SOIL HEALTH ASSESSMENT WITH A FOCUS ON BIOLOGICAL AND MICROBIOLOGICAL INDICATORS

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ABSTRACT

Soil health is a fundamental component for the sustainability of agricultural systems, being directly influenced by biological activity and the interactions among physical, chemical, and microbiological components. Among the various methods for evaluating soil quality, biological and microbiological bioindicators have gained prominence due to their sensitivity to changes caused by land use and management. Given the growing demand for tools that promote sustainable land use, this article aims to present a critical review of the main bioindicators used in soil quality assessment, with emphasis on soil fauna, enzymatic activity, microbial biomass carbon, microbial respiration, and the metabolic quotient. In addition, it seeks to highlight the importance of the BioAS technology as an innovative and accessible tool for monitoring soil health, providing technical and scientific support to guide more efficient and environmentally responsible agricultural practices. Soil fauna acts in the decomposition of organic matter, nutrient cycling, and improvement of physical soil properties. Meanwhile, microorganisms and their enzymes directly reflect the functionality and resilience of soil ecosystems. The integrated use of these indicators, combined with the standardization of methodologies such as BioAS, represents a promising path toward sustainable soil management and the conservation of natural resources.

KEYWORDS: Agroecological sustainability; Biogeochemical cycling; Conservationist management; Soil enzymes; Soil microorganisms.

AVALIAÇÃO DA SAÚDE DO SOLO COM FOCO EM INDICADORES BIOLÓGICOS E MICROBIOLÓGICOS

RESUMO

A saúde do solo é um componente fundamental para a sustentabilidade dos sistemas agrícolas, sendo diretamente influenciada pela atividade biológica e pelas interações entre os componentes físicos, químicos e microbiológicos. Dentre os diversos métodos de avaliação da qualidade do solo, os bioindicadores biológicos e microbiológicos têm ganhado destaque devido à sua sensibilidade às alterações causadas pelo uso e manejo do solo. Diante da crescente demanda por ferramentas que promovam o uso sustentável da terra, este artigo tem como objetivo apresentar uma revisão crítica dos principais bioindicadores utilizados na avaliação da qualidade do solo, com ênfase na fauna edáfica, atividade enzimática, carbono da biomassa microbiana, respiração microbiana e quociente metabólico. Além disso, busca destacar a importância da tecnologia BioAS como uma ferramenta inovadora e acessível para o monitoramento da saúde do solo, oferecendo suporte técnico e científico para orientar práticas agrícolas mais eficientes e ambientalmente responsáveis. A fauna do solo atua na decomposição da matéria orgânica, no ciclo de nutrientes e na melhoria das propriedades físicas do solo. Por sua vez, os microrganismos e suas enzimas refletem diretamente a funcionalidade e a resiliência dos ecossistemas do solo. O uso integrado desses indicadores, aliado à padronização de metodologias como o BioAS, representa um caminho promissor para o manejo sustentável do solo e a conservação dos recursos naturais.

PALAVRA-CHAVE: Sustentabilidade agroecológica; Ciclagem biogeoquímica; Manejo conservacionista; Enzimas do solo; Microrganismos do solo.

INTRODUCTION

Soil integrates mineral components, sand, silt, clay fractions, and stable organic matter that result from the activity of soil biota, forming the physico-chemical-biological base that sustains terrestrial ecosystems (ROCHA *et al.*, 2022). This biota ensures processes such as residue decomposition and nutrient cycling that connect the atmosphere, plants, and soil (MENDES *et al.*, 2019). In addition to providing nutrients, biologically active soil stores water, sequesters carbon, and mitigates pollutants, offering important environmental services (MENDES *et al.*, 2021). Therefore, the quality of soil, water, and air should be assessed in an integrated way: while water and air are primarily judged by the degree of pollution, soil involves solid, liquid, and gaseous phases and multiple ecological functions (BÜNEMANN *et al.*, 2018).

Authors such as Doran and Parkin expanded the concept of soil quality to include the health of plants, animals, and humans; Karlen *et al.* (2019) defined it as the capacity to sustain productivity and conserve water and air within ecosystem limits (SIMON *et al.*, 2022). Evaluating such functions requires well-integrated chemical, physical, and especially biological indicators. In recent years, methods that prioritize parameters capable of influencing ecological processes and that are easily applicable by technicians and farmers have advanced (SOLANGI *et al.*, 2024). The biodiversity of soil fauna stands out, as the variety of organisms maintains ecosystem stability and reflects the sustainability of management systems (OLIVEIRA *et al.*, 2024).

Bioindicators are defined as species or communities whose presence, abundance, or condition indicate environmental changes and, therefore, are considered more sensitive than many physicochemical indicators because they detect changes caused by soil use early (SILVA *et al.*, 2021). Among them, microorganisms and their enzymes figure as central tools in the rapid and low-cost diagnosis of soil quality.

Given the growing demand for tools that promote sustainable land use, this article aims to present a critical review of the main bioindicators used in soil quality assessment, with emphasis on soil fauna, enzymatic activity, microbial biomass carbon, microbial respiration, and the metabolic quotient. In addition, it seeks to highlight the importance of the BioAS technology as an innovative and accessible tool for monitoring soil health, providing technical and scientific support that can guide more efficient and environmentally responsible agricultural practices.

MAIN BIOINDICATORS USED IN SOIL QUALITY ASSESSMENT

Soil Fauna

Soil is a fundamental, active, and constantly transforming natural component that contributes significantly to the formation and stability of the atmosphere, assists in carbon sequestration, and enables and sustains the production of food and raw materials, in addition to housing a vast diversity of organisms of different sizes and metabolisms, forming the soil biota (SILVA *et al.*, 2021). This perspective is reinforced by Oberreich *et al.* (2024), who define soil as a living system: more than just a shelter for countless invertebrates, it is an environment in which this biological contingent performs crucial functions for ecosystem maintenance. Among these functions are nutrient recycling, organic matter decomposition, activation of microbiota, and improvement of physical characteristics such as aggregation, porosity, aeration, infiltration, and water retention (CHAMORRO-MARTÍNEZ *et al.*, 2022).

Soil fauna displays a wide range of forms, sizes, and ecological functions (Table 1). Regarding invertebrates, virtually all classes or orders are represented in the soil. Microfauna, with body size between 4 µm and 100 µm, includes protozoa, rotifers, copepods, tardigrades, nematodes, among others, which act indirectly in nutrient cycling and in the population regulation of fungi and bacteria (CHAMORRO-MARTÍNEZ *et al.*, 2022).

Mesofauna, ranging from 100 µm to 2 mm, includes mites, springtails, myriapods, arachnids, various insect orders, as well as some oligochaetes and crustaceans. Their trophic activities include the consumption of microorganisms and microfauna, contributing to the control of these populations and promoting the fragmentation of plant residues; although they depend significantly on soil moisture, they are typically terrestrial organisms (SILVA *et al.*, 2021).

Macrofauna representatives have body diameters between 2 mm and 20 mm, such as earthworms, beetle larvae and adults, centipedes, termites, ants, isopods, and arachnids (CHAMORRO-MARTÍNEZ *et al.,* 2022). This group positively influences primary productivity by occupying multiple trophic levels. Their action affects the transformation of organic matter and the supply of nutrients to plants, also influencing microbial communities involved in the processes of mineralization and humus formation (CHAMORRO-MARTÍNEZ *et al.,* 2022). Furthermore, they stand out for fragmenting organic residues and altering physical structure through burrowing and coprolite production. Macrofauna can also transport beneficial microorganisms, such as mycorrhizal fungi and nitrogen-fixers and select pathogens during digestion.

Invertebrates over 20 mm in length are classified as megafauna (CHAMORRO-MARTÍNEZ *et al.*, 2022).

The maintenance of soil quality is essential for the balance of biotic communities, the sustainability of production systems, and food security (SILVA *et al.,* 2021). However, inadequate management practices simplify ecosystems and reduce biodiversity, altering the composition of soil communities due to changes in habitat, resource availability, microclimate, and competition intensity.

TABLE 1. Main contributions of soil fauna to decomposition and soil structure.

Category	Body size	Nutrient recycling	Structural modification
Microfauna	4 µm – 100 µm	Regulates populations of bacteria and fungi; alters nutrient flows	Interacts with microflora, potentially affecting aggregation
Mesofauna	100 µm – 2 mm	Controls fungi and microfauna; fragments plant debris	Produces fecal pellets; creates biopores; promotes humification
Macrofauna	2 mm – 20 mm	Stimulates microbial activity; regulates fungi and mesofauna	Mixes organic and mineral materials; redistributes organic matter; forms aggregates; builds galleries

Source: Adapted from Chamorro-Martínez *et al.* (2022).

In order to characterize the distribution of soil fauna under different land uses in the Pampa Biome, Góes *et al.* (2021) analyzed three situations: Native Forest, Grazed Natural Grassland, and Ryegrass/Soybean Cropping. The Shannon index indicated greater diversity in the native forest and lower values in the monoculture system, a phenomenon attributed to the predominance of Collembola. Structural simplification and low input of plant residues in monoculture make the environment less favorable to diversity, even if it favors specific food sources for certain groups.

These findings reinforce that soil preparation and cultivation influence the abundance and diversity of soil biota. Oliveira Filho *et al.* (2016) showed that intense disturbance in conventional tillage reduces faunal activity, while no-tillage, by ensuring greater plant cover, promotes diversity. In line with this, Alves *et al.* (2022) found that systems with frequent addition of organic residues, such as poultry litter, increase organism abundance; however, high densities of springtails may reduce species evenness. The dominance of Collembola, however, is not negative, as several studies identify this group as a bioindicator of anthropic disturbances and soil quality (OLIVEIRA FILHO *et al.,* 2016).

In addition to services such as nutrient cycling, carbon sequestration, and pollutant retention, soil fauna directly influences plant productivity. Its diversity supports the decomposition and mineralization of residues, increasing nutrient availability for plants and other organisms. Soil biota shows sensitivity to biological, physical, and chemical variations induced by management and can be used as an indicator of quality (AQUINO; SAUTTER, 2022). Therefore, it is recommended to adopt conservationist practices that promote the preservation of soil fauna and ensure proper ecosystem functioning.

Soil Enzymatic Activity

In the soil, numerous metabolic reactions occur that promote the recycling and reuse of nutrients by plants and other organisms. Among the components of the soil are enzymes, protein macromolecules specialized in accelerating thermodynamically feasible chemical transformations (BALOTA *et al.,* 2004; ROCHA *et al.*, 2022). These proteins act as mediators of the catabolism of organic and mineral constituents of the soil, playing a decisive role in biogeochemical cycles: they convert organic matter, enable residue decomposition, accelerate the flow of elements, and contribute to the sustainability of ecosystems (ROCHA *et al.*, 2022).

The reactions catalyzed by enzymes present in the soil promote the recycling of carbon, phosphorus, nitrogen, and sulfur, essential to plant vigor, by transforming nutrients into assimilable forms for microorganisms and plants (MOREIRA; SIQUEIRA, 2006). Certain enzymatic activities are strongly linked to microbial biomass, reflecting the metabolic vitality of the soil microbiota (SOLANGI *et al.*, 2024).

Although most soil enzymes are synthesized by microorganisms, plants also exude them in the rhizosphere, in addition to originating from the degradation of plant, animal, and microbial biomass (ROCHA *et al., 2022*). However, for soil quality diagnoses, enzymes derived from microorganisms stand out (SOLANGI *et al.,* 2024). These proteins are classified as intracellular (endoenzymes) and extracellular (exoenzymes): the former remain inside the cell and regulate internal metabolic pathways, while the latter are secreted into the environment (DAMASO; COURI, 2018).

In the soil, enzymes are distributed among different compartments: they can be contained in living cells, released through exudation, or associated with dead cells, intact or after lysis (SOBUCKI *et al.*, 2021). Microbial death releases enzymes into the environment; many lose functionality, but some remain active adsorbed onto clay particles or organic matter (MENDES *et al.,* 2021).

Endoenzymes metabolize low-molecular-weight substrates, such as urea, inside the cell. Exoenzymes, in turn, degrade complex polymers—proteins, lipids, and polysaccharides—outside the cell; the resulting products, of lower molecular weight, are then absorbed (MOREIRA; SIQUEIRA, 2006; ROCHA *et al.*, 2022). Functionally, they are grouped into (i) carbon acquisition enzymes, (ii) nitrogen, (iii) phosphorus, and (iv) oxidative enzymes, the latter capable of breaking down recalcitrant molecules such as lignin (ROCHA *et al.,* 2022).

There are records of at least 500 enzymes involved in nutrient recycling (BALOTA *et al.*, 2004) however, β-glucosidase, arylsulfatase, alkaline phosphatase, and urease are widely adopted as markers of C, S, P, and N dynamics, as well as indicators of microbial activity. Some examples are cited in Table 2 (BALOTA *et al.*, 2004; SOBUCKI *et al.* 2021; ROCHA *et al.* 2022; OLIVEIRA *et al.,* 2024).

TABLE 2. Typical activity range of soil enzymes in the 0–10 cm layer.

Enzyme (indicator)	**Nutrient/Cycle**	**Reported range (µg PNP g^{-1} soil h^{-1})**	**Land use / management**
β-Glucosidase	Carbon	52–137	Flooded rice, conventional system
Arylsulfatase	Sulfur	12–223	From apple orchard to oak forest
Alkaline phosphatase	Phosphorus	98–213	Pasture vs. conserved area
Urease	Nitrogen	0.15–0.50 (µg NH_4 -N g^{-1} soil h^{-1}) -N	Potato crops and native field

Source: Adapted from Balota *et al.* (2004); Sobucki *et al.* (2021); Rocha *et al.* (2022); Oliveira *et al.* (2024).

The intensity of enzymatic activities usually decreases with depth (SOLANGI *et al.,* 2024; SOBUCKI *et al.,* 2021). To assess management impacts, surface sampling is recommended (0–5 cm or 0–10 cm); correlations with fertility use 0–20 cm (BALOTA *et al.,* 2004; ROCHA *et al.,* 2022). Air-drying, a common practice when there are many samples or prolonged transportation, can modify protein conformation and microbial viability; such effects should be considered (SOBUCKI *et al.,* 2021; ROCHA *et al.,* 2022).

Determinations of β-glucosidase and acid phosphatase are based on the release of p-nitrophenol, measured colorimetrically, while urease is quantified by the produced ammonia and titrated (BALOTA *et al.*, 2004; ROCHA *et al.*, 2022). Precise assays require optimization of temperature, pH, moisture content, substrate concentration, and incubation time (DICK, 1997).

In sugarcane, organic systems without tillage increased β-glucosidase, while burning reduced total activity compared to the cerrado (SOLANGI *et al.,* 2024). In a Red-Yellow Latosol, vine cultivation associated with live cover increased β-glucosidase and arylsulfatase, despite a 70% drop in microbial biomass compared to dense cerrado forest (ROCHA *et al.,* 2022). In the Cerrado of Minas Gerais, agroecological practices resulted in higher basal respiration and microbial colonies, reflecting better soil quality (ROCHA *et al.,* 2022).

Thus, soil enzymes constitute powerful indicators of soil condition: their activity reflects microbial diversity and, consequently, the health of the ecosystem (ROCHA *et al.,* 2022; SOLANGI *et al.,* 2024). Environmental variables—temperature, pH, moisture—modulate such activities; continuous monitoring and sustainable management are essential to preserve enzymatic functionality and the integrity of terrestrial ecosystems (SOBUCKI *et al.,* 2021; ROCHA *et al.*, 2022).

Microbial Biomass Carbon (MBC)

The most dynamic biologically active portion of organic matter present in the soil is formed by microorganisms, which correspond to approximately 60–80% of this total. Soil fauna and plant root systems complete this living part, comprising, respectively, between 15–30% and 5–10% of the total (ARAÚJO; MELO, 2010).

The conservation of productivity in agroecosystems and forests depends, to a large extent, on the continuous conversion of organic matter. Microorganisms play a fundamental role in residue degradation, in nutrient cycling and temporary retention, and in other chemical modifications of the soil matrix (BALOTA *et al.*, 1998). During such processes, microbial biomass decomposes and mineralizes plant residues, using them as energy and nutritional sources. Along this process, significant amounts of C, N, P, K, Ca, Mg, S, and micronutrients are temporarily immobilized and, after cell death, released, becoming once again accessible to plants (REIS JUNIOR; MENDES, 2007). Thus, the microbial pool acts as a key regulator of SOM transformations, functioning at times as a reservoir and at others as a drain of energy and elements (BALOTA *et al.*, 1998).

As it is readily decomposed, the microbial fraction represents a labile component of organic matter. Its size and activity are intensely modulated by seasonal variations in moisture, temperature, crop and residue management. Consequently, microbial biomass constitutes an early indicator of changes in organic matter content, even before global changes in physicochemical properties become detectable (SOUZA *et al.*, 2025).

Quantitatively, microbial biomass holds, on average, 2 to 5% of soil organic carbon and 1 to 5% of total soil nitrogen (BALOTA *et al.*, 1998). Routine determinations of this compartment are employed in research on C and N fluxes, nutrient cycling, and plant productivity in different terrestrial ecosystems. The values obtained allow the correlation of the amount of immobilized nutrients and the intensity of microbial activity with the productive potential and fertility of the land. As it is part of the active fraction of organic matter, biomass proves to be more sensitive than total C and N contents in identifying changes caused by management systems (SOUZA *et al.*, 2025).

Microbial biomass carbon is the first form of C to be metabolized after changes in land use or management, responding quickly and reliably to disturbances (Table 3). Thus, MBC constitutes an early indicator of changes in SOM and, consequently, in soil quality. High MBC values indicate greater potential capacity for microbial processes and represent a readily available carbon reserve (PEREZ *et al.*, 2004).

To estimate the size of this dynamic reservoir, the chloroform-fumigation-incubation (CFI) and chloroform-fumigation-extraction (CFE) methods stand out. Both continue to be widely used, although they have specific advantages and limitations, and have been periodically refined in recent investigations (GUIMARÃES *et al.*, 2017).

TABLE 3. Microbial biomass carbon (MBC) under different soil uses

Land use / Management	MBC (mg C kg $^{-1}$ soil)	Location / Condition
No-tillage (≥10 years)	517	Clayey soil, Brazil
Conventional tillage	419	Same site (control)
Native forest (0–10 cm)	471	Meta-analysis, 468 observations (China)
Dryland cropland (0–10 cm)	180	Meta-analysis, China
Eucalyptus (Oxisol)	224	Southern Brazil
Regenerating secondary forest	79	Southern Brazil

Source: Adapted from Perez *et al.* (2004); Araújo; Melo (2010); Guimarães *et al.* (2017); Goés *et al.*, (2021); Souza *et al.* (2025).

Regardless of the adopted technique, it is widely recognized that organic carbon and microbial biomass activity are privileged quality metrics in agroecosystems, as they promptly reflect the changes imposed by different managements. Araújo *et al.* (2019) verified these attributes in soils cultivated with corn intercropped with various legumes and compared them with native forest areas at two depths (0–10 cm and 10–20 cm). In the surface layers, intercropping with sunn hemp and dwarf pigeon pea led to MBC increases of over 40% compared to monocropped corn, reaching levels similar to the natural environment, highlighting the benefits of introducing legumes into the system.

In Dourados (MS), Pereira *et al.* (2008) investigated the impact of green manures on microbial biomass and its activity in a very clayey dystroferric Red Latosol. Cover crops of jack bean, pigeon pea, forage sorghum, and an agroforestry system were compared to bare soil and native forest. The natural ecosystem exhibited the highest values of MBC, basal respiration, and organic matter; however, all cover crops stimulated microbial biomass in comparison to the exposed soil, reinforcing the positive influence of conservation practices (ARAÚJO; MELO, 2010).

The increase in SOM is extremely advantageous. Examining its dynamics, especially through MBC quantification, provides support for understanding the storage of nutrients and carbon in the soil. These biogeochemical cycles are closely related to the health of the soil ecosystem, favoring plant growth and greater input of residues into the soil (SOUZA *et al.*, 2025).

In summary, MBC is configured as a sensitive and reliable bioindicator of soil quality. Its determination allows the direct evaluation of the size and vigor of the microbial community, enabling efficient monitoring of soil performance under different use practices. In addition to regulating carbon cycling, this fraction influences soil structure, fertility, and pathogen resistance. Therefore, continuing to investigate and apply MBC is essential for sustainable land management and environmental protection.

Microbial Respiration

The most changeable vital fraction of soil organic matter is composed of microorganisms, which account for around 60–80% of this contingent. Soil fauna and plant roots complete this living portion, totaling, respectively, between 15–30% and 5–10% (ARAÚJO; MELO, 2010).

The permanence of agroecosystem and forest productivity largely depends on the uninterrupted conversion of organic matter. Microorganisms play a central role in residue degradation, in the recycling and temporary immobilization of nutrients, in addition to other chemical soil transformations (BALOTA *et al.*, 1998). During these processes, microbial biomass decomposes and mineralizes plant residues, using them as energy and nutrient sources. Thus, significant amounts of C, N, P, K, Ca, Mg, S, and micronutrients are temporarily retained and, after cell death, made available to plants again (REIS JUNIOR; MENDES, 2007). In this way, the microbial pool acts as an essential regulator of SOM changes, functioning either as a reservoir or as a sink for elements (BALOTA *et al.*, 1998).

Because it is readily decomposed, the microbial fraction represents a labile component of organic matter. Its volume and activity are strongly modulated by seasonal fluctuations in moisture, temperature, cultivation practices, and residue management. Consequently, microbial biomass constitutes an early marker of variations in SOM content, even before global changes in physical-chemical properties become detectable (SOUZA *et al.*, 2025).

In absolute terms, microbial biomass contains, on average, 2 to 5% of soil organic carbon and 1 to 5% of total soil nitrogen (BALOTA *et al.*, 1998). Routine assessments of this fraction are used in studies of C and N flow, nutrient cycling, and plant productivity in various land formations. The data obtained allow associating the amount of retained nutrients and the intensity of microbial activity with the productive potential and fertility of the land. As it is part of the active fraction of organic matter, biomass is more sensitive than total C and N contents in detecting impacts caused by management practices (SOUZA *et al.*, 2025).

Microbial Biomass Carbon (MBC) is the first form of C to be metabolized after land use or management changes, reacting quickly and reliably to disturbances. Thus, MBC is an early indicator of changes in SOM and, therefore, in soil quality. High values indicate greater potential for microbial processes and represent a readily accessible carbon reserve (PEREZ *et al.*, 2004).

To estimate the size of this dynamic reservoir, the chloroform-fumigation-incubation (CFI) and chloroform-fumigation-extraction (CFE) methods stand out. Both remain widely used, although they have specific merits and limitations, and are continuously improved in current research (GUIMARÃES *et al.*, 2017). Regardless of the technique, it is recognized that organic carbon and microbial activity are privileged metrics of soil quality, as they rapidly reflect changes imposed by management.

Metabolic Quotient

The MBC expresses the fraction of carbon that the soil microbial biomass fixes in its cellular structures. Microbial respiration, a reflection of the soil microbiological dynamics, can be assessed by measuring the CO_2 released as a consequence of the activity of microorganisms operating under aerobic and anaerobic conditions (EMBRAPA, 2020).

In order to offer a more elucidative reading and establish dynamic links between biomass and activity, Anderson and Domsch (1993) apud Ashraf *et al.*, (2022) conceived the metabolic quotient (qCO_2) as a unique metric of metabolic activity. This indicator combines microbial biomass values with respiration rates, representing the CO_2 flux per unit of microbial biomass (ASHRAF *et al.*, 2022). The qCO_2 corresponds to CO_2 emission per biomass unit over a given interval ($C\text{-}CO_2$ $C\text{-}mic^{-1}$ h^{-1}) and constitutes a sensitive parameter of microbial biomass efficiency in utilizing available carbon, being useful to infer biological vitality and the sanitary state of the soil (ANDERSON; DOMSCH *et al.*, 1993).

The adoption of qCO_2 as an indicator of changes in soil quality is based on the theory of microbial community respiration described by Odum (1971) apud Ashraf *et al.*, (2022), according to which:

> “[…] the increase in community respiration may be the first sign of stress, as the repair of damage caused by soil disturbances diverts energy from growth and reproduction to cell maintenance. Thus, during stress, more energy is allocated to maintenance, so that part of the biomass carbon is released as CO_2.” (ASHRAF *et al.*, 2022, p. 1796).

Several investigations show that this quotient helps assess soil quality, as it reflects the biomass stress level (ANDERSON; DOMSCH, 1993). Biomass considered “efficient” exhibits a lower respiration rate than biomass considered “inefficient.”

Thus, stressing factors — such as contamination by heavy metals, nutrient scarcity, and low pH — as well as sudden disturbances (intensive cultivation or burning) can reduce microbial biomass efficiency (FERREIRA *et al.*, 1999). In short, low qCO_2 values suggest economical energy use and a more stable environment, whereas high values indicate stressed or disturbed ecosystems. Soils with high qCO_2 tend to be dominated by fast-growing colonizer organisms, characterizing systems with high energy demand and far from equilibrium (DORNELLES, 2017; SILVA *et al.*, 2021). However, values need to be compared within the same soil type, as additional variables also affect microbial biomass (ASHRAF *et al.*, 2022).

Vieira *et al.* (2016) analyzed the immediate impact of fire on physical, chemical, and microbiological properties in pasture soils in southern Minas Gerais. The authors found that the use of fire as a cleaning method increased the qCO_2 of the burned soil by almost seven times compared to the unburned soil, showing a strong negative impact of this management on microbial biomass and confirming qCO_2 as an excellent bioindicator of fire effects.

Dadalto, (2015), evaluating microbial activity under different tillage systems in Rolim de Moura (RO), found lower qCO_2 in native forest soil and higher in conventional tillage. No-till showed greater efficiency in carbon incorporation, reflected in lower qCO_2 and, therefore, lower CO_2 emission. The higher qCO_2 value observed in conventional tillage indicates that microorganisms face environmental stress. The lower ratio observed under native forest shows that less CO_2 is released by respiration and more carbon is incorporated into microbial tissues, reducing qCO_2. These microbiological attributes prove to be consistent indicators of changes in soil quality due to management and sampling depth.

Measuring qCO_2 constitutes a valuable tool for estimating soil health and its capacity to support vegetation (Table 4). Soils with high biological activity tend to be more fertile and productive, as microorganisms decompose organic matter and make essential nutrients available to plants. Furthermore, monitoring qCO_2 makes it possible to evaluate the impact of agricultural practices — such as intensive applications of fertilizers and pesticides — that may suppress biological activity, compromising soil quality and crop productivity. Continuous monitoring of qCO_2, along with other bioindicators, helps in the early detection of problems and in the adoption of measures to preserve soil health, promoting more sustainable and healthier agriculture for all.

TABLE 4. Metabolic quotient (qCO_2) in different soil ecosystems.

Ecosystem / Condition	**qCO_2 (µg CO_2-C mg^{-1} Cmic h^{-1})**	**Interpretation***
Chernozem – residential area	0.50	High efficiency, stable soil
Organic vineyard (summer)	2.31 – 2.49	Moderate efficiency, sustainable management
Chernozem – industrial zone	6.83	High stress, low efficiency

*Values < 1 suggest balanced ecosystems; 1–3 reflect moderate disturbance; > 3 indicate strong anthropic pressure or adverse conditions.

Source: Adapted from Aanderson; Domsch (1993); Dadalto, (2015); Vieira *et al.* (2016); Ashraf *et al.* (2022)

Bioas Technology

Considering the relevance of the biological integrity of the soil for excellence and productivity, numerous studies have been carried out with the aim of creating procedures that standardize and simplify the biological evaluation of soil. These procedures seek to gather relevant data so that agronomists and farmers can make informed decisions about production models. Among the methods resulting from these efforts, bioanalysis, developed by Embrapa, stands out as it provides a comprehensive overview of the soil's biological components (MENDES *et al.*, 2021).

The Embrapa Soil Bioanalysis Technology (BioAS) was made official on July 23, 2020, as a result of more than two decades of research. It constitutes a pioneering initiative on the international scene by integrating the biological component into routine soil analyses, which were traditionally limited to chemical and physical aspects (MENDES *et al.*, 2021). BioAS is based on the quantification of the activity of the enzymes arylsulfatase and β-glucosidase. Since these are directly associated with productive potential and the sustainability of land use, these enzymes act as bioindicators of soil quality. Through studies conducted by Embrapa, reference values for these enzymes have been established for different soil classes. High levels of enzymatic activity suggest that the cropping systems and/or management practices adopted in the sampled soil are appropriate and sustainable; conversely, low levels indicate that the employed management does not support productivity or soil resource conservation (MENDES *et al.*, 2021).

Arylsulfatases convert organic sulfur compounds into inorganic species readily assimilated by plants, directly influencing their development. These enzymes are highly sensitive to changes in fertility and can therefore be used as markers of soil quality (ROCHA *et al.*, 2022).

The activity of β-glucosidase is fundamental in the cellulose degradation process, as it hydrolyzes cellobiose residues, releasing glucose — an important energy substrate for the microbiota. The quantity and quality of plant residues, as well as soil pH, strongly influence this enzyme (MENDES *et al.*, 2021).

The quantity and quality of plant residues incorporated into the soil, along with the input of nutrients and other management inputs, leave their "fingerprint," that is, a biological signature, in the soil environment. As emphasized by Mendes *et al.* (2021), "the soil's ability to store in its memory the type of management it has undergone is closely linked to its living fraction, to its biological component." Thus, measurements of enzymatic activity constitute one of the ways to access this memory, since the obtained value reflects the overall activity of enzymes from living organisms (microorganisms, plants, and animals) and from past generations that remained in the soil as an abiotic component.

As enzymes are organic molecules, the soil's ability to store and stabilize organic matter (OM) — as well as related structural attributes such as aggregation and porosity — is closely linked to its capacity to preserve and safeguard enzymes (DAUNORAS *et al.*, 2024). However, changes in OM or in structural attributes often take years to be detected, whereas variations in enzymatic activity can be perceived over much shorter intervals (MENDES *et al.*, 2021). Therefore, an increase in biological activity, especially enzymatic activity, can indicate that the management system favors the accumulation of soil organic matter (SOM) over time, although in the initial phases, such an increase in activity does not always correspond to actual increases in SOM levels (MENDES *et al.*, 2021).

BioAS presents a significant differential, as enzymes are more sensitive than chemical and physical parameters, anticipating changes in soil health due to land use and supporting management decisions (MENDES *et al.*, 2021).

Arylsulfatase and β-glucosidase, analyzed together or separately, demonstrate high sensitivity in identifying management-related variations (MENDES *et al.*, 2019). These enzymes are closely linked to SOM — a key parameter of soil quality — as well as to grain yield. Additionally, they are directly involved in SOM cycling and are not significantly influenced by the application of fertilizers or soil amendments such as lime (MENDES *et al.*, 2021). Many microbiological attributes, such as microbial biomass carbon, basal respiration, and activities of acid phosphatase, cellulase, and dehydrogenase, correlate with arylsulfatase and β-glucosidase; for this reason, these two markers were chosen to represent the soil's biological "machinery" in bioanalysis (EMBRAPA, 2020).

The BioAS method broadly assesses soil biological health and, when combined with traditional analyses, provides robust information. The protocol is performed after the second-crop harvest, collecting samples from the 0–10 cm layer in both rows and inter-rows of the last crop. The laboratory sends the results to an Embrapa online platform, which then issues the final report containing interpreted enzymatic activity values and soil quality indices; these numbers are compared with specific reference values established for each soil class (EMBRAPA, 2020).

From the perspective of soil quality assessment, both chemical and biological indicators have been employed, with enzymatic activity quantification being especially notable. In this context, Batista *et al.* (2023) compared the activity of arylsulfatase and β-glucosidase in different cropping systems; activity was higher under no-tillage than under conventional tillage and in natural fields, a result attributed to the greater capacity of conservationist systems to increase organic matter in the surface layers of the soil.

Meanwhile, Cavalcante *et al.* (2020) monitored Caatinga soils over three years (2014–2016) in preserved areas and sites under anthropogenic intervention. The activities of β-glucosidase, urease, and arylsulfatase declined under human influence, and the strongest association between accumulated rainfall and enzymatic activity occurred in the preserved area in both seasons of the year, highlighting the vulnerability of these soils to anthropogenic intervention.

Bioanalysis represents an innovative technology with great potential to standardize soil biological evaluation methodologies. The tool provides minimum, average, and maximum values for several parameters, assisting technicians and producers in selecting management strategies. Furthermore, the high sensitivity of the selected enzymes serves as an additional refinement, ensuring more accurate results.

FINAL CONSIDERATIONS

The discussions presented throughout this article highlight the central role of bioindicators in assessing soil health, especially those based on microbial activity. Soil microorganisms, due to their sensitivity and rapid response to environmental changes, stand out as effective tools for monitoring the effects of land use and management. Their involvement in essential processes such as organic matter decomposition, nutrient cycling, and biological nitrogen fixation underscores their importance in maintaining fertility and the sustainability of agroecosystems.

Among the main indicators analyzed, enzymatic activity stands out as a sensitive parameter that allows inferences about the functional capacity of the soil.

Techniques such as BioAS, which utilize specific enzymes, have proven promising due to their practicality, sensitivity, and ability to detect biological changes that would not be identified by conventional chemical methods. Standardizing and expanding the use of these methodologies can strengthen biological monitoring and contribute to more sustainable agricultural practices.

Indicators such as microbial biomass carbon (MBC), microbial respiration, and the metabolic quotient (qCO_2) provide complementary and valuable diagnostics about the soil's functional state. They offer insights into the efficiency of microbial activity, nutrient availability, and the level of stress the system is under. The integrated use of these indicators enhances the capacity to interpret soil quality and the effects of management practices, contributing to the adoption of more resilient and adaptive strategies.

Finally, the combination of different bioindicators, along with the development of more accessible and low-cost methodologies, represents a promising path to broaden the use of these tools by farmers, especially those in family-based agriculture. Continued research in this area is essential to consolidate biological indicators as an integral part of soil management, promoting not only increased productivity but also the conservation of natural resources and the sustainability of agricultural systems.

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