C-loss determined on a weekly basis, described by Anderson and Domsch (1990).

Specific enzymatic activities have been used to compare and discuss values of enzymatic activities in soils with different organic matter contents (Barriuso et al., 1988) and could be considered as simple indexes of soil quality. The concept of specific enzymatic activity is derived from classic biochemistry in which the values of enzymatic activity are usually expressed according to the protein content, this being a way of normalizing analytic enzymatic results, although the use of this concept in soil biochemistry refers to the values of enzymatic activity expressed in units of C or total N. Kandeler et al. (1999a) have successfully used these types of expression to study the influence of organic fertilisation and soil use on enzymatic activities within particle size soil fractions. However, Trasar-Cepeda et al. (2000), in both field and laboratory experiments, have shown erratic behaviour in the values of specific dehydrogenase-, urease-, β-glucosidase- and phosphomonoesterase activities when these parameters were used to evaluate the quality of contaminated soils, suggesting that the use of specific activities is also very limited.

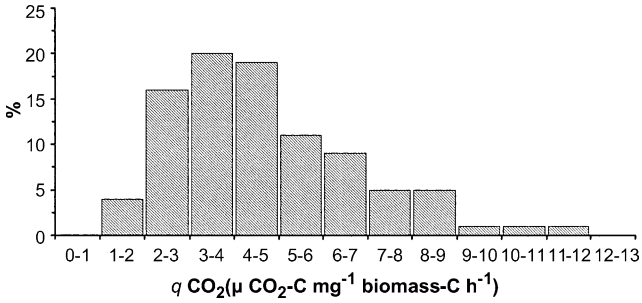


Fig. 2. Frequency percentage distribution of metabolic quotient values (qCO_2, μg CO_2–C mg^{-1} biomass C h^{-1}) obtained for 80 native soils under climax vegetation of Atlantic oakwood (unpublished results from the authors).

According to other authors, ecological information on specific aspects of microbial activity may be obtained by calculating the ratio of soil enzyme activity to microbial biomass. An increasing ratio may indicate either increasing enzyme production and enzyme release by microorganisms or an enhanced release of enzymes immobilized on clay or humic colloids to the soil solution (Kandeler and Eder, 1993). The ratios increase for enzymes of the nitrogen cycle in soils receiving high quantities of cattle slurry over long periods of time, due to the application of slurry inducing higher levels of enzyme production by microbial biomass (Kandeler and Eder, 1993). It has also been used in laboratory incubations to study the influence of the application of cadmium on certain biochemical and microbiological properties, in which it has been shown that this metal reduces the values of dehydrogenase and phosphomonoesterase per unit of biomass (Landi et al., 2000). Nevertheless, the scant use of this type of expression means that there are presently no data with which to confirm whether the ratios between enzyme activity and microbial biomass are useful for evaluating biochemical soil quality.

In summary, the use of simple indexes for soil quality suffer from the same limitations as individual biochemical properties: lack of reference levels, lack of coherent behaviour of the same ratio in the experiments of different authors, and lack of objective criteria for relating a variation in the index with the modification of a given process

occurring in the soil. In other words, one would have to accept (albeit reluctantly) the notion of Sojka and Upchurch (1999) that the search for a simple, affordable, workable soil quality index is unattainable.

4.3. Complex indexes

Another option for the evaluation of soil quality is to use complex indices calculated by the combinations of different biochemical properties of soil. Some of these attempts were first carried out in the 1980s, such as the indexes of Stefanic et al. (1984) and of Beck (1984), designed for soil fertility. Stefanic's index, the biological fertility index, uses a mathematical expression of the values of both dehydrogenase and catalase activities. Beck's index, or enzymatic activity number, includes dehydrogenase, catalase, phosphatase, protease and β-glucosidase. Some of these expressions were reformulated during the following decade (Stefanic, 1994). These expressions are essentially polynomial *formulae* in which the values of the enzymatic activities are multiplied by empirical factors. Their weakness lies in the lack of a constant and objective reference value.

In 1994 a different approach was undertaken by Sinsabaugh et al. (1994), who considered the degradation processes of plant remains in the soil involving a great number of enzymes that worked in cascade. One problem is that any one of these enzymes generates a short-circuit in the decomposition process, which leads to a loss in the soil degrading function, and therefore, in soil quality. Researchers that promote this idea consider six enzymes to be fundamental in the characterization of the overall biochemical state of soil: β-1-4-glucosidase, β-1-4-endoglucanase or endocellulase, β-1-4-exoglucanase or exocellulase, β-xylosidase, phenol-oxidase and peroxidase activities. Using principal component analysis (PCA) Sinsabaugh et al. (1994) obtained a so-called lignocellulosic factor which, when integrated throughout the whole litter decomposition process, was significantly correlated with the percentage of weight-loss of the plant-remains over time. The cascade of enzyme activities approach (i.e. the lignocellulosic factor) has been considered by Nannipieri et al. (2002) as one of the best among those using biochemical properties as indicators of soil quality due to its accurate and focused selection of enzyme activities. In any case, one criticism that could be posed to this approach is that only considers enzymes of the C cycle, ignoring the enzymes from the cycles of the rest of the biophilic elements (N, P and S). Consequently, it can behave as a good indicator of the soil capacity to degrade lignocellulosic material, but not as an indicator of the global capacity of the soil to degrade organic compounds.

A similar option was used by Monreal and Bergstrom (2000), who obtained a decomposition factor by PCA that was capable of explaining 96% of variation in soil enzymatic activity as a consequence of cropping system and tillage, and it was mainly due to the changes undergone by β-glucosidase, dehydrogenase and L-glutaminase activities due to soil management. Nevertheless, and with the aim of simplifying the evaluation of the process, Fioretto et al. (2001) preferred to consider only the determination of α-amylase and β-amylase among all the enzymes implicated in the litter decomposition process.

The search for a decomposition factor, based on the measurement of the activity of different enzymes working in cascade, could be considered as a good option. Unfortunately, however, no data are available to evaluate the response of these decomposition factors to the presence of contaminants or any other distorting agent in soils, which means that it is not possible to judge the validity of these approaches with any degree of certainty.

Trasar-Cepeda et al. (1998) showed that in native soils under climax vegetation, and without human distortion, the biochemical equilibrium that is characteristic of a mature stable ecosystem (Fedoroff, 1987) can be expressed mathematically as a combination of several microbiological and biochemical properties:

$$\begin{aligned}\text{Total N} = {} & 0.38\times10^{-3}\,\text{Microbial biomass C} \\ & + 1.40\times10^{-3}\,\text{Mineralized N} \\ & + 13.60\times10^{-3}\,\text{Phosphomonoesterase} + 8.90\times10^{-3}\,\beta \\ & - \text{glucosidase} + 1.60\times10^{-3}\,\text{Urease}\end{aligned}$$

Taking into consideration that this equation was obtained using climax soils, and being statistically very significant ($n=80$, $R^2=0.98$, $P<0.001$), Trasar-Cepeda et al. (1998) suggested that any disturbance or stress on the soil will modify this relation in such a way that the total N calculated by this equation (Nc) will be lower or higher than the total soil N content measured by Kjeldhal (Nk). Therefore, the Nc/Nk ratio could be used quantitatively as an index of the degree of soil degradation or disturbance. The same authors have employed this approach to evaluate the loss of quality in soils affected by management, by contamination with organic effluents and with heavy metals (Trasar-Cepeda et al., 2000). They observed that this relation differentiated between biochemically stimulated soils under stress (i.e. soils with more biochemical activity than could be expected on the basis of its total N content, for example situations brought about by the application of fertilizers or products that stimulate microbial activity) and those that are truly degraded. Furthermore, with this approach it is also possible to distinguish the effects of the pollutant from any prior degradation that the soil might have suffered, and to differentiate between the pollutants in relation to the different degrees of soil degradation that they may cause (Fig. 3). In spite of these promising results, this equation has only been used for soils from a certain geographic area, which currently makes it difficult to verify whether its validity is more general. Furthermore, and as a criticism of this option, Nannipieri et al. (2002) consider this equation to be too empirical and that the reasons for choosing the

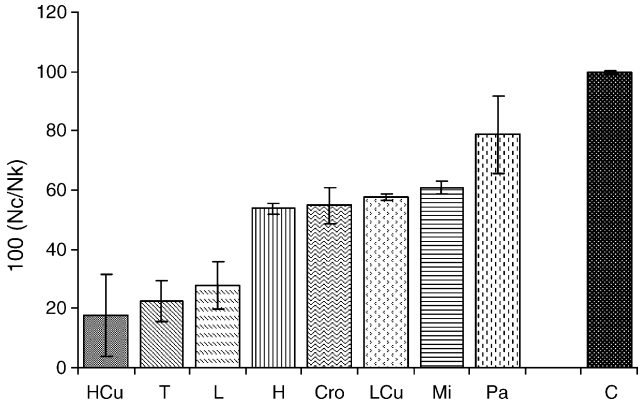


Fig. 3. Values of 100 (Nc/Nk) obtained for different cropped soils (Cro), pasture soils (Pa), minesoils (Mi), and soils contaminated by high and low dose of copper (HCu and LCu, respectively), landfill effluents (L), tannery effluents (T) and diesel spills (H) and native soils under climax vegetation (C). The represented values are average of three replicates in each case.

biochemical and microbiological parameters that appear in it are not always clear.

Dilly and Blume (1998) suggest that the use of indexes which combine data from a small number of biochemical properties could mask important microbial features in the soil ecosystem. For this reason, they suggest that it is preferable to select a set of 10 basic biochemical properties related to: (a) the content in microbial biomass (biomass C determined by fumigation–extraction and by substrate induced respiration, SIR); (b) the general microbial activity (basal respiration, dehydrogenase activity, arginine ammonification, DMSO reduction); and (c) the specific microbial activity (β-glucosidase, protease, and both alkaline and non-buffered phosphatase). These authors suggest the use of sun-ray plots or star plots, in which the variability in the form of the star reflects the diversity or abundance of the micro-organisms involved, and the area of the star is associated with vitality (Bachmann and Kinzel, 1992). The integrated signal resulting from the combination of both diversity and vitality can be used for the assessment of sustainable land use regarding soil microbiology (Dilly and Blume, 1998). For this representation the scale of each line needs to be adapted to the range of all values, and its use by the aforementioned authors has clearly shown the negative effects of single-crop farming and crop rotation on the biochemical quality of soil as opposed to the natural forest or meadow ecosystems. Apparently there would seem to be no great advantage to this approach, as similar conclusions were obtained by different authors using single properties or simpler indexes.

Another option in the use of a set of biochemical properties and currently in widespread utilisation involves statistical techniques (parametric and non-parametric). This option was already alluded to by Larson and Pierce (1991) when they described (even though they did not make specific formulations) mathematical functions involving minimum data sets and pedotransfer functions, and by applying statistical concepts for the evaluation of soil quality. Regarding the use of statistical techniques exclusively employing biological and biochemical soil properties, the following approaches are worthy of mention.

Bentham et al. (1992) use principal component analysis (PCA) in order to evaluate the degree of recovery of mine soils. The analysis cluster, based on the PCA, discriminates between the different habitats, the clustering based on three different biochemical indexes (dehydrogenase activity, ATP and ergosterol content) being especially useful. A classification of the habitats based on a three-dimensional arrangement of the biochemical properties of the soil could be implemented in order to verify its recovery potential and progress in recovered soils. Alternatively, multiple-variable indicator kriging (MVIK) developed by Smith et al. (1993), elegantly integrates different properties into a single, joint index with the aim of predicting the probability of an area satisfying a given quality standard. Indicator variograms are generated, which are the basis for estimating the quality of unsampled areas. The results are presented in the form of maps of the probability of having a given quality requirement. Approaches of this type have also been employed by Yakovchenko et al. (1996), using parameters deduced from the relation between the amount of N taken by the crop, and the mineralized nitrogen (microbial respiration plus net mineralized N) during the growth season. Halvorson et al. (1996) combine six variables, transforming them with a threshold-based indicator variable transform, into one single variable (MVIT). If this is created by adhering to soil-quality criteria, then the kriging could produce maps that indicate the probability of a soil being high/low quality. Wander and Bollero (1999), also using advanced statistical techniques, clearly demonstrate that the use of non-tillage practices improve the biological and physical condition of soil, and that a new step in the evaluation of soil quality could be to determine the biological and environmental relevance of soil quality measures together with soil processes at regional level. These authors suggest that only by developing efficient and consistent strategies can the determination of soil quality be of practical use to farmers and decision makers.

In short, the use of complex expressions in which various biochemical properties are involved appears to be a promising line of work. The inclusion of different biochemical properties makes it possible to better reflect the complexity of the soil system, and complex expressions also seem to be suitable for evaluating soil quality, at least for the condition in which they have been designed. The main drawback of these complex indexes is that they have not been tested in locations or under conditions other then those for which they were designed, and due to this at present there is no way of verifying whether they are or not universally valid. This tell us that in order to progress in the development of a general expression for the evaluation

of soil quality it will be necessary to carry out an intense and coordinated work at an international level.

5. Final remarks

The biochemical properties of soil have been used widely to evaluate soil quality, both individually and combined in simple indexes and in more complex ones, which stresses the fact that the scientific community recognises their potential value. Nevertheless, up until now, published results show that biochemical properties can still not be considered as efficient diagnostic tools, since, as has been stressed throughout the present paper, neither the individual properties nor the indices obtained with them can be considered to be of general use as those properties or indexes that appear to be valid in certain situations are totally ineffective in others.

There are a number of reasons that can explain the inability of biochemical approaches to quantify soil quality. From a methodological perspective, the lack of standard analysis methods accepted by all laboratories is a fundamental problem when interpreting the values of biochemical properties. Differences in sample collection, storage, pre-treatment, protocols for determining enzymatic activities (in which temperature, substrate concentration, incubation time, etc. are crucial) make it practically impossible to compare data obtained from different laboratories. Moreover, we should take into account the high degree of variability between biochemical properties, both seasonal and regarding edaphic factors, as well as the lack of reference values or broad databases for high-quality soils that could be used to make comparisons. All this leads to the often contradictory conclusions reached by different researchers, when describing the effects of a contaminant or of a given type of soil management on the soil quality.

These methodological problems, along with the inherent complexities of a dynamic soil system mean that with the knowledge currently available, no estimation of soil quality, using simple indexes or ratios, can be considered reliable. As Sojka and Upchurch (1999) point out, the use of one or two biochemical properties is not sufficient to demonstrate the complexity of the functioning of the soil system. Efforts in the use of biochemical properties as indicators of soil quality should be focussed on the search for complex expressions that are capable of describing the complexity of the soil much more accurately. Even though the different approaches mentioned in this review seem to be useful, it will be necessary to test them in areas and situations other than those in which they were generated in order to verify whether they are universally valid. In other words, it is obvious that the scientific community should make greater efforts to understand the behaviour of a broad group of soil properties and how they relate to each other and their role in the functioning both undisturbed and degraded soils.

Apart from the above considerations, one should bear in mind the present drive to formulate and introduce laws that make it possible to decide whether a soil is contaminated or not as well as to estimate the degree of recovery of a previously degraded soil. In the particular case of the European Union, these laws will have to be adhered to in 25 countries (constituting nowadays the European Union) with different land use types and within very different geographic and climatologic scenarios. However, at present there is no official methodology for estimating the level of soil quality; the laws currently in force only recommend the use of simple tests (e.g. soil respiration, root germination and elongation) whose generic application under the different conditions prevailing in Europe may be unreliable. This legislative interest in soil protection must give rise to a coordinated effort to search for a common methodology for the estimation of soil quality, in which biological and biochemical soil properties will play a very important role due to their high sensitivity to distorting agents. This implies that the standardisation of the methods and the construction of databases of soil biological and biochemical properties will have to be carried out. Unfortunately, these tasks are unattractive for the majority of research teams, as they require an enormous analytical effort and are presently unsuitable for financial support because they form part of basic research. Nevertheless, the attainment of these basic tasks is so important that only after they have been carried out will the search for globally applicable indexes for the evaluation of soil quality be possible.

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