

Biological soil quality indicators: a review

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The definition of soil quality encompasses physical, chemical and biological characteristics, and it is related to fertility and soil health. Many indicators can be used to describe soil quality, but it is important to take into account sensitivity, required time, and related properties, than can be explained. Properties related to organic matter content, such as C/N ratio, organic carbon fractions (humic acids, fulvic fraction); enzymatic activity (β glucosidase, urease, aryl sulfatase, phosphatases) or aggregate stability, can be used as soil quality indicators. They provide early information about mineralization processes, nutrient availability and fertility, as well as effects resulting from changes in land use, or agricultural practices (e.g. tillage or application of different types of organic matter). In this context, biological properties have been used as soil quality indicators, because of their relationship with organic matter content, terrestrial arthropofauna, lichen, microbial community (biomass or functional groups), metabolic products as ergosterol or glomalin and soil activities as microbial respiration and enzyme production.

Keywords: Biological indicators, soil quality

Introduction

The interest in soil quality can be traced back to the ancient Roman civilization. Through the time, the use of agricultural residues, application of organic matter, rotation, and tillage practices has been fundamental in maintaining soil fertility. One important discovery, at the end of the nineteenth century, was the nitrogen fixing microorganisms, associated with roots that opened the door to a better understanding of rhizosphere and the development of soil ecology as related to soil fertility.

Traditional soil management in agriculture is based on temperate crop rotations with grass crops for livestock production, improving soil structure and increasing fertility, with an important role of animals and natural fertilizers. After the Second World War, this traditional system was reduced, increasingly separating livestock from arable land, which led to the elimination of grass and animal manure application in many arable crop systems. Soil management was neglected, leading to growing concerns about the physical condition of the soil, which was evident in the report "Modern agriculture and the earth" (1); soil erosion (2,3) and leaching of nutrients. These concerns triggered definitions of national policies in Canada, United States (4) and England (5) aiming at land conservation and recovery of soil's ability to meet its multiple functions, concepts that finally met in "soil quality".

This concept of soil science dates back to the 1970s. when Warkentin (1977) suggested the development of a concept of soil quality that encompasses the following facts (6):

1. Land resources are being evaluated for different uses
2. Multiple stakeholder groups are concerned about resources
3. Priorities of society and the demands on land resources are changing
4. Soil resources and land use decisions are made in a human or institutional context.

The Soil Science Society of America (SSSA), after much discussion about the subject, came with a broad definition: "The ability of a specific type of soil to function within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or improve air quality and water to support human health and livable" (7).

Other definitions have been proposed by different groups and authors (8, 9). All share relevant elements summarized as the sustainability of the soil as a resource for food production, to support human life and to preserve or improve the soil for future generations. According to Doran (2000), the biggest challenge is to maintain the balance between ecosystems and to establish priorities between food production, energy and fiber production (10).

In this context, soil quality acquires an important dimension related to the strategies for conservation, health, good agricultural practices and agroecosystems sustainability. The problem is how to measure this soil quality if it depends on climate, soil characteristics, vegetation, anthropic influence and interactions among them?

1. Soil quality indicators

Indicators of soil quality have been defined, from the ecological, economic, and social development standpoints; they usually take into consideration soil properties or associated crops that can be used in response to the dynamic changes in agroecosystems. These indicators are not well defined, nor accepted or approved parameters to characterize or to define soil quality exist (11). Changes in soil quality can be measured through indicators which include physical, chemical and biological processes and characteristics so it is necessary to provide quality indices including different indicators, to determine soil quality.

According to USDA soil quality indicators are classified into four categories that include visual, physical, chemical, and biological indicators. Visual indicators can be obtained through field visits, perception of farmers, and local knowledge. These are identified through observation or photographic interpretation, subsoil exposure, erosion, presence of weeds, color, type of coverage, and through comparison between systems operated with the unaudited interim anthropogenic, which gives a clear idea whether the soil quality has been affected positively or negatively (12). Mairura (2007), reported the integration between scientist's and farmer's evaluation in Kenya and showed how the local knowledge used as indicators was valid for consistent classification of soil quality (13).

The physical indicators are related to the organization of the particles and pores, reflecting effects on root growth, speed of plant emergence and water infiltration; they include depth, bulk density, porosity, aggregate stability, texture and compaction.

Chemical indicators include pH, salinity, organic matter content, phosphorus availability, cation exchange capacity, nutrient cycling, and the presence of contaminants such as heavy metals, organic compounds, radioactive substances, etc. These indicators determine the presence of soil-plant-related organisms, nutrient availability, water for plants and other organisms, and mobility of contaminants.

Finally, biological indicators include measurements of micro- and macro-organisms, their activities or functions. Concentration or population of earthworms, nematodes, termites, ants, as well as microbial biomass, fungi, actinomycetes, or lichens can be used as indicators, because of their role in soil development and conservation; nutrient cycling and specific soil fertility (14). Biological indicators also include metabolic processes such as respiration, used to measure microbial activity related to decomposition of organic matter in soil, and a commonly used index: the metabolic quotient (qCO_2), defined as the respiration to microbial biomass ratio, which is associated to mineralization of organic substrate per unit of microbial biomass (15).

Other biological indicators that have been widely studied are the chemical compounds or metabolic products of organisms, particularly enzymes such as cellulases, arylsulfatase, phosphatases, related to specific functions of substrate degradation or mineralization of organic N, S or P. Soil enzymatic activity assays act as potential indicators of ecosystem quality being operationally practical, sensitive, integrative, described as "biological fingerprints" of past soil management, and relate to soil tillage and structure (16). Determination of rates of decomposition of plant debris in bags or measurements of the numbers of weed seeds, or the presence and quantification of the population of pathogenic organisms can also serve as biological indicators of soil quality (17).

1.1 Selection of soil quality indicators

Soil quality is estimated by observing or measuring different properties or processes, and, several of these indicators can be used to determine soil quality indices.

According to different authors (10, 18), indicators should be limited and manageable in number by different types of users, simple and easy to measure, cover the largest possible situations (soil types), including temporal variation, and be highly sensitive to environmental changes and soil management (16).

The selection of indicators thus depends on the soil and functions being assessed. These features include, among others: support for the development of living organisms, water and nutrient flows, diversity and productivity of plants and animals, elimination or detoxification of organic and inorganic contaminants. Likewise, the selection depends on the sensitivity of these properties to soil management or changes in climate, as well as the accessibility and usefulness to producers, scientists, conservationists and policy makers (19, 20).

The selection of indicators implies knowing research needs, and the power to interpret the indicator: the land use, the relationship between the indicator and the soil function that is being evaluated, the easiness and reliability of the measurement, the variation in time of the crop, application of organic matter or crop rotation in relation to sampling, the sensitivity of the soil property to be measured against changes in the ecosystem (20).

Moreover, many soil ecosystems functions are difficult to infer directly and, consequently, soil quality must often be inferred from other easily measurable soil properties (21), bearing in mind that soil quality should be directed mainly towards the detection of changes or trends that can be measured in time; however some indicators may change faster than others; thus not only the changes detected must be real but also sufficiently sensitive in short periods of time, so a quick action on the agroecosystem can be taken to correct problems before undesirable situations or irreversible losses of soil quality occur. General properties as aggregate stability, bulk density, pH, salinity, cation exchange capacity, microbial biomass, enzymatic activity, and basal respiration are used as indicators of soil quality (21).

In fact, some authors suggest that a soil quality indicator is not adequate if it is not directly related to the target user. If the goal is a quality index for soil crop production, then soil organic matter, infiltration, soil aggregation, pH, microbial biomass, N forms, bulk density, electrical conductivity or salinity, and available nutrients, represent a group of indicators that can be used to describe most of the soil basic functions like the ability to accept, hold and release water to plants, maintain productivity, and respond to management and erosion processes (20).

In the same way, for a better interpretation of soil quality indicators, Segnestam (2002) expressed the need of using a baseline for comparison and to determine whether positive or negative impacts on environment have occurred. Besides, variations in time and rates of change as well as local indicators should be determined to define potential models for larger scales (22). For this reason the indicators associated to organic matter are considered to determine soil quality; they can be correlated with different chemical, physical and biological properties, some of them having high sensitivity, and their changes can offer stakeholders, policy or research institutions, correlated results in short time and make decisions timely for a given agroecosystem.

2. Role of soil organic matter (SOM) and associated indicators

Soil organic carbon (SOC) is a soil property considered one of the most important indicators of soil quality; it has positive effects on soil physical properties and promotes water infiltration, storage and drainage (23, 24). It is directly related to the maintenance of soil structure, presence of different groups of microorganisms, mineralization of organic matter, and nutrient availability.

Soil properties associated with soil organic matter (SOM) have been recognized as key indicators (8) and to have an effect on other properties. Soil organic matter defines the energy supply to microorganisms, availability and quality of substrates, and the biodiversity necessary to sustain many soil functions. However, SOM content varies with changes in climate, soil and crop management, being higher in places with larger average annual precipitation, lower mean annual temperature, and higher clay content (25, 26). Similarly, SOM content is affected by intermediate grazing intensity, incorporation of crop residues or the addition of organic matter fractions and by soil management practices such as minimum or conservation tillage. Franzluebbers *et al.* (2002), proposed stratification ratios of soil properties, i.e. N and C pools, including total and particulate organic C and N, soil microbial biomass C, and potential C and N mineralization to explain differences respect to soil quality in soils with conventional tillage and no tillage (27).

Regarding SOM decomposition, there are factors such as N and P concentration, clay or polysaccharides content that affect its decay, altering soil properties associated with soil quality. Some fractions, like starch or protein, are easily metabolized while humic substances are more resistant to decay (28); the latter participates in nutrient exchange processes, formation of aggregates between organic substances and mineral particles, and in the immobilization of toxic materials (29).

Haynes (2000) studying arable and pastoral soil in New Zealand, indicated that soil organic matter content is affected by soil management, but changes in total SOC content from land use may be difficult to detect; therefore labile organic fractions, represented by dissolved organic carbon (DOC), N or P, are more sensitive properties (30) to change, since the easily mineralizable carbon fraction is the direct source of energy for microorganisms in soil. Chan *et al.* (2002), found that particulate organic matter is more responsive to changes in management practices, than the total organic carbon, being related to aggregate stability and nitrogen mineralization (31).

3. Biological indicators

Biological indicators also include properties associated with biological activity on organic matter, such as microbial biomass carbon (32) and soil respiration (17, 33); they also include various indices such as abundance, diversity, food chains, stability of communities (10), and organisms associated to mesofauna such as earthworms, nematodes and arthropods which are used as soil restoration or ecological indicators (34). Finally, biological activities such as enzyme activity (35), potentially mineralized nitrogen or CO₂ production are associated to this group of biological indicators (10, 36).

Soil organisms are sensitive indicators, and reflect the influence of human management and climate changes. Similarly, soil organisms are considered indicators of quality and health because the diversity and abundance may be related to functions such as decomposition of organic matter, plant and root development (competition), sequestration and detoxification of heavy metals (37), pesticides and other pollutants, disease-suppressive soil, and presence of pathogens in soil and plant (38, 42).

3.1. Earthworms

For many people the organisms are also a way of predicting changes in soil functions. They help to understand the causes and effects of management practices and land use on plant productivity. Earthworms for example, have been shown to be important in organic matter decomposition, particularly by soil movement that results in incorporation and mix of residues.

Earthworms are recognized as a key factor in the way many terrestrial ecosystems work (43). They help in aggregate stability, to improve water-holding capacity, pore size and infiltration rate (44).

Studies by Lapped *et al.* (2009) in the Southern agricultural region of Saint-Lawrence Valley-Canada, described the relation between two endogen earthworm species and soil aggregate stability as soil quality indicators; there was a relationship between the presence of *Aporrectodea caliginosa* and silt and clay contents, while *Allolobophora chlorotica* presence was related with OM amendments (45). The overall earthworm population was low, which is common for tilled agricultural soils, but benefited from the OM input from the urban compost, a proof that fertilization based on organic residues, as implemented in organic agriculture, is highly beneficial for earthworms and soil quality.

When organic matter is incorporated to the soil as amendments or organic waste, earthworms can help regulating temperature and aeration. Dick (1983), indicated that earthworms reduce negative effects caused by increased temperature when organic matter is added in surface soils, during spring. Edwards and Noble (1982) described that earthworm populations peak in late autumn and again in spring allowing a good mix of organic matter with a positive impact on soil aeration (46, 47).

Earthworms have been demonstrated as indicators of anthropogenic land use. In Netherlands they are used as a biological indicator of soil quality and in Germany as a factor for soil biological site classification, a practical form to define the use of soil, based on the structure of the earthworm community, their abundance and biomass (48). Experiments by Mackay and Kladvko (1985) and by Edwards and Lofty (1982), indicated that reducing soil tillage practices and pesticide applications, and increasing organic matter in soil, resulted in increased earthworm populations (46, 49). These organisms have been also recognized as easy and simple (for land managers) indicators of soil health due to high sensitivity to pesticides and to high concentrations of heavy metals such as Cu, Cd, Hg, Pb, and Zn (50), metal organic compounds (pentachlorophenol) and PCBs (51, 52); Wang *et al.* (2009), studied the earthworms and soil microbial biomass carbon (SMBC), as support of chemical analysis and bioindicators of soil pollution with heavy metals in an abandoned copper mine in eastern Nanjing, China. Earthworms from families *Megascolecidae*, *Moniligastridae*, and *Lumbricidae* were present. There was a poor correlation between earthworm densities or biomass and chemical parameters. The authors developed several linear regression models based on the soil physical and chemical characteristics and metal concentrations in earthworm bodies, proposing the use of soil-dwelling earthworms as an indicator of metal availability (53).

The knowledge of earthworms and their role in soil is increasing. The research techniques are an important tool to facilitate meaningful analysis from the micro-scale within a soil profile (e.g. drilosphere effects) to a field scale or landscape scale. Furthermore, an additional framework of understanding is required to investigate the role of earthworms in the biogeochemical cycles. Authors like Barlett *et al.* (2010), propose the integration of technologically advanced methods in combination with systems based on models in order to understand at the landscape scale, the functions of earthworms as individuals and populations within their ecosystems (43).

3.2. Ants and termites

Likewise, other invertebrates in soil, ants, have been used as biological indicators of soil quality and soil management changes. The ants have been studied because of their easy collection, abundance (biomass), relevance in food chains, and sensitivity to environmental disturbances.

Boulton *et al.* (2003) studied the effect of ant (*Messor andrei*) nests and adjacent, non-ant, soil from a semi-arid, serpentine grassland in California, on major soil organism groups (bacteria, fungi, nematodes, miscellaneous eukaryotes, and microarthropods). They found that all groups were more abundant and diverse in ant nests than in the non-ant soil, and all soil nutrients were similarly enriched inside nests, suggesting that these ant nests exert significant effects on the resident soil biota through the movements of nutrients to the soil surface. Andersen *et al.* (2002), explained the importance of ants as biological indicators, especially in restoration processes after impacts (i.e. mining industry), because their presence depends on the diversity and composition of plant communities making them a better predictor of microbial biomass than plant species diversity (54, 55).

On the other hand, ants along with termites have been recognized as indicators of land recovery, as their presence probably increase carbon and nutrient levels, especially nitrogen, phosphorus and potassium, as well as calcium and magnesium exchange due to transport and decomposition of fresh material by their enzymatic system (56, 57). Cammeraat *et al.* (2002), studied the effect of seed harvesting ants (*Messor bouvieri*) on fertility, rainfall infiltration, structural properties, and water repellency of top semi-arid soils in Spain; the soils of the ant nests had a lower pH, higher concentrations of organic carbon and inorganic nutrients, higher structural stability, significant higher infiltration and were more water repellent than the control areas. The ant nests act as sinks for water under slightly humid to wet conditions, whereas under extremely dry conditions, which prevail in summer and the beginning of autumn, infiltration is strongly reduced (58).

Termites have also been used as soil quality indicators. In tropical rain forest of Lopé - Gabón Africa, Roose-Amsaleg *et al.* (2005), working with soil-feeding termite nests of *Cubitermes* of different ages (fresh to mature to old), and soils that originated from three forests differing in terms of age and vegetative cover, demonstrated that mature

nests increase significantly the concentration of potassium and phosphorous, clay and fine silt, pH, organic matter, water retention capacities and cationic exchange capacity compared to control soils (59).

Jimenez *et al.* (2008), in savanna ecosystems in Colombia, demonstrated that termites and ants affect the availability of tropic and spatial resources for other organisms, increasing OC and NH₄ average in relation to soil without influence of biogenic structures. In the same region, Mora *et al.* (2005), evaluated the changes in 10 soil enzymes (xylanase, amylase, cellulase, α -glucosidase, β -glucosidase, β -xylosidase, *N*-acetyl-glucosaminidase, alkaline and acid phosphatases, and laccase), to characterize the functional diversity in soil-feeding termite (*Ruptitermes* sp.) and soil pellets produced by two species of leaf-cutting ants (*Acromyrmex landolti* and *Atta laevigata*) in relation to a control soil; it was demonstrated that some of the soil pellets have different enzymatic profile that the control soil and the diversity of these structures and species are related to different pathways for the decomposition of organic matter (60, 61).

3.3. Microorganisms

Microorganisms are widely used as soil quality indicators. Soil contains a large variety of microbial taxa with a wide diversity of metabolic activities (62). Soil microbial biomass compared with that of superior organisms is a more sensitive indicator and is influenced by different ecological factors like plant diversity, soil organic matter content, moisture, and climate changes. Microorganisms play a key role in nutrient cycling and energy flow (63) and provide information on the impact of intercropping, incorporation of organic matter, management practices (64), and tillage activities contributing to soil structure and stabilization (65). Microbial communities respond to environmental stress or ecosystem disturbance, affecting the availability of energetic compounds that support microbial population (66).

Nitrogen is the most limiting crop nutrient; thus biological N fixation, particularly the development of productive associations of plant and N-fixing bacteria and possibly other free living N fixers, has the potential to provide an endless and low cost source of N. Another important nutrient for many soils worldwide is phosphorus (P); many soils are inherently low in P and have high P adsorption capacity. This is exacerbated by the lack of P replacement when removed by crop harvesting and soil erosion. Unlike N deficiency, which potentially can be corrected by biologically fixing atmospheric N₂, P deficiencies need to be corrected by adding P sources to soils and increasing P uptake efficiency. Besides biological N₂ fixation and P solubilization in the rhizosphere, microorganisms can enhance nutrient use efficiency by increasing root surface area e.g. mycorrhizal, fungi, promoting other beneficial symbioses of the host plant, and microbial interactions. Nitrogen fixation and P solubilization are possible under low N and P concentrations, which means that the presence of a symbiotic process can be used as indicator of N or P availability in soil (67).

However, it is important to know that microbial indicators have advantages and disadvantages and should be selected based on easiness of measurement, reproducibility and sensitivity to variables that control quality and soil health. In addition, many of the microbial groups are culture-independent, making essential the use of molecular techniques, which complement the traditional culture techniques (68).

3.4. Metabolic substances

There are many metabolic substances that can be used as soil quality indicators; they include sterols, antibiotics, protein, enzymes, etc.

- Ergosterol

Ergosterol, is the main endogenous sterol of fungi, actinomycetes, and some microalgae; its concentration is an important indicator of fungal growth on organic compounds and mineralization activity (69). Barajas *et al.* (2002) demonstrated that heavy metals (Cu 80 ppm, Zn 50 ppm or Cd 10 ppm) and fungicides (Thiram 3 ppm or pentachlorophenol 1.5 ppm) at concentrations that reduce the metabolic activity between 18% and 53% (pollutant-stressed cultures) did not affect the ergosterol content, while the fungicide Zineb (25 ppm) reduced significantly the ergosterol content in biomass basis. Likewise, Molope *et al.* (1987), working with pastures and arable soils, found a correlation between fungi hiphae and ergosterol quantity and soil aggregates stability demonstrating by electronic microscopy the importance of fungi in Thixotropy, a purely physical process involving rearrangement of the clay micelles, in soil (70, 71).

Puglisi *et al.*, (2003) analyzed the content of cholesterol, sitosterol, and ergosterol in agricultural soils (hazelnut in Napoles irrigated with contaminated water and intensive horticulture in Bari, Italy), determining that crop rotation does not affect the presence of these sterols, but ergosterol plays the most active role in soil metabolic activity in soils with industrial contamination (72).

In the Pacific region, Joergensen and Castillo (2001), determined positive correlations between qCO₂ and ergosterol-to-biomass C ratio and between biomass C-to-soil C ratio and clay content and suggested as key problems of soil fertility and soil quality in Nicaragua, the low availability of soil organic matter and phosphorus to soil microorganisms, which are magnified by a low percentage of fungi, probably reducing the ability of soil to provide nutrients for plant growth (73).

- Glomalinalin

Among these fungal components, glomalin, an insoluble and hydrophobic proteinaceous mix of substances (74), is of particular interest. Glomalin as glomalin-related soil protein (GRSP) has been proposed to improve the stability of soil by avoiding disaggregation by water (75, 76). A strong relationship between glomalin concentration and the amount of water stable aggregates (WSA) has been demonstrated based on research made by Harner *et al.* (2004), with soil chronosequence, and observations made by Wright and Upadhyaya (1998); Rillig (2004) proposed that aggregates (and soils) with high glomalin-related soil protein (GRSP) concentrations may be fairly “saturated” with GRSP, perhaps because most pores in these macro-aggregates have already been partially “sealed” by deposition of this substance, slowing down penetration of water into the aggregate (77, 78)

Glomalin-related soil protein (GRSP), has been studied by many researchers as a biochemical marker in soil (79), specially because of stability below a certain steady-state level, despite potentially negative management effects on mycorrhizal fungi, such as tillage, inclusion of fallow into crop rotation and fertilization with inorganic phosphorus (80). Bedini *et al.* (2009), using *Medicago sativa* plants, inoculated with different isolates of *Glomus mosseae* and *Glomus intraradices* in a microcosm experiment, found significantly higher aggregate stability (as mean weight diameter (MWD) of macro aggregates of 1–2 mm diameter), in mycorrhizal soils compared to non-mycorrhizal ones and the GRSP concentration and soil aggregate stability were positively correlated with mycorrhizal root volume and weakly correlated with total root volume (81).

– Enzyme activities

For many years, studies on soil microbiology have been based only on the analysis of the microflora present in the soil, ignoring the influence of extracellular enzymes produced by microorganisms on the decomposition of organic matter and the continuous flux of different elements in the soil (82). Soil enzymes play biochemical functions in the overall process of organic matter decomposition in the system; they are important in catalyzing several reactions, necessary for the life processes of microorganisms in soils, the stabilization of soil structure, the decomposition of organic wastes, organic matter formation, and nutrient cycling, providing an early indication of the history of a soil and its changes in agricultural management (83, 84). Thus, they have been studied as indicators of soil quality from the decade of the 80's.

Enzyme activities have been associated with indicators of biogeochemical cycles, degradation of organic matter and soil remediation processes, so they can determine, together with other physical or chemical properties, the quality of a soil (85). Authors such as Dick (1996), Nielsen and Winding (2001), and Eldor (2007), report enzymes as good indicators because: a) they are closely related to organic matter, physical characteristics, microbial activity and biomass in the soil, b) provide early information about changes in quality, and are more rapidly assessed. Nevertheless, due to enzyme origin (from bacteria, fungi, plants, and a range of macroinvertebrates), different enzyme locations (intra or extracellular), matrix association (alive or dead cells, clays or / and humic molecules) and assay laboratory conditions, it has been demonstrated that it is of great importance to optimize the procedures for enzymatic activity determination in order to obtain the best values and indices according to intrinsic soil properties. Because enzymes are difficult to extract from soils and regularly lose their integrity (86-88), enzyme activity determination must be made under strict laboratory conditions paying particular attention to temperature control, incubation time, pH buffer, ionic strength of the solution, and substrate concentration (89, 90).

○ β – Glucosidase

It is widely distributed in the environment, and its activity has been detected in soil, fungi and plants. It has been used as a key soil quality indicator due to its importance in catalytic reactions on cellulose degradation, releasing glucose as a source of energy to maintain metabolically active microbial biomass in soil (88, 91). At the same time, it plays an important role in energy availability in the soil which is directly related to labile C content and with the ability to stabilize soil organic matter, showing low seasonal variability (92). On the other hand, it has been reported that enzyme activity could be inhibited by the presence of heavy metals like Cu and Cd (93).

As a free enzyme in soil solution, it normally has a short – lived activity, because they can be rapidly degraded, denatured or irreversibly inhibited. However, a certain proportion of these free enzymes may lose stabilization because of adsorption on soil minerals or incorporation into humic material, which, despite affecting their catalytic potential it may enable enzyme activity to persist in soil (94, 95).

○ Phosphatase

Phosphorus is an essential nutrient for plant growth and crop yields, however a large portion is immobilized because of intrinsic characteristics of soils such as pH that affects the availability of nutrients and the activity of enzymes, altering the equilibrium of the soil solid phase (96). Soil microorganisms play a key role on phosphate solubilization with the release of low molecular weight organic acids (97) and production of extracellular enzymes as phosphatases.

Phosphatases are a group of enzymes that catalyze hydrolysis of esters and anhydrides of phosphoric acid. Its activity depends on extracellular enzymes, which can be free in the soil water phase or stabilized in the humic fraction or clay soil content (97, 98). In soil, phosphomonoesterases have been the most studied enzymes probably because they have activity both under acidic and alkaline conditions, according to its optimal pH, and because they act on low molecular P-compounds including nucleotides, sugar phosphates and polyphosphates (93); thus they can be used as soil quality indicators. Turner and Haygarth (2005), evaluated phosphatase activity in temperate grassland, and found a strong correlation between enzyme activity and soil properties such as pH, total N, organic P and clay content.

○ Dehydrogenase

Dehydrogenase enzyme activity determination is attractive due to the fact they are an integral part of microorganisms and are involved in organic matter oxidation; nevertheless, this activity is not consistently correlated with other properties of biological systems such as O₂ consumption, CO₂ production or microbial biomass (88). However, it has been considered as a soil quality indicator, because it is involved in electron transport systems of oxygen metabolism and requires an intracellular environment (viable cells) to express its activity (99).

Consistently, the activity of this enzyme is not present in extracellular form as hydrolases (β - Glucosidase, urease, phosphatase), which suggests, that it is not an enzyme that can be used to evaluate the processes of soil degradation, since its activity fluctuates as microbial activity does, in response to management practices and/or climatic effects (99).

Nevertheless, dehydrogenase activity is bound to living and active cells, but it depends on the presence of interferences with heavy metals, catalysis of the assay procedure by extracellular phenol oxidase and other alternative electron acceptors as nitrate and humic substances (100, 101). Studies by Speir and Ross (2002) and Kandeler and Dick (2007) suggest that Cu presence could affect enzyme activity when being assessed.

○ Urease

These enzymes are involved on urea hydrolysis into CO₂ and NH₃ and consequently with soil pH increase and N losses by NH₃ volatilization. Due to the role of urea as a fertilizer, focus has been placed on urease in order to evaluate N supply to plants, however, fertilization practices have been reported as being very inefficient due to large N losses to the atmosphere by volatilization mediated by these enzymes (93). On the other hand, new enzymes involved in N-cycle have been subject of study; in this aspect, there is not a much data available about ammonia monooxygenase (AMO) activity in soil (102); even though it is not included as quality indicator, this membrane-bound enzyme could be useful in determining nitrification rates and the effect of nitrification inhibitors, faster than quantifying nitrate as end product. Gutiérrez *et al.* (2009) studied the spatial variability of three hydrolytic enzymes including AMO activity in a rice soil in Chile; they found very low activities because conditions on which soil samples were collected (fallow phase, dry soils) but they show a positively correlation with N availability which indicates that this enzyme can be used to make some inferences about the nitrification process in soil and determine if nitrogen losses are due to volatilization, nitrification or denitrification.

Urease has been widely used to evaluate changes on soil quality related to management, since its activity increases with organic fertilization and decreases with soil tillage (103). This enzyme, mostly the cases is an extracellular enzyme representing up to 63% of total activity in soil. It has been show that its activity depends on microbial community, physical, and chemical properties of soil (104), and its stability is affected by several factors: organo-mineral complexes and humic substances make them resistant to denaturing agents such as heat and proteolytic attack (93).

Urease activity is used as a soil quality indicator because it is influenced by soil factors such as cropping history, organic matter content, soil depth, management practices, heavy metals and environmental factors like temperature and pH (105). The understanding of urease activity should provide better ways to manage urea fertilizer, especially in warm high rainfall areas, flooded soils and irrigated conditions (93).

Final remarks

There are several biological soil properties that can be used as soil quality indicators, alone or in combination with other chemical or physical properties. However they are far from being universal and should be chosen according to the situation under consideration. On the other hand there are several properties difficult to determine and to interpret that many times explain about the same as simpler and less costly measurements; similarly, only properties that are sensitive to management changes should be used; Proper sampling strategies and multivariate analysis of the results are key factors to consider when using biological soil indicators.

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