MICROBIAL METABOLIC ACTIVITY AS LEGACY OF AGRICULTURAL MANAGEMENT IN MAIZE AGROECOSYSTEMS FROM MEXICO HIGHLANDS †

[ACTIVIDAD METABÓLICA MICROBIANA COMO LEGADO DEL MANEJO AGRÍCOLA EN AGROECOSISTEMAS DE MAÍZ DE VALLES ALTOS MÉXICO]

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SUMMARY

Background. Soil ecological functions such as C mineralization, enzyme activity, and microbial biomass determine the maintenance of soil fertility in the short and long term. Microbial activity is a sensitive indicator of changes in soils under agricultural management. **Objective**. Evaluate the metabolic response of soil microbial communities in two temperate maize agroecosystems with different management intensities. **Methodology**. This study evaluated total soil nutrient concentrations, C mineralization, and microbial metabolic activity by comparing two agricultural regimes. The first one is an intensive regime (IR) characterized by the exclusive use of synthetic fertilizers in a maize monoculture. The second one is a traditional regime (TR) characterized by the use of mixtures of organic matter (maize and bean residues and manure) with synthetic fertilizers in a rotation system of maize and beans. Physical, chemical, and biological properties were tested in the laboratory, and the specific enzyme activity (SEA) and metabolic quotient (qCO_2) were calculated. **Results.** Total soil C concentration was 19% higher in TR (26.6 mg g^{-1}) than in IR (5.1 mg g^{-1}); total C biomass was 30% higher in TR (279 mg C g^{-1}) versus IR (83.9 mg C g^{-1}), and potential C mineralization was 40% higher in TR (356 µg C g^{-1} d^{-1}) than IR (214 µg C g^{-1} d^{-1}); in contrast, SEA and qCO2 were lower in TR versus IR. These results support the hypothesis that the microbial community is more efficient under TR than IR because it produces extracellular and intracellular enzymes while growing in biomass. **Implications**. The present study provides new information about the effect of agricultural management on microbial activity, which is important for farmers not only in Mexico Highlands but also in any agricultural scenario exposed to changes in management practices. **Conclusions**. Assessment of biological soil properties is a sensitive indicator of changes in soil properties induced by management. Metabolic indices are suitable for the evaluation of ecological functions in cultivated soils.

Key words: carbon cycle; enzyme activity; maize; metabolic quotient; soil microbial activity.

RESUMEN

Antecedentes. Las funciones ecológicas del suelo como la mineralización de C, la actividad enzimática y la biomasa microbiana determinan el mantenimiento de la fertilidad del suelo a corto y largo plazo. La actividad microbiana es un indicador sensible de cambios en los suelos bajo manejo agrícola. **Objetivo.** Evaluar la

† *Submitted August 8, 2021 – Accepted May 5, 2022.* https://doi.org/10.56369/tsaes.3911

ISSN: 1870-0462.

respuesta metabólica de la comunidad microbiana en dos agroecosistemas de maíz con diferente intensidad de manejo. **Metodología**. Este estudio evalúa la concentración total de nutrientes del suelo, la mineralización potencial de C y la actividad metabólica mediante la comparación de dos regímenes de manejo agrícola. El primero, es un régimen intensivo (RI) caracterizado por el uso exclusivo de fertilizantes sintéticos en un monocultivo de maíz. El segundo, es un régimen tradicional (RT) caracterizado por utilizar mezcla de materia orgánica (residuos de maíz, frijol y estiércol) con fertilizantes sintéticos como fertilizantes, en un sistema de rotación de maíz y frijol. Las propiedades físicas, químicas y biológicas se evaluaron en laboratorio y se calcularon la actividad enzimática específica (AEE) y el cociente metabólico (qCO2). **Resultados**. La concentración total de C en el suelo fue 81% mayor en el RT (26.6 mg g^{-1}) que en RI (5.1 mg g^{-1}); la biomasa de C total fue un 70% más alta en el régimen tradicional (279 mg C g^{-1}) que en el RI (83,9 mg g^{-1}) y la mineralización potencial de C fue del 40% mayor en RT (356 µg C g^{-1} d^{-1} que en RI (214), pero SEA y qCO_2 fueron menores en RT. Estos resultados apoyan la hipótesis de que la comunidad microbiana es más eficiente en RT que RI porque produce enzimas extracelulares e intracelulares y al mismo tiempo crece en biomasa. **Implicaciones**. Este estudio presenta información nueva del efecto del manejo agrícola sobre la actividad microbiana del suelo en suelos de origen volcánico del centro de México. **Conclusiones**. La evaluación de las propiedades biológicas del suelo es un indicador sensible de los cambios en las propiedades edáficas inducidos por el manejo agrícola. Los índices de metabolismo microbiano son precisos para evaluar las funciones ecológicas de suelos cultivados.
Palabras clave: actividad enzimática, actividad microbiana del suelo, agroecosistemas; ciclo de carbono; coeficiente metabólico.

INTRODUCTION

The shift from traditional to modern agriculture derived from the introduction of intensive technologies such as inorganic fertilization, monocultures, mechanized tillage, hybrid seeds, and pesticides that compromise food production and soil productivity (Curtaz *et al*., 2014; García-Oliva, 2005; Etchevers *et al*., 2015). Since the green revolution, agricultural policies at the global level have supported the implementation of these technologies without considering the loss of soil ecological functions (Khaliq *et al*., 2014). Additionally, the pace of food production has exceeded the capacity of the soil to self-regulate its fertility, leading to crop yields that depend on the use of fertilizers (Khaliq *et al*., 2014; Curtaz *et al*., 2014; Tarrasón *et al*., 2016; Etchevers *et al*., 2015). This issue has become one of the major environmental issues facing mankind. As a result of inadequate agricultural management practices, FAO (2015) estimated that nearly 12 million hectares of fertile soil had been heavily degraded.

Long-term soil degradation reduces agricultural productivity because agricultural soils lack a natural organic matter turnover to maintain fertility (Squire *et al*., 2015). For these reasons, the fertility of agricultural soils depends on agricultural practices (Squire *et al*., 2015). The above is remarkable in volcanic soils because their physical and chemical properties confer high fertility, holding the capacity to support crop growth and productive agroecosystems (Ugolini and Dahlgren 2002; Takahashi and Dahlgren 2016). This type of soil has andic characteristics, reflecting a high phosphate retention capacity. However, these soils are fragile and easily degradable through changes in land use and intensified management (Perret and Dorel 1999; Muñoz *et al*. 2011; Beck- Broichsitter *et al*. 2016).

A sustainable agricultural system maintains ecological functions (Etchevers *et al*., 2015). Both microbial biomass and enzymatic activity are sensitive indicators that immediately record changes in soil properties induced by management (Acosta-Martínez *et al*., 2019). Enzyme activity contributes to the decomposition of soil organic matter and nutrient dynamics. Therefore, it can be a suitable indicator of the intensity of nutrient transformations in different environments (Ciarkowska *et al*., 2014). The metabolic quotient (qCO2) is an index of the efficiency of microorganisms in the use of C. qCO2 allows interpreting changes in microbial metabolic efficiency to use C for growth (incorporation into biomass) or nutrient acquisition (C mineralization). In addition, qCO2 has the potential to reflect the effects of tillage, land-use changes, and cultivation on soil fertility (Raiesi and Beheshti 2014).

In Mexico, agriculture covers 16% of the land (27 million hectares), allocated mainly to maize as the dominant crop. Recently, in addition to small-scale maize farming for subsistence purposes, highly intensified large-scale methods have been adopted by farmers for commercial purposes (Gómez-Tovar *et al*., 2005). However, intensive land use is a consequence of the misinterpretation of agricultural issues in the context of globalization. Technological development has increased the number of chemicals used in crops without reference to any specific condition (Hernández-

Xolocotzi, 1988). This condition happened in Mexican agricultural systems starting in the 1940s, when the foundations of the so-called Green Revolution were laid through the implementation of public policy programs promoted by the Secretariat of Agriculture and the Rockefeller Foundation without considering the diversity of agricultural regimes in the country (Appendinni, 2001; Hernández-Xolocotzi, 1990). Based on this policy, agriculture development progressed through three operational axes: (i) the massive introduction of technology-dependent foreign investment; (ii) the establishment of a credit network; and (iii) subsidies for the fertilizer industry (Hernández-Xolocotzi, 1990). Such practices caused profound impacts on the soil at the national level; 45% of the territory has soil features reflecting physical, chemical, or biological degradation (INEGI, 2015). In formal national assessments, there has been little recognition of soil degradation in terms of the metabolic activity of microorganisms. This focus is essential for sustainable agriculture. The aim of this study was to evaluate the metabolic response of soil microbial communities in two temperate maize agroecosystems with different management intensities as examples of diverse production methods in volcanic soils from Mexico Highlands.

MATERIALS AND METHODS

Study Area

This study was conducted in the Mexican Highlands (50 km east of Mexico City), a mountainous volcanic area with natural temperate forest ecosystems (19°14′10″ N; 98°39′48″ W) (Figure 1). The climate is temperate-humid (Bw), with a mean annual temperature of 14 ºC and mean annual precipitation of 930 mm concentrated between May and September. In this area, the stratovolcanoes Iztaccíhuatl (5,220 m a.s.l.) and Popocatépetl (5,450 m a.s.l.) dominate the landscape (Bobbink and Heil 2003). The prevailing soil type is a silandic Andosol developed over pyroclastic fall materials such as tephra, ash, and pumice (Bobbink and Heil 2003; WRB 2014).

The surrounding area is highly populated, and industry, forestry, and agriculture are currently practiced in areas once covered by temperate mountain forests dominated by pines and oaks (INEGI 2007). 'White maize' (Zea mays spp.) is the most common crop, cultivated through different agricultural practices, including a variety of mechanization and fertilization schemes (synthetic/organic), with or without herbicide application. It is grown either as monoculture or under crop rotation. To compare the effect of agricultural management on soil properties and microbial activity, two study sites were selected to represent contrasting agricultural scenarios within the region. One of them is traditional management (TR) in the rural locality Manuel Ávila Camacho, municipality of Ixtapaluca. This locality is characterized by manual tillage and crop rotation and uses organic or synthetic fertilizers. The other area involves Intensive Management and is located in Amecameca de Juárez, a peri-urban area that borders the Izta-Popo National Park. In this area, white maize cultivation practices include mechanized tillage, hybrid seeds, monoculture, synthetic fertilization, and herbicides. Both sites have been under agricultural management for approximately 50 years.

Experimental Design and Soil Sampling

The experiment was organized in a 2 × 2 factorial design, selecting two types of agricultural management: intensive (I) and traditional (T), and two cultivation statuses: active (A) and fallow (F), thus assessing four treatment combinations: intensive active (IA), intensive fallow (IF), traditional active (TA), and traditional fallow (TF). For the analyses, each treatment was assessed through five composite samples collected at 0–30 cm depth in five- different plots (20 m × 20 m). Twenty composite soil samples were taken for laboratory procedures and another 20 unaltered soil samples for bulk density, with a core of 127 cm^3. Each sample was stored separately in a tightly sealed bag, then stored at 4 °C in the dark to inhibit microbial activity until further laboratory analysis.

A soil profile (1 m depth) of each treatment was open for qualitative description according to Siebe *et al*. (2006). Both profiles in the intensive treatments showed a sequence of Ap-AC-C horizons. Ap horizons encompassed from 0cm to 30 cm depth, evidencing brown color (10YR 3/2 Munsell), while AC horizons ranged from 30 cm to 50 cm depth and C horizons from 50 cm to 100 cm depth. AC and C horizons showed dark brown color (10YR 2/2 and 10YR 3/3 Munsell). Both profiles from the traditional cultivation regime showed a sequence of Ap-2Ap-Bw-C horizons. Ap horizons ranged from 0 cm to <15 cm and reflected black-dark brown color (7.5YR Munsell). The 2Ap-horizons comprised from <15 cm to 30 cm and showed a brown color (10YR 3/2 Munsell). Bw horizons were evident from 30 cm to 50 cm depth and C horizons from 50 to 100 cm in depth. Bw and C horizons showed a dark brown color (10YR 2/2 and 10YR 3/3 Munsell).

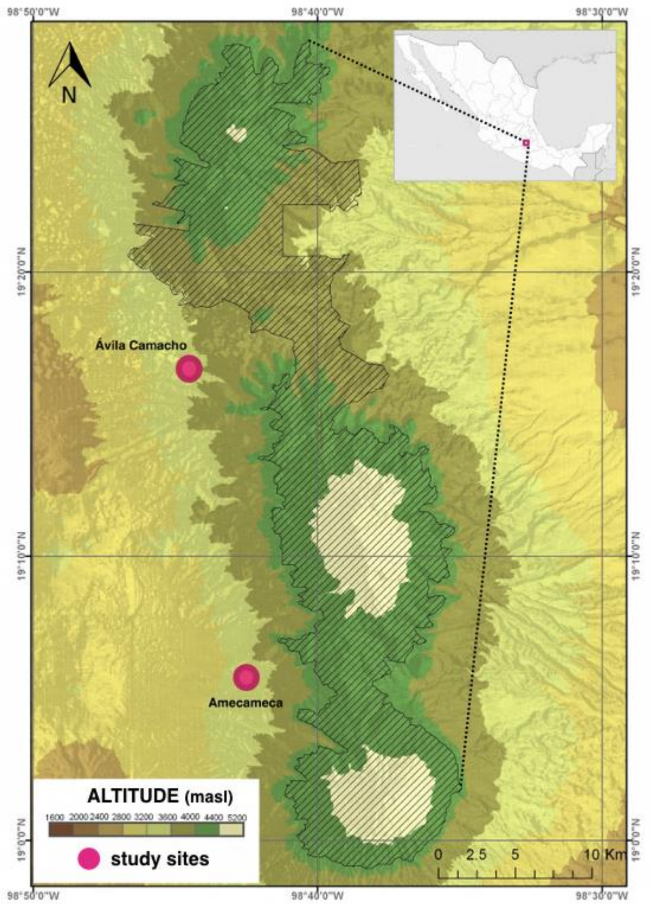

Figure 1. Study area in Mexico Highlands. The dark grey shade corresponds to the Izta-Popo National Park. Pink circles mark sample sites.

The shape and size of soil aggregates were similar in surface horizons in all treatments, but the size changed from fine (<1 mm) to coarse (5–20 mm) in deeper horizons. To evaluate the stability of aggregates, ten soil aggregates were placed in water for 10 seconds (Schlichting *et al*. 1966). According to this criterion, the stability of aggregates was classified as 'very low' under intensive management (10 out of 10 soil aggregates in the surface horizons disintegrated in water). In contrast, the stability of aggregates was higher in the traditional regime (7 out of 10 in the surface horizons disintegrate in water). Then, all profiles were classified as silandic Andosols (WRB 2014). All treatments were considered comparable because soils were similar in natural pedogenesis.

Laboratory Analyses

Physical variables

The gravimetric moisture content was determined by the method described by Van Reeuwijk (1992), drying each soil sample at 105 °C for 72 h to a constant weight. bulk density was measured with the cylinder method (Schlichting *et al*. 1996) and texture by hydrometry (Bouyocus 1962).

Chemical variables

Following a conventional procedure, soil samples were sieved through a 2 mm mesh, and collected 10 g of soil from each sieved sample for chemical testing. Then, the sieved sample was mixed with deionized water at a 1:2.5 w/v ratio for measuring pH and electrical conductivity with an Aqualytic SensoDirect pH24 potentiometer (Van Reeuwijk 1992). Exchangeable bases were extracted with 1N ammonium acetate pH 7 and Ca2+ and Mg2+ were analyzed by atomic absorption spectrophotometry (Perkin Elmer 3100). K^+ and Na^+ were analyzed with a flame photometer using the method proposed by Van Reeuwijk (1992). Total C and N contents were determined with a Perking Elmer 2400 Series II CNHS Elemental Analyzer equipped with a thermal conductivity detector, using helium as carrier gas; the temperature required was 975 °C for combustion and 640 °C for reduction. The calibration used a certified acetanilide standard (Costech C, 71.09 %; N, 10.36 %). NH_4^+ was determined using the indophenol blue method, as suggested by Rodier (1981), and NO_3^- using the extraction method with sodium salicylate ($C_7H_5NaO_3$) (Monteiro *et al*., 2003). Then, NH_{4+} and NO_{3-} were determined colorimetrically using a Genesys 20 spectrophotometer (Thermo Scientific) at 640 and 420 nm, respectively. Available P was reduced with stannous chloride and then read in a spectrophotometer at 820 nm, according to the method proposed for acid and slightly acid soils (Bray and Kurtz 1945).

Biological variables

C in microbial biomass (C_{mic}) was measured with the fumigation-extraction method (Vance *et al*. 1987), fumigating with ethanol-free $CHCl_3$, extracted with 0.5 M K_2SO_4, and quantifying the extracted C (Chávez- Vergara *et al*. 2014). Then, Cmic was calculated as the difference between C extracted from fumigated samples and C from non-fumigated samples divided by a KEC value of 0.45 (Jenkinson *et al*. 2004). Potential C mineralization was measured on days 3, 7, 12, 18, 21, and 25 of incubation. Soil samples were placed in polyvinyl chloride tubes with a 0.17 mm mesh at one end. Each sample was moistened to field capacity by capillarity and introduced into a 1 L jar with a CO_2 trap (a vial with 10 mL of 0.5N NaOH) (Robertson *et al*. 1999). Each sample was incubated at 25 °C and analyzed on five dates, adding 5 mL of 1N $BaCl_2$ to the CO_2 trap to precipitate Na_2CO_3; later, the residual NaOH was quantified with 0.5N HCl using phenolphthalein as an indicator (Robertson *et al*. 1999). The specific activities of exoenzymes were determined. In particular, β-glucosidase (β-gl) and polyphenol oxidase (POX) for the C cycle and phosphomonoesterase (Pho) for the P cycle. In this context, dehydrogenase (DHG) was selected as an indicator of the overall metabolic activity of the microbial community (Sinsabaugh and Follstand, 2011; Chávez-Vergara *et al*., 2014). The enzyme activity was determined according to Fioretto *et al*. (2009) using particular enzyme-related substrates and colorimetric measurements of the pNP produced during incubation. For the determination of β-gl, POX, and Pho, 2 g of fresh soil were added to 30 mL of modified universal buffer solution (MUB) at pH 5.8; afterward, three 0.67 mL aliquots per enzyme were obtained from this extract by adding 0.67 mL of a specific enzyme substrate. Samples were incubated for 2 h at 17 °C and then centrifuged for 2 min, adding 0.75 mL of 1N NaOH and 3 mL of deionized water to stop the reaction in all samples. Solutions were vortexed and light absorbance was measured in a spectrophotometer at 410 nm for β-gl and POX, and 460 nm for Pho (Chávez-Vergara *et al*. 2014). In the case of DHG, 1 g of fresh soil was added to a 1 % TTC solution and incubated for 24 h at 17 °C, reading DHG at 546 nm (Alef and Nannipieri 1995). The absolute activity of all enzymes is reported on a dry-weight basis and expressed in μmol g^{-1} h^{-1}.

Soil microbial metabolism indicators

Two indicators of soil ecological functions were used: specific enzyme activity (SEA) and metabolic quotient (qCO_2). SEA was calculated from a modification of the formula reported in Chávez-Vergara *et al*. (2014) based on Waldrop *et al*. (2000):

$$SEA = A / (C_{mic} / 0.001)$$

where *SEA* is expressed in μmol of pNP or tyrosine released per milligram of the nutrient in the microbial biomass per hour, *A* is the activity of the enzyme (μmol), and C_{mic} is the microbial C concentration in the soil (mg g^{-1}).

The metabolic quotient of the microbial community was calculated from

$$qCO_2 = CO_2 / C_{mic}$$

where qCO_2 is the relationship between accumulated CO_2 from the potential mineralization of C (μg C g^{-1}) and C_{mic} is the microbial C concentration in the soil (mg g^{-1}).

Statistical Analyses

The STATISTICA-12 program was used to run a factorial ANOVA on the data (physical, chemical, and biological properties of the soils), with the management regime (traditional and intensive) and cultivation status (active and fallow) as factors. A post-hoc Tukey HSD analysis was applied when the differences were significant ($p < 0.05$). A Principal Component Analysis (PCA) was applied to evaluate the soil properties and microbial metabolic indicators.

RESULTS

Soil Nutrient Concentrations

The physical and chemical properties of soil showed significant differences between intensive and traditional management (Table 1), except as specified. Soil bulk density and soil acidity were higher in intensive versus traditional management. The plot cultivation status (fallow or active) exhibited no significant effects on soil bulk density and pH.

The total soil C concentration was higher under the traditional regime and not affected by cultivation status. The total soil N concentration was higher under the traditional regime versus the intensive regime, and not affected by the cultivation status. Last, the soil C:N ratio did not differ between management regimes. In contrast, soil P concentration was higher under the intensive versus the traditional regime. The soil C:P ratio was significantly higher under the traditional versus the intensive regime. No significant effects of cultivation status on nutrient ratios were observed (Table 1).

Soil NO_3^- concentration was not affected by the management regime or crop status. In contrast, soil NH_4^+ concentration was higher under the traditional than the intensive regime. Soil NO_3^- and NH_4^+ were not affected by cultivation status. The NH_4^+:NO_3^- ratio was significantly higher under the traditional regime than under the intensive one. Dissolved inorganic P was significantly higher under intensive versus traditional management. Dissolved inorganic P was not affected by the cultivation status (Table 1).

Microbial activity

The total microbial C concentration and the potential soil C mineralization were significantly higher in soils under the traditional versus the intensive regime. Cultivation status did not affect total microbial C concentration or potential soil C mineralization. The enzymatic activities of β-glucosidase and DHG were significantly higher under the traditional versus the intensive regime. POX activity was not affected by the agricultural regime or cultivation status. In contrast, Pho activity was not affected by the management regime and was significantly higher under active cultivation than in the fallow status.

The specific enzyme activity of β-glucosidase (SEA-Bg) was significantly higher under the intensive than the traditional regime. SEA-Bg was affected by the cultivation status, displaying a higher activity in the active than the fallow cultivation status. The specific enzymatic activity of DHG (SEA-DHG) was significantly higher under traditional than under intensive management. The specific enzymatic activity of POX (SEA-POX) was significantly higher under intensive versus traditional management. The specific enzymatic activity of Pho (SEA-Pho) was affected by the interaction between the management regime and cultivation status (Table 2). Last, qCO_2 was significantly lower under the traditional regime with fallow cultivation (Table 2).

Table 1. Physical and chemical soil properties affected by the management regime in maize agroecosystems in central Mexico.

	Intensive active (n = 5)	Intensive fallow (n = 5)	Traditional active (n = 5)	Traditional fallow (n = 5)
Physical properties				
Bulk density (g cm^{-3})	1.09 ± 0.01^{Aa}	1.08 ± 0.01^{Aa}	0.82 ± 0.01^{Ba}	0.77 ± 0.03^{Ba}
Humidity (%)	11.4 ± 0.01^{Ba}	10.8 ± 0.01^{Ba}	28 ± 0.01^{Aa}	29.2 ± 0.01^{Aa}
Chemical properties				
pH	5.2 ± 0.006^{Ba}	4.8 ± 0.1^{Ba}	6.1 ± 0.2^{Aa}	6.9 ± 0.1^{Aa}
Conductivity (dS m^{-1})	0.22 ± 0.01^{Aa}	0.37 ± 0.2^{Aa}	0.26 ± 0.1^{Aa}	0.29 ± 0.1^{Aa}
Total nutrients (mg g^{-1})				
Carbon	5.1 ± 0.06^{Ba}	4.7 ± 0.06^{Ba}	26.6 ± 0.16^{Aa}	34.8 ± 0.41^{Aa}
Nitrogen	0.4 ± 0.008^{Ba}	0.4 ± 0.04^{Ba}	2 ± 0.018^{Aa}	2.6 ± 0.004^{Aa}
Phosphorus	1.6 ± 0.03^{Aa}	1.4 ± 0.14^{Aa}	0.49 ± 0.04^{Ba}	0.54 ± 0.03^{Ba}
Available inorganic nutrient (µg g^{-1})				
NH_4^+	36.7 ± 7.1^{Ba}	65.2 ± 3.5^{ABa}	92.8 ± 9.7^{Aa}	81.5 ± 9.4^{Aa}
NO_3^-	15.7 ± 2.0^{Aa}	14.1 ± 1.0^{Aa}	13.2 ± 2.0^{Aa}	14.9 ± 2.4^{Aa}
PO_4^+	145.1 ± 11.7^{Aa}	107.6 ± 6.7^{Aa}	7.7 ± 0.2^{Ba}	9.6 ± 0.6^{Ba}
Nutrient ratios				
C:N	12.5 ± 0.6^{Aa}	11.1 ± 1.0^{Aa}	13.3 ± 0.1^{Aa}	13.4 ± 0.7^{Aa}
C:P	10.4 ± 0.4^{Ba}	8.5 ± 0.8^{Ba}	16.2 ± 9^{Aa}	24.2 ± 5.3^{Aa}
NH_4^+:NO_3^-	2.3 ± 0.4^{Ba}	4.6 ± 0.8^{Ba}	6.9 ± 0.9^{Aa}	5.4 ± 1.3^{Aa}

Notes: Mean ± SD. Different capital letters indicate significant differences ($p < 0.05$) between management regimes; different lowercase letters indicate significant differences ($p < 0.05$) between active and fallow plots.

Table 2. Microbial activity of soils as affected by the management regime in maize agroecosystems in central Mexico.

Soil microbial activity	Intensive active $n = 5$	Intensive fallow $n = 5$	Traditional active $n = 5$	Traditional fallow $n = 5$
Microbial immobilization (mg C g^{-1})				
Cmic	83.93 ± 27.1^{Ba}	111.53 ± 25.0^{Ba}	279.03 ± 38.5^{Aa}	483.53 ± 12.3^{Aa}
C mineralization (µg C g^{-1} d^{-1})				
CO_2	214.27 ± 3.38^{Ba}	217.36 ± 4.88^{Ba}	356.23 ± 42.28^{Aa}	360.74 ± 7.27^{Aa}
Microbial enzyme activity (µmol g^{-1} h^{-1})				
β-gl	0.02 ± 0.00^{Ba}	0.02 ± 0.004^{Ba}	0.03 ± 0.004^{Aa}	0.03 ± 0.005^{Aa}
POX	0.74 ± 0.07^{Aa}	0.65 ± 0.048^{Aa}	0.81 ± 0.073^{Aa}	1.16 ± 0.100^{Aa}
DHG	3.04 ± 0.46^{Ba}	3.99 ± 0.528^{Ba}	50.10 ± 4.69^{Aa}	96.51 ± 4.119^{Aa}
Pho	0.06 ± 0.00^{Aa}	0.04 ± 0.006^{Ab}	0.05 ± 0.008^{Aa}	0.05 ± 0.006^{Ab}
Specific enzymatic activity (µmol mg $Cmic^{-1}$ h^{-1})				
SEA-β-gl	0.49 ± 0.00^{Aa}	0.21 ± 0.03^{Ab}	0.09 ± 0.01^{Ba}	0.05 ± 0.00^{Bb}
SEA-POX	11.4 ± 1.02^{Aa}	8.73 ± 1.75^{Aa}	3.08 ± 0.37^{Ba}	2.7 ± 0.24^{Ba}
SEA DHG	52.07 ± 16.3^{Ba}	55.06 ± 21.8^{Ba}	223.21 ± 55.4^{Aa}	221.5 ± 23.2^{Aa}
SEA Pho	1.1 ± 0.2^{Aa}	0.33 ± 0.09^{Ab}	0.23 ± 0.02^{Ba}	0.12 ± 0.01^{Bb}
qCO_2	4.86 ± 0.62^{Aa}	3.53 ± 0.61^{Aa}	3.13 ± 0.58^{Aa}	1.61 ± 0.18^{Bb}

Notes: Mean ± SD. Different capital letters indicate significant differences ($p < 0.05$) between management regimes; different lowercase letters indicate significant differences ($p < 0.05$) between active and fallow plots.

Analysis of Metabolic Microbial Indicators

The plot resulting from the PCA indicated that the variability in management regimes was associated with two principal components (PC). Together, PC1 and PC2 explained 65.6 % of the total variability. PC1 accounted for 52.81 % of the variance and showed a clear separation of plots from the intensive and traditional regimes. PC2 explained 12.81 % of the variance and separated the active from fallow cultivation statuses within the traditional regime (Figure 2).

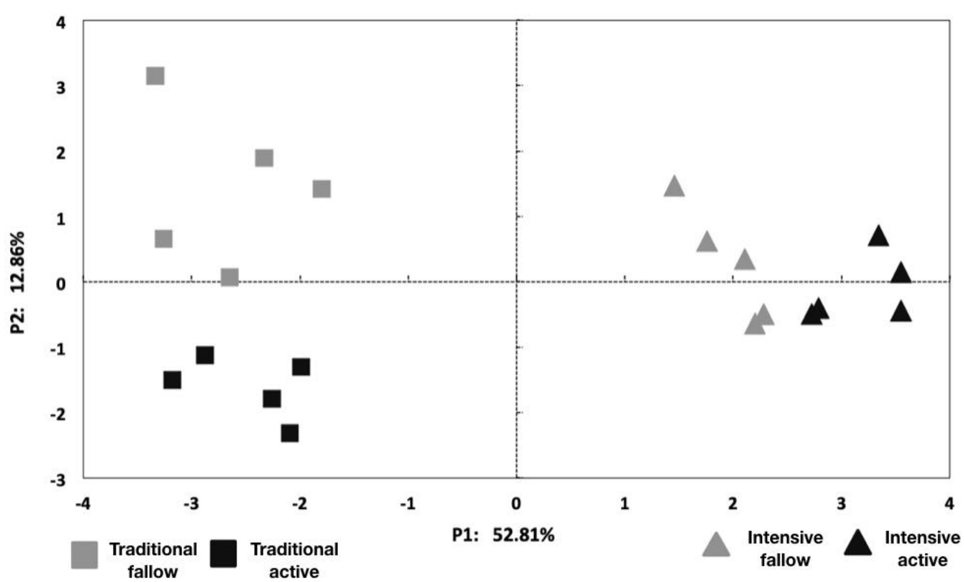

Figure 2. Principal Component Analysis (PCA) includes the physical, chemical, and biological properties evaluated in the four treatments of the experimental design: intensive fallow, intensive active, traditional fallow, and traditional active.

DISCUSSION

Physical and Chemical Changes Derived from Management Practices

Our results indicate that soil bulk density is strongly affected by agricultural management. The use of mechanized tillage reduces the volume of soil, breaks soil structure, and reduces porous space (Lima *et al*., 2017). The soil under intensive management is more compacted and, in the long term, mechanical tillage affects soil fertility because it reduces the stabilization of organic matter and the availability of soil nutrients (Martín *et al*., 2017). Demessie *et al*. (2013) found that the bulk density of volcanic soils is lower when total C and N concentrations are higher. In Mexico Highlands, bulk density is lower in soils under traditional management, suggesting that the addition of fresh organic matter could improve soil fertility and conserve the volume of soil. The addition of fresh organic matter to soils under intensive management may be an option for regulating the effects of mechanized tillage and would enhance the availability of soil nutrients.

The total concentrations of soil nutrients in agroecosystems depend on the type of fertilizer (organic or synthetic), and the intensity of application (amount per unit area) (Liu *et al*., 2017). The mechanisms that account for the higher C and N content in soils under traditional management are probably related to the combined use of organic amendments and synthetic fertilizer. In this study, mixed fertilization with organic amendments and synthetic fertilizer showed a positive effect under traditional management, where organic amendments are composed of maize and bean litter and manure that provide nitrogen and organic matter to the soil, in turn promoting the accumulation of organic C in the upper layer of soil. Flores-Sánchez *et al*. (2013) pointed out that in volcanic soils, the combined use of synthetic fertilizers and fresh organic matter has a positive effect on soil fertility. In contrast, in areas where only synthetic fertilizers are used, as soils under intensive management, total C and N concentrations in soil are lower, which is also a key factor in soil chemical degradation. This investigation found that the volcanic soils of Mexico Highlands in crops with exclusive use of synthetic fertilizers and mechanized tillage also use more fertilizers for agricultural production. The relationship between fertilizers and yield deserves further evaluation as this information can be used to modify agricultural management using the most efficient doses of fertilizer or organic amendments to mimic the positive effects of mixed fertilization that characterize traditional management.

Inorganic nitrogen in soils plays a key role in plant growth; ammonium (NH_4^+) is scarcer than nitrate (NO_3) but is immediately used by plants and microorganisms (Addiscot and Dexter, 1994). Our results showed that differences between NH_4^+ and NO_3^- levels can be associated with the type of fertilization. For example, soil acidification is the most common form of chemical soil degradation resulting from N fertilization (Brady and Weil, 2010, Li *et al*., 2016). Acidification in agricultural soils occurs when intensive N fertilization favors the oxidation of NH_4^+ to NO_3^-, releasing H^+ into the soil (Guo *et al*., 2010; Beltrán-Paz, 2017). Jordanova (2017) pointed out that volcanic soils are naturally acidic (pH < 6), but our findings showed that the accumulation of NO_3^- under intensive management reflects that nitrification is stronger than mineralization. NH_4^+ was always higher in the traditional regime and may be associated with the use of fresh organic matter that promotes the dominance of mineralization over nitrification.

Microbial Metabolic Activity as a Legacy of Agricultural Management Practices

Microbial metabolic activity is a sensitive indicator of changes in soil properties induced by agricultural management (Yi *et al*., 2021). The activity of soil microorganisms depends mainly on carbon availability because this element maintains the metabolic functions of the microbial community, including C and N mineralization (Chávez-Vergara *et al*., 2015, Kallenbach and Grandy, 2011). In agricultural soils, microbial activity depends on the addition of organic matter because it helps to counter the negative effects of over-fertilization. In this study, microbial activity indicators (biomass, mineralization, and enzyme activity) reflect a more efficient microbial metabolism in the traditional regime, where carbon availability is higher, promoting more C and N mineralization. The opposite occurs under an intensive regime where metabolic activity is lower due to limitations in C.

Enzyme activity is a functional strategy of microorganisms for the acquisition of specific compounds to perform their metabolic processes and can be interpreted as an indicator of nutrient transformations (Panettieri *et al*., 2014, Ciarkowska *et al*., 2014). The higher DHG and β-glucosidase activities showed a more efficient microbial metabolism in soil under the traditional regime and also reflects the addition of organic amendments because organic C enhances the activity and growth of heterotrophic microorganisms (Rao *et al*. 2014). Hence, the lower enzymatic activity of DHG in soils under an intensive regime is unsustainable in the long term because the exclusive use of synthetic fertilizers inhibits the growth of microorganisms (Chávez-Vergara *et al*. 2015). Moreover, ß-glucosidase measures the transformation of labile C compounds, and greater activity in soils under the traditional regime reflects the efficient use of organic C by the soil microbial community. This may be related to higher soil C concentrations and the stability of upper soil layers due to manual tillage.

Enzyme activity is widely used as a soil quality indicator (Raiesi and Beheshti, 2014, Arcand *et al*., 2017, Chavarria *et al*., 2018). However, the interpretation of enzyme activity can be unclear in soils with nutrient limitations. Both indices, qCO_2 and SEA, contribute to clarifying the changes in microbial efficiency related to carbon use (incorporation into biomass) or nutrient acquisition (C mineralization). The SEA-Bg activity and the SEA-POX activity support the hypothesis that the microbial community was more efficient in soils under the traditional regime because the microbial community not only produces extracellular and intracellular enzymes, but it grows in biomass at the same time (Raiesi and Beheshti, 2014, Chávez-Vergara *et al*., 2014). The SEA-Pho activity reflects an inefficient microbial community as regards phosphorus use in soils under an intensive regime because it produces Pho and does not grow in biomass. In soils under the traditional regime, SEA-Pho activity reflects a microbial community that produces Pho and grows in biomass. Low qCO_2 values indicate a more efficient microbial community that maintains C mineralization and accumulates C biomass. The evaluation of SEA and qCO2 has shown that microbial communities are more efficient in incorporating carbon into soils under organic fertilization (Arcand *et al*., 2017) and in reducing tillage (Chavarria *et al*., 2018) than soils under synthetic fertilization and mechanized tillage.

CONCLUSIONS

The present study provides new information for central Mexico concerning agricultural management regimes, by reporting details of agricultural management such as tillage, crops, and fertilization intensity. Agricultural management regimes can affect the physical, chemical, and biological properties of soils; however, the magnitude of these effects depends on the type of practices associated with each management regime. Intensification, such as synthetic fertilization and mechanical tillage, reduces nutrient content, the size of the microbial community, and the efficiency of use of specific substrates associated with C, N, and P dynamics. Furthermore, the addition of organic matter through organic fertilizers in the agroecological regime promotes the physical, chemical, and biological fertility of the soil and helps conserve ecological functions.

Our evaluation of the effect of agricultural management on nutrient dynamics and microbial community activity revealed some of the effects of the usage of synthetic fertilizers and mechanized tillage on the conservation of the ecological functions of soil. One of the strengths of the experimental design in this work is that it evaluated the soil under the same environmental conditions under different intensities of agricultural management. For this reason, the soil under intensive management reflects what could happen to the soil under a traditional regime over a period of 20–35 years if the management regime changes to synthetic fertilization, introduces mechanized tillage, and eliminates the addition of organic matter. This information is important for farmers, not just in this locality, but also in any agricultural scenario exposed to changes in production technologies.

Acknowledgments
The first author gratefully acknowledges the Sustainability Sciences Post-graduate Program at UNAM (Posgrado en Ciencias de la Sostenibilidad, Universidad Nacional Autónoma de México) and the Consejo Nacional de Ciencia y Tecnología (CONACyT) for the fellowship granted (723835/607566). All the authors wish to thank the peasants in Ávila Camacho and Amecameca, Estado de México, for their cooperation and hospitality during fieldwork. María Elena Sánchez-Salazar edited the English manuscript.

Funding. This project was funded by Dirección General de Asuntos de Personal Académico de la Universidad Nacional Autónoma de México trough the project identifies with the code IN104515. The Consejo Nacional de Ciencia y Tecnología (CONACyT) funded this investigation trough a fellowship identifies with the number 723835/607566.

Conflict of Interest Statement. Authors declare that there is no conflict of interests.

Compliance with ethical standards. There are no ethical issues related to this research.

Data Availability. Data are available with the corresponding author Leopoldo Galicia (lgalicia@igg.unam.mx)

Author contribution statement (CRedit). L. Rodríguez-Bustos-Conceptualization, Data curation, Formal Analysis, Methodology, writing original draft, writing review, and editing. **L. Galicia**- Conceptualization, Funding acquisition, Project administration, Supervision, Validation. **B. Chávez-Vergara**- Conceptualization, Formal Analysis, Methodology, Resources, Validation. **O. Beltrán-Paz**- Formal Analysis, Resources, Supervision, Validation, Writing original draft.

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Supplementary data

Suppl. Table 1. F (p) values of factorial ANOVA of soil parameters; significant values are marked in bold.

Soil parameters	Source of variation		
	regimes	cultivation status	interaction
Physical variables			
Bulk density	**170. 7 (< 0.001)**	1.7 (0.2)	0.52 (0.4)
Humidity	**66.79 (< 0.001)**	0.01 (0.8)	0.17 (0.6)
Chemical variables			
pH	**48.8 (< 0.001)**	2.46 (0.1)	1.64 (0.2)
Conductivity	0.24 (0.6)	**17.63 (< 0.001)**	7.75 (0.1)
Total Nutrients			
Carbon	**537.7 (<0.001)**	12.1 (0.001)	**15.5 (<0.001)**
Nitrogen	**591.6 (<0.001)**	**15.34 (< 0.001)**	**14.7 (<0.001)**
Phosphorus	**161.1 (<0.001)**	0.83 (0.3)	2.4 (0.1)
Dissolved inorganic nutrients			
NH_4^+	**15.4 (<0.001)**	0.85 (0.3)	4.5 (0.04)
NO_3^-	0.18 (0.6)	0.68 (0.4)	0.01 (0.9)
PO_4^+	**201.35 (<0.001)**	0.21 (0.6)	0.03 (0.8)
C:N	3.12 (0.09)	0.57 (0.4)	0.85 (0.3)
C:P	**119.9 (<0.001)**	0.62 (0.4)	0.50 (0.4)
NH_4^+: NO_3^-	**14. 8 (<0.001)**	3.7 (0.06)	0.59 (0.4)
Immobilized C			
C_{mic}	**42.2 (< 0.001)**	9.43 (0.007)	3.33 (0.08)
C mineralization			
CO_2	**46.7 (<0.001)**	0.06 (0.8)	0.007 (0.9)
Metabolic quotient			
qCO_2	10 (0.005)	3.07 (0.09)	**21.5 (<0.001)**
Enzyme activity			
ß-gl	**24.5 (<0.001)**	0.66 (0.41)	0.78 (0.37)
POX	**15.4 (0.001)**	3.19 (0.09)	8.81 (0.009)
DHG	**27.1 (<0.001)**	3.38 (0.08)	2.99 (0.1)
Pho	4.25 (0.04)	**12.3 (< 0.001)**	5.03 (0.02)
SEA			
β-gl	**14.83 (0.001)**	5.73 (0.02)	1.9 (0.18)
POX	10.47 (0.005)	0.89 (0.35)	0.11 (0.73)
DHG	**35 (<0.001)**	0.15 (0.6)	0.24 (0.6)
Pho	**123.3 (<0.001)**	**110.9 (<0.001)**	**63.5 (<0.001)**