

## Assessment of Soil Properties of Organic and Conventional Agroecosystems in Laguna, Philippines

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### ABSTRACT

Soil is a natural resource influenced by farming systems. Organic farming (O<sub>1-3</sub>) and conventional farming (C<sub>1-3</sub>) areas were assessed to determine the effect of agroecosystem on soil properties. The data on soil chemical parameters and microbial biodiversity were subjected to t-test/Mann-Whitney U test, and Spearman Rank Correlation analyses. Significant differences for pH ( $p<0.001$ ), electrical conductivity ( $p<0.001$ ), organic matter ( $p=0.04$ ), total nitrogen ( $p=0.04$ ), total iron ( $p<0.01$ ), and total sulfur ( $p=0.01$ ), were observed between O<sub>1-3</sub> and C<sub>1-3</sub>, while no significant differences were observed for moisture content ( $p=0.37$ ), available phosphorus ( $p=0.08$ ), and exchangeable potassium ( $p=0.40$ ). An unidentified bacterial phylum with no recognized kingdom classification was observed to be present in both agroecosystems comprising 14% of the microbial community. No significant differences were observed for alpha diversity indices between O<sub>1-3</sub> and C<sub>1-3</sub>. The association between the soil chemical properties and microbial biodiversity (OTUs) in two agroecosystems was also not significant. In conclusion, variation in farming system was found to have influenced soil chemical composition and the soil microbial biodiversity. It is highly recommended that further studies and trials be conducted to evaluate the influence of farming systems to soil properties.

**Keywords:** conventional, organic, soil chemical properties, soil microbial biodiversity

### INTRODUCTION

The agriculture sector is dependent on natural resources for its productivity and sustainability. One of the most fundamental natural resources on earth necessary for agricultural production is the soil. Soil is made up of physical, chemical, and biological components that interact in a variety of way and affect both belowground and aboveground ecosystems (Ocampo *et al.*, 2020; Buot, 2020). The interplay among the physical, chemical, and biological components in the soil contributes to the quality and health of the soil.

Soil's intricate functions and dynamics are influenced by a variety of environmental elements, both natural and man-made. Natural factors may include moisture, temperature, organic matter content, and other inherent elements present in the soil. One of the most significant human-induced factors is the agricultural land use and management which influences physical, chemical, and biological components of the soil. Agricultural management has an impact on soil microorganisms and

microbial activities by altering the quantity and quality of plant wastes entering the soil, as well as their spatial distribution, through nutrition and input modifications (Garcia-Orenes *et al.*, 2013). Conventional farming practices, through excessive fertilizer and pesticide usage and heavy tillage activities, can disrupt the function and structure of soil microbial communities, affecting the normal functioning of terrestrial ecosystems as well as soil health and quality (Ella *et al.*, 2012; Pampulha and Oliveira, 2006). Organic farming systems, on the other hand, has been clamored to maintain fertility and quality soils. The use of organic soil amendments has promoted the activities of soil microbial communities which in turn influences nutrient cycling and crop growth (Enwall *et al.*, 2007). The environmental risk with the repeated application of manures in organic farming systems is the introduction of fecal microbial flora into soil which could affect the endogenous microbial structure (Soupir *et al.*, 2006). All these factors have the potential to alter the biodiversity in the soil ecosystems resulting to direct and indirect effects on plant growth, nutrient cycling, and soil formation.

Few studies have been conducted to assess the influence of organic and conventional farming systems on soil dynamic and properties. Farming systems should be built based on the capacity and suitability of the soil resources by knowing its properties and processes. Understanding these properties and processes is essential for identifying appropriate components of long-term farming systems that also improve and maintain soil quality and biodiversity (Emami *et al.*, 2012).

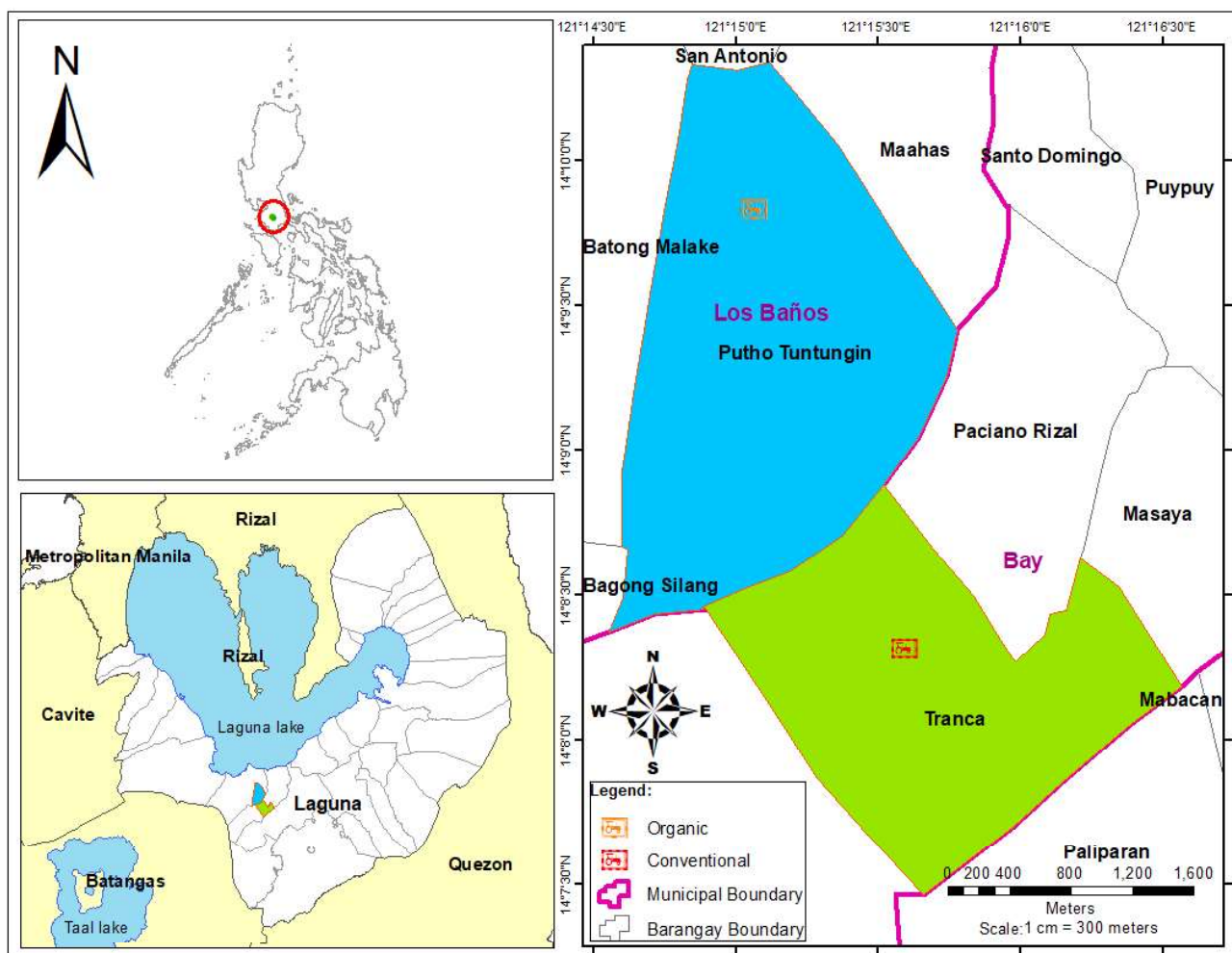
The general objective of the study was to assess how farming systems or agroecosystems can potentially modify the soil dynamics and soil properties including chemical and biological diversity. This study was conducted using soil samples gathered from the Binhing Pagkaing Alalay sa Mag-anak na Nagsisikap (PAMANA), Pili Drive, Los Baños, Laguna and the National Seed Foundation, Barangay Tranca, Bay, Laguna, Philippines. The duration of the study was from July to September 2021.

## MATERIALS AND METHODS

### Study Site

Two agroecosystems were assessed for this study, the organic agroecosystem and conventional farming sites located at Pili Drive, University of the Philippines Los Baños (UPLB), Los Baños, Laguna, Philippines, and at Barangay Tranca, Bay, Laguna, Philippines, respectively (Fig. 1). Both sites are under the supervision of the Institute of Plant Breeding (IPB) - UPLB.

**Organic Farming Area (O<sub>1,3</sub>).** The first site is the Binhing PAMANA, located along Pili Drive inside the UPLB campus, Municipality of Los Baños, Laguna. It is a 0.02 ha open field enclosed with trees and plants that serve as buffer to adjacent experimental farms. The topography of Los Baños ranges from rolling to steep and mountainous, with good drainage. Textural classification and soil series of the area belongs to Macolod clay loam (Glorioso and Ella, 2015). The area was previously



**Figure 1.** The two (2) study areas in Laguna: Organic Farming Area in Los Baños, Laguna and Conventional Farming Area in Bay, Laguna (Source: ArcGIS Coordinate Reference System: WGS 84).

an experimental station for pesticide trials under the management of the National Crop Protection Center. It was in 2008 when the area started to convert from conventional farming system to organic farming system through a new collaborative project with the Philippine Amusement and Gaming Corporation (PAGCOR) aimed to produce high quality seedlings and provide trainings on food and natural farming input production to low income generating families in the City of Manila. The project was known as Gulay Manok ATBP: Pagkaing Alay sa Mag-anak na Nagsisikap (GMA PAMANA), a project organized by the former UPLB Chancellor Luis Rey Velasco, and former UPLB College of Agriculture Dean Domingo Angeles with the involvement of Dr. Rodel G. Maghirang. Since its conception in 2008, organic and natural farming practices was implemented in the area as an advocacy and social responsibility. It was changed to its current name in 2015 and serves now as vegetable breeding station developing low input organic vegetable varieties, as training facility for farmers and study area for thesis students. The station has been sustained through various project collaborations and funding assistance from the Department of Science and Technology – Philippine Council for Agriculture, Aquatic and Natural Resources Research and Development (DOST-PCAARRD), Department of Agriculture - Bureau of Agricultural Research (DA-BAR), and the UPLB. Currently, the station has 4 regular research and administrative staff, and 2 contractual personnel. Before the soil collection, the area was planted with round (Domino F1 and Ilocos Round) and cylindrical (Fortuner) varieties of eggplant. Yield obtained during the last cropping season was 253 kg for round eggplant and 110 kg for cylindrical eggplant. Land preparation and mulching is done every 2–3 cropping; while weeding is done when needed. The station has a waterline system sourced from the university. Watering of the area is done by manual watering using hose or sprinkler every other day. Approximately 600 liters of water was used to irrigate the crops and the soil. Organic soil amendments such as vermicompost, fermented plant juice, fermented fruit juice, and processed chicken manure are the main source of fertilization for the area. Fifty (50) kg of vermicompost was applied as basal fertilizer. Another 50 kg of vermicompost as side dressing was incorporated in the soil 45 and 60 days after transplanting. Sixteen (16) L of diluted fermented plant juice was applied every 2 weeks from transplanting; and another 16 L of diluted fermented fruit juice every 2 weeks from flowering to harvesting. In addition, 200 g of processed chicken manure was applied in the area for fertilization. Crop protection management practices implemented during the cropping season were pruning of affected shoots and the application of Aztron biological pesticide only

when needed during the reproductive stage. Generally, the area is adhering to the organic farming practices as provided by the Philippine National Standards for Organic Agriculture (PNS/BAFS 07:2016).

**Conventional Farming Area (C<sub>1,3</sub>).** The second site, a 0.63-ha open area, is the National Seed Foundation (NSF) located in Barangay Tranca, Bay, Laguna. Topography of Bay is characterized from level to undulating and consisting of flood plains with wide terraces. The textural classification and soil series in this area belongs to Calumpang clay that serves as one of the best lowland rice areas in Laguna (Glorioso and Ella, 2015). The NSF, a marketing and extension arm of the IPB-UPLB, manages the area with the main purpose of developing quality and affordable seeds and seedlings to farmers and the public. Main fund source for NSF's activities is from regular funds allocated by the UPLB. It recently got funding allocation from the DA BAR for the implementation of the project entitled "Enhancing the availability of foundation seeds of superior open-pollinated, public vegetable varieties to small scale farmers". Before the soil collection, the area was planted with long purple eggplant that yielded 16 kg in March 2021. No crops were planted from March 2021 until July 2021 since the area was fallowed, harrowed, and plowed in preparation for the next cropping season in August 2021. Weeding and under brushing were done 2–3 times per crop cycle. Mulching was not done due to unavailability of materials. Irrigation is done once a week by flooding of rows. For fertilization and crop protection, synthetic fertilizers and pesticides were applied during the production. Two (2) bags of complete (14-14-14) fertilizer were used for basal application, while 2 bags each of urea + complete and urea + muriate of potash were applied as side dressings. Insecticides (*i.e.*, Lannate, Furadan, and Sevin) and fungicide (Dithane) were applied on crops when there were signs of pest and disease damage. Roundup, a glyphosate-based herbicide was applied on areas 1–2 days after sowing. Volume and application rate was based on the direction provided in the label. Fruits were harvested for seeds which were subjected to wet processing to ensure germination. Processed seeds were put in packets and then sold to farmers and other clients.

### Soil Sampling

Both study areas were divided into three transects to ensure that all portions of the farm were well represented. In each transects, six bulk soil cores were randomly sampled. The coordinates for each sampling points were identified and recorded using GPS receiver (Garmin GPSMAP 78s). Soil cores were collected from a depth of 10–20 cm using an auger. The auger was washed with



distilled water and disinfected with alcohol to prevent contamination. This step was repeated each time before sampling a new transect. The soil samples were taken from the auger using sterile gloves and placed in a sterile plastic bag. Soil samples were then air-dried for 3 days then sieved on 2 mm mesh. Homogenization of the soil samples were done at the Binhing PAMANA. The six bulk soil cores collected in each transect were mixed where three replicates were taken for physicochemical analyses and one replicate for soil microbial analysis. A total of nine replicates for physicochemical analyses, and three replicates for microbial analyses per study area were collected. Soil samples for physicochemical analyses were placed in clean polypropylene plastic bags, while soil samples for microbial analysis were placed in plastic tubes and stored at 4° C before transport to laboratory for soil microbial biodiversity analysis.

### Soil Physicochemical Analysis

The physicochemical properties of the soil such as soil texture, MC, pH, EC, OM, available P, exchangeable K, and total Fe were analyzed by the Department of Agriculture - Regional Soils Laboratory, Lipa, Batangas. S content of the soil was analyzed by the Department of Agriculture - Bureau of Soils and Water Management, Diliman, Quezon City, and determined from the result of sulphate analysis by conversion method. The result was then converted to Total Sulfur by multiplying the Total sulfate by 0.334.

The hydrometer method developed by Bouyoucos (1962) was used in identifying the % composition of sand, silt, and clay of the soil samples. Soil texture was determined using the soil texture triangle. Gravimetric method was used to measure MC of soil samples. Soil pH level was determined using the potentiometric method and EC by using the conductimetric test method. Organic matter was determined using Walkley and Black method, UV-Vis and % N was computed from the organic matter using the formula: % N = %OM x 0.05. The resulting %N was converted to ppm. Available P was determined using the Olsen, UV-Vis test method (Jackson 1958; Bray & Kurtz 1945). Exchangeable K was identified using the Ammonium Acetate Extraction and Atomic Emission Spectroscopy method, and Total Fe by using DTPA-TEA Extraction and Flame Atomic Absorption Spectroscopy (Flame AAS) method. Turbidimetric method was used to determine Total Sulfate.

### Soil Microbial Analysis

DNA and NGS Library quality checking, library construction, DNA sequencing, and Operational Taxonomic Units (OTU) Analysis/Bioinformatics were done by Macrogen, Inc., Seoul, South Korea. This

was made possible through the assistance of Kinovett Scientific Solutions, Co., Quezon City, Philippines.

**DNA Quality Checking.** Quantity of DNA was identified through the picogreen method using Victor 3 fluorometry, which employ a double-stranded DNA specific dye, and specifically and accurately quantitate dsDNA even in the presence of many common contaminants (Turner Biosystems, Inc. 2007; Macrogen, Inc. n.d). This was followed by gel electrophoresis to assess the condition of the DNA. Finally, size of DNA was checked using 2100 Bioanalyzer for DNA fragments with < 1.kb, and Pulsed-field gel electrophoresis (PFGE) method for larger DNA fragments which are < 150kb.

**NGS Library Quality Checking.** To verify the size of PCR enriched fragments, the template size distribution was assessed by running on an Agilent Technologies 2100 Bioanalyzer using a DNA 1000 chip. It was then followed by library quantity check through Illumina Library and PacBio Library. Optimum cluster densities across every lane of every flow cell were created to achieve the highest quality of data on Illumina sequencing platforms. The process required accurate quantitation of DNA library templates. Afterwards, prepared libraries using qPCR according to the Illumina qPCR Quantification Protocol Guide were then quantified. Furthermore, Qubit standard Quantification solution and calculator were used to generate a standard curve of fluorescence readings and calculate the library sample concentration.

**Library Construction.** After performing quality control (QC), qualified samples were subjected to library construction. The sequencing library was prepared by random fragmentation of the DNA or cDNA sample, followed by 5' and 3' adapter ligation. Alternatively, "tagmentation" combines the fragmentation and ligation reactions into a single step that greatly increases the efficiency of the library preparation process. Adapter-ligated fragments were then PCR amplified and gel purified.

**DNA Sequencing.** For cluster generation, the library was loaded into a flow cell where fragments were captured on a lawn of surface-bound oligos complementary to the library adapters. Each fragment was then amplified into distinct, clonal clusters through bridge amplification. When cluster generation was completed, the templates were readied for sequencing.

**OTU analysis.** FLASH V1.2.11 was used for assigning, cutting, and merging of paired end reads. CD-HIT-OTU and rDNATools were used to perform OTU finding and



identification as well as to export, filter, and cluster 16s sequences (Magoc and Salzberg 2011; Li *et al.*, 2012; Schloss *et al.*, 2009). On the other hand, QIIME was utilized for identification of taxonomic composition for each sample from phylum to level species, construction of phylogenetic trees from aligned and filtered representative sequences of OTUs, and production of publication-quality graphic results through statistical analysis and visualization (Caporaso *et al.*, 2010).

### Statistical Analyses

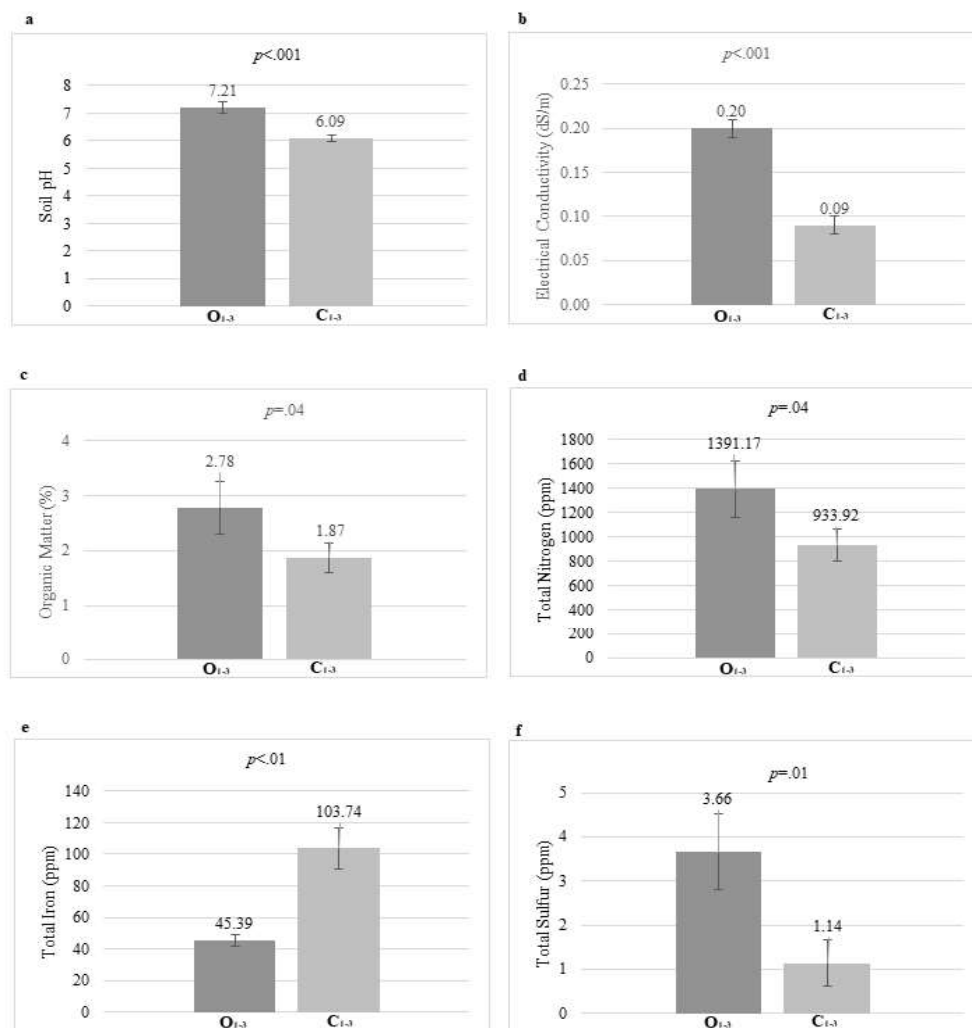
Shapiro-Wilk's test was used to assess the normality of the data before subjecting the data to variance test, t-test, and correlation analysis. Variance test was then conducted to identify the assumption of variance for t-test. Afterwards, the results of the soil analyses were statistically analyzed using a t-test analysis for the comparison of the chemical properties and soil microbial biodiversity of the two farming systems. Mann-whitney U test, on the other hand, was used for the analysis of parameters that resulted to a non-normal distribution.

Spearman's correlation coefficient analysis was then used to assess the relationship of soil microbial diversity with the soil chemical properties. Statistical tool used for the assessment of the result was the R software.

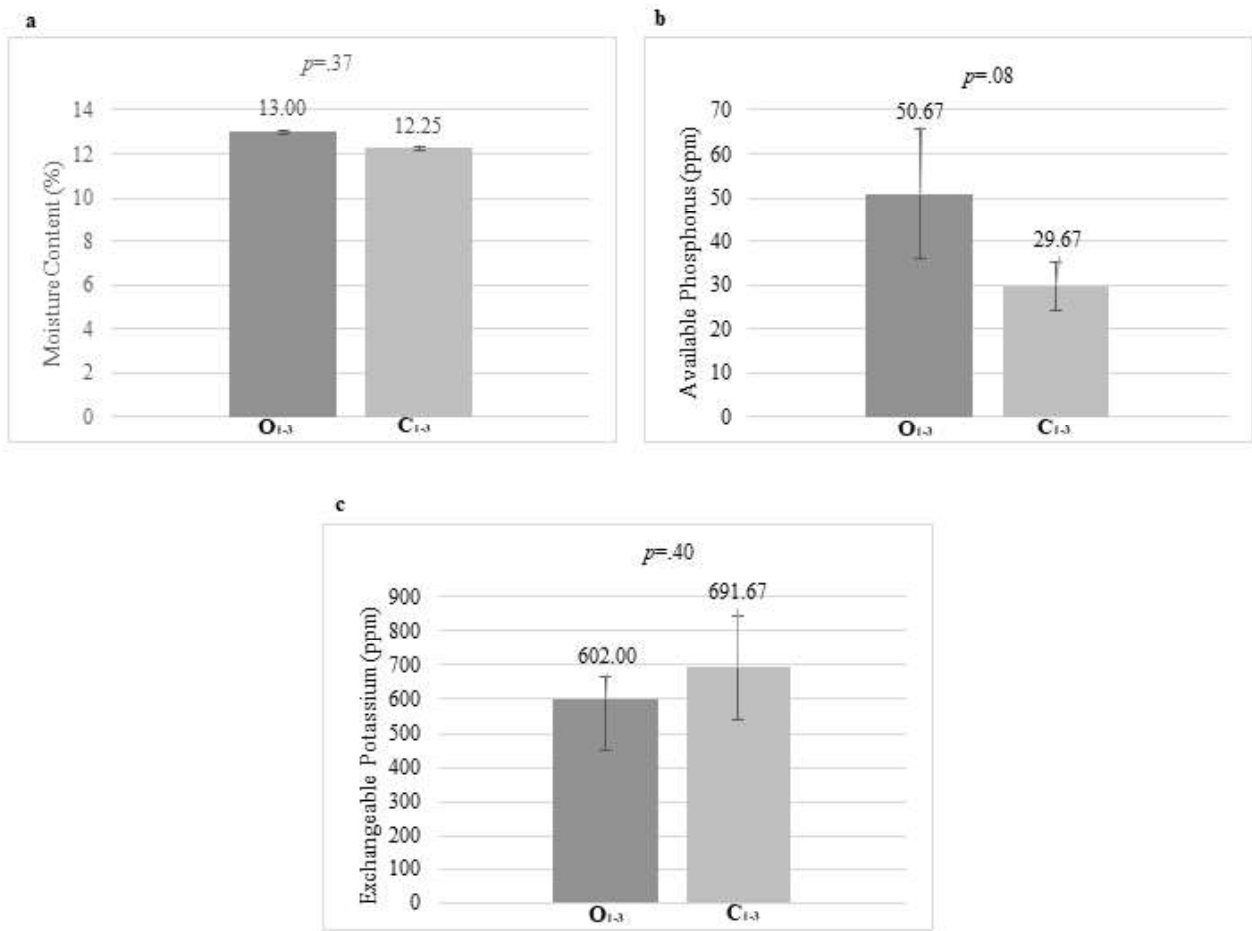
## RESULTS AND DISCUSSION

### Soil Physicochemical Properties of the Two Agroecosystems

Soil chemical properties such as soil pH, MC, EC, OM, total N, available P, and total S were higher in  $O_{1-3}$  than in  $C_{1-3}$ . Exchangeable K and Total Fe were the only exceptions as they were found to be higher in  $C_{1-3}$  than in  $O_{1-3}$ . Statistical significance was observed for soil pH ( $p<0.001$ ), EC ( $p<0.001$ ), total Fe ( $p<0.01$ ), total S ( $p=0.01$ ), OM ( $p=0.04$ ), and total N ( $p=0.04$ ) (Fig. 2). On the other hand, no statistical significance was observed for MC ( $p=0.37$ ), available P ( $p=0.08$ ), and exchangeable K ( $p=0.40$ ) (Fig. 3). Both agroecosystems were dominantly loam and clay loam in texture.



**Figure 2.** Soil chemical properties of  $O_{1-3}$  and  $C_{1-3}$  with observed statistical significance: (a) soil pH level, (b) EC (dS/m) (c) OM content, (d) total N, (e) total Fe, (f) Total S. N=3.



**Figure 3.** Soil chemical properties of O<sub>1-3</sub> and C<sub>1-3</sub> with no statistical significance: (a) MC, (b) available P, (c) exchangeable K. N=3.

The absence of statistical significance in the MC for both agroecosystems can be associated with the soil textural classification and water availability (Savci, 2012; Owens and Rutledge, 2005) Both agroecosystems were classified as loam and clay loam, soil textural classifications that have moderate moisture saturation and moisture depletion rate (Pragyan and Arulmozhiselvan, 2019; Sudha and Sinha, 2017). Irrigation and water supply are made available daily in both agroecosystems which in return did not add to the differences in O<sub>1-3</sub> and C<sub>1-3</sub>. This implies that the MC for organic farming systems (13%) was comparable with conventional farming systems (12.25%).

The resulting high soil pH of 7.21 for O<sub>1-3</sub> can be attributed to the use of processed poultry manure as fertilizer. Rayne and Aula (2020) reported that the elevated level of calcium carbonate in poultry manure contributes to the increase in soil pH. Similarly, Azeez & Van Averbek (2012) mentioned in their studies that buffering effect from CaCO<sub>3</sub> in manures influenced pH increase. Another explanation relevant to the increase in soil pH in organic farms was made by Aziz *et al.*

(2016) where it was reported that humic and fulvic acids produced from OM in organic soil amendments were able to form complex compounds with Al<sup>3+</sup> in soil. On the other hand, lower pH value of 6.09 can be observed for C<sub>1-3</sub> due to the use of urea-based fertilizers. Although urea is least expected to cause soil acidity, the long-term application of this type of fertilizer can contribute acidification of soils (Lungu and Dynoodt, 2008; Barak *et al.*, 1997). Nonetheless, the soil pH of soil under C<sub>1-3</sub> are within the optimum range of 5.5–7.0 which makes nutrients readily available for crops.

Electrical conductivity is an indication of the amount of nutrients that are made available for absorption by crops (Brady, 1974). Higher mean EC can be seen from soil samples under organic farming practices (0.20 dS/m) than soil samples under conventional farming practices (0.09 dS/m). Similar observation was reported by Aban (2014) and Capewell (2013). More so, organic farming utilizes animal manure for fertilization. With manures incorporated in the soil, nutrients and salts are mineralized and release to the soil, which in return increases the EC of the soil (Azeez

and Van Averbek, 2012). EC results for both farming systems are considered non-saline.

OM was higher for  $O_{1-3}$  (2.78%) than  $C_{1-3}$  (1.87%) and consequently with higher total N of  $O_{1-3}$  (1391.17 ppm) than in  $C_{1-3}$  (933.92 ppm) since the value was computed based on the OM content. These high values can be associated with the implementation of organic farming system through the incorporation of organic soil amendments. In a study conducted by Zaman *et al.* (2015) relative to the effects of vermicomposting on post-harvest soils, OM content of soils incorporated with vermicompost was 2.39% while OM content of soils without vermicompost was 1.70%. Mondelaers *et al.* (2009) explained that organic farms implement integrated farming system approach and manure and crop residues recycling, which in turn improves OM content. Studies by Benoit *et al.* (2016) and Benoit *et al.* (2014) reported that lower leaching of nutrients as nitrate ( $NO_3^-$ ) and nitrous oxides ( $N_2O$ ) were influenced by continued implementation of organic farming practices and by increased OM content in the soils. Increase in total N can also be attributed to the incorporation of vermicompost and animal manure as fertilizers. It was observed by Azarmi *et al.* (2008) that total N in soil was significantly affected by vermicompost due primarily to the ability of vermicompost to produce residual N in soil. Rayne and Aula (2020) observed as well that an increasing trend for total N in soils as the rate of animal manure is increased, and that the release of N or any other nutrient from animal manure depends on the rate of mineralization.

Despite the non-statistical significance for available P, a higher mean value was observed with soils samples under organic farming system (50.67 ppm) than with conventional farming system (29.67 ppm). Presence of humus and the incorporation of organic amendments in soil could have influenced an increase in available P in soil. Capewell (2013) and Doran *et al.* (1996) explained that humus is more apparent in organic farms which help in holding P in the soil. Azarmi *et al.* (2008) and Marinari *et al.* (2000) reported in their studies that vermicompost application in soils has significantly increased P compared to plots with no vermicompost application which implies that the continuous inputs of phosphorus into the soil were probably from slow release from vermicompost application, while the release of P was due primarily to the soil microbial activity of soil microorganisms (Arancon *et al.*, 2006). On the other hand, the utilization of organic soil amendments such as plant and manure composts could possibly influence P level in soils. Herencia *et al.* (2008) and Laboski and Lamb (2003) explained that organic soil amendment

degradation resulted in organic acid concentrations which then successfully reduced P adsorption to the soil and enhanced its availability.

In contrast to available P, exchangeable K was higher in soils under conventional farming systems (691.67 ppm) than soils under organic farming systems (602.00 ppm). The resulting values for Exchangeable K in  $C_{1-3}$  could be attributed to the soil texture classification and the farming input. Complete (NPK) fertilizers, Urea + Muriate of Potash, and Urea+ NPK were used as soil amendments in  $C_{1-3}$  and served as source of K. More so, Buol (1987) explained that clay loam and loam soil with 24–38% clay could retain exchangeable K at higher concentrations than sandy soils with 2% clay; thus the high numerical mean value for  $C_{1-3}$ .

The high significant difference in numerical mean values for total Fe could be attributed with pH value of the soil. Jelic *et al.* (2010) reported that the solubility of iron was reduced when soil pH has been increased which favored the oxidation of ferrous ion ( $Fe^{2+}$ ) to ferric ion ( $Fe^{3+}$ ) and influenced the precipitation of Fe (III) salts and oxides. It was also reported that iron in acidic soils is reduced to ferrous ion and made iron more available in soils for plant uptake. These findings explained the higher mean value in  $C_{1-3}$  (103.74 ppm) than in  $O_{1-3}$  (45.39 ppm).

Higher mean value for total S can be observed for  $O_{1-3}$  (3.66 ppm) than in  $C_{1-3}$  (1.14 ppm) which can be attributed to OM content and use of organic soil amendments for fertilization. It was observed by Rayne and Aula (2020) that organic soil amendments such as animal manure significantly increased OM content in soils. With the accumulation of OM content in soils, total S is more pronounced and made available for transformation into forms available for plant uptake (Jordan and Reisenauer 1957). Moreso, Eriksen (2009) reported that S is present in both organic and inorganic forms of manure. With these observations, this signified that organic farming influences the presence of total S in soils.

### Soil Microbial Biodiversity of the Two Agroecosystems

Five alpha diversity indices *i.e.*, OTUs, Chao1, Shannon, Inverse Simpson, and Good's Coverage were used as basis in the identification and interpretation of soil microbial communities of the two agroecosystems. In general, higher mean values can be observed for those samples under organic farming system than those under conventional farming practice. However, no significant differences were observed between the



two agroecosystems in all the alpha diversity indices (Table 1). The study was limited to three replicates per area; thus, there was no chance to eliminate outliers.

Moreso, non-significant results can be attributed to high variation in the obtained results.

**Table 1.** Alpha diversity indices of the soil microbial community of the two agroecosystems.

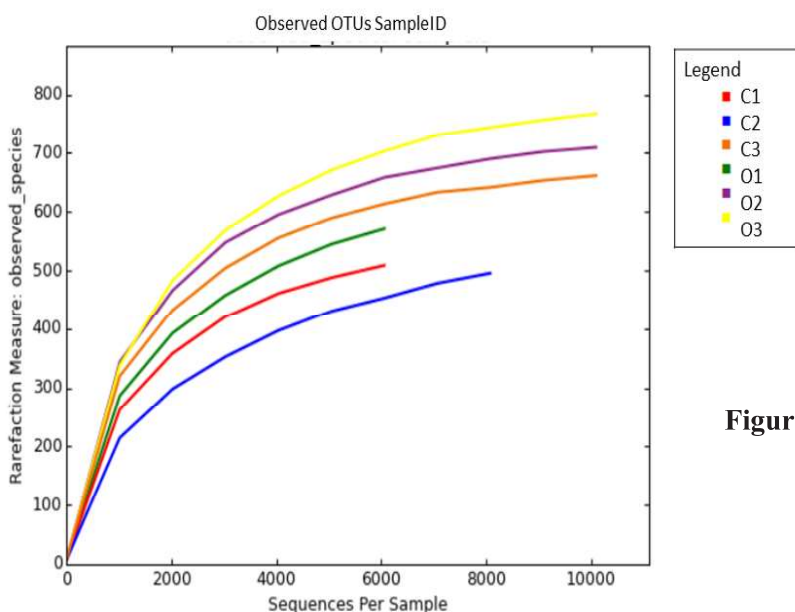
Indexes	Organic Farming Area				Conventional Farming Area				p-value
	O <sub>1</sub>	O <sub>2</sub>	O <sub>3</sub>	Mean	C <sub>1</sub>	C <sub>2</sub>	C <sub>3</sub>	Mean	
OTUs	576.00	718.00	779.00	691.00	517.00	496.00	678.00	563.67	0.20
Chao1	690.73	739.62	812.25	747.53	582.00	625.14	690.69	632.61	0.07
Shannon	7.53	8.03	7.87	7.81	7.17	5.91	7.72	6.93	0.19
Inverse Simpson	0.99	0.99	0.99	0.99	0.98	0.91	0.99	0.96	0.38
Goods Coverage	0.98	1.00	1.00	0.99	0.98	0.98	1.00	0.99	1.00

Note: O<sub>1</sub>, O<sub>2</sub>, and O<sub>3</sub>: Replicates of organic farming area C<sub>1</sub>, C<sub>2</sub>, and C<sub>3</sub>: Replicates of conventional farming area

High throughput of DNA sequences classified as OTUs with 97% similarities were yielded through Illumina sequencing. A mean average of 691 OTUs was observed in O<sub>1-3</sub> compared to the yielded mean average of 563.67 in C<sub>1-3</sub>. O<sub>3</sub> recorded the highest number of OTUs at 779 followed by O<sub>2</sub> at 718. C<sub>2</sub> was observed to have the lowest recorded OTUs at 496. The higher mean average for O<sub>1-3</sub> based on OTUs could be attributed to the provision of organic soil amendments and plant supplements that enriches OM content, and the indigenous bacteria from vermicompost and processed animal manure. Chao1 is an estimator of abundance and a parameter to determine community richness (Orlina 2020). Higher Chao1 mean values were observed in O<sub>1-3</sub> (747.53) compared to C<sub>1-3</sub> (632.61). It can also be observed that Chao1 mean values were greater than

OTU mean values for both agroecosystems implying that additional reads can be done to discover other species. This was well justified by the result of the rarefaction curve (Fig. 4). The rarefaction curve for OTUs was observed to have not reached plateau, thus additional reads is likely to discover more OTUs for the samples.

Shannon indices for both O<sub>1-3</sub> and C<sub>1-3</sub> were 7.81 and 6.93, respectively. These indices implied that there is a great diversity of microbial populations in the two agroecosystems. Higher Inverse Simpson index of 0.99 was observed for O<sub>1-3</sub> compared to C<sub>1-3</sub> with an index of 0.96. Since both values are close to 1, this is a good indication of a high microbial population in an agroecosystem. On the other hand, both agroecosystems



**Figure 4.** Rarefaction curve of observed OTUs.

have an average of 0.99 for Good's Coverage that yields a good estimate of the number of species present in the entire population detected in the soil samples.

Based on the taxonomic abundance count, there were 19 phyla, 53 classes, 110 orders, 206 families, 379 genera, and 526 species found in the soil samples obtained from the two agroecosystems. It was observed

that the most common bacterial phyla in the two agroecosystems were Proteobacteria, Actinobacteria, Acidobacteria, Firmicutes, Nitrospirae, Verrucomicrobia, Gemmatimonadetes, Chloroflexi, and Bacteroidetes (Table 2). The most dominant phyla under different soils were Proteobacteria and Actinobacteria accounting for 23.96% and 14.24% of the total microbial population, respectively.

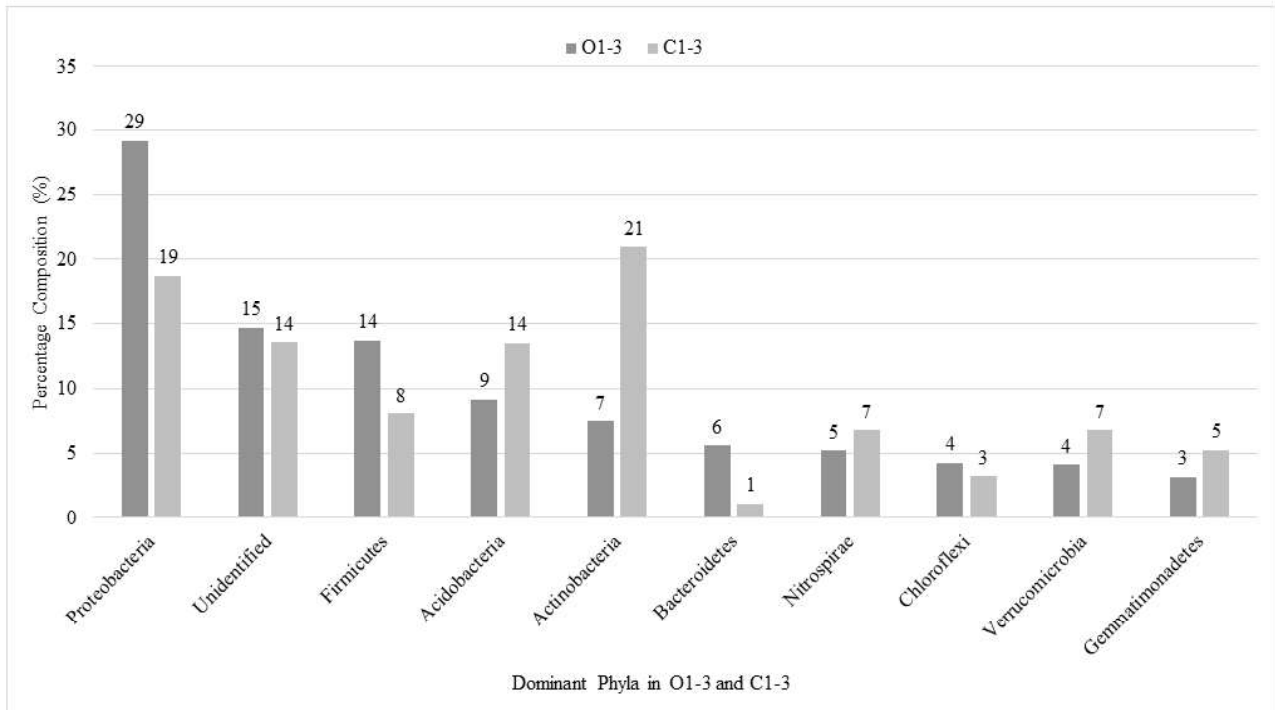
**Table 2.** Percentage composition of the microbial community of the two agroecosystems at the phylum level.

Phylum	Relative Abundance (%)									
	Total Mean	Standard Deviation	O <sub>1</sub>	O <sub>2</sub>	O <sub>3</sub>	Mean O <sub>1-3</sub>	C <sub>1</sub>	C <sub>2</sub>	C <sub>3</sub>	Mean C <sub>1-3</sub>
<b>Kingdom: Archaea</b>										
<i>Candidatus Thermoplasmatota</i>	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<b>Kingdom: Bacteria</b>										
<i>Acidobacteria</i>	11.37	3.12	8.45	7.80	11.35	14.72	11.66	12.45	16.49	13.57
<i>Actinobacteria</i>	14.24	11.34	6.88	8.74	6.87	9.20	15.39	36.45	11.10	20.98
<i>Armatimonadetes</i>	0.00	0.01	0.00	0.00	0.02	0.01	0.00	0.00	0.00	0.00
<i>Bacteroidetes</i>	3.34	3.13	4.57	3.27	8.94	5.59	0.69	0.73	1.86	1.09
<i>Chlamydiae</i>	0.16	0.17	0.08	0.15	0.43	0.22	0.00	0.00	0.29	0.10
<i>Chloroflexi</i>	3.76	0.66	4.23	4.07	4.52	4.27	3.27	2.73	3.76	3.25
<i>Cyanobacteria</i>	0.04	0.06	0.00	0.00	0.12	0.04	0.12	0.00	0.00	0.04
<i>Firmicutes</i>	10.88	4.20	9.75	13.65	17.76	13.72	9.84	8.36	5.95	8.05
<i>Fusobacteria</i>	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.01
<i>Gemmatimonadetes</i>	4.22	1.64	3.66	3.77	2.14	3.19	6.97	3.70	5.07	5.25
<i>Ignavibacteriae</i>	0.02	0.02	0.05	0.04	0.01	0.03	0.00	0.00	0.00	0.00
<i>Nitrospirae</i>	5.96	2.18	8.14	4.10	3.28	5.17	8.53	4.92	6.82	6.76
<i>Planctomycetes</i>	2.24	0.94	2.51	2.94	2.52	2.66	1.83	0.55	3.11	1.83
<i>Proteobacteria</i>	23.96	7.07	32.30	29.75	25.51	29.19	22.76	12.37	21.07	18.73
<i>Spirochaetes</i>	0.03	0.07	0.00	0.16	0.00	0.05	0.00	0.00	0.00	0.00
<i>Thermodesulfobacteria</i>	0.20	0.22	0.43	0.52	0.14	0.36	0.02	0.06	0.01	0.03
<i>Verrucomicrobia</i>	5.43	1.91	3.56	4.64	4.04	4.08	5.60	8.91	5.80	6.77
<b>Kingdom: unknown</b>										
<i>Unidentified</i>	14.15	3.46	15.41	16.40	12.34	14.72	13.32	8.77	18.64	13.57

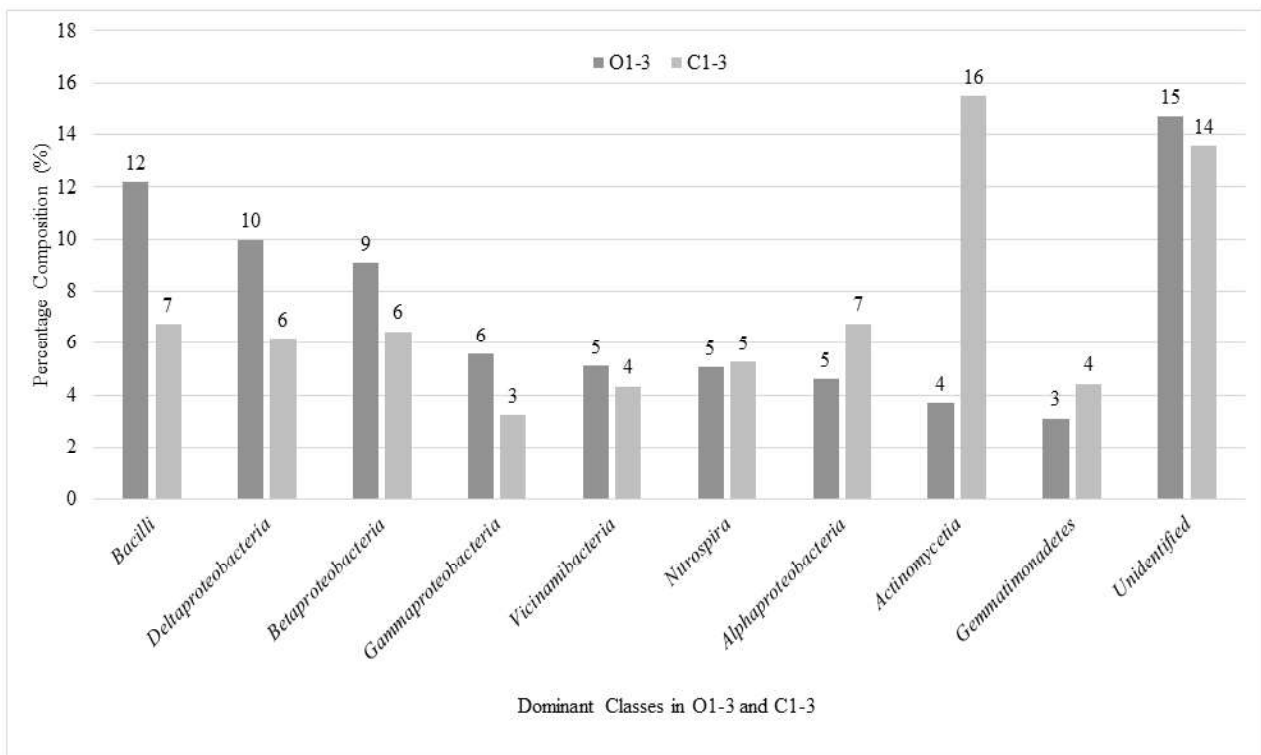
Proteobacteria, a Gram-negative bacterium, has been observed to be of higher percentage in O<sub>1-3</sub> (29%) than in C<sub>1-3</sub> (19%); while Actinobacteria, a Gram-positive bacterium, has been found to be more dominant in C<sub>1-3</sub> (21%) than in O<sub>1-3</sub> (7%) (Fig. 5). Similar observation was reported by Liao *et al.* (2018) and Pershina *et al.* (2016) that organic farming systems tend to be dominated by Proteobacteria where its survival and growth was attributed to the incorporation of organic amendments, manure, and silage. Chen *et al.* (2012) also mentioned that Proteobacteria is more dominant in soils that are less disturbed, hence the higher percentage of the said phylum in O<sub>1-3</sub> with low tillage practices. More so, Shange *et al.* (2012) reported dominance of Actinobacteria in conventional fields due to its propensity for soil environments that are highly tilled and cultivated. This type of phyla plays a key role in the decomposition of plant biomass by producing cellulolytic enzymes to breakdown simple sugars and cycle of soil minerals (Zhang *et al.*, 2019; Lewin *et al.*, 2016). An unidentified bacterial phylum with no

recognized kingdom classification was also observed to be present in both agroecosystems comprising 14% of the microbial community. The relative abundance percentage was revealed to be second (15%) in O<sub>1-3</sub> and third (14%) in C<sub>1-3</sub>. Furthermore, *Candidatus Thermoplasmatota* was the only phylum under Kingdom Archaea that has been identified for soil samples cultivated under conventional farming practice.

Actinomycetia belongs to the Phylum Actinobacteria that has been identified as the most dominant class for C<sub>1-3</sub> (Fig. 6). The dominance of this class could be attributed primarily to land use since Hill *et al.* (2011) reported that microbial population of Actinomycetia favors physical disturbance and shows higher relative abundance in cultivated system. Moreover, the smaller size particles of loam and clay loam soils in C<sub>1-3</sub> offered a protective habitat for actinomycetes through pore size exclusion of predators and assisted the growth and survival of this class (Sheoran *et al.*, 2018). With actinobacteria and Actinomycetia favoring physical



**Figure 5.** Percentage composition of dominant phyla in  $O_{1-3}$  and  $C_{1-3}$ .

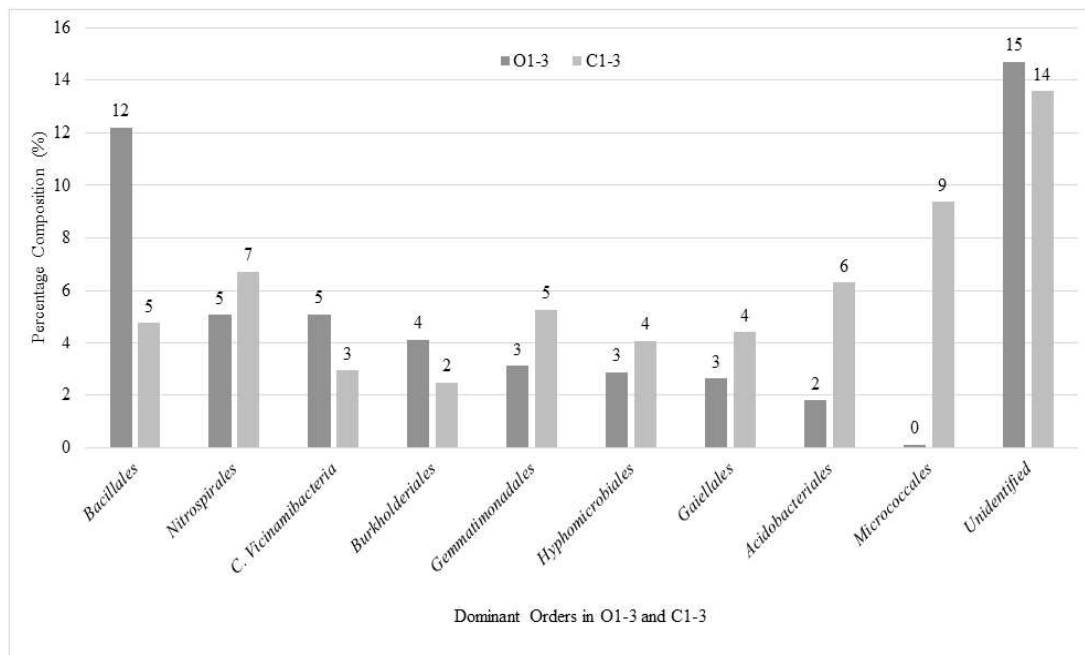


**Figure 6.** Percentage composition of dominant classes in  $O_{1-3}$  and  $C_{1-3}$ .

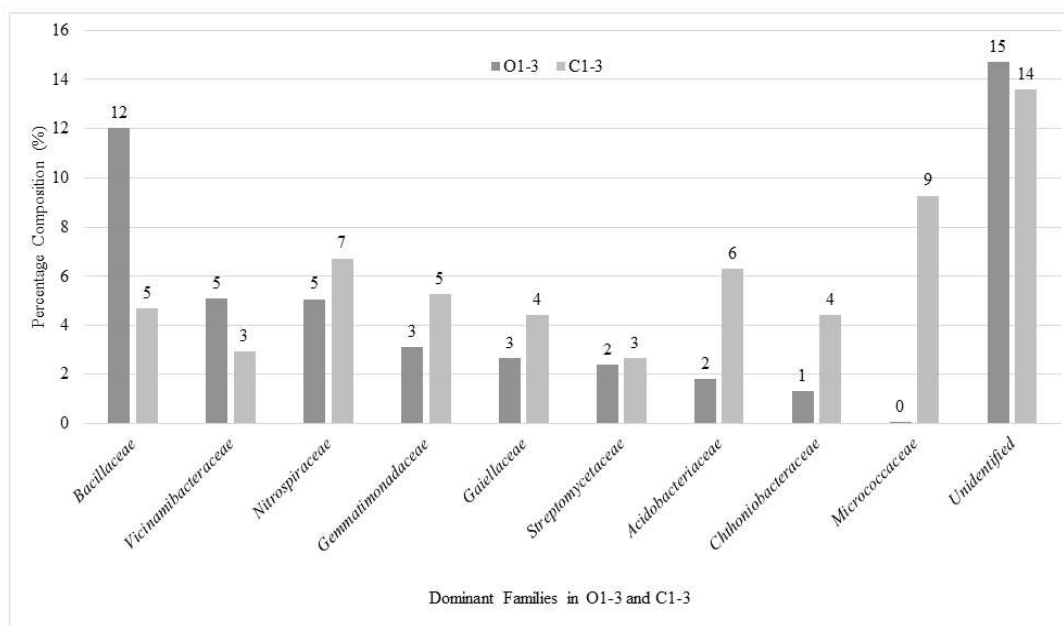


disturbance in soils, this justified the lowest relative abundance observation in  $O_{1-3}$  for Micrococcales (order), Micrococcaceae (family), Pseudarthrobacter (genus), and Pseudarthrobacter phenanthrenivorans (species) (Fig. 7-9). Interestingly, the results presented a high relative abundance of an unrecognized microorganism from class, order, family, genus, and species level in  $O_{1-3}$ . The second most dominant class for  $O_{1-3}$  was Bacillus. Bacillus under the Phylum Firmicutes are known to be a beneficial bacteria and as versatile plant growth-

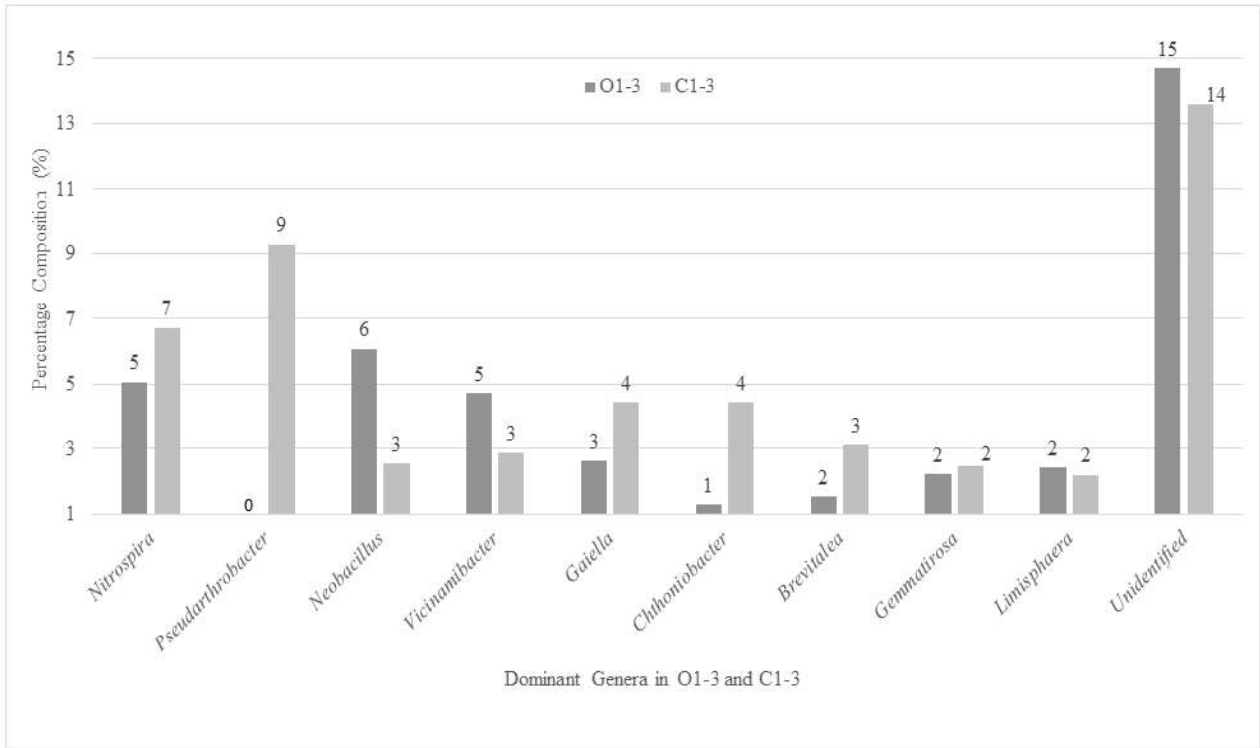
promoting bacteria (PGPB) by assisting in the production of auxin, surfactin, and phosphatase (Bacon *et al.*, 2015). The dominance of this Gram-positive bacteria has been reported by Ye *et al.* (2016) to be influenced by organic soil amendments that favors the microbial growth and abundance. This has been made evident since vermicompost, fermented fruit juices, fermented plant juices, fish amino acids, and processed animal manure were incorporated in the  $O_{1-3}$ .



**Figure 7.** Percentage composition of dominant orders in  $O_{1-3}$  and  $C_{1-3}$ .



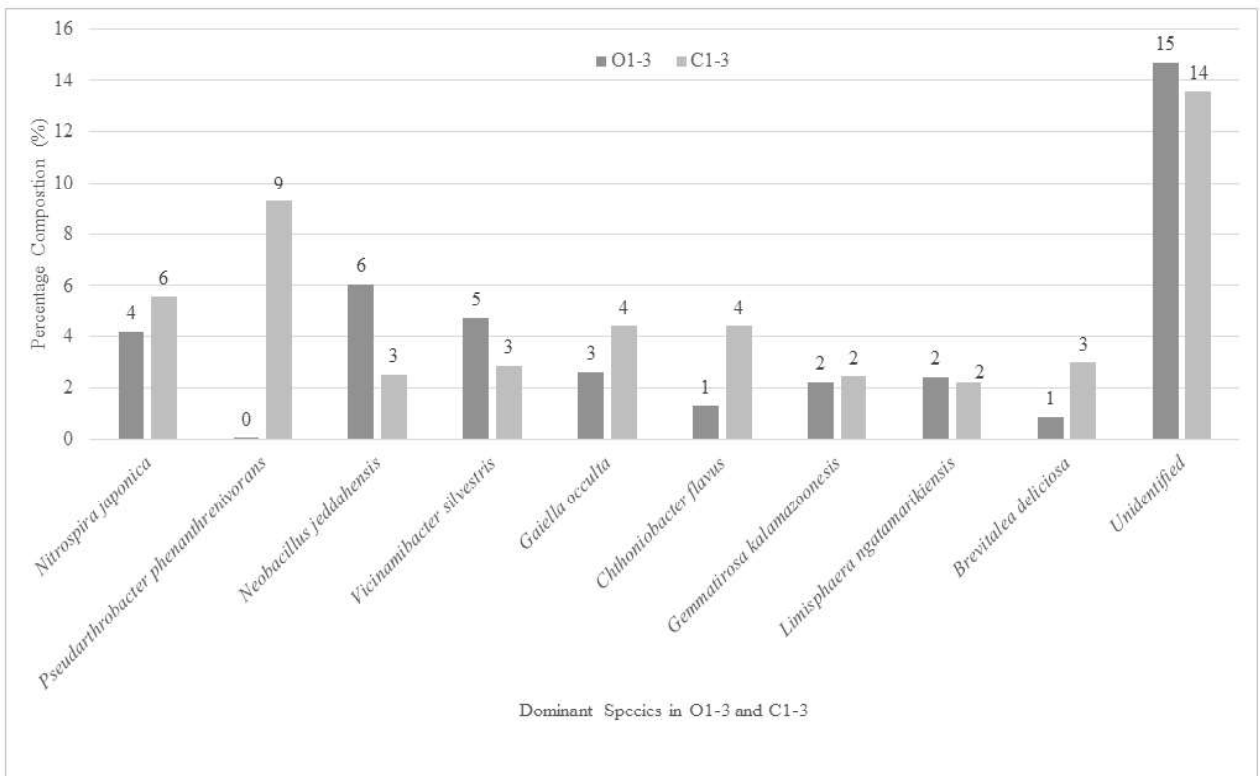
**Figure 8.** Percentage composition of dominant families in  $O_{1-3}$  and  $C_{1-3}$ .



**Figure 9.** Percentage composition of dominant genera in O<sub>1-3</sub> and C<sub>1-3</sub>.

Both agroecosystems were observed to be dominated by an unrecognized species with no kingdom, phylum, class, order, family, and genus classification (Fig. 10);

hence an implication that the DNA sequencing of this species has yet to be recognized by any known bacterial taxonomy. The second dominant species for O<sub>1-3</sub> with



**Figure 10.** Percentage composition of dominant species in O<sub>1-3</sub> and C<sub>1-3</sub>.

an average contribution of 6.07% was *Neobacillus jeddahensis*, known also as *Bacillus jeddahensis*. This species was first isolated in the manure of an obese man originating in Jeddah, Saudi Arabia. This was also observed to grow in axenic media between 28 and 45° C (Bittar *et al.*, 2015). The third dominant species in O<sub>1-3</sub> was the *Vicinamibacter silvestris* with an average of 4.73% contribution in the microbial community of the area. This species was observed to transform organic phosphorus to inorganic phosphorus which makes phosphorus available for plant use (Huber *et al.*, 2016; Kumar *et al.*, 2011).

On the other hand, the second and third most dominant species in C<sub>1-3</sub> were *Pseudarthrobacter phenanthrenivorans* and *Nitrospira japonica*, respectively. *Pseudarthrobacter phenanthrenivorans* is an aerobic, non-motile Gram-positive bacterium that can internalize phenanthrene by growing cells on glucose, and in phenanthrene as a sole carbon source (Kallimanis *et al.*, 2011). It was further reported by the authors that the recorded growth was in pH 6.5–8.5 and temperature 37° C–40° C. Ushiki *et al.* (2013) explained *Nitrospira japonica* as a bacterium containing nitrite and bicarbonate as the sole energy and sole carbon

source, respectively. This type of species under the Class Nitrospira was also reported to convert urea to ammonia and carbon dioxide and contribute to nitrogen cycling. (Koch *et al.*, 2015). However, there are limited studies discussing the influence of the above-mentioned species to the farming systems.

### Association between soil chemical properties and soil microbial diversity

The correlation between the soil chemical properties and the soil microbial biodiversity of O<sub>1-3</sub> and C<sub>1-3</sub> is presented in Table 3. It was observed that MC, soil pH, EC, OM, total N, available P, and total S have positive correlation with the OTUs. Highest positive spearman correlation coefficient (0.83) was observed between MC and OTUs which could imply that MC is a significant factor in the microbial abundance. Borowik and Wyszowska (2016) concluded in their study that the excessively dry and wet soils may cause a decrease in the microbial abundance as these situations create adversity to the growth of aerobic gram-positive and gram-negative bacteria. Therefore, it is important to identify the optimum moisture content necessary for the growth and survival of the microorganisms in the soil.

**Table 3.** Correlation ( $r^2$ ) of alpha indexes with the soil chemical parameters.

Parameters		OTUs	Chao1	Shannon	Inverse Simpson	Goods Coverage
Moisture	$r^2$	0.83	0.60	0.77	0.54	0.77
Content (%)	$p$ -value	0.06	0.24	0.10	0.30	0.10
pH	$r^2$	0.66	0.77	0.60	0.37	-0.09
	$p$ -value	0.18	0.10	0.24	0.50	0.92
Electrical Conductivity (dS/m)	$r^2$	0.60	0.83	0.54	0.18	-0.14
	$p$ -value	0.24	0.06	0.30	0.66	0.80
Organic Matter (%)	$r^2$	0.31	0.54	0.37	0.60	-0.43
	$p$ -value	0.56	0.30	0.50	0.24	0.42
Total Nitrogen (ppm)	$r^2$	0.31	0.54	0.37	0.60	-0.43
	$p$ -value	0.56	0.30	0.50	0.24	0.42
Available Phosphorus (ppm)	$r^2$	0.37	0.60	0.31	0.43	-0.49
	$p$ -value	0.50	0.24	0.56	0.42	0.36
Exchangeable Potassium (ppm)	$r^2$	-0.66	-0.60	-0.77	-0.54	-0.77
	$p$ -value	0.18	0.24	0.10	0.30	0.10
Total Iron (ppm)	$r^2$	-0.83	-0.94	-0.89	-0.77	-0.31
	$p$ -value	0.06	0.02	0.03	0.10	0.56
Total Sulfur (ppm)	$r^2$	0.83	0.83	0.77	0.30	0.14
	$p$ -value	0.06	0.06	0.10	0.54	0.80

Legend:  $r^2$ : Spearman Correlation Coefficient; \*Low significance ( $0.05 \geq p$ -value > 0.01)



On the other hand, a negative correlation for OTUs was observed with exchangeable K and total Fe. It can also be observed that the spearman correlation coefficient between OTUs and Total Fe was -0.83. Interestingly, significant differences between total Fe and Chao1 ( $p=0.02$ ) and between total Fe and Shannon Index ( $p=0.03$ ), which imply negative association between the parameters. Fe is largely oxidized and precipitated as ferric oxides in cultivated soils. With the deficit in Fe, exudates are released by roots which significantly affect microbial abundance and enzymatic activity. Soil microbes also influence the weathering of soil and release of Fe through the process of redox, complexation, acidification (Colombo *et al.*, 2014). In general, no significant differences were observed between the soil chemical and soil microbiological properties.

## CONCLUSIONS AND RECOMMENDATIONS

This study was conducted to assess how agroecosystems can potentially modify soil chemical properties and microbial biodiversity. Two agroecosystems located in the Municipalities of Los Baños and Bay in the Province of Laguna were assessed to determine the effects to soil chemical properties and soil microbial diversity from July to September 2021. Significance differences for pH, EC, OM, total N, total Fe, and total S were observed between  $O_{1-3}$  and  $C_{1-3}$ , while no significant differences were observed for available P and exchangeable K. The two agroecosystems each have a distinct and diverse microfauna that housed novel and unknown microbial species. Proteobacteria, a gram-negative bacterium, has been observed to be of higher percentage in  $O_{1-3}$  than in  $C_{1-3}$ ; while Actinobacteria, a gram-positive bacterium, has been found to be more dominant in  $C_{1-3}$  than in  $O_{1-3}$ . An unidentified bacterial phylum with no recognized kingdom classification was also observed to be present in both agroecosystems comprising 14% of the microbial community. No significant differences were observed for alpha diversity indices between  $O_{1-3}$  and  $C_{1-3}$  and the association between the chemical properties and soil microbial diversity (OTUs) in the two agroecosystems was not significant. Interestingly, significant differences between total Fe and Chao1 and between total Fe and Shannon Index were observed which imply negative monotonic association between the parameters. It is concluded that the variation in farming system or agroecosystem was found to have influenced to varying extents the soil chemical composition and the soil microbial community. It is highly recommended that further studies and trials be conducted to evaluate the influence of farming systems to soil properties. Effects of soil depth as well as the volume and frequency of soil amendments can be done to further evaluate its effect to

soil chemical properties and soil microbial biodiversity. Influence of soil properties to crop productivity and efficiency can be set up with consideration to the choice of crops and relevant cropping seasons. Thus, there is a need to conduct a detection of the effect of climate to soil properties, from planting to harvesting, as well as monitoring of growth yield, quality, and sales as affected by farming system. In addition, 18sRNA gene sequencing can also be considered to describe new species that have never been successfully cultured.

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