

ORIGINAL RESEARCH ARTICLE

Abundance and Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores Isolated from the Rhizosphere of papaya and other Different Cropping Systems in Central Kenya

¹Jacinta Muiruri, ¹Fredah K. Rimberia, ¹Mwajita R. Mwashasha and ¹Agnes Kavoo.

¹Department of Horticulture and Food Security, School of Agriculture and Environmental Sciences, Jomo Kenyatta University of Agriculture and Technology (JKUAT), Nairobi, Kenya

Corresponding author: cintawm@yahoo.com

ABSTRACT

Arbuscular mycorrhiza fungi (AMF), obligate symbionts, are important in the majority of cultivated plant species in colonizing roots and supporting plant growth in adverse climatic conditions. However, the abundance and quality of mycorrhizal colonization is affected by land-use types, cropping systems and climate change. On the other hand, rhizospheric mycorrhizae present in rhizospheric soils can be isolated for enhancing plant performance. One such opportunity arises in the acclimatization of seedlings for adaptation to depleted field conditions. Isolation and characterization of rhizospheric AMF species is important in evaluating the efficiency of colonization especially in plants that have not been previously evaluated such as papaya, which is well known for its high nutritive value. In this study, soils were sampled from grass, banana and papaya plants' rhizosphere, from three different papaya growing regions in Kenya; Mwea, Mitunguu and Juja. Spores were isolated using the sucrose method. Spore abundance was done using a gridded Petri dish and morphologically characterized using the International Culture Collection of Vesicular Arbuscular Mycorrhizal Fungi (INVAM) database. At least 4 families, 10 genera and 41 species of glomeromycota phylum were isolated from the 3 sampling sites. The families of glomeraceae (16 species) and acaulosporaceae (14 species) dominated in Juja and Mwea Sub Counties. Glomus spp. isolated from the rhizosphere of banana and grass plants were the most abundant, at $p \le 0.05$, in Juja while *Diversispora spp* (diversisporaceae) was the least abundant, at $p \le 0.05$. Mwea Sub County had the most spore abundance, at $p \le 0.05$, compared to Mitunguu and Juja. Mycorrhizal spores isolated from the grass family were the most abundant, at $p \le 0.05$, (Mwea, 41; Juja, 37; Mitunguu, 35.2) as opposed to banana and papaya plants. The results showed that AMF spore abundance and diversity varies with different locations and the associating plants.

Keywords: Arbuscular mycorrhiza fungi, rhizospheric, spores, sucrose.

1.0 Introduction

Arbuscular mycorrhizal fungi (AMF) belongs to glomeromycota phylum/division, from the kingdom of fungi. They are mostly found in terrestrial ecosystems and are obligate root symbionts that establish a mutualistic symbiosis with quite a number of plant species worldwide (Lekberg *et al.*, 2013). The AM fungi play an important role in plant nutrition and especially phosphorous and nitrogen uptake, as well as water absorption and this, is attributed to the presence of arbuscules that they produce inside the host plant's roots



(Smith and Read 2008). According to Augé (2001), this aspect results in the enhancement of plant growth and the ability to withstand abiotic and biotic stresses. The colonization of the roots is arbitrated by genetic, morphological and functional relations between the AMF species and the plant (Kiriacheck *et al.*, 2009).

According to studies conducted by Pawlowska and Taylor (2004), the morphology of spores may comprise one million nuclei and they also vary in size, shape and colour. The diversity, as well as distribution of AMF species, could be affected by the farming systems of the plants and their communities (Jefwa *et al.*, 2006, Dalpe *et al.*, 2000). To accomplish adequate levels of productivity including desirable food quality, sustainable agricultural systems have been enhanced. This systems enables decreased fertilizer usage which that leads to precluded environmental pollution and on the other hand, input costs are minimised (Harrier and Watson 2004; Siddiqui *et al.*, 2008).

Various studies have been conducted in exploring the importance of microorganisms in boosting the soil fertility and the improved crop production and substitutes to the use of commercialised synthetic pesticides (Igiehon, N. O., and Babalola, O. O., 2017). More attention is being devoted on improving the exploitation of indigenous soil microorganisms which will lead to improved soil fertility (Hamel and Strullu, 2006), as well as plant protection against soil borne diseases especially within the rhizosphere (Jawson *et al.*, 1993; Knudsen *et al.*, 1995).

Crops respond and benefit from AMF depending on various agricultural factors such as the inoculation potential of the mycorrhizal fungi, fertilization, tillage practices as well as the reliability of the host crop on mycorrhizal colonization (Auge, 2004). Cultivation in soils with decreased levels of fertilizer enhances numerous AMF beneficial effects such as retaining of nutrients in the soil (Djuuna *et al.*, 2009).

Papaya (*Carica papaya* L.) is a tropical fruit but its high productivity is affected by over fertilization using chemicals and other synthetic products (Campostrini and Glenn, 2007). According to Alarcón *et al.*, 2002, papaya orchards in Mexico are established on soils with low fertility, and fertilizers constituting on mainly phosphorus are applied especially during the reproductive stage to improve its productivity. However, in most soils, phosphorous is unavailable to the plants' roots (Holford, 1997).

There is high diversity of AMF observed around the rhizosphere in the natural habitat (Opik *et al.*, 2008). The distribution of the AMF is due to their ability to withstand high levels of nutrients in various types of habitats (Porras-Alfaro *et al.*, 2007). Even under the same climatic conditions and ecosystem, the AMF communities will be different (Meadow & Zabinski, 2012).

Due to the diversity of AMF spores, there is need to isolate, obtain the spore abundance and characterize their various genus and species even under the same climatic conditions in order to improve on the soils with lesser AMF spores through boosting it with inoculum obtained from isolated spores. The increased amounts of available AMF spores in the soils



will have an improved overall performance of crops.

2.0 Material and Methods

2.1 Description of study sites

The research was conducted at Mwea, Mitunguu and Juja in Kenya, since the areas have existing established papaya orchards, which is the crop of focus in this study. The site at Mwea, in Kirinyaga County, is located at 00 42' 0" S latitude, 370 22' 0" E longitude and lies at 1093 M above sea level. It has an annual mean temperature of 22.71oC and mean annual rainfall of 930 mm. Mitunguu, in Meru County, is located at 00 60' 0" S latitude and 370 47' 0" E longitude and lies at 1498 M above sea level. It has an annual mean temperature of 20.60C and mean annual rainfall of 550 mm. Juja, on the other hand, is in Kiambu County and has geographical coordinates of 10 11' 0" S latitude and 370 7' 0" E longitude. It lies at 1519 M above sea level. The average annual temperature is 19.60C while the mean annual rainfall is 1014 mm

2.2 Sampling procedure and soil collection

Soil samples were collected using soil auger from the papaya, banana and grass plants, located in Mwea, Mitunguu and Juja, around the plants' rhizosphere at a depth of 0-20 cm and packed separately in sterilized polyethene bags.

2.3 Isolation and characterization of arbuscular mycorrhizal fungi (AMF)

In the JKUAT laboratories, spore isolation was carried out as described by Schenck and Perez, 1990. Fifty grams of soil was sampled out and placed in a 250 ml conical flask. About 100ml of tap water was added and the flask was capped with a rubber cork. The mixture was agitated vigorously and left to decant for 30 s and then washed through 250 μ m, 100 μ m and 45 μ m pore sieves. The contents of the 45 μ m pore sieve were backwashed into a small sized beaker and swirled. The contents were then quickly decanted into 50 ml centrifuge tubes and balanced by weight then centrifuged for 5 min at 1750 rpm. The supernatant was discarded and the tubes were filled with 48% sucrose solution (sucrose-227 g dissolved in 500 ml water), balanced by weight and stirred vigorously to re-suspend the precipitate then centrifuged for 15 sec at 1750 rpm. The supernatant sucrose was emptied through a 45 micromesh sieve. The spores retained on the sieve were rinsed thoroughly with distilled water to wash out the sucrose. The spores were then washed away with distilled water into gridded Petri dishes for examination.

2.3.1 Spore abundance

The number of spores were counted under a dissecting microscope (Labomed), using 10X objective lens hence a total magnification of 100 X using a gridded Petri dish and a laboratory needle. The population of the spores was expressed according to the number of spores per 50 g of the soil according to the plant and location. Six spores per treatment were randomly isolated using a needle and a 5 ml transfer pipette to a microscope slide. Melzer's reagent was added onto the spore and the slide was covered with a microscope coverslip. Since the microscope was fitted with a camera and connected to the computer, the spores observed were captured and measured.



2.3.2 Identification of fungal structures

Characterization of the spores was carried out morphologically based on the color, shape, melzer's reaction on the spore color, spore surface and size of the spore and distinguished according to descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005).

2.4 Data management and analysis

Soil was sampled from the root's rhizosphere of the papaya, banana and grass plants using a zig zag pattern across a paddock. The tabulated data was subjected to one way analysis of variance (ANOVA) using Genstat statistical package 15th edition, while the means found to be significantly different at $p \le 0.05$ were separated using Tukey's HSD test.

3.0 Results

3.1 Spore abundance

The mean spore obtained from the 3 locations and among the plants was significantly ($p \le 0.05$) different. Plants obtained from Mwea exhibited the highest number of spores from the 3 plants in comparison with Mitunguu and Juja areas. Soils obtained from plants' rhizosphere in Mwea exhibited the highest number of spores in comparison with Mitunguu and Juja areas. However, there was no significant ($p \le 0.05$) difference between the number of spores isolated from grass plants'rhizosphere from Mwea and Juja areas, 37.73 and 37 respectively. Spores isolated from the grass plants' rhizosphere were most abundant in all areas. Papaya plant had the least mean number of spores and was significantly ($p \le 0.05$) different among the areas with Juja and Mitunguu displaying the lowest overall number of spores, 18.5 and 21.2 respectively (figure 1).

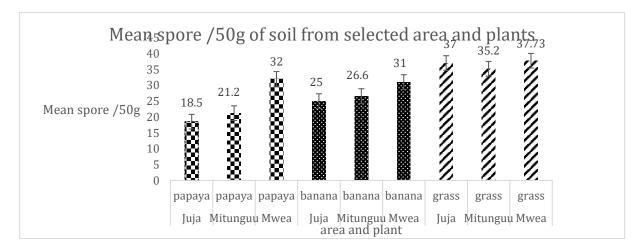


Figure1: Mean spore abundance per 50g of soil from the rhizosphere of banana, papaya and grass plants obtained from Mitunguu, Mwea and Juja areas



3.2 Spore characterization

3.2.1 Glomeromycota families distribution

Majority of the isolated spores belonged to the family of aculosporaceae (23), followed by glomeraceae (19), gigasporaceae (10), while diversisporaceae (2), had the least number of genus and species (Table 1). Five out of the six spores characterized from the rhizosphere of papaya plants from Mitunguu area belonged to aculosporaceae family while the other spore belonged to glomeraceae family. Five out of the six spores characterized from the rhizosphere of the rhizosphere of banana plants from Juja area belonged to glomeraceae family while the other spore belonged to aculosporaceae family. Only the rhizosphere of banana from Mwea and the rhizosphere of papaya from Juja had spores belonging to diversispora family.

Table 1: Glomeromycota families' distribution in the rhizosphere of banana, papaya and grass plants obtained from Mitunguu, Mwea and Juja areas

Area	Plant	Acaulosporaceae	Glomeraceae	Gigasporaceae	Diversisporaceae
Mitunguu	Рарауа	5	1		
	Banana	2	3	1	
	Grass	1	2	3	
Juja	Рарауа	3	2		1
	Banana	1	5		
	Grass	1	4	1	
Mwea	Рарауа	4	1	1	
	Banana	3	1	1	1
	Grass	3		3	

3.2.2 Botanical classification of the spores

The isolated spores obtained from the rhizospheric soil of papaya, banana and grass from Mitunguu, Mwea and Juja areas per 50 g of soil were classified according to their shapes, color, melzer's reaction, size of the spore's diameter and the spore surface, based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005) as shown in tables 2 to 10 below. The colors of the isolated spores varied, and consisted of brown, red-brown, orange brown, bright greenish yellow among others. The diameter of the spores ranged between 42 to 84 μ m while the dominant shapes of the spores were globose and sub globose (Tables 2 to 10).

Table 2: Morphological Miturguu area based o	pical chanacterizatio red on the descriptio	bble 2; Morphological characterization of arbucular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of papapa from Miturguu area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Micorrhizal Fungi (INVAM, 2005)	acular mycorrhizal fungi (AMF) ob kel by the International Culture Col Mycorrhizal Fungi (INVAM, 2005)	F) obtained from th s Collection of Arb 005)	te rhisorpheric so uscular and Vesi	oil af papapa fi cular-Arbusoul	8.8
Image of spore	Shape	Color	Melzers reaction	Size (diameter in µm)	Size (diameter Spore surface in µm)	Genus	
7	Globase	Brown	Red brown	45	Granular	Acaulospora	
6	Globose	Dark greenish	Pinkish red	43.5	Granular	Acaulospora	
	Sub globose		No reaction				

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Miturguu area o	aced on the descriptio	Miturguu area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesscular-Arbuscular Miscorhizal Fungi (INIAM, 2005)	led by the International Culture Col Mycorwhisal Fungi (INTAM, 2005)	: Collection of Arb 005)	ucular and Vesic	cular-Arbuscula	
Image of spore	Shape	Color	Melzers reaction	Size (diameter in µm)	Spore surface	Genus	Species
7	Globose	Brown	Red brown	<u>5</u>	Grannlar	Acaulospora	tubenculata
6	Globose	Dark greenish	Pinkish red	43.5	Granular	Acaulocpona	tubenculata
6	Sub globose	Brown	No reaction	к	Granular	Acaulocpona	foreata
0	Elliptical	Brown	No reaction	51.5	Granular	Acaulocpona	sp3
0	Globose	Brown	Dark brown	66	Granular	Giomus	ţ
	Ovoid	Yellow-brown	Dark red purple	61.5	Irregular	Acaulospora	colombiana

Abundance and Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores



	Speci
^t bananas from ar-Arbuscular	Genus
hizospheric sail af saular and Vesiaul	Spore surface
abtained from the r e Collection of Arbu 05)	Size (diameter Spore surface
wlar myoarrhizal fungi (AMF) abti ed by the International Culture Co Wyoarrhizal Fungi (INWAM, 2005)	Melzers
haracterization of arbuscular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of bananas from n the descriptions provided by the international Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005)	color
00	strape
Table 3; Morphologioal Mitunguu area based	nage of spore

Abundance and Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores

++ +	Image of spore	Shape	Color	Melzers reaction	Size (diameter in µm)	Spore surface	Genus	Species
	0	Globose	Yellow	uight purple	64.5	Granular	Glomus	aggregatum
		Elliptical	Yellow	Light purple	57	Irregular	Glomus	manihotis
	0	Elliptical	Orange-brown	Darker orange	585	Granular	Acaulaspora	capsicula
		Irregular	Brown	Pinkish purple	12	Granular	Dentiscutata	erythropa
		Globose	Dark red brown	Darker orange brown	555	Granular	Acaulospora	capsicula
	8	Globose	Orange brown	No reaction	8	Granular	Glomus	g ds

Table 4: Marpho Mitunguu area b	logical characterizati ased on the descripti	Table 4: Morphological characterization of arbuscular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of grass from Mitunguu area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INNAM, 2005)	usaular myoorrhizal fungi (AMF) of ded by the international Culture Co Myoorrhizal Fungi (INNAM, 2005)) obtained fram th e Collection of Arbu 05)	e rhizospheric sai iscular and Vesicu	l of grass from Ilar-Arbuscular	
Image of spore	shape	Color	Melzers reaction	Size (diameter in µm)	spore surface	Genus	species
	Globose	clear brown	Light pinkish red	8	Granular	Soutellospora	biornata
	Irregular	Dark brown	Light pinkish red	69	Granular	Dentiscutata	reticulata
	Elliptical	Vellow-brown	Red brown	48	Irregular	Acaulospara	scrobiculata
	Sub globose	Vellow orange	No reaction	525	Smooth	Glomus	sp2
	Globose	Vellow brown	Dark red purple	75	Granular	Gigospora	decipiens
0	Globose	Pale yellow brown	Dark red	8	Granular	Rhizophagus	fasoioulatus



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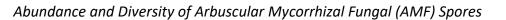
Abundance and Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores

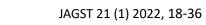
Table 5, Marphok Mwea area bas	ogical characterizati sed on the descriptio	Table 5: Morphological characterization of arbuscular mycorrhizal fungi (AMF) abtained from the rhizospheric soil of papaya from Mwea area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005)	scular myoorrhizal fungi (AMF) ob d by the International Culture Coli Myoorrhizal Fungi (INVAM, 2005)	obtained from the clilection of Arbu- 15)	e rhizospheric soil scular and Vesiou	of papaya from lar-Arbuscular	
Image of spore	Shape	Color	Meizers reaction	Size (diameter in µm)	Spore surface	Genus	Species
	Irregular	Yellow-brown	No reaction	42	Irregular	Acoulospora	kaevis
0	Globose	Dark greyish green	Red brown	52.5	Granular	Acoulospora	elegans
	Sub glabase	Orange-brown	Darker orange	8	Granular	Acoulospora	excavate
	Elliptical	Brown	No reaction	58.5	Granular	Acoulospora	1 qe
	Suib glabase	Bright greenish yellow	Dark red brown	73.5	Irregular	Gigaspora	gigantean
	Sub glabase	Orange brown	No reaction	8	Granular	Funnelijformus	mosece

Table 6. Morphol Mwea area ba	ogiool characterizatio sed on the description	Table 6: Morphological characterization of arbuscular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of banana from Mwea area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005)	sovlar myoorrhizol fungi (AMF) obt d by the International Culture Colli Myoorrhizal Fungi (INVAM, 2005)	obtained from th Collection of Arbu 05)	e rhizaspheric sail scular and Vesicu	l af banana fram Ior-Arbuscular	
Image of spore	Shape	Color	Meizers reaction	Size (diameter in µm)	Spore surface	Genus	Species
	Sub glabase	Orange brown	No reaction	B	Granular	Acaulospora	asounooj
	gnoldo	Yellow brown	Red brown	2	Granular	Glomus	มการอิสังครับ
	Sub glabase	Orange brown	Red purple	ᅜ	Granular	Acaulospora	colombiana
	Irregular	Red brown	Dark red brown	78	Irregular	Dentiscutata	heterogama
	Sub glabase	Red brown	No reaction	70.5	Granular	Diversispara	paoloida
	Globose	Light brown	Pinkish red	5.55	Granular	Acaulospora	spi



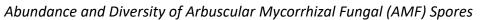
Table 7: Morpholo area based an th	Table 7: Morphological characterization of orbuscular mycorrhizal fungi (AMF) obtoined from the rhizospheric soil of grass from Mwe area based on the descriptions provided by the International Culture Collection of Arbusovlar and Vesicular-Arbusovlar Mycorrhizal Fungi (INVAM, 2005)	of arbuscular mycorrhizal fungi (AMF) abtained from the rhizospheric soil of grass from Mwea ed by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizol Fungi (INVAM, 2005)	icorrhizal fungi (AMF) a tional Culture Collection Fungi (INVAN, 2005)	btained from the r of Arbusavlar ano	hizosphenic sail aj ¹ vesicular-Arbusc	f grass from Mw Wor Mycorrhizol	
Image of spore	Shape	Color	Melzers reaction	Size (diametar in µm)	Spore surface	Genus	species
	Sub glabase	Orange-brown	Red brown	8	Granular	Acaulaspara	serobioulata
8	Globose	Orange brown	Red purple	8	Granular	Scutellaspora	scutatia
	Sub glabase	Orange brown	No reaction	51	Granular	Acaulaspara	laewis
0	Globose	Red brown	Red purple	ţ	Granular	Acaulaspora	dentioulata
	Ovoid	Red brown	No reaction	72	Granular	Soutellospora	sp1
0	Sub globase	Red brown	Red purple	73.5	Granular	Sortellospora	biomata

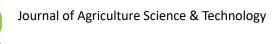






Elliptical Yellow brown Sub globose Orange brown Elliptical Orange brown Globose Dark grevish green	reaction	Size (diameter in µm)	spore surface	Genus	Species
	own Red brown	46.5	Irregular	Acaulospora	scrobiculata
	own No reaction	46.5	Granular	Acaulospora	foveata
	own Hyalline	54	Granular	Diversispara	eburneo
	ı green Red brown	8	Smooth	Acaulospora	elegans
Globose	n Red brown	585	Granular	Glomus	Sąż





ambisporum

Glomus

Granular

G,

No reaction

Orange brown

Globose

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Image of spore	strape	Color	Meizers reaction	Size (diametter in µm)	spore surface	Genus	Species
	Elliptical	Yellow	Light purple	82	Granular	Glamus	manihotis
	Elliptical	Yellow	Light purple	78	Granular	Glamus	manihotis
	Globose	Orange brown	Darker orange	615	Granular	Glamus	ooronatum
	Sub glabase	Reddish brown	Darker red brown	73.5	Granular	Septoglomus	desertioola
6	Sub globose	Brown	Light brown	8	Granular	Glamus	5p3
0	Elliptical	Orange-brown	No reaction	8	Granular	Glamus	ambisporum

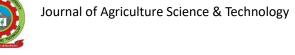
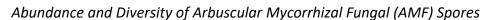


Table 9; Marphological characterization of arbuscular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of banana from Juja

Abundance and Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores

Table 10: Morpholo area based on th	ogical characterizatio e descriptions provide	Table 10: Morphological characterization of arbuscular mycorrhizal fungi (AMF) obtained from the rhizospheric soil of grass from Juja area based on the descriptions provided by the International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM, 2005)	nyaarrhizal fungi (AMF) a tional Culture Collection Fungi (INVAM, 2005)	btained from the Jf Arbuscular and	rhizospheric soil c Vesicular-Arbusci	of grass from Juja Mar Myaarrhizal	
Image of spore	Shape	color	Melzers reaction	Size (diameter in µm)	Spore surface	Genus	Species
6	Globose	Yellow brown	Dark purple	9	Irregular	Glomus	intraradices
0	Sub globose	Brown	Red brown	615	Smooth	Glamus	sp1
	Globose	Brown	Red brown	72	Granular	Septoglomus	constructum
	Irregular	Bright orange	Darker orange	8	Irregular	Acaulaspora	sp2
0	Globose	Red brown	Dark purple	55.5	Granular	Septoglomus	desertioola
0	Globose	Cream yellow brown	Hyaline	79.5	Irregular	Rooostra	sp2





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4.0 Discussion

Arbuscular mycorrhizal fungi (AMF) are sturdily reliant on their host plants (Fitter, 2005) and have different levels of specific hosts pairing (Johnson *et al.*, 2003; Klironomos, 2003). According to Fitter (2005), the AMF are incapable of developing independently whether in axenic culture or naturally. AMF requires root colonization of a vascular plant for their life cycle to be complete (Brundrett, 2004).

There were variations in the number of spores recorded in this study among the plants'rhizospheric soil from different areas. The spore abundance dissimilarities could be due to abiotic factors and especially the type of soil and the amount of water in the soil. For example, Mwea area had the highest number of spores and this could be due to poor drainage of soils (black cotton soils) resulting to flooding compared to Mitunguu and Juja areas. Arbuscular mycorrhizal fungi are not only present in wet areas but also ubiquitous there (Ipsilantis *et al.*, 2007). However, the ubiquitous occurrence of AMF in wet areas has been attributed to the aerenchyma present in such plants, which is well-developed (Miller, 2000). Seasonal variations can lead to fluctuations in the number of AMF spores in a given soil sample, although species like Glomus can be found in all seasons and more regularly as revealed by Tabassum *et al.*, 2016.

In the current study, papaya plants' rhizospheric soil had the least mean number of spores compared to grass and banana rhizospheric soil. According to Whipps, 2001, papaya plants, being a terrestrial plant, are colonized naturally by AMF. Various organisms in the soil however compete for their territory and this can lead to other different types of fungi colonizing this plants thus reducing the advantageous effects of the AMF. Grass plants' rhizospheric soil had the most number of spores and this concurred with the soil spore count results of Sankaralingam *et al.*, (2016), which were highest in the maize field, which belongs to grass family, poaceae, in comparison to rice, sugarcane and banana. According to Brundrett and Abbott (2002), hyphae, are structures found in mycorrhiza and their combination forms mycelium, the source of inoculum. They are however susceptible to soil disturbance as this pulls down the AMF infectious ability. Among the plants involved in the current study, grass plants' rhizosphere was the least disturbed and this could have attributed to the high number of spores compared to banana and papaya plants' rhizosphere.

Schüßler and Walker (2010) have described and named most of the AMF species according to the morphology of their spores. In the current study, spores of different genera were isolated from different plants and locations. Acaulosporaceae and glomeraceae families dominated the 3 areas and still among the plants. In similar environmental conditions, *Acaulospora species* and *Glomus species* have been observed to produce more spores than *Gigaspora species* and *Scutellospora species* (Bever *et al.*, 1996). Studies administered in agricultural fields have further concluded that soil disturbance leads to drastic relocation of the AMF community (Schnoor *et al.*, 2011). Maherali and Klironomos (2007) indicated that glomeraceae and gigasporaceae families distribute most of their biomass in the intraradical and extraradical hyphae respectively while acaulosporaceae produce low biomass both intra and extraradically. Majority of the spores isolated from papaya plants'



rhizospheric soil belonged to acaulosporaceae family. According to Chagnon *et al.*, 2013, this AMF families have characteristic life history with most species under Acaulosporaceae being stress tolerators, glomeraceae being associated with ruderals while those in gigasporaceae are competitors.

AMF spores vary in their wall characteristics such as colour, size, shape germination structures and formation mode, and these characteristics perform species identification using keys to taxa Glomeromycota (Oehl *et al.*, 2006, 2008).

The lowest, highest and the mean of the AMF size is described according to the species and is necessary for the study of the taxonomy and ecology of spores (Oehl *et al.*, 2008). Higher sizes of spores, more than 200 μ m, can easily differentiate gigasporaceae family from others such as glomeraceae whose sizes are less than 200 μ m (Oehl *et al.*, 2006). In the current study, the size of the diameter of the spores ranged between 42 to 84 μ m at X10 magnification, with the larger sizes being from gigasporaceae family while the smaller sizes from acaulosporaceae family.

The shapes of spores may vary among and within species as in the case of *Funneliformis mosseae*, whose shapes vary from globose, sub-globose and irregular (Al-Qarawi *et al.*, 2013). Redecker *et al.*, 2013 has described nine different shapes which include: irregular, elliptical, triangular, globose, oblong, pulvinate, subglobose, ovoid and knobby. Some spores have similar ornamentation and colours but their altered shapes differentiate their species, e.g. *Scutellospora calospora* and *Scutellospora dipurpurescens* (Oehl *et al.*, 2008). Most of the observed shapes in this study were globose, irregular, elliptical, oblong, and ovoid.

The colours observed in this study using the Munsell colour chart ranged from brown, redbrown, orange-brown and light brown. The colours of the AMF spores vary between their families, genus and sometimes species from white to black and variants such as dark, bright, pale or light (Oehl *et al.*, 2008). These colours also depend on the maturity of the spore and their integrity e.g. *Acaulospora capsicula* has colours ranging from orange-brown, red-brown and dark red-brown (Schüßler and Walker, 2010). Addition of melzer's reagent on the spore changed the inner and outer colour of most of the spores while other spores did not react. During the identification of AMF species, the color of the spore is a very suitable characteristic to unravel many uncertainties of the taxonomy (Oehl *et al.*, 2008).

5.0 Conclusion

This current study showed the diversity of Arbuscular Mycorrhizal Fungi (AMF) spores isolated from papaya, banana and grass rhizosphere from Mitunguu, Mwea and Juja areas in terms of the genus and species of the glomeromycota phylum. The difference in AMF genus and species could be attributed to the different climate conditions and the plants associated with the spores. The study also revealed that acaulosporaceae and glomeraceae families were dominant in the rhizosphere of the 3 plants obtained from the 3 different areas. Since the agricultural management practices varied among the 3 plants and on the 3 areas, this could affect spore abundance in the plants. Papaya plants' rhizospheric soil had low spore count and majority of the observed spores belonged to acaulosporaceae family



which is associated with stress tolerance.

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