



# Article Importance of Soil Health for *Coffea* spp. Cultivation from a Cooperative Society in Puebla, Mexico

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**Abstract:** The cultivation systems of *Coffea* spp. in a cooperative society in Puebla, Mexico, include Rustic, Traditional Polyculture, Commercial Polyculture, Unshaded Monoculture and Shaded Monoculture. In this work, the properties of the soil were analyzed through physical, chemical and biological analyses to determine its nutritional status. Composite sample analyses were conducted to determine physical, chemical and microbiological parameters (fungi, actinomycetes, mesophilic bacteria, nitrifying and denitrifying bacteria). Leaf nutrients were determined. Rustic was the cropping system with the highest amount of K in the soil and nutrient assimilation in the leaf (N, P, K and Fe) (p = 0.001); in addition, it had high populations of mesophilic bacteria, fungi and actinomycetes and very low nitrification and denitrification rates. The principal component analyses (PCA) (>3.25%) indicated that actinomycetes and K in soil favor the assimilation of Fe, K and P. This *Coffea* spp. cultivation system generated a lower impact on soil health than the rest of the systems and favored forest ecosystem conservation.

Keywords: soil quality; soil fertility; crop management; agroecosystem

# 1. Introduction

Primary activities are the exploitation of natural resources that have not undergone a prior transformation; in Mexico, they contribute MXN 864 681.8 million of the gross domestic product (GDP) [1] and the cultivation of *Coffea* spp. occupies twelfth place [2] with a cultivated area of 702,686 ha [3]. Worldwide, Mexico is ranked tenth as a producer of *Coffea* spp., with Chiapas, Veracruz and Puebla being the main producers [4].

Since its introduction, the cultivation of *Coffea* spp. has been influenced by sociocultural and environmental factors [5], developing five cultivation systems: Rustic or mountain, Traditional Polyculture, Commercial Polyculture, Unshaded Monoculture and Shaded Monoculture [6].

Agricultural systems in the cultivation of *Coffea* spp. depend on chemical fertilization [5], which intensifies soil chemical characteristic degradation, especially soil acidification, in addition to modifying pH, loss of exchangeable bases, reducing microbial activity, increasing aluminum (Al) and manganese (Mn) toxicity, and soil degradation [7–9]. Over time, soil health has been negatively impacted and is less able to fulfill its ecosystem function, which is to satisfy the needs of the organisms present [10,11]. Its evaluation allows us to obtain a complete position of the state of the soil through the analyses of its physical, chemical and biological properties [11].



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**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). Physicochemical indicators such as moisture, pH, CEC (cation exchange capacity), P, OC (organic C),  $N_t$  (total N), K and micronutrients (Zn, Mn, Cu, Fe) can be considered, as can the biodiversity of micro and macroorganisms involved in the soil's biogeochemical cycles [12].

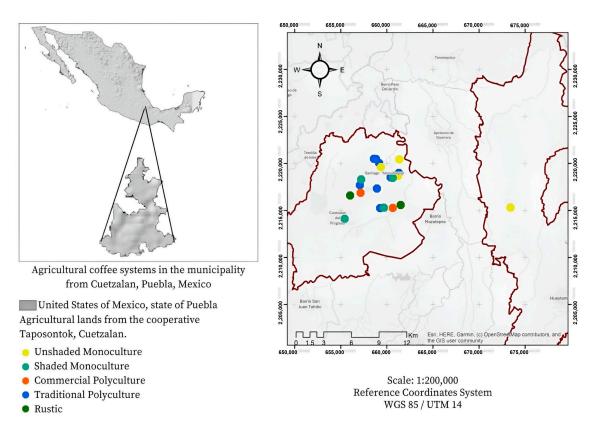
In the municipality of Cuetzalan del Progreso, Puebla, Mexico, agricultural soils dedicated to the production of *Coffea* spp. are susceptible to changes in moisture and temperature, which hinders high yields of coffee cherries, along with the change in land use, expensive inputs and migration [13]. Due to the poverty and marginalization in which the majority of producers find themselves, it is necessary to generate information on the health of the soil in the different cultivation systems.

In this context, considering the economic importance of cultivating *Coffea* spp. in Mexico and the impact it can have on soil health, this research proposes to study the properties of the soil through physical, chemical and biological analyses to determine its nutritional status under different cultivation systems of *Coffea* spp. in a cooperative society in Puebla, Mexico.

# 2. Materials and Methods

## 2.1. Sampling Site

Sampling was carried out in 15 plots of producers located in the "Taposontok" cooperative society of the municipality of Cuetzalan del Progreso, Puebla, with a latitude between  $19^{\circ}57'00''$  N and  $20^{\circ}05'18''$  N, and a longitude between  $97^{\circ}24'36''$  W and  $97^{\circ}34'54''$  W [14]. The altitude varies between 180 and 1600 m above sea level. The predominant climate is semi-warm and humid, with year-round rain and an average temperature of 20.3 °C [15] (Figure 1).



**Figure 1.** Sampling sites in the cooperative society, Puebla, Mexico. Cultivation methods: Unshaded Monoculture, Shaded Monoculture, Commercial Polyculture, Traditional Polyculture, and Rustic.

Sampling was carried out in February 2023. In each cropping system of five to ten years, randomized complete block design plots selected (n = 3): Unshaded Monoculture

(without the presence of shade), Shaded Monoculture (with species introduced for shade), Commercial Polyculture (with the introduction of plant species for shade and production), Traditional Polyculture (with shade from the forest ecosystem and introduction of valuable species), Rustic (with shade from the forest ecosystem) [6]. For the physical and chemical analyses of the soil, a composite sample to a depth of 0 to 30 cm was taken. For biological analyses, rhizospheric soil was sampled in sterile plastic containers. Leaf samples from coffee plantations without pests and diseases were used for the analyses of micronutrients. All samples were stored at 4 °C until further analyses.

#### 2.2. Physical and Chemical Analyses

The soil samples were dried and sieved through a 2 mm mesh, and the analyses were carried out in accordance with the Official Mexican Standard NOM-021-RECNAT-2000 [16]. The parameters analyzed in soil were moisture (gravimetric method), apparent density (test tube method), pH (electrometric method), EC (electrical conductivity by conductimetric method), OC (organic carbon), OM (organic matter by method of Walkley and Black), soil texture (Bouyoucos method), exchangeable bases (Ca, Na, Mg, K) and CEC (Cationic Exchange Capacity) (ammonium acetate method). Nt (total nitrogen) (Kjeldahl method) and extractable P (Bray and Kurtz method) were determined in soil and leaves [16].

Zn, Mn, Cu and Fe were quantified in soil by extraction with diethylenetriamine penta-acetic acid (0.005 M); the solution was shaken for 2 h at 120 rpm and filtered through the Whatman No. 42 paper (Merck, Darmstadt, Germany). For the determination of Ca, Mg, Na, K, Zn, Mn and Fe in leaf, an extract was prepared with  $H_2O_2$  (30% w/w) and concentrated HNO<sub>3</sub> (analytical grade); digestion was carried out at 200 °C for 10 min in a Mars Xpress microwave (CEM Corporation, North Carolina, United States of America). It was subsequently filtered with Whatman No. 42 paper and volumetric to 50 mL.

Finally, 0–8 mL of extract was injected into a flame atomic absorption spectrometer (Agilent 55B AA, Agilent Technologies, California, United States of America). An N<sub>2</sub>O/acetylene flame, a hollow cathode lamp with a current intensity of 10 mA and wavelength of 239.9 nm, was used for Ca quantification. An air/acetylene flame was used for the rest of the elements. For Mg and Cu, a hollow cathode lamp with a current intensity of 4 mA was used; Mg was read at a wavelength of 202.6 nm and Cu at 327.4 nm. A hollow cathode lamp with a current intensity of 5 mA was used to determine Na (330.2 nm), Zn (213.9 nm), Mn (279.5 nm), Fe (372 nm), K in sheet (404.4 nm) and K in soil (769.9 nm).

Calibration curves were developed for K in leaf (0, 50, 100 and 250 mg L<sup>-1</sup>), K in soil (0, 1 and 3 mg L<sup>-1</sup>), Ca in leaf (0, 50, 100 and 250 mg L<sup>-1</sup>), Ca in soil (0, 50 and 100 mg L<sup>-1</sup>) and Cu in soil (0, 2.5 and 5 mg L<sup>-1</sup>). For the following elements in both soil and leaf: Mg (0, 5, 10 and 20 mg L<sup>-1</sup>), Na (0, 50 and 100 mg L<sup>-1</sup>), Zn (0, 0.5, 1 and 2 mg L<sup>-1</sup>), Mn (0, 2, 3 and 5 mg L<sup>-1</sup>) and Fe (0, 10, 15 and 20 mg L<sup>-1</sup>).

#### 2.3. Microbiological Analyses

The total mesophilic bacterial population was determined by the most probable number (MPN) technique in nutrient broth medium in triplicate of serial dilutions from  $10^{-5}$  to  $10^{-7}$  at 48 h, 120 rpm and 30 °C [17].

Nitrifying bacteria were quantified in a Nitrosomonas medium (g L<sup>-1</sup>): (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 1.7; MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.2; CaCl<sub>2</sub>·2H<sub>2</sub>O, 0.02; K<sub>2</sub>HPO<sub>4</sub>, 0.015; Ferric EDTA, 0.001; pH 7.5; with 1 mL of trace element solution (g L<sup>-1</sup>): MnCl<sub>2</sub>·4H<sub>2</sub>O, 0.02; Na<sub>2</sub>MoO<sub>4</sub>·2H<sub>2</sub>O, 0.01; ZnSO<sub>4</sub>·7H<sub>2</sub>O, 0.01; CuSO<sub>4</sub>·5H<sub>2</sub>O, 0.002; CoCl<sub>2</sub>·6H<sub>2</sub>O, 0.0002. The nitrifying populations were determined using the MPN technique of  $10^{-5}$  to  $10^{-7}$  serial dilutions in triplicate for 8 d, 120 rpm and 30 °C.

For the denitrifying bacteria, Nitrate reduction Broth Clark medium was used (g  $L^{-1}$ ): Peptone, 20; KNO<sub>3</sub>, 2; pH 7.0. Dilutions were made from  $10^{-1}$  to  $10^{-4}$  and monitored for 5 days at 30 °C; Durham hoods were used for gas production as growth by MPN. The Sodium Salicylate and Sulfanilamide methods quantified the production of nitrates and nitrites to calculate nitrification and denitrification rates [18,19]. For the quantification of fungi and actinomycetes, the pour-plate method was used using malt mineral medium (g L<sup>-1</sup>): NH<sub>4</sub>NO<sub>3</sub>, 7; K<sub>2</sub>HPO<sub>4</sub>, 1; KH<sub>2</sub>PO<sub>4</sub>, 1; Malta, 0.2; pH 5.6; enriched with 1 mL of micronutrient solution (g L<sup>-1</sup>): MgSO<sub>4</sub>·7H<sub>2</sub>O, 4; FeSO<sub>4</sub>·7H<sub>2</sub>O, 0.2; CaCl<sub>2</sub>·2H<sub>2</sub>O, 0.2. Then, 100  $\mu$ L was plated from 10<sup>-3</sup> to 10<sup>-6</sup> dilutions, incubated in triplicate for 6 days at 30 °C and quantified as Colony Forming Units (CFU) g<sup>-1</sup> dry soil [20].

## 2.4. Statistical Analyses

The data generated were analyzed in RStudio software version 4.2.2, and the normality of the data was evaluated using the Shapiro–Wilk test with the "stats" package. A Pearson correlation analysis (p = 0.001, 0.01 and 0.05) was performed with the "corrr" package for leaf properties. Subsequently, analyses of variance (ANOVA) and Tukey's multiple comparison tests of means were performed using the "agricolae" package to determine if there were statistically significant differences (p < 0.05) in the physical, chemical and biological properties of the soil and the leaf between the different cultivation methods. Finally, principal component analyses (PCA) were used to indicate different relationships between the variables using the "prcomp" and "FactoMineR" packages. The optimal number of clusters for the data set was determined by the methods "mcclain", "tau", "ptbiserial", "frey", "Elbow" and "Silhouette". Once the optimal number of clusters (two) was selected, the k-means algorithm was applied to both the variables and the individuals (cropping systems) [21].

## 3. Results

## 3.1. Comparison of Physical, Chemical and Microbiological Characteristics in Soil

The rustic cultivation system had the silt sandy loam and silty loam texture; the rest of the *Coffea* spp. cultivation systems had a generally clayey texture. The pH values ranged between 3.9 and 4.93, with an apparent density of 0.81 to 0.91 mg cm<sup>-3</sup>, a moisture percentage range of 40.27 to 66.36% and an EC range of 0.13 to 0.28 dS (Table 1). For macronutrients, Shaded Monoculture had the highest percentage of N<sub>t</sub> (0.47%, *p* = 0.001) and OM (10.89%, *p* = 0.001). However, for K, the Rustic system had the highest value (0.76 cmol kg<sup>-1</sup>, *p* = 0.001), while P was highest in Unshaded Monoculture (7.96 mg kg<sup>-1</sup>, *p* = 0.001).

For the micronutrients, the Rustic system had significantly lower concentrations of Cu (1.05 mg kg<sup>-1</sup>), Zn (0.50 mg kg<sup>-1</sup>) and Mn (0.86 mg kg<sup>-1</sup>) and a higher concentration of Na (1.14 cmol kg<sup>-1</sup>). The Traditional Polyculture had the highest values (p = 0.001) of Mg (4.26 cmol kg<sup>-1</sup>), Zn (2.88 mg kg<sup>-1</sup>), Mn (8.96 mg kg<sup>-1</sup>) and Cu (3.45 mg kg<sup>-1</sup>).

Shaded Monoculture had the highest CEC (18.40 cmol kg<sup>-1</sup>) and Ca (6.83 cmol kg<sup>-1</sup>). For Fe, the Unshaded Monoculture system had the highest concentration (140.45 mg kg<sup>-1</sup>) and significantly lower values for Ca (1.93 cmol kg<sup>-1</sup>), Mg (1.13 cmol kg<sup>-1</sup>) and CEC (7.88 cmol kg<sup>-1</sup>). In Commercial Polyculture, the significantly lowest values were Fe (11.46 mg kg<sup>-1</sup>), OM (4.79%), K (0.33 cmol kg<sup>-1</sup>) and Na (0.88 cmol kg<sup>-1</sup>) (Table 1).

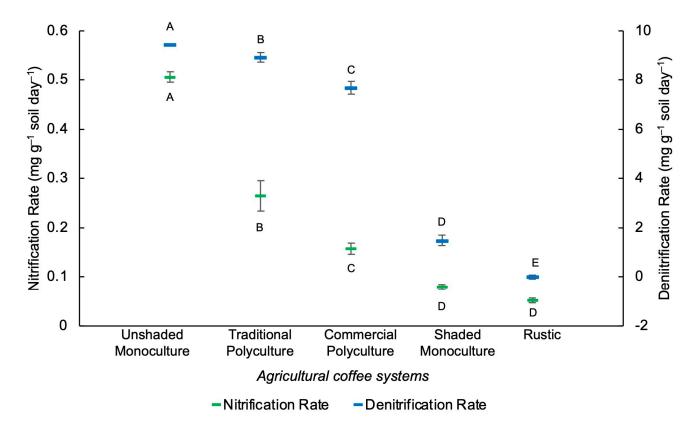
In the microbial population analyses, the Commercial Polyculture had a significantly 118-fold lower population of fungi and 3.6-fold lower actinomycetes with respect to Rustic. The Rustic system had the significantly largest population of mesophilic bacteria ( $30.16 \times 10^6$  cells g<sup>-1</sup> soil), fungi ( $0.59 \times 10^6$  CFU g<sup>-1</sup> soil) and actinomycetes ( $0.83 \times 10^6$  CFU g<sup>-1</sup> soil) and a lower population of nitrifying bacteria and denitrifying bacteria. On the other hand, in Unshaded Monoculture, the population of mesophilic bacteria ( $2.46 \times 10^6$  cells g<sup>-1</sup> soil) was significantly lower, with a greater population of nitrifying bacteria ( $336.54 \times 10^6$  cells g<sup>-1</sup> soil) and denitrifying bacteria ( $0.03 \times 10^6$  cells g<sup>-1</sup> soil) (Table 1).

Parameter		Unshaded Monoculture			Traditional Polyculture			Commercial Polyculture			Shadeo	Shaded Monoculture			Rustic		
Textural Class		Clay, Silty Clay			Silty Loam, Clay			Silty Loam, Loam Clay			Loam, Clay			Silt Sandy Loam, Silty Loam			-
pН		3.90	$\pm 0.49$		4.93	$\pm 0.55$		4.58	$\pm 0.24$		4.78	$\pm 0.52$		4.63	$\pm 0.38$		0.128
	Bulk Density (mg cm <sup>-3</sup> )		$\pm 0.01$		0.91	±0.05		0.88	±0.09		0.81	±0.07		0.87	$\pm 0.08$		0.495
Moisture (%	)	58.06	$\pm 3.75$	ab	40.27	$\pm 1.89$	b	66.36	$\pm 1.48$	а	55.72	$\pm 2.42$	ab	48.31	$\pm 2.77$	ab	0.010
EC (dS)	EC (dS)		$\pm 0.03$	а	0.28	$\pm 0.01$	а	0.14	$\pm 0.02$	b	0.24	$\pm 0.01$	а	0.13	$\pm 0.01$	b	0.001
$ m P~(mg~kg^{-1}$	$P (mg kg^{-1})$		$\pm 0.28$	а	0.33	$\pm 0.05$	b	0.68	$\pm 0.38$	b	0.20	$\pm 0.01$	b	0.26	$\pm 0.02$	b	0.001
OM (%)	OM (%)		$\pm 0.39$	bc	5.46	$\pm 0.90$	с	4.79	$\pm 0.40$	с	10.89	$\pm 0.40$	а	7.14	$\pm 0.19$	b	0.001
OC (%)	OC (%)		$\pm 0.23$	bc	3.17	$\pm 0.52$	с	2.78	$\pm 0.23$	с	6.31	$\pm 0.23$	а	4.14	$\pm 0.11$	b	0.001
N <sub>t</sub> (%)	N <sub>t</sub> (%)		$\pm 0.05$	bc	0.15	$\pm 0.01$	с	0.32	$\pm 0.03$	b	0.47	$\pm 0.07$	а	0.30	$\pm 0.02$	b	0.001
	K	0.41	$\pm 0.02$	b	0.36	±0.02	bc	0.33	±0.02	c	0.38	±0.02	bc	0.76	$\pm 0.05$	a	0.001
Exchangeable Bases	Ca	1.93	$\pm 0.38$	с	6.79	$\pm 1.10$	а	2.96	$\pm 0.47$	bc	6.83	$\pm 0.35$	а	4.12	$\pm 0.41$	b	0.001
$(\text{cmol kg}^{-1})$	Mg	1.13	$\pm 0.37$	с	4.26	$\pm 0.64$	а	1.43	$\pm 0.18$	bc	2.29	$\pm 0.04$	b	1.58	$\pm 0.08$	bc	0.001
	Na	1.02	$\pm 0.02$	ab	0.91	$\pm 0.01$	bc	0.88	$\pm 0.02$	с	0.90	$\pm 0.01$	bc	1.14	$\pm 0.11$	а	0.001
CEC (cmol kg	 )	7.88	$\pm 0.25$	c	15.56	$\pm 1.48$	b	15.02	±0.31	b	18.40	±0.32	 a	14.29	$\pm 0.24$	b	0.001
	Zn	0.66	$\pm 0.04$	b	2.88	±0.18	 a	0.53	±0.21	b	2.55	$\pm 0.47$	 a	0.50	$\pm 0.12$	b	0.001
Micronutrients	Mn	1.40	$\pm 0.20$	с	8.96	$\pm 0.20$	а	2.05	$\pm 0.54$	bc	3.67	$\pm 1.47$	b	0.86	$\pm 0.10$	с	0.001
$(\mathrm{mg}\mathrm{kg}^{-1})$	Cu	2.48	$\pm 0.46$	b	3.45	$\pm 0.23$	а	3.14	$\pm 0.08$	а	2.80	$\pm 0.12$	ab	1.05	$\pm 0.05$	с	0.001
	Fe	140.45	$\pm 1.95$	а	50.50	$\pm 0.90$	b	11.46	$\pm 0.26$	с	51.60	$\pm 0.20$	b	20.79	$\pm 0.59$	с	0.001
Mesophilic Bact	Mesophilic Bacteria <sup>1</sup>		$\pm 0.07$	e	22.56	±0.24	c	19.60	±0.31	d	27.59	±0.34	b	30.16	$\pm 0.08$	a	0.001
Nitrifying Bacte	Nitrifying Bacteria <sup>1</sup>		$\pm 3.00$	а	279.45	$\pm 1.88$	b	226.73	$\pm 2.57$	b	133.12	$\pm 5.72$	с	1.51	$\pm 0.05$	d	0.001
Denitrifying Bac	Denitrifying Bacteria <sup>1</sup>		$\pm 0.01$	а	0.02	$\pm 0.01$	b	0.023	$\pm 0.01$	b	0.02	$\pm 0.01$	b	0.01	$\pm 0.01$	с	0.001
Actinomycete	Actinomycetes <sup>2</sup>		$\pm 0.05$	b	0.39	$\pm 0.01$	b	0.23	$\pm 0.03$	с	0.38	$\pm 0.02$	b	0.83	$\pm 0.07$	а	0.001
Fungi <sup>2</sup>		0.28	$\pm 0.01$	с	0.39	$\pm 0.01$	b	0.05	$\pm 0.01$	d	0.26	$\pm 0.03$	с	0.59	$\pm 0.03$	а	0.001

Table 1. Physical,	chemical and	microbiological	analyses in soils	under coffee systems.

Data represent the mean  $(n = 3) \pm SD$ . Different letters in the column indicate significant statistical differences between cultivation systems according to the Tukey test (p < 0.05). EC: Electrical conductivity, OM: organic matter, OC: organic carbon, N<sub>t</sub>: total nitrogen, CEC: cation exchange capacity. <sup>1</sup>: values  $\times 10^6$  cells g<sup>-1</sup> soil dry; <sup>2</sup>: values  $\times 10^6$  CFU g<sup>-1</sup> soil dry.

Differences were found in the nitrification and denitrification rates of the five cultivation systems. The Unshaded Monoculture had the highest rate of nitrification (0.506 mg g<sup>-1</sup> soil day<sup>-1</sup>) and denitrification (9.430 mg g<sup>-1</sup> soil day<sup>-1</sup>). In contrast, the Rustic system had the lowest nitrification rates (0.051 mg g<sup>-1</sup> soil day<sup>-1</sup>), and nitrite production was not detected (Figure 2).



**Figure 2.** Nitrification and denitrification rates by cropping system in *Coffea* spp. Different letters indicate significant statistical differences between cultivation systems according to the Tukey test (p < 0.05).

#### 3.2. Nutrient Analyses in Coffea spp. Leaves

The Rustic production system yielded the significantly highest concentrations of foliar macronutrients and micronutrients (p = 0.001) in *Coffea* spp., including P, K, Zn and Fe (Table 2). On the other hand, N (2.98%), Ca (15,160.23 mg kg<sup>-1</sup>) and Mg (5362.29 mg kg<sup>-1</sup>) were significantly higher in Shaded Monoculture and significantly lower for Rustic. Rustic also had the significantly lowest values for Mn (33.33 mg kg<sup>-1</sup>). The Na concentration for the five culture methods did not differ (p = 0.6).

A Pearson total correlation analysis (Table 3) was performed to determine the relationship among nutrients in the leaves of *Coffea* spp. The positively related macronutrients were P and K (0.7609674, p < 0.001) and N and K (0.576633, p = 0.02). The macronutrients confithat were positively related to the micronutrients were P and Fe (0.8290195, p = 0.0001). K and Fe (0.9139314, p = 0.00000) and P and Mn (-0.54382089, p = 0.04) were negatively correlated. Micronutrients such as Ca and Mg (0.7383237, p = 0.002) and Na and Mg (0.655838, p = 0.008) also correlated positively.

Parameter		Unshade	Unshaded Monoculture			Traditional Polyculture			Commercial Polyculture			Shaded Monoculture			Rustic		
N (%)		1.20	±0.16	с	2.60	±0.11	ab	2.30	±0.26	b	2.98	±0.13	а	2.79	±0.12	а	
$P (mg kg^{-1})$		290.20	$\pm 5.19$	b	156.02	$\pm 3.65$	с	156.02	$\pm 4.56$	с	295.18	$\pm 4.78$	b	660.01	$\pm 2.80$	а	
	Κ	9659.61	$\pm 27.63$	с	11,558.43	$\pm 27.80$	b	8866.94	$\pm 11.44$	с	12,029.73	$\pm 25.15$	b	17,297.22	$\pm 23.73$	а	
	Ca	10,021.27	$\pm 10.00$	с	14,233.60	$\pm 14.97$	b	16,233.93	$\pm 12.88$	а	15,160.23	$\pm 17.44$	b	10,967.89	$\pm 14.96$	с	
	Mg	2804.05	$\pm 17.49$	с	3745.70	$\pm 19.14$	b	3478.99	$\pm 20.78$	b	5362.29	$\pm 15.82$	а	2833.22	$\pm 19.99$	с	
Micronutrients (mg kg $^{-1}$ )	Zn	10.42	$\pm 0.42$	с	12.92	$\pm 0.42$	а	12.92	$\pm 0.42$	а	11.45	$\pm 0.46$	b	12.50	$\pm 0.43$	а	
	Mn	37.50	$\pm 0.33$	С	53.33	$\pm 0.30$	b	50.73	$\pm 0.49$	b	60.21	$\pm 0.54$	а	33.33	$\pm 0.40$	с	
	Na	248.88	$\pm 2.09$		244.71	$\pm 1.49$		262.07	$\pm 3.25$		262.07	$\pm 3.42$		248.32	$\pm 1.67$		
	Fe	100.83	$\pm 1.83$	с	91.66	$\pm 1.67$	С	96.25	$\pm 1.10$	С	144.79	$\pm 1.12$	b	271.24	$\pm 1.08$	а	

Table 2. Analyses of micronutrients in *Coffea* spp. leaf.

Data represent the mean  $(n = 3) \pm SD$ . Different letters in the column indicate significant statistical differences between cultivation systems according to the Tukey test (p = 0.001).

Variables	Ν	Р	Ca	Na	Mg	К	Zn	Fe	Mn
NT	1	0.2467462	0.15408777	0.20881933	0.4175778	0.576633	0.46497838	0.4765052	0.15264905
Ν		(0.37532)	(0.58349)	(0.45513)	(0.12145)	(0.02443) *	(0.08074)	(0.07254)	(0.58705)
п	0.2467462	1	-0.34567145	-0.17495034	-0.3109498	0.7609674	0.31167246	0.8290195	-0.54382089
ľ	(0.37532)	1	(0.20696)	(0.53287)	(0.25929)	(0.00098) ***	(0.25812)	(0.00013) ***	(0.03612) *
6	0.1540878	-0.3456715	1	0.35947898	0.7383237	$-\bar{0}.\bar{3}4\bar{6}0\bar{0}6\bar{4}$	-0.00839027		0.3478592
Ca	(0.58349)	(0.20696)	1	(0.18818)	(0.00167) **	(0.20649)	(0.97633)	(0.26525)	(0.20390)
NT.	0.2088193	-0.1749503	0.35947898	1	0.655838	-0.1032731	0.22520867	-0.1655947	0.08639506
Na	(0.45513)	(0.53287)	(0.18818)	1	(0.00794) **	(0.71417)	(0.41966)	(0.55532)	(0.75949)
Mg	0.4175778	-0.3109498	0.73832374	0.65583796	1	-0.2822739	0.35225978	0.2968784	0.27643548
-	(0.12145)	(0.25929)	(0.00167) **	(0.00794) **	1	(0.30805)	(0.19785)	(0.28259)	(0.31859)
V	0.576633	0.7609674	$-\bar{0}.\bar{3}4\bar{6}0\bar{0}6\bar{4}5$	-0.10327305	-0.2822739	1	$-0.4\overline{6}\overline{5}4\overline{0}0\overline{8}2$	0.9139314	-0.3976754
K	(0.02443) *	(0.00098) ***	(0.20649)	(0.71417)	(0.30805)	1	(0.08043)	(0.00000) ***	(0.14212)
7	0.4649784	0.3116725	-0.00839027	0.22520867	0.3522598	0.4654008	1	0.3405685	-0.51123327
Zn	(0.08074)	(0.25812)	(0.97633)	(0.41966)	(0.19785)	(0.08043)	1	(0.21419)	(0.05145)
г	0.4765052	0.8290195	-0.30728059	-0.16559474	-0.2968784	0.9139314	0.34056846		-0.49853466
Fe	(0.07254)	(0.00013) ***	(0.26525)	(0.55532)	(0.28259)	(0.00000) ***	(0.21419)	1	(0.05855)
Ма	0.1526491	$-0.5\overline{4}3\overline{8}\overline{2}0\overline{9}$	0.3478592	0.08639506	0.2764355	$-\bar{0}.\bar{3}9\bar{7}6\bar{7}5\bar{4}$	$-0.51\overline{1}2\overline{3}3\overline{2}7$	$-\bar{0}.\bar{4}9\bar{8}5\bar{3}4\bar{7}$	1
Mn	(0.58705)	(0.03612) *	(0.20390)	(0.75949)	(0.31859)	(0.14213)	(0.05145)	(0.05855)	1

Table 3. Pearson correlation matrix of micronutrients in *Coffea* spp. leaf.

\* The correlation is significant at a value of p < 0.05. \*\* The correlation is significant for p < 0.01. \*\*\* The correlation is significant for p < 0.001. Two-sided significance values are in parentheses.

#### 3.3. Principal Component Analyses (PCA)

The principal components analyses showed an association of treatments defining two distinct groupings. The analysis showed that 59.6% of the variability can be explained by two main dimensions, Dim 1 (33.6%) and Dim 2 (26%). Furthermore, the plots belonging to the Rustic system had similar properties to each other, which were grouped in Cluster 1 (20%), and are different from the plots of the Unshaded Monoculture systems, Shaded Monoculture, Traditional Polyculture and Commercial Polyculture, which were grouped in Cluster 2 (80%). The differences between the *Coffea* spp. cultivation systems are visualized, separating the Rustic system, with minimal intervention in its ecosystem, from those with some modification in Figure 3.

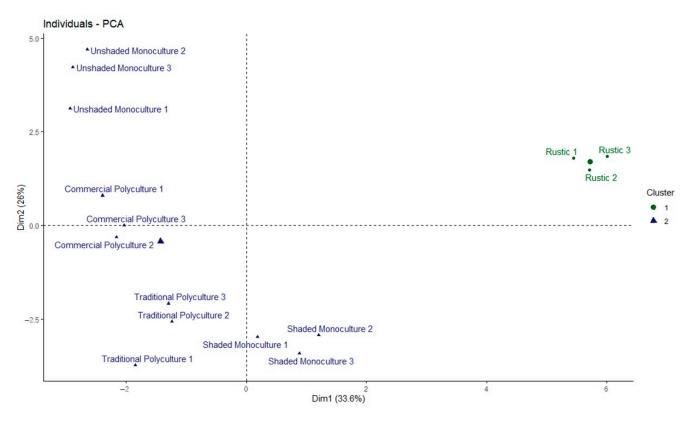
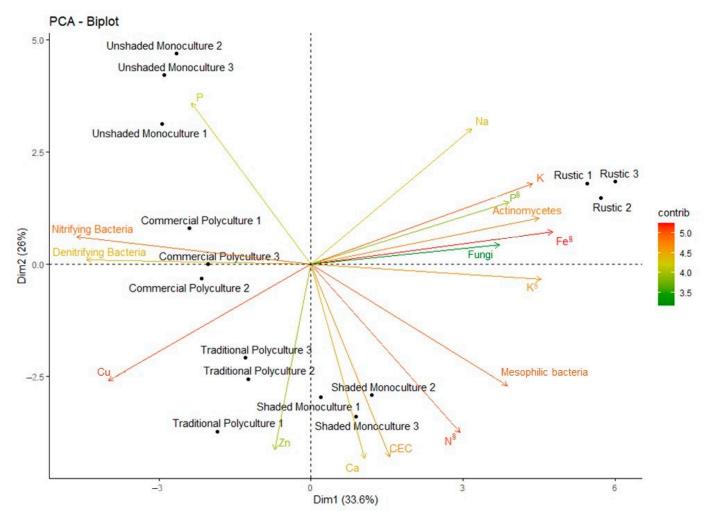


Figure 3. Principal component analyses (PCA) for the five Coffea spp. cultivation systems.

The PCA was carried out with all the study variables. The results indicated that the most important variables to explain the variability of the main dimensions (Figure 4) are the most explanatory of the variability to the average value of 3.25% for Dim 1 and Dim 2 as Fe (leaf) > N (leaf) > Cu > K (soil) > mesophilic bacteria > nitrifying bacteria > actinomycetes > CEC > K (leaf) > Ca > denitrifying bacteria > Na > P (soil) > Zn > P (leaf) > fungus.

The Rustic *Coffea* spp. cultivation system was influenced by Fe (leaf) > K (soil) > actinomycetes > K (leaf) > Na > P (leaf) > fungus. In Shaded Monoculture, the variables of N (leaf) > mesophilic bacteria > CEC > Ca were involved. Commercial Polyculture and Unshaded Monoculture systems were influenced by nitrifying bacteria > denitrifying bacteria > P (soil), and Traditional Polyculture was influenced by Cu > Zn.

For the Rustic *Coffea* spp. cultivation system, the variables with the most significant contribution were actinomycetes (8.10%), fungi (5.55%), mesophilic bacteria (5.99%) and five macroelements such as K (7.63%). Furthermore, this system favors the assimilation of nutrients such as K (8.27%), P (6.11%) and Fe (9.11%) in the leaf.



**Figure 4.** Principal component analyses (PCA) for leaf nutrients and physical, chemical and biological properties of soil. §: Nutrients in *Coffea* spp. leaf. CEC: Cation exchange capacity.

The Shaded Monoculture of *Coffea* spp. is influenced by the N (7.28%) and CEC (9.50%) due to the amount of N and Ca in the soil.

Traditional Polyculture is influenced by the micronutrients Cu (6.39%) and Zn (8.82%), which were found in a higher proportion in the soil, and Na (4.70%) is not influenced by the low amount reported.

The Commercial Polyculture and Unshaded Monoculture systems behaved similarly. The high participation of nitrifying (8.57%) and denitrifying (7.82%) bacterial populations related to N loss and P availability (6.70%) in the soil. Fungi and actinomycetes compete for nutrients, and Fe and K are unavailable for *Coffea* spp. plants.

#### 4. Discussion

The *Coffea* spp. crop in Taposontok is susceptible to diseases (rust, cercosporiosis, phoma leaf spot and bacterial blight) and pests favored by climate change [22]. Its different cultivation systems (Rustic, Unshaded Monoculture, Shaded Monoculture, Commercial Polyculture and Traditional Polyculture) have negatively affected coffee cherries' yield related to soil health [3]. This is associated with other factors such as climate, soil fertility and plant nutrition [23].

Soil physical and chemical characteristics impact plant nutrients via the availability of organic matter, water, texture and pH, to mention a few [24,25]. Nutrients are essential in several metabolic pathways that involve defense mechanisms [26], so quantifying soil

nutrients in the study system is essential because the cooperative did not have information on the fertility status of its soils.

Soil health results from the interactions between physical, chemical and biological properties that determine its function [27,28]. In the cultivation of *Coffea* spp., the nutrients that are most demanded are N and K, followed by Ca, P, Mg, S, Fe, Mn, Zn, Cu and B [29]. However, the nutritional requirements of the *Coffea* spp. crop can change depending on factors such as variety, yield, plant age and crop management [30]. These conditions were observed in the field, and the soil health of each cultivation system was diagnosed for the region's producers.

In the study site, the Rustic cropping system had high amounts of K in the soil (0.76 cmol kg<sup>-1</sup>), considered high according to the Official Mexican Standard NOM-021-RECNAT-2001 (>0.6 cmol kg<sup>-1</sup>) [16] and the recommended dose of the culture (>0.4 cmol kg<sup>-1</sup>) [31]. Actinomycete, fungi and mesophilic bacteria populations are active in the mobilization of K towards the plant for resistance processes against fungal diseases and for photosynthesis [32–34]. The amounts of K in the Rustic cropping system are in the optimal production range (15,800–21,499.99 mg kg<sup>-1</sup>) [29].

On the other hand, this Rustic system had the most significant amount of assimilable P (660.01 mg kg<sup>-1</sup>). However, the P is below the optimal range of the crop (1400–2000 mg kg<sup>-1</sup>), so the plant is deficient in P and the yield of coffee cherries decreases [29,35]. Among the sampled systems, Rustic *Coffea* spp. had the highest leaf concentration of Fe (271.24 mg kg<sup>-1</sup>), which is important for photosynthesis processes in fruit-bearing crops [36], with an optimal range of 54–121 mg kg<sup>-1</sup> [29]. These data show that the Rustic system favors the assimilation of Fe.

Other micronutrients, such as Cu, had in all cropping systems in amounts of  $1.05-3.45 \text{ mg kg}^{-1}$ , suitable for this soil type  $(1.0-3.0 \text{ mg kg}^{-1})$  [16,31]. Cu is important for plant respiration and photosynthesis, carbohydrate and N metabolism, antioxidant activity and lignification processes [37].

The CEC can affect nutrient availability in the soil for the plant since it influences the retention of cations such as Ca, as observed in the Shaded Monoculture system, where the CEC (18.40 cmol kg<sup>-1</sup>) is considered medium (15–25 cmol kg<sup>-1</sup>) [16,31]. Also, this cultivation system had the highest concentration of N by the plant (2.98%), which is considered high according to the recommended dose of the crop (2.36–2.78%) [29].

In modified cultivation systems (Commercial Polyculture and Unshaded Monoculture) in the study area, P is low in availability for mesophilic bacterial populations and in the cultivation of *Coffea* spp. P in all cultivation systems is considered deficient according to the recommended dose for *Coffea* spp. (10 mg kg<sup>-1</sup>) [31]. N data were deficient due to nitrification, denitrification, leaching and crop export, mainly documented in other intensive farming systems [38].

Small producers of *Coffea* spp. may often need to change crop management to diversify their production and ensure food sovereignty [39]. Carrying out this diversification under agroecological principles can reduce the use of inputs, increase crop yields, promote sustainability and conserve biodiversity and soil health [40,41]. However, diversified agroecosystems such as the Rustic system are not the most effective in crop yield but contribute with ecosystem services, such as preserving soil health and biodiversity,  $CO_2$  capture, improving the landscape, resilience to climate change, water conservation and maintaining biogeochemical cycles [40,42,43]. The Taposontok cooperative reported average coffee cherry yields of 3780 kg ha<sup>-1</sup> in rustic, 2850 kg ha<sup>-1</sup> of traditional polyculture, 2219 kg ha<sup>-1</sup> in commercial polyculture, 3166 kg ha<sup>-1</sup> in unshaded monoculture and 3534 kg ha<sup>-1</sup> of shaded monoculture.

The Rustic system had low activity of bacterial populations in denitrification processes. This can be attributed to the Rustic system's minimal intervention in the forest ecosystem. Only *Coffea* spp. is introduced, and the remaining plant layer is preserved in its entirety. This minimal modification allows soil health conservation and the fulfillment of its functions in the ecosystem, such as diversification of production, resilience against external factors and maintaining long-term soil fertility. In addition, by guaranteeing the health of the soil, benefits can be obtained in the plant's nutritional status.

## 5. Conclusions

The different cultivation systems of *Coffea* spp. in a cooperative society in Puebla, Mexico, indicated that the Rustic cultivation system stands out for conserving soil health. This is due to its relationship with the availability of K, Fe, P and the activity of different microbial populations (mesophilic bacteria, fungi and actinomycetes) that influence the maintenance of *Coffea* spp.

The rest of the coffee system (Traditional Polyculture, Commercial Polyculture, Unshaded Monoculture and Shaded Monoculture) had a K deficiency mainly in the soil and fewer properties associated with preserving soil health.

Systems conserved under agroecological management can become a sustainable ecosystem for small producers of *Coffea* spp.

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