

Soil Physicochemical Characteristics and Spore Density of Indigenous Arbuscular Mycorrhizal Fungi (AMF) in Different Vegetation Patches of a Marginal Upland in Central Philippines

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Date received: February 5, 2021

Revision accepted: July 18, 2023

Abstract

*To show the importance of plant-microbe association in stressed tropical landscapes, the study evaluated the physicochemical characteristics and indigenous arbuscular mycorrhizal fungi (AMF) spore density in selected vegetation patches of marginal upland soils in Central Philippines. Five quadrats were used in the study, namely *Andropogon aciculatus* (control plot), *Imperata cylindrica*, *Chromolaena odorata*, *Melastoma malabathricum* and mixed vegetation patch. Results showed that soils in the studied vegetation patches had different physicochemical characteristics and AMF spore density. Although all patches had clay soil texture and comparable water-holding capacity, mixed vegetation patch had moderate compaction (1.33 g cm^{-3}) and higher porosity (50.31%) compared with the other patches, which had extreme compaction ($1.37\text{-}1.65 \text{ g cm}^{-3}$) and lower porosity (37.74-48.76%). In terms of chemical properties, soils in different patches had moderate (5.69) to strongly acidic pH (5.13), moderate organic matter (2.69-2.91%) levels, low P (2.67-3.94 ppm) content and above critical K levels ($0.2\text{-}1.2 \text{ cmol}^+ \text{ kg}^{-1}$). Results also revealed that *C. odorata* patch had the highest spore density count ($11.33 \text{ spores } 100 \text{ g}^{-1}$), followed by mixed vegetation ($9 \text{ spores } 100 \text{ g}^{-1}$), *M. malabathricum* ($8.33 \text{ spores } 100 \text{ g}^{-1}$), and *A. aciculatus* patch having the lowest spore density count ($4 \text{ spores } 100 \text{ g}^{-1}$). The significant spore density variability was attributed to the differences in soil's physicochemical characteristics among the different vegetation patches and AMF host specificity. Spores detection, therefore, indicated AMF presence and helped the adaptation mechanism of the natural vegetation in this degraded upland.*

Keywords: AMF, marginal upland, spore density, vegetation island

1. Introduction

Marginal or degraded uplands are hilly or mountainous lands with low crop productivity due to poor soil quality, limited water availability and unfavorable socioeconomic conditions (Tyler, 2004; Asio *et al.*, 2009, 2013). Dwivedi (2002) suggested that degraded uplands are widespread in Southeast Asia and other humid tropical climates where resource-poor farmers habituate. However, intensive cultivation predominates in the Philippines despite its fertility constraints and ecological instability (Cramb, 2001).

Of the total land area of the Philippines, 60% is classified as uplands and is vulnerable to soil erosion, particularly when denuded of its forest cover (Oldeman, 1994; Briones, 2009). Uplands in Central Philippines, particularly in Leyte, are subjected to massive soil degradation due to deforestation and increasing demands for arable lands for various agricultural activities (Asio, 2006). These scenarios lead to soil degradation, which forms a fragile environment mainly portrayed by the marginal uplands of Central Philippines. Asio *et al.* (2013) observed that this upland constitutes a specific vegetation distribution pattern that significantly influenced the landscape's ecological stability. This vegetation exhibits patches or strip patterns (Facelli and Temby, 2002) of grasses and shrubs. Rango *et al.* (2006) often called these patches fertility or hydrologic islands that are a reservoir of nutrients or pools of fertility reserve (Engler and Guisan, 2009). These patches are comparable to the vegetation patches of the drylands in the tropics, which perform a dynamic role in the dryland environment stability (Nelson, 1994). These vegetation patches are also vital in regulating hydrological processes (Vásquez-Méndez *et al.*, 2010), improving soil fertility (Muñoz-Robles *et al.*, 2011), and help control plant microbes' interactions (Martinez-Garcia *et al.*, 2011) that are essential components of the ecosystem in arid and semiarid areas.

Mendez *et al.* (2010) explained the vital function of these patches in the fragile ecosystem with an emphasis on regulating hydrological processes such as reduced erosion and surface sealing. These vegetation patches often contain high organic matter (OM) reserves, where intense biological activity occurs, and the hotspot of microbial diversity and physical activity (Nannipieri *et al.*, 2017). The vegetation patches are frequently associated with microorganisms that help the vegetation create a mechanism for adaptation in the disturbed ecosystem (Bautista *et al.*, 2007). Such vegetation means to modify and engineer its root's structure to enhance water and nutrient absorption, which is possible through a mutual association with fungi in the natural ecosystem.

This association, which is frequently called the association of life, is termed mycorrhiza. Mycorrhiza is rooted in two Greek words, *myco* meaning fungi and *rrhiza* meaning root (O'Connor *et al.*, 2002), and its meaning, in reality, is the symbiosis between a fungus and root, which Frank first introduced in 1885 (Alizadeh, 2011). Linderman (1991) and Alizadeh (2011) explain that mycorrhiza is a mutual sharing of life. The fungi, as the associate and vital partner of the plant, have the responsibility to supply food, growth hormones, and safety of plant's roots from pathogens attack. In contrast, the vigorous plant offers nutrients for the energetic requirement of the fungus. Its role in the natural and disturbed ecosystem is valuable and indispensable as its function is unique and incomparable to other microbes and plant associations (Alizadeh, 2011). There are seven recognized types of mycorrhizal associations involving different groups of fungi and host plants and distinct morphology patterns (Brundrett *et al.*, 1996). The most popular and highly studied association is the vesicular-arbuscular mycorrhizas in which zygomycete fungi produce arbuscules, hyphae and vesicles within root cortex cells (Brundrett *et al.*, 1996; Chen *et al.*, 2005). Alizadeh (2011) explained that terrestrial ecosystems have a wide distribution of fungus-root associations, and almost 80-90% of terrestrial plant species are associated with mycorrhizal fungi.

Degraded lands harbor low arbuscular mycorrhizal fungi (AMF) diversity and abundance (Asmelash *et al.*, 2016). Many studies have found that disturbance of the soil conditions due to extreme land use measures decreased AMF spores' density and root colonization (Oehl *et al.*, 2003; Azevedo *et al.*, 2019; Ríos-Ruiz *et al.*, 2019; Prayudyarningsih *et al.*, 2021). Since the physicochemical characteristics of soils are attributed to the number of spore variability in soils, degraded soil quality exacerbates the variability and, to an extreme extent, decreases spore density. Compaction due to extensive grazing and human disturbances has been observed to decrease spore density and colonization in the soil of Northern Ethiopia (Birhane *et al.*, 2017). Since soil aggregation is sensitive to changes in management practices, excessive land use measures significantly decrease AMF diversity and spore production (Azevedo *et al.*, 2019). Some land use measures that threaten AMF include modern intensive farming practices (Oehl *et al.*, 2003). The declining soil fertility levels, the rapid decomposition of OM, the loss of topsoil, and the extreme compaction are among the soil physicochemical properties that endanger AMF activity and adaptation (Douds and Miller, 1999).

On the other hand, the acidic soil condition ($\text{pH} < 5$) and deficient P content in the soils have been found to stimulate AMF spore density and species occurrence (Ríos-Ruiz *et al.*, 2019). AMF thrives well in acidic soil conditions, and AMF activity promotes P solubilization; therefore, the low P levels in the soils enhance their activity and diversity (Chen *et al.*, 2007). Interestingly, degraded lands are associated with soil acidity and P deficiency. As such, AMFs are well adapted in these areas contributing to the adaptation of the natural vegetation despite extreme soil conditions. Chen *et al.* (2007) have stressed that this could be how vegetation survives even minute nutrient resources are available.

Only a few studies have been done concerning the interaction among vegetation cover, physical soil status, and the hydrological processes of soil erosion and runoff in the degraded lands of the Philippines. As such, this study aimed to determine soils' physical and chemical characteristics under vegetation patches of marginal upland in Central Leyte. The spore density of mycorrhizal fungi is also known to demonstrate the vegetation patches' association with indigenous AMF. This study followed the hypothesis that the vegetation patches are associated with indigenous AMF, and spore production and proliferation can detect its presence. Detection of spores will give a clue to the biological fertility of this ecosystem and may express factors of its increase. Furthermore, it can contribute to the scanty information about the vegetation patch's structure and function in the ecosystem, which can further explain the patch formation and development in the marginal upland ecosystem.

2. Methodology

2.1 Sampling Site

The study site was located in Sitio Batuan, Barangay Linao Inopacan, Leyte, Philippines (Figure 1). It is degraded or marginal upland under intensive cultivation and consists of specific patterns of vegetation distribution (Figure 2). Inopacan is a fifth district and the fourth income class municipality in the province of Leyte, which marks the boundary between the town of Hindang in the south, Baybay City in the north, and the Camotes Sea in the west. It has a total land area of 9,462 ha with a population of 21,389 as of 2020 (Philippine Statistics Authority, 2021). It comprises several mountains and hills with about 300 masl local relief. This area is known to have a wide range of

degraded or marginal uplands used for agricultural activities. This upland consists of unique vegetation distributed in the area either locally or in patches. Among these are ‘amorsecó’ (*Andropogon aciculatus*), cogon (*Imperata cylindrica*), melastoma (*Melastoma malabathricum*), ‘hagonoy’ (*Chromolaena odorata*), Carabao grass (*Paspalum conjugatum*), guava (*Psidium guajava*), and ‘talahib’ (*Saccharum spontaneum*). These vegetations are considered the bio-indicator of a degraded or marginal landscape and are vital in preserving the fragility of this ecosystem.

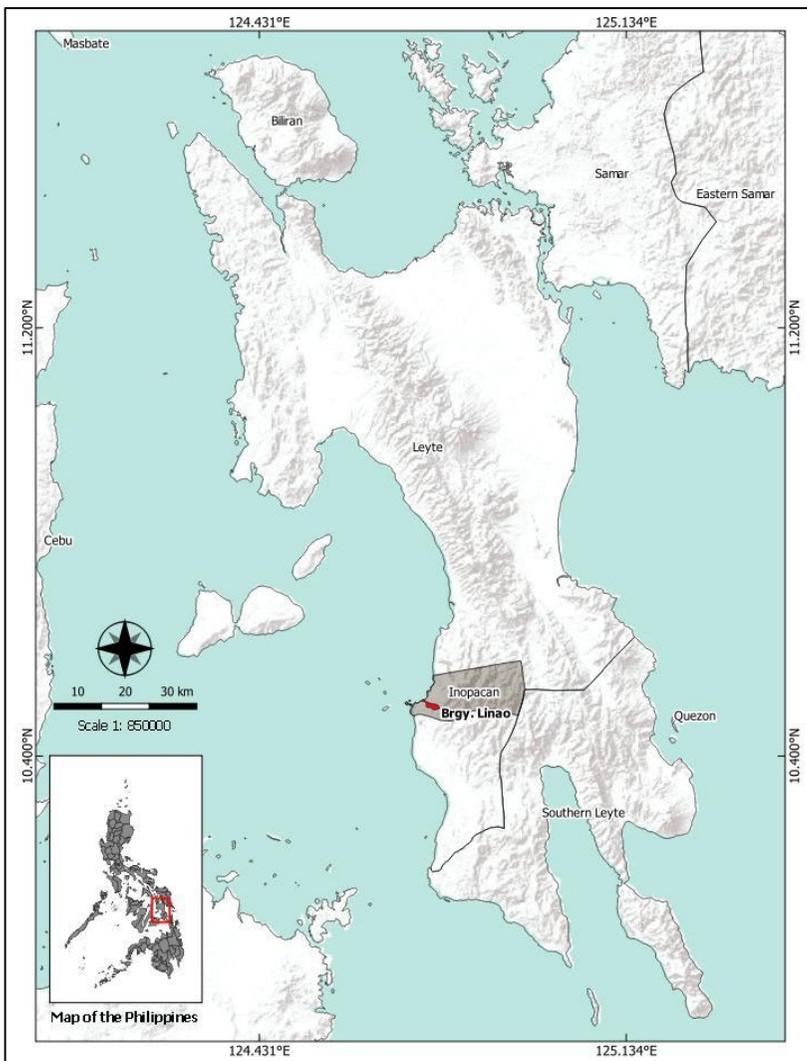


Figure 1. Map showing the study site in Inopacan, Leyte, Central Philippines

2.2 Selection of Vegetation Patches

Four vegetation patches were selected in the study site. These patches were situated in the landscape of *A. aciculatus* as it contains a mixture of vegetation such as *I. cylindrica*, *C. odorata* and *M. malabathricum*. The selected vegetation patch contained the following: patch 1 – mixed Vegetation; patch 2 – *M. malabathricum*; patch 3: – *I. cylindrica*; patch 4 – *C. odorata*; and control vegetation patch – *A. aciculatus*. These vegetation patches had a dimensional area ranging from 15 to 30 m². Three quadrats with a dimension of 1.5 x 1.5 m were measured per vegetation patch. Soil samples for analysis and spore density determination were obtained in these quadrats.

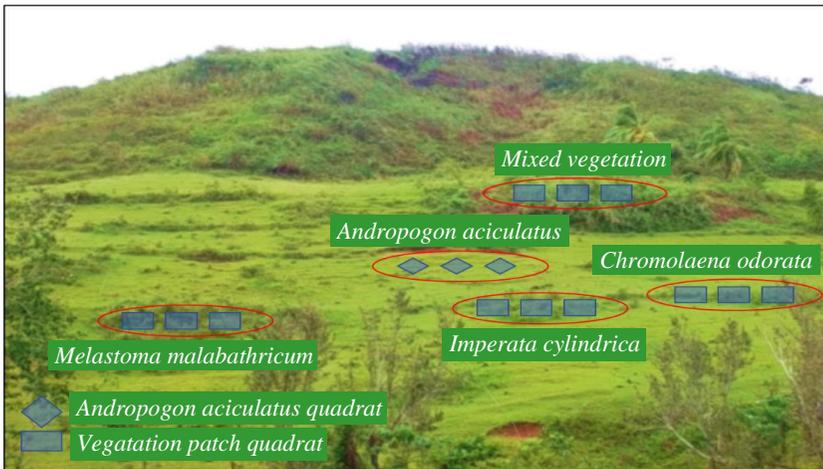


Figure 2. The sampling site showing the vegetation patches of a degraded upland in Central Philippines

2.3 Soil Sample Collection and Preparation

Soil samples were obtained from the soil surface to 20-cm depth in the quadrats of each vegetation patch. This was done by collecting 1 kg composite soil sample that came from four soil subsamples taken from every quadrat of each vegetation patch. As there are a total of five patches on the site, 15 composite samples were collected altogether. Immediately after sampling, the soil samples were placed in a properly labeled plastic bag and brought to the Department of Agronomy and Soil Science, Visayas State University (VSU), Visca, Baybay City, Leyte for processing and analyses.

Each soil sample was divided into two parts. The first part was refrigerated for special analyses requiring fresh samples such as water holding capacity while the second part was air-dried, pulverized using a wooden mallet, and sieved in a 2-mm wire mesh to get the fine earth for the determination of most soils' chemical and physical properties. For OM determination, enough soil samples were grounded and allowed to pass through a 0.425-mm wire mesh.

2.4 Soil Analyses

2.4.1 Particle Size Analysis

This was determined using the pipette method (International Soil Reference and Information Center [ISRIC], 2002). Twenty grams of fine earth soil was treated with sodium hypochlorite at pH 8 to destroy organic matter. It was heated gently until reaction subsided. Dispersion was done by shaking the samples for exactly 4 h and further dispersed using an ultrasonic disintegrator (UP100H, Hielscher Ultrasonics, Germany) for 3 min with the addition of 10-mL calgon solution (sodium hexametaphosphate). Percent silt and clay was determined by pipetting at specific time interval. The soil fractions such as sand (2-0.02 mm), silt (0.02-0.002 mm) and clay (< 0.002 mm) were separated. Percent sand, silt and clay was computed using standard formulas.

2.4.2 Bulk Density

The paraffin clod method by Blake and Hartge (1986) was used in determining bulk density (g cm^{-3}). Briefly, undisturbed air-dry soil clod, approximately 5.0 cm in diameter, was tied securely and weighed in air. Then the soil clod was dipped in melted (hot) paraffin. The paraffin-coated clod was weighed in air and then weighed in water. The moisture content of the clod was also determined. The bulk density was obtained using Equation 1.

$$BD (\text{g/cm}^3) = \frac{DW \times ODS}{SA - SPW + PA - (PA \times DW/DP)} \quad (1)$$

where BD is the bulk density; DW is density of water at a temperature of determination (g cm^{-3}); ODS is oven-dry weight of soil sample (g); SA is the net weight of soil sample in air (g); SPW is the net weight of soil sample plus paraffin in water (g); PA is the weight of paraffin coating in air (g); and DP is the density of paraffin (g cm^{-3}).

2.4.3 Porosity

Porosity (%) was calculated from the bulk density value and a constant particle density of 2.65 g/cm³ using Equation 2.

$$f(\%) = 1 - \left(\frac{\rho_b}{\rho_s} \right) \times 100 \quad (2)$$

where f is porosity (%); ρ_b is the bulk density (g cm⁻³); and ρ_s is the particle density (g cm⁻³).

2.4.4 Water Holding Capacity

The water holding capacity (WHC) (%) was obtained using the method prescribed by Forster (1995). Twenty grams of field moist soil was used and placed in a funnel with ordinary filter paper. Afterwards, 100 g of distilled water was added and stand overnight. Pre-weighed beakers held the percolated water and was weighed after. Percent water holding capacity was calculated using Equation 3.

$$WHC(\%) = \left(\frac{W_i - W_p}{ODW} \right) \times 100 \quad (3)$$

where WHC (%) is the percent water holding capacity; W_i is the initial weight of the water; W_p is the weight of percolated water; and ODW is the oven-dry weight of the soil.

2.5 Soil pH

This was analyzed potentiometrically using a soil-water solution ratio of 1:2.5 (ISRIC, 2002). A 20-g fine earth soil was weighed in a plastic cup. After which, 50-mL distilled water was added, and the solution was stirred thoroughly to form a suspension. It was allowed to stand for 30 min and was stirred again before reading with a precalibrated pH meter (Orion Star A210, Thermo Fisher Scientific, United States).

2.6 Soil OM

The Modified Walkley-Black method by Nelson and Sommers (1982) was used to determine the percent soil OM (%). Briefly, 0.5-g soil that passed through a 0.425-mm sieve (no. 40) was placed in a 500-mL Erlenmeyer flask. Using a volumetric pipette, the soil was added with 10-mL 1N K₂Cr₂O₇ and

swirled gently to disperse the solution. Under the fumehood, 10 mL of concentrated H₂SO₄ was added rapidly and the flask was swirled immediately until the soil and reagents were mixed. The mixture was allowed to stand under the fume hood for 1 h before adding 200-mL distilled water. Four drops of O-phenanthroline indicator were added into the solution, stirred using a magnetic stirrer and titrated with 0.5 N FeSO₄.7H₂O until the solution turned into a greenish cast to dark green as endpoint. The organic carbon and organic matter were calculated using Equations 4 and 5, respectively.

$$OC (\%) = N_{FeSO_4.7H_2O} \times \left(\frac{B-S}{w} \right) \times 0.39 \times mcf \quad (4)$$

$$OM (\%) = OC (\%) \times 1.724 \quad (5)$$

where *OC (%)* is percent organic carbon; *N* is the normality of FeSO₄.7H₂O; *B* is the volume (mL) of ferrous sulfate used in blank; *S* is the volume (mL) of ferrous sulfate used in sample; *w* is the weight (g) of soil used; *mcf* is the moisture correction factor; *OM (%)* is the percent OM.

2.7 Available Phosphorus

Available P (mg/kg) was analyzed according to the Bray method (Jackson, 1958). Exactly 2.5 g of fine earth soil sample was weighed, added with 25-mL extracting solution (0.1 N HCl and 0.03 N NH₄F), and shaken for 5 min using a reciprocating shaker at 180 oscillations per min. Filtrate was collected by filtering the solution through Whatman #42 filter paper. Two milliliter aliquot of the filtrate was added with 10-mL Reagent C (mixture of ascorbic acid and ammonium molybdate), mixed through the vortex mixer (EW-04726-01, Cole Parmer, United States) and allowed to stand for 1 h for the blue color development (Murphy and Riley, 1962). Absorbance was read using a spectrophotometer (Spectronic 20D⁺, Cole Parmer, United States) at 880-nm wavelength. Extractable P was calculated using Equations 6 and 7.

$$ppm P \text{ in solution} = Ods \times K \quad (6)$$

$$ppm P \text{ in soil} = ppm P \text{ in solution} \times (25/2.5) \times dilution \quad (7)$$

where *Ods* is the optical density of samples; *K* is the slope of the standard curve (average *K*); 25 is the volume (mL) of extracting solution; and 2.5 is the weight of soil used.

2.8 Exchangeable Potassium

Exchangeable potassium (K) (mg/kg) were extracted using 1N NH₄OAc (pH 7.0) method (ISRIC, 2002). Exactly 2.5-g soil was weighed in a 125-mL Erlenmeyer flask, and added with 25-mL 1N NH₄OAc (pH 7.0). The flask was covered with parafilm and shaken for 5 min in a reciprocating shaker. Solutions were filtered with Whatman #42 in a 50-mL beaker. The extracts were brought to the Central Analytical Services Laboratory, PhilRootcrops, VSU, Visca, Baybay City for the quantification of exchangeable Ca, Mg, K and Na with the use of an Atomic Absorption Spectrophotometer (Spectra AA 220 FS, Varian, United States).

2.9 Spore Density Determination

One hundred gram of soil sample (2 mm) was placed into 2-L capacity beaker. The soil was suspended with about 1 L of tap water and was vigorously stirred using plastic spoon for 1.5 min. The suspension was allowed to settle for 30 s. After settling down of soil particles, the upper layer of soil suspension was poured into a stack of sieves (2, 1, 0.5 and 0.425 mm); the finest sieve being at the bottom of the stack. The sieving and decanting procedure was repeated seven to eight times using the same soil or until the suspension was clear. The materials remain in the sieve (1, 0.5 and 0.425 mm) was suspended and transferred to centrifuge tubes and was centrifuged for 5 min at 3,000 rpm. Spores remain at the bottom of the tube while organic materials remained in suspension. After removing the supernatant, the sediment was re-suspended in 50% sucrose solution and was centrifuged again for 3 min at 2,000 rpm. After this, the spores were in the supernatant or in the sugar-water interface. The supernatant fluid containing the spores was poured into a 28 µm sieve and was poured with a small amount of tap water gently because too much exposure of spores to high concentrations of sugar can dehydrate them. The number of spores in a suspension was determined under a microscope (MSU-200, Cole Parmer, United States) by transferring small amount of the suspension into a small plastic petri dish with gridlines to facilitate easy counting.

2.10 Statistical Analyses

Correlation analyses were performed to evaluate the relationships between different soil properties and spore numbers. Analysis of variance (ANOVA) and pair tests at 0.05 level of significance were also carried out. All statistical

analyses were performed using Statistical Tool for Agricultural Research (STAR) 2.0.1 (International Rice Research Institute, 2016).

3. Results and Discussion

3.1 Physical Characteristics of Different Vegetation Patch

3.1.1 Particle Size Distribution

The particle size distribution of the sand, silt, and clay determines the texture of the soil. The texture represents the stable soil property and influences many physical and chemical processes in the soil such as the ease of tillage operation, the amount of air and water the soil can hold, and the rate of water movement (Hillel, 2004). In this study, the soil of the different vegetation patches was clay soil texture (Table 1). This is similar to the report of Asio *et al.* (2014) stating that the soil texture of the soil profiles evaluated at the different topographic positions of the same area had silty clay to clay soil texture.

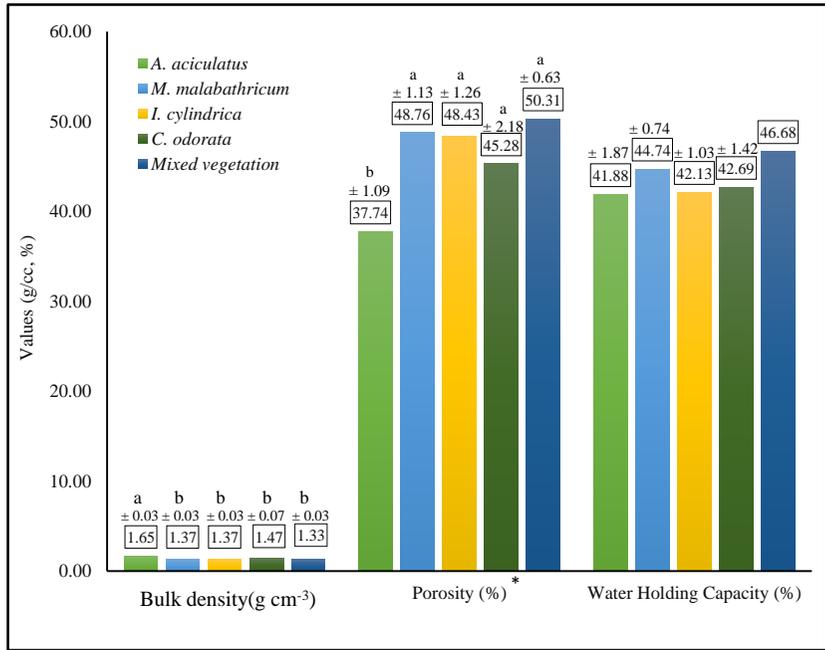
Table 1. Particle size distribution of soils in the different vegetation patches of a degraded upland in Central Philippines

Vegetation path	Particle size distribution (%)			Soil textural class
	Sand	Silt	Clay	
<i>A. aciculatus</i>	28	30	42	Clay
<i>M. malabathricum</i>	30	25	45	Clay
<i>I. cylindrica</i>	28	29	43	Clay
<i>C. odorata</i>	29	29	42	Clay
Mixed vegetation	34	22	45	Clay

3.1.2 Bulk Density

Bulk density (Db) refers to the mass (net) of soil per unit bulk volume of undisturbed soil expressed in g cm^{-3} (Tan, 2005). It is used as an index of compaction and porosity as it directly influences root development and air diffusion in the soil environment (Hillel, 2004). According to Hossain *et al.* (2015), most mineral soils have Db ranging from 1.0 to 2.0 g cm^{-3} . In this study, the bulk density of different patches ranged from 1.33 to 1.65 g cm^{-3} (Table 2), wherein *A. aciculatus* had significantly higher ($p < 0.05$) Db values (1.65 g cm^{-3}) in comparison with the other vegetation patches, which had

comparable Db values ($p > 0.05$) with one another, as mixed vegetation with the lowest Db values (1.33 g cm^{-3}) (Figure 3).



± values refer to standard error; * significantly different at < 0.05 ; means overhead with the same letter are not significantly different at 0.05; coefficient of variation (%): bulk density (11.69); porosity (13.87); WHC (19.80).

Figure 3. Physical properties of soils in the different vegetation patches of a degraded upland in Central Philippines

To make use of Db values, Canarache (1991) devised the packing density (Pd) index to determine the compactability of the soil using the Db values and the % clay of the soil (Table 1), categorizing the Db index into three classes ($< 1.45 \text{ g cm}^{-3}$ no compaction; 1.45-1.75: moderate compaction; and > 1.75 : extreme compaction). Among the vegetation patches, only the mixed vegetation had moderate compactability, while the remaining patches had extreme compaction. These values suggest the moderate to extreme compaction of the soils in the studied area. Asio *et al.* (2015) and Sabijon and Asio (2022) reported comparable ranges of Db values (1.35 to 1.66 g cm^{-3} and 1.30 to 1.50 g cm^{-3} , respectively) of selected degraded soils in Samar Island. Soil compaction may not be well studied in the Philippine soils (Asio *et al.*, 2009); these values indicate pressing concerns since, at this condition, it will favor surface run-off, exacerbating soil erosion that removes fertile topsoil and decreasing the soil fertility and productivity (Lal, 1990) eventually. Meanwhile, the lower Db values of other vegetation patches ($p > 0.05$) were

worth noting compared with the *A. aciculatus* patches. This result can be attributed to the amount of OM (Figure 3) introduced through leaf litter decomposition of the vegetation in the patches. As it decomposes, OM releases organic compounds that glue or bind soil particles improving aggregation and decreasing compaction.

Table 2. Packing density index and compactability of soils in the different vegetation patches of a degraded upland in Central Philippines.

Vegetation Path	D _b	% Clay	P _d index	Compactability
<i>A. aciculatus</i>	1.65	42	2.03	Extremely compact
<i>M. malabathricum</i>	1.37	45	1.78	Extremely compact
<i>I. cylindrica</i>	1.37	43	1.76	Extremely compact
<i>C. odorata</i>	1.47	42	1.85	Extremely compact
Mixed vegetation	1.33	45	1.74	Moderately compact

3.1.3 Porosity

Porosity is an index of the relative pore volume in the soil (Weil and Brady, 2017). Hillel (2004) explains that it is the overall amount of pore spaces or that part of the soil volume occupied by air and water. The porosity of the degraded soils in the different vegetation patches ranged from 37.74 to 50.31%, wherein *A. aciculatus* had significantly lower ($p < 0.05$) porosity values (37.74%) in comparison with the other remaining vegetation patches. Similar to the trend of Db, the mixed vegetation had the highest porosity (50.31%) but was comparable ($p > 0.05$) with the porosity of the vegetation patches dominated by *I. cylindrica*, *M. malabathricum* and *C. odorata*. As Db values reflect compaction, porosity indicates the number of pore spaces; hence, as bulk density increases, porosity decreases, and vice versa. In this study, although the vegetation patches dominated other than *A. aciculatus* showing higher porosity values, these values range (37.74-50.31%) still indicated poor porosity. Asio *et al.* (2015) have reported a porosity value of 37.32 to 52.83% of degraded soil in Samar Island. They attributed their findings to soil disturbance due to farm operation and increased OM decomposition. Although currently abandoned for crop production activities, soil disturbance was observed in the studied area since it served as the grazing site for farm animals such as carabao, cows and goats.

3.1.4 WHC

The ability of soils to hold a given volume of water is referred to as WHC. It has a direct relationship with porosity and an indirect relationship with bulk density. Soil texture has a significant impact since the pore volume takes time to fill and store a specific amount of water. This study observed no clear trend ($p > 0.05$) in WHC values between the different vegetation patches (Figure 3). Although WHC ranged from 41.88 to 46.68%, and within the WHC range of clayey (Moody and Cong, 2008) and degraded soils (Asio *et al.*, 2015; Sabijon and Asio, 2022), results corroborated the observation of Asio *et al.* (2015) as WHC being not a desirable indicator of soil degradation due to its dependence to soil texture and a stable soil property.

3.2 Soil Chemical Properties of Different Vegetation Patches

3.2.1 Soil pH

Soil reaction (pH) expresses the activity of the hydrogen ions in the soil solution. It affects the availability of mineral nutrients in plants and many soil physicochemical processes (Bohn *et al.*, 2001). The pH of the soils in the studies degraded upland ranged from 5.13 (*A. aciculatus*) to 5.69 (*I. cylindrica*) (Figure 4). However, the pH of the soils from the patch of *A. aciculatus* was significantly lower ($p < 0.005$) than the other vegetation patches; the pH of the soils in the *A. aciculatus* patch and other vegetation patches corresponded to the pH range of a strong acidic condition (5.0-5.5) (Landon, 1991); only the pH of the soils from patch dominated with *I. cylindrica* (5.69) had moderate acidity (5.5-6). Nevertheless, the pH values in this study corresponded to acidic soil conditions regardless of the vegetation patches. This pH range was comparable to the observation of Asio *et al.* (2014), who discovered that the soils in the same area had a pH near 5. Navarrete *et al.* (2013) also reported pH levels of 5.12 and 5.48 from the studied degraded soils in Ormoc and Baybay, Leyte, respectively. Calubaquib *et al.* (2016) and Sabijon and Asio (2022) reported a pH range of 4.5 to 5.4 and 5.0 to 7.0 in the studied degraded soils in Luzon and Samar, respectively. The moderate to strong acidity condition means that this degraded upland had soil fertility problems regarding nutrient availability, micronutrient toxicity and microbial activity (Bohn *et al.*, 2001).

3.2.2 OM Content

Organic matter refers to all decomposed, partly decomposed and undecomposed materials of plant and animal origin (Bohn *et al.*, 2001). It is

the critical component of soil to carry out necessary environmental functions by supplying plant nutrients, facilitating soil aggregation, and driving microbial activities.

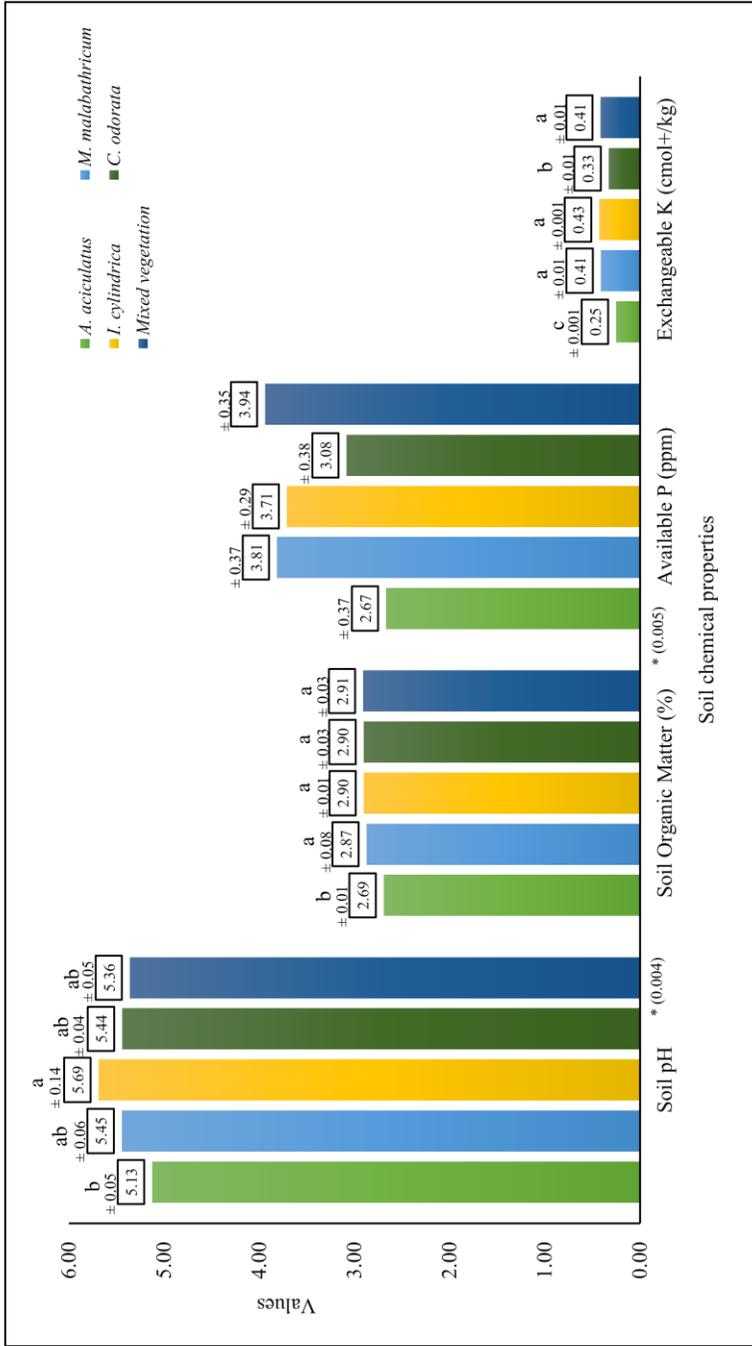
The OM content of the soils in the different vegetation patches ranged from 2.69 to 2.91%. Based on the Hazelton and Murphy (2016) proposed rating levels for soil carbon/organic matter to assess soil health or soil condition, these OM value range corresponded to moderate (M3), band 7 level suggesting the improving soil health/condition manifested with structural stability, pH buffering capacity, soil nutrient levels and WHC. Asio *et al.* (2014) reported a high OM content in the same studied soil reaching nearly 4%. While the area is characterized as degraded (Asio *et al.*, 2014), the OM values were high. However, soil from *A. aciculatus* patch has significantly lower ($p < 0.05$) OM content compared to the soils from the remaining vegetation patches, which have similar OM content.

Coyne and Thompson (2006) suggested that plant litter is a vital source of organic matter in the soil. Leaf litter decomposition is the primary process that releases organic matter into the soil environment (Brady, 1990). The soils from different vegetation patches received higher leaf litter biomass than soils from *A. aciculatus* patch attributing the 7% higher organic matter content ($p < 0.005$) of the soil from the different vegetation patches than the soils from the *A. aciculatus*. Furthermore, soil erosion was active in the *A. aciculatus* patch leading to the loss of topsoil rich in organic matter. Erosion rates of the same site were reported by Villamayor *et al.* (2017) at 64.68 to 98.81 t ha⁻¹ year⁻¹ to the areas of no crop, while with natural vegetation strips, erosion rates of 52.60 to 61.34 t ha⁻¹ year⁻¹ were accounted. The lower erosion rates in areas of no crops with natural vegetation strips are due to canopy cover, preventing soil erosion and removal of OM-rich topsoil.

3.2.3 Available P

Phosphorus is a critical element in natural and agricultural ecosystems (Bohn *et al.*, 2001). Tropical soils are generally phosphorus deficient, and in many cases, it is the limiting nutrient in agriculture (Sanchez, 2019). Meanwhile, available P refers to the P available for plant utilization.

Available P of the soils from the vegetation patches were not significantly different ($p > 0.05$) as P content ranged from 2.67 to 3.94 ppm. According to Landon (1991), these P values corresponded to low/deficient (< 5 ppm).



± values refer to standard error; * significantly different at < 0.05; means overhead with the same letter are not significantly different at 0.05; coefficient of variation (%): Soil pH (4.22); SOM (2.40); available P (18.94); and exchangeable K (3.68).

Figure 4. Chemical properties of soils in the different vegetation patches of a degraded upland in Central Philippines

Asio *et al.* (2014) reported comparable P content (< 10 ppm, low or deficient) of the soils in the same area. Accordingly, these values were expected due to moderate to extreme acidity and the degraded nature of the area, leading to P fixation; hence, the low P content detected among the different vegetation patches.

3.2.4 Exchangeable Potassium

Exchangeable K is one of the interchangeable bases in the soil aside from exchangeable Ca, Mg and Na. Sposito (2008) stresses that these bases regulate the soil pH and thus are an essential factor in soil fertility. Exchangeable bases reflect the contribution of parent material and soil management practices' influence on the soil environment.

Exchangeable K of the soils from the different patches ranged from 0.25 to 0.43 cmol+ kg⁻¹. Similar to the result of Asio *et al.* (2014) (0.2 to 1.2 cmol+ kg⁻¹), exchangeable K values were above the critical levels (> 0.20 cmol+ kg⁻¹) as reported by Landon (1991). Asio *et al.* (2014) mentioned that the above critical value of the exchangeable bases such as K is attributed to the abundance of rock fragments of basalt and andesite in the area. The exchangeable bases are released during the weathering of these rocks. Meanwhile, exchangeable K from the soils in the patches dominated with mixed vegetation (*M. malabathricum* and *I. cylindrica*) was significantly higher ($p < 0.05$) compared with the patch dominated with *C. odorata*. In contrast, the patch dominated by *A. aciculatus* had the lowest exchangeable K content. Strawn *et al.* (2020) explained organic matter and parent material as a good source of exchangeable bases in the soil environment. However, these are prone to massive losses in the tropical climate (Nelson, 1994) and a significant fertility problem in crop production (Brady, 1990) and the marginal upland ecosystem (Briones, 2009). These features explain the variation of exchangeable K in the vegetation patch and the surrounding *A. aciculatus*. Similar to the OM content, soils from *A. aciculatus* were subjected to soil erosion, leading to the removal of nutrient-rich topsoil, whereas soils dominated with the other vegetation were protected with canopy cover; hence, the higher K content.

3.3 Spore Density of Indigenous AMF

Spore density is the ratio of the number of spores in a given weight of soil. It is an indicator of AMF presence in a particular environment. It serves as the

fungus's vital reproductive structure and a precious organ that holds the genetic material making it possible to store life and sustain fungus reproduction, especially in a disturbed ecosystem.

Zhao *et al.* (2001) underscored the importance of spore density assessment and its contribution to understanding ecosystem structure and function, diversity protection, and ecosystem sustainability. According to their study, many factors could influence spore proliferation in a given host rhizosphere and values for AMF spore density associated with different plants and sites are also variable. They added that climo-edaphic factors, host dependence, the age of the host plants, the sporulation abilities of AMF, and the dormancy and distribution patterns of AMF spores in the soils might be significant influences on the spore proliferation in a given soil ecosystem. In this study, the spore density of the soils in the different vegetation patches ranged from 4 to 11.33 spores/100 g soil, being high in the *C. odorata* patch and low in the patch dominated by *A. aciculatus* (Figure 5). The significant variability ($p < 0.05$) and the spore density among the different vegetation patches were worth noting. Spore density of soils dominated with *C. odorata* had the highest spore density (11.33 spores/100 g soil), comparable with the spore density of soils in the vegetation patch dominated with mixed vegetation (9 spores/100 g soil) and *M. malabathricum* (8.33 spores/100 g soil); *I. cylindrica* patch had comparable spore density (7.33 spores/100 g soil) with mixed vegetation and *M. malabathricum*, while *A. aciculatus* had the lowest spore density (4 spores/100 g soil) than the spore density of *I. cylindrica*. The differences in the spore density can be attributed to the uneven spatial distribution (clumped distribution) of AMF spores and the complex structure of the degraded soil environment (Zhao *et al.*, 2001). This also suggests that AMF spores did not coincide with vegetation distribution's zonation (patch) pattern. These differences may be related to the different behavior of AMF spore production in each vegetation since some species of AMF need longer to germinate. In contrast, others had no capability of germination. Other authors (Oehl *et al.*, 2003; Azevedo *et al.*, 2019; Ríos-Ruiz *et al.*, 2019) found identification of AMF species imperative as they have different behavior in spore production.

Variation in the number of spores is affected by the physical and chemical characteristics of the soil (García-González *et al.*, 2016). In this study, the soils of the different vegetation patches showed variability in terms of bulk density, porosity, WHC, soil pH, OM, and nutrient content such as P and K. These physicochemical characteristics, therefore, are factors that could be affecting the variation in the number of spores of the soils from the different vegetation patches. However, the response was so variable that no clear effect trend was

distinct. Table 3 indicates the relationship between spore density and the soil's physicochemical characteristics in the chosen vegetation patches among the parameters; only silt and bulk density resulted from a negative correlation, while others are positively correlated to the number of spores in a given vegetation patch.

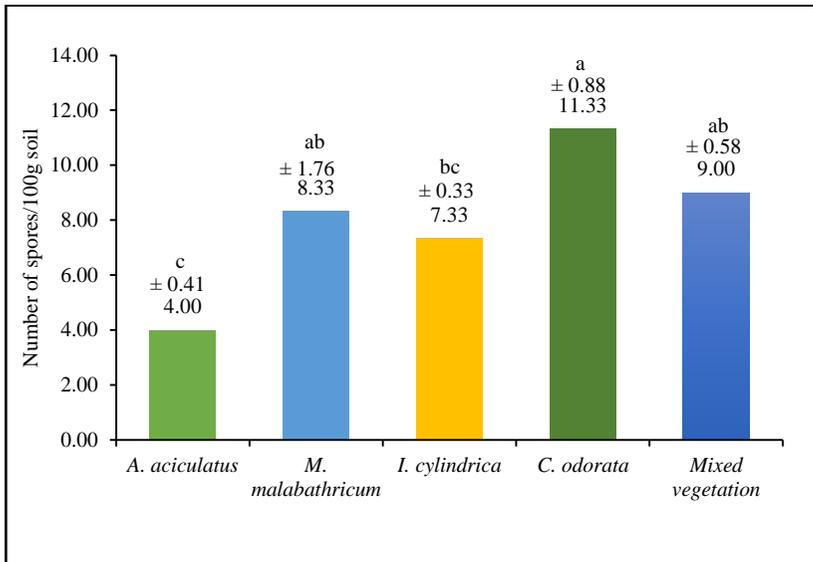
On the other hand, soil pH, organic matter, available P, and exchangeable K affect AMF metabolism and activity. The acidic soil condition ($\text{pH} < 5$) and deficient P content in the soils have been found to stimulate AMF spore density and species occurrence (Ríos-Ruiz *et al.*, 2019). Although the spore density detected was low, this could also explain the detection of AMF spores in the soils of the different vegetation patches. According to Chen *et al.* (2005) AMF thrives well in acidic soil conditions, and AMF activity promotes P solubilization. Therefore, the low levels of P in the soils enhance their activity and diversity. Moreover, AMF promotes phosphate uptake under deficient soils by elongating external hyphae around the roots' phosphate depletion zone. This behavior may have helped vegetation in the degraded uplands to survive and adapt despite the adverse edaphic conditions due to soil degradation.

Meanwhile, regardless of the vegetation patches, the detection of the mycorrhizal spores in the soils of the different patches indicated the presence of mycorrhizal association with the natural vegetation in this degraded upland. Although the spores detected were low, some works have reported comparable spore density ranges with this study. Azevedo *et al.* (2019) found an average of 8-12 spores of AMF per 100 g of soils under different farm management practices in Brazil. Oehl *et al.* (2003) detected even < 5 spores per 100 g in soils under different land use intensity in Central Europe; and Prayudyaningsih *et al.* (2021) discovered < 50 spores per 100 g in disturbed soil conditions such as the landslide-impacted area in Indonesia. However, Ríos-Ruiz *et al.* (2019) revealed > 100 spores per 100 g in degraded soils in Peru. Zhao *et al.* (2001) and Khakpour and Khara (2012) also found that spore density ranged from 55 to 1,908 spores per 100 g of soil in tropical rainforest and 13-30 spores per 100 g in northern Iran soil, respectively. The low spore detected was regarded as likely due to the degraded nature of the soils in the studied area. Welemariam *et al.* (2018) stressed that degraded lands harbor low levels of AMF abundance and diversity. They added that even livestock and human disturbances (present in the studied area) decreased the AMF spore density including root colonization and nutrient availability. Livestock and human disturbance disturb the natural soil conditions affecting the abundance and richness of AMF (Guadarrama and Alvarez-Sanchez, 1999).

Table 3. Pairwise correlation coefficients of the physicochemical properties and spore density of the soils in vegetation patches of degraded upland in the Central Philippines

Variables	Spore density	Sand	Silt	Clay	Bulk density g/cm ³	Porosity (%)	Water holding capacity (%)	Soil pH	Soil organic matter (%)	Available P (ppm)	Exchangeable K (cmol _c /kg)
Spore Density	1										
Sand	0.3677	1									
Silt	-0.351109	-0.9490**	1								
Clay	0.2475	0.7658**	-0.9281**	1							
Bulk Density (g/cm ³)	-0.4245	-0.4788	0.5950*	-0.6584*	1						
Porosity (%)	0.5138*	0.5662*	-0.6728*	0.7084*	-0.9671**	1					
Water holding capacity (%)	0.2949	0.6213*	-0.6407*	0.5760*	-0.1870	0.2207	1				
Soil pH	0.4423	-0.0247	-0.0877	0.2134	-0.6697	0.6245	0.0080	1			
Soil organic matter (%)	0.6023*	0.4007	-0.4444	0.4264	-0.6719*	0.6943*	0.2863	0.5106*	1		
Available P (ppm)	0.1831	0.4377	-0.5421*	0.6009*	-0.6190*	0.5816*	0.3708	0.5006*	0.6609*	1	
Exchangeable K (cmol _c /kg)	0.4686	0.4644	-0.6243*	0.7405*	-0.9115**	0.8951**	0.3433	0.7060**	0.7200*	0.6833*	1

* Significant at 0.05; ** highly significant, ^{ns} not significant



± values refer to standard error and mean overhead with the same letters are not significantly different at 5%.

Figure 5. AMF spore density of soils under different vegetation patches

4. Conclusion and Recommendation

The patch of *C. odorata*, *I. cylindrica*, *M. malabathricum* and mixed vegetation contained spores higher than the surrounding *A. aciculatus* landscape. The spore numbers of the marginal soil in the upland of Inopacan, Leyte were affected by the soil's physicochemical characteristics, particularly the high bulk density, low porosity, moderate to strong acidity and low P and K levels.

The nature of the research requires various soil parameters to quantify and analyze the association of AMF to the vegetation patches. Hence, further analyses of soil's physical, chemical and biological properties are required to understand the behavior of these vegetation patches and the overall landscape. More parameters, such as AMF genetic and functional diversity, should be included to thoroughly understand the patch occurrence, formation and development, which can help understand the marginal upland ecosystem.

The results of this study can raise questions vital to our comprehensive understanding of the role of mycorrhiza in the ecology of marginal uplands of

the Philippines. Answering these questions needs other intensive investigations, such as determining the adaptive AMF species and harnessing their potent potential to aid the management of degraded lands.

5. Acknowledgement

The author acknowledges the guidance of Dr. Victor B. Asio in exploring the dynamics of degraded uplands; the help of Dr. Angela A. Ferraren in the microbiological perspectives of the study; the soil analysis made by Ms. Cynthia Dolores V. Godoy; and the permission of the inhabitants in the research area given to the author to conduct this study. The author also recognizes the Accelerated Science and Technology Human Resource Development Program – National Science Consortium of the Department of Science and Technology for the funding.

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