Study on Arbuscular Mycorrhizal Fungi Related to Soil P, Soil OM and Soil pH in cassava field from Thailand

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Abstract Two-hundred and ninety-five soil samples were collected from cassava field in Thailand to study distribution of arbuscular mycorrhizal (AM) fungi under root zone of cassava. The AM fungal spores were counted for spore number 100g^{-1} soil, morphotyped classified and AM fungal colonization determined after pot culture. Soil P, soil OM and soil pH were analyzed to calculate the correlation between each parameter and AMF spore number. The result showed that cassava has been planted in wide soil pH in Thailand. They could be grown in acidic soil to alkaline soil (pH 3.0-8.5). Soil P in cassava field was 0.05- 91.91 mg kg^{-1}. Soil OM was 0.22-4.49%. AMF spore number under cassava field was 25-1,880 spores 100g^{-1} soil classified as eighteen AMF species. *Glomus* sp. and *Acaulospora* sp. were always found under cassava field. This research showed no relationship between Soil OM and AMF spore number and no relationship between Soil pH and AMF spore number. However, we found negative relationship between Soil P and AMF spore number (AMF spore number = 426.11– 4.12Soil P, r^2 = 0.075**).

Keywords: AMF, Cassava, Soil P, Organic matter, pH

Introduction

In Thailand, cassava (*Manihot esculenta* Crantz) is considered one of the most important economic crops with annual production of around 25 million tons (Piyachomkwan and Tanticharoen, 2011). Arbuscular mycorrhizal (AM) fungi promote many plants in particular improved nutrition, better growth, stress tolerance, and disease resistance. In addition, the hyphal networks of AM fungi improve soil characters such as soilparticle aggregation, improving the resistance of soil erosion by wind and water. AM fungi are also decrease nutrient leaching from the soil, thereby contributing to the retention of nutrients in the soil and decreasing the risks of contamination of ground water. (Chen *et al.,* 2018). AMF diversity in Thailand has been studied and reported in different plant community, In the forest restoration area of Doi Suthep-Pui National Park, northern Thailand, twenty-one AM species, were founded and identified into Acaulospora (6 species), Glomus (12 species) and Scutellospora (3 species). AM fungi belonging to the genera Glomus and Acaulospora were dominant. Abundant species present were *Acaulospora elegans*, *Glomus multicaule* and *Scutellospora pellucida* (Nandakwang *et al.,* 2008). The composition of AMF communities associated with rubber tree roots in Northeast Thailand differed between cassava and rubber tree plantations and was influenced by soil texture and nutrient content (sand, K, P, Ca). AMF community composition gradually shifted with the age of the trees (Herrmann *et al.,* 2016). Twenty-two species of AM fungi were found associated with tangerine in orchads of Chiang Mai province. AMF colonization and spore density in rhizoshere significantly depressed when available P more than 500 mg P kg^{-1} soil. (Youpensuk *et al.,* 2008). Sarr *et al.,* (2019) studied the composition of AMF community in cassava field soils in two agro-ecologies of Cameroon using Illumina MiSeq of the ITS2 region. Fifteen AMF species were identified from the 27 OTU, and they were dominated by *Glomus sinuosum* and *Paraglomus occultum* in both sites. However, AMF species in cassava field of Thailand have been never reported. The research has focused on AMF spore number founded in the cassava field related to soil chemical properties to study possibility of the native AMF improving cassava cropping.

Materials and methods

Soil sampling

Two-hundreds and ninety-five soil samples were collected from cassava field across all region of Thailand during the year of 2007. One kilograms of soil was collected in root zone of cassava plant, air dried, grinded through 2 mm sieve.

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AMF spore density and classification

AM spores were extracted from 100 g air-dried soil samples by wet sieving and decanting method (Gerdeman and Nicolson, 1963) followed by 40%sucrose certification (Danial and Skipper, 1982). Spores were recovered by filtering through a 250 and 45#m sieve. The AM spores was counted and identified according to morphological characteristics by INVAM species guide and manual for identification of AMF (Schenck and Perez, 1988). The frequency of each AMF spore characteristic was calculated by the percentage of the number of the samples in which the genus was observed. Each AMF spore characteristic was studied AMF colonization in cassava root.

Analysis of soil chemical properties

Soil pH was determined using a pH meter. Available phosphorus in soil (soil P) was determined by Bray II method (Bray and Kurtz, 1945) and Ammonium molybdate-Ascorbic acid method (Watanabe and Olsen, 1965). And soil organic matter (soil OM) was determined by Walkley and Black Method (Walkley and Black, 1934).

Statistical analysis

Relationship between AMF spore number and chemical properties computed using Pearson's correlation coefficient and simple regression was also calculated.

Results

Cassava plant could be grown in various soil chemical properties in all region of Thailand except southern. They could be grown in very strongly acid to moderately alkaline soil (pH = 3.0-8.5). The soil from cassava field contained 0.05-91.91 mg kg^{-1} of available P and 0.22-4.49% of soil OM. The spore number of AMF in cassava field was about 25-1,880 spores 100 g^{-1} soil (Table 1).

Eighteen AMF species was found and classified into Gigaspora, Scutellospora, Glomus, Acaulospora, Entrophospora and Glomus genera, Acaulospora genera. *Glomus* sp. and *Acaulospora* sp. were high frequency found under cassava field. After pot culture study, fifteen AMF species showed colonization in cassava root (Table 2).

The result of this research found negative relation between AMF spore number (y) and soil P (x) as equetion y = 426.11-4.1211x (r^2 = 0.0748**, n = 295). While AMF spore number in cassava field was not related to soil OM ($r^2 = 0.003^{ns}$) and soil pH ($r^2 = 0.001^{ns}$). However, when soil OM increased trended to increase AMF spore number whereas soil pH increased trended to decrease AMF spore number (Figure 1).

Table 1. Range and average for soil chemical properties, AMF spore number in soil from cassava field of Thailand.

Region of Thailand	No. of sample	Soil pH	Soil P (mg g^{-1})	Soil OM (%)	AMF spore number (spore 100 g^{-1} soil)
1. Northeastern	153	4.5-8.0 (6.1)	0.16-81.57(21.9)	0.22-4.49 (1.3)	60-1,880 (334.5)
2. Central	89	3.0-8.5 (6.6)	0.05-91.91(24.2)	0.43-4.23 (1.8)	40-1,220 (315.9)
3. Northern	53	4.8-7.5 (6.7)	0.84-91.91(24.2)	0.43-3.37 (1.6)	25-1,320 (359.9)
Total	**295**	**3.0-8.5 (6.4)**	**0.05-91.91 (22.5)**	**0.22-4.49 (1.5)**	**25-1,880 (333.4)**

Table 2. Characteristic of AMF spore, classification, frequency and colonization of AMF founded in root zone of cassava.

	Characteristic of AMF spore founded in root zone of cassava	Classification by spore morphology	Frequency in field (%)	Colonization (%)
1	Globose shape with hypha, 150-200µm, shiny brown color.	*Glomus* sp.1	91	√
2	Globose, 80-100µm, white to cream color, single chlamydospore with hypha.	*Glomus* sp.2	98	√
3	Globose, 80-115 µm, white to cream color, single chlamydospore with hypha and content in spore present.	*Glomus* sp.3	92	√
4	Sporocarp formation without peridium, subglobosechlamydospore 60-90µm, grey color.	*Glomus* sp.4	48	√
5	Sporocarp formation with peridium, subglobosechlamydospore, dark brown color.	*Glomus* sp.5	15	×
6	Globose shape with hypha, 220-250µm, dark orange and grey color.	*Glomus* sp.6	78	×
7	Globose shape with hypha, 200-240µm, shiny white to green color.	*Glomus* sp.7	66	√
8	Subglobose, 80-90µm, pale yellow to green color, single chlamydospore with hypha.	*Glomus* sp.8	59	√
9	Globose shape, 75-130 µm, shiny yellow to green color to orange color.	*Acaulospora* sp.1	98	√
10	Globose shape, 70-110 µm, green to grey color.	*Acaulospora* sp.2	80	√
11	Globose shape, 90-120 µm, shiny white to orange color, content in spore present.	*Acaulospora* sp.3	99	√
12	Globose shape, 100-120 µm, shiny creamy to orange to grey color.	*Acaulospora* sp.4	87	√
13	Subglobose, 60-90µm, dark orange color, single chlamydospore.	*Acaulospora* sp.5	74	√
14	Globose to subglobose shape, 250-280µm, dark orange color.	*Acaulospora* sp.6	94	×
15	Globose shape with suspensor, 220-250µm, dark orange color.	*Gigaspora*sp.2	45	√
16	Globose shape with suspensor, 220-250µm, white to cream color.	*Scutellospora* sp.	33	√
17	Globose shape, 150-200 µm, orange to dark orange brown, and hyaline, subglobosesporiferoussuccule and thick spines present.	*Entrophospora* sp.1	41	×
18	Globose, 75-90µm, white color, single chlamydospore	*Entrophospora* sp.2	84	√

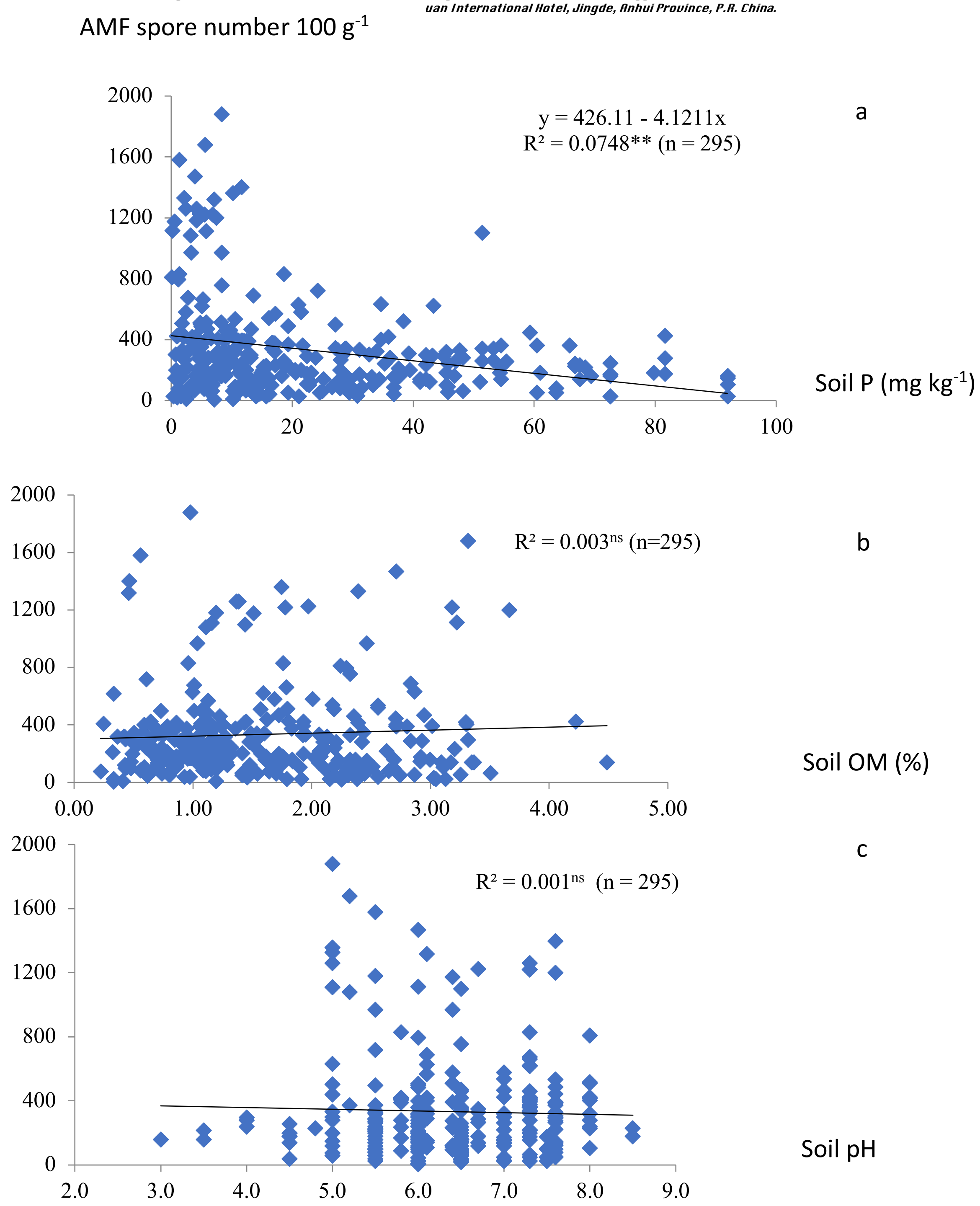

Figure 1. Relationship between AMF spore number 100 g^{-1} soil and soil P (a) between soil OM (b) and between soil pH (c)

Discussion

The result indicated that cassava plant could be grown in very strongly acid to moderately alkaline soil (pH = 3.0-8.5). The spore number of AMF in cassava field was about 25-1,880 spores 100 g^{-1} soil. The pH of the soil in cassava field with high AMF spore number ranged from 5 to 8. Isobe *et al*. (2007) found high spore density of AMF in the samples with soil pH ranged from 6 to 8, and soil samples with a pH lower than 6 and higher than 8, the spore density was lower than 5 spores g^{-1} DW. Thus, in the acid or alkaline soil, the sporogenesis of AM fungi is suppressed. Youpensuk *et al*. (2006) studied AMF colonization in upland rice and *Macaranga denticulate* in soil pH at 4.5, 5.9 and 7.8. They found highest AMF colonization percentage at 5.9 of soil pH. Althought AMF spore number in cassava field was not related to soil OM but AMF spore number trended to increase with increasing of soil OM supported by Vaidya *et al.,* (2007) found that organic matter addition can improve AMF biomass and spores as well as plant survival. The negative relation between AMF spore number and soil P have been found in this research. According to AM fungi colonization and spore number had negative effects by phosphorus fertilization (Jasper *et al*., 1979) (Bhadalung *et al*., 2005). Sarr *et al.,* (2019) also found agro-ecologies of Cameroon which had less available P showed a higher AMF diversity and contained 10 site-specific species, compared to agro-ecologies which more than available where only 2 site-specific species were found. Khakpour and Jalil. (2012) found relationship between soil physical and chemical factors and number of mycorrhizal spores, as well as between these factors and percentage root colonization. Furthermore, there was negative correlation between number of spores with phosphorus content and electric conductivity of soil samples and positive correlation between number of spores and root colonization. However, some research showed positive relation between numbers of AMF spore and available P in soil. Effendy and Wijayani (2011) found a linear corelation between numbers of AMF spore and available P in soil. The greater the number of AMF spore, the higher the available P in soil. Ong *et al.,* (2012) found soil P in forest rehabilitation was positively related to the spore count ($r > 0.68$, $P < 0.001$) while the most probable number (MPN) was negatively influenced by soil K ($r = -0.632$, $P < 0.01$). In this study confirmed that soil chemical properties, especially available soil P have a direct negative effect on the abundance of AM in the root zone of cassava. And AMF could produce spore in strongly acid condition to slightly alkaline condition and soil OM trended to increase AMF spore.

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