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Composition and spore abundance of arbuscular mycorrhizal fungi in sweet potato producing areas in Uganda

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Introduction: Farming systems influence composition and abundance of microbial communities.

Methodology: A study was conducted using morphotyping and enumeration methods to determine the composition and spore abundance of Arbuscular Mycorrhizal Fungi (AMF) in sweet potato producing regions in eastern Uganda. Sampling was done from fields with crop types (CTs) including legumes (groundnuts, common beans, cowpea, soybeans, green grams), sorghum, sweet potato, and fallowed fields which were used as a control. Three agro-ecological zones (AEZs) i.e., Mt. Elgon High Farmlands (MEHF), Lake Victoria Crescent (LVC), and Southern and Eastern Lake Kyoga Basin (SELKB) were considered.

Results and discussion: A total of 6 AMF genera comprising of Glomus, *Acaulospora*, Scutellospora, Entrophospora, Archaeospora, and Gigaspora were isolated from the study sites. Agro-ecological zones had a significant ($p<0.05$) effect on Entrophospora spp. while crop types had a significant ($p<0.05$) effect on *Gigaspora* spp. although all the AMF genera were present in all AEZs and CTs. Spore abundance was similar across the AEZs except for MEHF (177) which was lower while spore abundance lowest in sweet potato (177) and largest in fallow (224), attributed to soil properties and similar crops included in the crop rotation program. The AMF can be isolated, identified, and multiplied to produce bioinoculants for the regions.

KEYWORDS

arbuscular mycorrhiza, composition, spore abundance, agroecological zone, cropping type

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1 Introduction

Arbuscular mycorrhizal fungi are obligate endosymbionts of up to 90% terrestrial plants (1) belonging to the phylum Glomeromycota (2) and sub-phylum Glomeromycotina (3). They are gaining prominence in agriculture for increasing crop production. Therefore, understanding their biogeographical patterns and drivers for maintaining ecosystem services amidst changes in farming systems aimed at agro-ecological adaptability is a prerequisite. Various biotic and abiotic factors and biological processes are reported to impact on microbial biogeographical patterns (4, 5). Several studies have identified different edaphic variables that influence soil bacterial and fungal community compositions (6). Abiotic factors include soil type (7), soil texture (8), soil acidity (9), soil temperature (10), soil moisture (11), soil available P (12) and cropping systems (tillage, crop rotation, fallowing) (13–16). Öpik et al. (17) reported that AMF communities vary in composition due to differences in ecosystems under different disturbance regimes. Other factors include organic matter (18), soil biota (19, 20), and plant communities (21, 22).

Some studies in sub-Saharan Africa have focused on the diversity and spore abundance of AMF (e.g., 23–26) but have not emphasized agricultural zoning and cropping systems moreover in Uganda. Cropping systems influence the biological, physical, and chemical properties of soils, as well as the geographical distribution of plants, which greatly impact on the diversity and spore abundance of AMF (23). Crops such as cereals, legumes, coffee, bananas, cassava, and other root and tuber crops that benefit from AMF associations dominate Africa's landscape (27). Less frequently cultivated fallow fields always have higher spore abundance than fields under frequent conventional cultivation (23). In the drier areas of the Maasai-Mara Ecosystem in Kenya, maize and wheat monocrops recorded significantly lower AMF diversity, species richness, and spore density in the wet and dry season than the maize-bean intercrops dominated by *Scutellospora* and *Acaulospora* species (28). Similarly, the dominance of Scutellospora and *Acaulospora* had been reported earlier in Western Kenya (29). Sporulation of genus *Acaulospora* was high in acidic soils (26, 30), increasing their dominance in soils of a pH range of 5.51 to 6.77 (28). Castillo et al. (31) reported high sporulation of *Acaulospora* spp. Under conventional tillage than no-tillage. On the other hand, Jansa et al. (32) reported higher sporulation of Scutellospora spp. In undisturbed and moderately disturbed soils. The limited occurrence of *Gigaspora* spp. In various ecosystems compared to other AMF genera has been confirmed by their low density reported by Schalamuk et al. (33) Jefwa et al. (34), and Muchane et al. (28).

In the study of Belay et al. (24) a total of 15 AMF genera (*Glomus*, *Acaulospora*, *Funneliformis*, *Gigaspora*, *Scutellospora*, *Septoglomus*, *Claroideoglomus*, *Entrophospora*, *Rhizophagus*, *Paraglomus*, *Diversispora*, *Pacispora*, *Racocetra*, *Sclerocytis*, and *Ambispora*) were isolated from both field and trap culture soils. A total of 31 species was observed in the study in an irrigated mixed fruit cropping system that received manure followed by 23 species in a crop rotation field with teff, sesame, and sunflower that received 50 kg Urea ha^{-1} and 100 kg DAP ha^{-1} (24). In the same study, 15 AMF species were noted in a 30-year-old natural forest with acacia, fig, and stinkwood trees, 14 species in an acacia plantation, and up to 11 species in fields of sorghum or maize monocrops receiving 50 to 100 kg Urea ha^{-1} and 100 to 150 kg DAP ha^{-1} (24). *Glomus* and *Acaulospora* AMF were the most diverse groups represented by 9 species each, followed by *Funneliformis* and *Gigaspora*. *Glomus* and *Acaulospora* spp. Produce more spores in a shorter time than *Scutellospora* and *Gigaspora* in the same environment (23, 35, 36). Seven genera comprising *Acaulospora*, *Ambispora*, *Glomus*, *Claroideoglomus*, *Pacispora*, *Gigaspora*, and *Scutellospora* were isolated from cassava cropping fields in Abengourou, East Côte d'Ivoire of which the genus Glomus was dominant (37). Earlier on in South Africa, studies had shown that the rhizosphere of cassava in Limpopo contained *Acaulospora* scrobiculata, Glomus rubiforme, and Gigaspora sp. Whereas the Mpumulanga soils had *Acaulospora* scrobiculata, *Acaulospora* mellea, *Acaulospora racticede*, *Glomus etunicatum*, *Glomus rubiforme*, *Gigaspora* sp, and *Scutellospora* sp (38).

However, no studies have been carried out on the composition and spore abundance of AMF in fields under sweet potato production. Therefore, the objective of the study was to determine the composition and spore abundance of AMF as influenced by different crop types (CTs) in three sweet potato producing AEZs in eastern Uganda.

2 Materials and methods

2.1 Study sites description

The study was conducted in eastern Uganda covering three agro-ecological zones (AEZ) namely, Mt. Elgon High Farmlands (MEHF), Lake Victoria Crescent (LVC), and Southern and Eastern Lake Kyoga Basin (SELKB). The sites were distributed, in Magola and Rubongi sub-counties of Tororo district (MEHF); Busware and Banda sub-counties of Namayingo district (LVC) and, in Bukedea, Ongi'no, Malera, Kolir, and Kachumbala sub-counties of Bukedea district (SELKB). The landscape of MEHF has steep slopes and is divided by many valleys. The climate is cool and wet with the southern part being warmer with less rain in July than the northern part. Rainfall peaks in April and May but is generally more than 100 mm per month from March to November. In the LVC, the landscape of West of the Nile River in the LVC is an old land surface marked by ridges or laterite-capped hills, long slopes, and wide, often swampy valleys while on the East of the Nile, the landscape is rolling with wide valleys and relatively less rolling. Soils are often acidic and low in K, but with moderate levels of organic matter. Southern and eastern Lake Kyoga Basin (SELKB) has a gently rolling landscape with wide valleys draining to Lake Kyoga. The soils of the western part of this zone are generally loamy on the ridges and upper slopes and sandy loam on the lower slopes. This sub-humid AEZ has two cropping seasons with almost equal average rainfall intensity, 560 mm during March-June, and 540 mm during July-November (Figure 1).

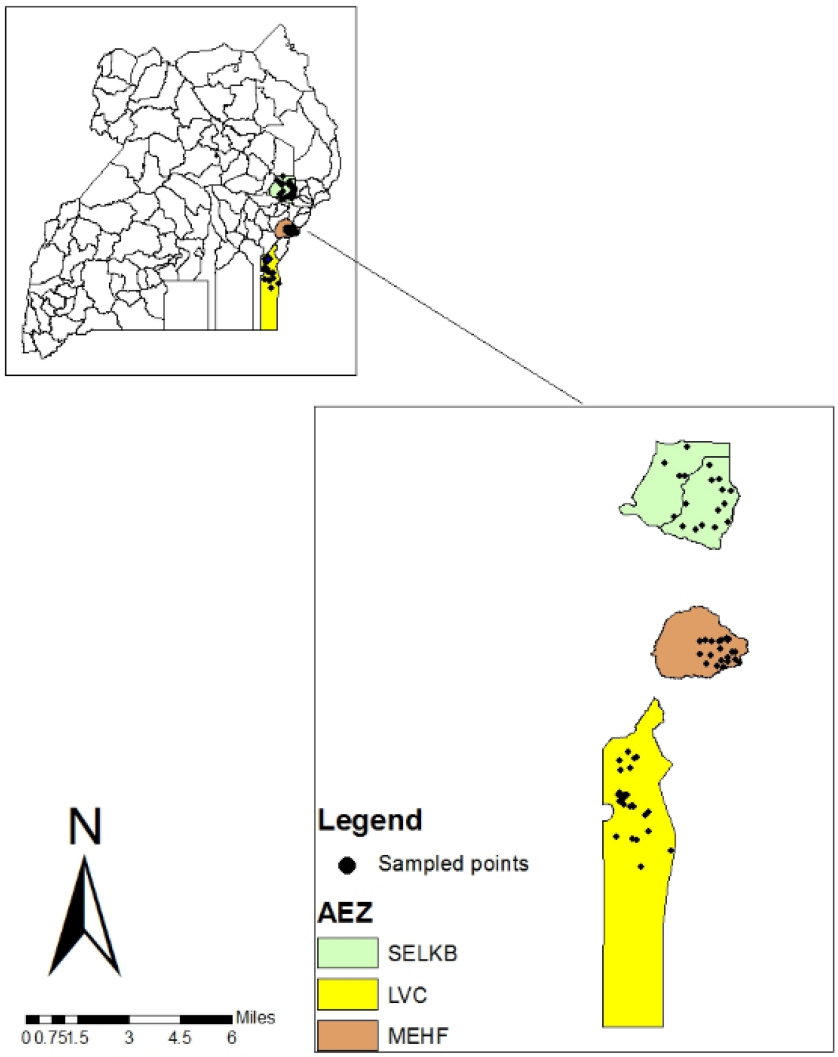

FIGURE 1
A map showing study areas in eastern Uganda. SELKB, Southern and Eastern Lake Kyoga Basin; MEHF, Mt. Elgon High Farmlands; LVC, Lake Victoria Crescent.

The area under sweet potato production in eastern Uganda is 159,948 hectares and the average yield is 5.3 t ha^{-1} while in central Uganda 98,054 hectares are under sweet potato production with an average yield of 3.2 t ha^{-1} (39). Further description of the AEZs is shown in Table 1.

2.2 Soil sampling procedure

A multistage sampling frame was used to select study sites. The study sites were selected considering the major sweet potato producing regions targeting different crop types and hence the selection of Mt. Elgon High Farmlands (MEHF), Lake Victoria Crescent (LVC), and Southern and Eastern Lake Kyoga Basin (SELKB). Districts that experience moisture stress conditions were considered and hence Tororo, Namayingo and Bukedea were selected. Occurrence of moisture stress conditions in these study sites is attributed to the high temperature recorded and rainfall that is unreliable and highly variable in terms of its onset, cessation, amount, and distribution (44). Sub counties i.e., Tororo (Magola and Rubongi sub-counties), Namayingo (Busware and Banda sub-counties) and Bukedea (Ongi'no, Malera, Kolir, and Kachumbala sub-counties) were randomly selected. In the selected sub-counties, a list of smallholder farmers (producing the targeted crops) was proposed by contact farmers in the respective sub-counties and farmers were randomly selected from the list. Farmers' fields were used as replicates and the samples were obtained following the existing crop types and soil amendments on that field. The geographical location for each field was captured using a Geographical Positioning System (GPS). Farmers provided more details on the fields that were sampled from for the previous 5 years (Table 2). Nineteen (19) samples were obtained from SELKB, 20 from MEHF and 25 from LVC. It is worth noting that the farmers had racticed mono-cropping, inter-cropping, and crop rotation including cassava, groundnuts, sweet potato, maize, soybean, green grams, millet, sesame, oranges, eggplant, green pepper,

TABLE 1 Agro-ecological characteristics of the study areas.

Charateristic	SELKB [1]	MEHF [1]	LVC [1]
Population density (pers km^2)	129 [3]	345 [3]	280 [3]
Altitude (masl)	1143 [4]	1400-1800 [3]	1106 [4]
Mean annual temperatures (° C)	28-31 [4]	≤20 [1]	22.0 [1]
Rainfall pattern (mm $year^{-1}$)	>1200 [1; 3;4]	>1200 [1; 3]	>1200 [1]
Soil textural classes and other characteristics	The Western part (loam on ridges and upper slopes, sandy loam on lower slopes); East and North West (sandy soils, occasionally acidic, often low in organic matter) [1]	North part (red clay loam, well drained, highly leached, often acid, good nutrient supply); south (surface soil-high sand content, lower nutrient supply, very low soil erosibility, moderately high rainfall erodibility[1]	High clay content, common soils (sandy-clay-loam); clay-loam, acidic, low in K with moderate levels of organic matter [1]
Main crops in order of priority	Finger millet, banana, maize, cotton, rice, sorghum, cassava, sweet potato, and groundnut [1]	Beans, banana, maize, groundnuts, Arabica coffee, sweet potato [1]	Banana, beans, sweet potato, cassava, maize, rice, Robusta coffee, [1]
Cropping practices	No inorganic fertilizer use, crop rotation, mono-cropping/inter-cropping of cassava, groundnuts, sweet potato, maize, soybean, green grams, millet, sesame, oranges, eggplant, green pepper, cabbage, mangoes, onions, cowpea, upland rice, sunflower, and watermelon, while fallowing on a few of the fields were maintained for ≤ 6 months	No inorganic fertilizer use, crop rotation, mono-cropping/inter-cropping of maize, beans, soybean, cassava, millet, cotton, tomatoes, coffee, banana, sweet potato, groundnuts, sorghum, napier grass, brinjals, eggplants, and cowpea, while fallowing on a few of the s was maintained for ≤ 6 months	No inorganic fertilizer use, crop rotation, mono-cropping/inter-cropping of banana, millet, maize, beans, cassava, soybean, sweet potato, sorghum, tomatoes, and Irish potato, while fallowing on a few of the fields was maintained for ≤ 6 months

Source: 40[1]; 41[2]; 42[3]; 43[4].

cabbage, mangoes, onions, cowpea, upland rice, sunflower, and watermelon and, short-term fallows. A soil auger was used to obtain 12 samples from each site in a zigzag pattern at a depth of 0 – 20 cm, which were pooled together, and a 2 kg composite homogeneous sample obtained. The homogeneous sample was placed in a strong polythene bag and sealed securely to prevent further drying and labeled clearly for ease of identification. Samples were collected in a dry season, a period when sporulation increases (45). The samples were split for use in AMF identification and soil physical and chemical analysis.

2.3 Soil physical and chemical analysis

Each soil sample for physico-chemical analysis was air-dried, sieved through 2 mm sieve, homogenized, and analyzed for pH (1 soil:2.5 H_2O ratio), soil organic carbon (SOC), total and available P, exchangeable cations (Ca^{2+}, Mg^{2+}, K^+, and Na^+) and texture at Soil, Water and Plant Analytical Laboratory of Makerere University following procedures outlined in Okalebo et al. (46). Soil pH was measured in a soil-water solution at a ratio of 1:2.5 (w/v) using a pH meter (Mettler-Toledo, AG 8603) after mixing on a rotary shaker for 30 minutes at 150 rpm (47). Total N was assessed after wet digestion of air-dried soil samples with a mixture of concentrated sulphuric acid (H_2SO_4) and selenium powder and salicylic acid and measured using a spectrophotometer (Jenway, 6405 UV/Vis) (48). Available P was extracted using Bray 1 method in a mixture of ammonium fluoride and hydrochloric acid, shaken for 1 minute at 150 rpm. The available P was complexed in a mixture of ascorbic acid and ammonium molybdate (49) and measured using a spectrophotometer (Jenway, 6405 UV/Vis). Exchangeable bases were extracted with ammonium acetate by shaking soil samples in ammonium solution for 20 minutes and measured using a flame-photometer (K^+, Na^+) (Jenway, Essex CM6 3LB) and atomic absorption spectrophotometer (Ca^{2+}, Mg^{2+}) (50) (Jenway, 6405 UV/Vis). Soil organic carbon was determined by wet oxidation with potassium dichromate under concentrated sulphuric acid at 150 °C for 30 minutes. The unreacted dichromate was titrated against standardized ferrous ammonium sulphate solution using ferroin indicator to determine the end point (51). Soil texture was determined using a Bouyoucos (Gallenkamp Bouyoucos) method (52).

2.4 Spore isolation and morphological identification of arbuscular mycorrhizal fungi genera

Arbuscular mycorrhizal fungi spores were isolated according to Jenkins (53) procedure with modifications by Ingleby (54) from 50 g of air-dried soil samples by wet sieving through 710 and 45 µm sieves, followed by sucrose gradient centrifugation. After centrifugation, spores and spore clusters were transferred into Petri dishes. The spores were distinguished into genera under the reflected light on stereomicroscope with the color of spore, spore size, hyphal attachments on spore and surface appearance of spore used as the diagnostic features and enumerated. Slide specimens were prepared for each AMF genus and further described under a

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TABLE 2 Details of the fields obtained at sampling time.

Field no.	Site/Farmer's name	AEZ	Latitude	Longitude	CT	Fertilizer/Manure application
1	DATIC	MEHF	0.69300	34.18090	Maize, Cassava	None
2	MUARIK	LVC	0.46370	32.60990	Fallow	None
3	Margaret Ochieng'	MEHF	0.71412	34.12003	Green grams	NPK foliar feed
4	Nociata Akello	MEHF	0.71424	34.12348	Groundnuts	None
5	Samuel Sunday	MEHF	0.71427	34.12454	Groundnuts	None
6	Omita Opiyo	MEHF	0.70722	34.11651	Soyabeans	None
7	Wilson Ochieng'	MEHF	0.62775	34.05937	Groundnuts	None
8	Wilson Ochieng'	MEHF	0.62784	34.05995	Cowpeas	None
9	Betty Owour	MEHF	0.62782	34.07033	Soyabeans	None
10	Jennifer Obaa	MEHF	0.62635	34.06430	Soyabeans	Manure
11	Jennifer Obaa	MEHF	0.62623	34.06410	Groundnuts	None
12	Joseph Otieno	MEHF	0.71429	34.12097	Sorghum	None
13	DoroRoza Alowo	MEHF	0.71447	34.12473	Sorghum	None
14	Nociata Awino	MEHF	0.71431	34.12348	Sweet potato	None
15	DoroRoza Alowo	MEHF	0.71474	34.12443	Sweet potato	None
16	Betty Owour	MEHF	0.62771	34.07032	Sweet potato	None
17	Japheth Ofwono	MEHF	0.60544	34.07655	Fallow	None
18	Dennis Odoyi	MEHF	0.71416	34.10313	Fallow	None
19	Joseph Otieno	MEHF	0.71429	34.12097	Fallow	None
20	Nociata Akello	MEHF	0.71418	34.12322	Fallow	None
21	Yafesi Okoth	MEHF	0.62561	34.09459	Fallow	None
22	Emmaculate Nyapendi	MEHF	0.63960	34.08994	Fallow	None
23	Charles Ochoo	MEHF	0.64705	34.08577	Fallow	None
24	James Okumu	LVC	0.27108	33.54018	Soyabeans	Rhizobia
25	James Okumu	LVC	0.27108	33.54018	Soyabeans	Rhizobia+DAP
26	Fred Mbageya	LVC	0.27207	33.54147	Common beans	None
27	Fred Mbageya	LVC	0.27207	33.54147	Cowpeas	None
28	Simon Mbageya	LVC	0.27190	33.54246	Soyabeans	None
29	Moses Wafula	LVC	0.27191	33.54250	Common beans	None
30	Constant Wanyama	LVC	0.27621	33.54440	Groundnuts	None
31	Patrick Sifuna	LVC	0.26278	33.54113	Green grams	None
32	Contant Wandera	LVC	0.26886	33.54313	Soyabeans	None
33	Juma Nyegenye	LVC	0.26863	33.54274	Groundnuts	None
34	Frasko Maende	LVC	0.26837	33.54190	Soyabeans	None
35	Friday Mang'eni	LVC	0.26717	33.54199	Common beans	None
36	Wison Wanyama	LVC	0.27560	33.54104	Common beans	None
37	Fred Mbageya	LVC	0.27168	33.54092	Sorghum	None
38	Sam Mulino	LVC	0.27221	33.54160	Sorghum	None
39	Odinga Mbageya	LVC	0.27190	33.54208	Sorghum	None

(Continued)

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TABLE 2 Continued

Field no.	Site/Farmer's name	AEZ	Latitude	Longitude	CT	Fertilizer/Manure application
40	Constant Wanyama	LVC	0.27257	33.54170	Sorghum	None
41	Francis Hagaba	LVC	0.27034	33.54238	Sorghum	None
42	Fred Mbageya	LVC	0.27213	33.54115	Sweet potato	None
43	Sam Mulino	LVC	0.27221	33.54160	Sweet potato	None
44	Faisi Natocho	LVC	0.26981	33.54166	Sweet potato	None
45	Stephen Bwire Anania	LVC	0.12203	33.53931	Fallow	None
46	Barua Ogong'la	LVC	0.10556	35.33000	Fallow	None
47	Yohana Osenga	LVC	0.10066	33.53433	Fallow	None
48	Marsala Nafula	LVC	0.11502	33.51145	Fallow	None
49	Ochieng' Odoki	LVC	0.11838	33.50678	Fallow	None
50	Gilbert Ilomu	SELKB	1.33543	34.05953	Groundnuts	None
51	Justin Aide	SELKB	1.49936	34.01362	Cowpeas	None
52	Charles Orungo	SELKB	1.39368	34.09563	Cowpeas	None
53	Charles Orungo	SELKB	1.39368	34.09563	Green grams	None
54	Julius Ariko	SELKB	1.39660	34.11750	Common beans	None
55	Julius Ariko	SELKB	1.39660	34.11750	Green grams	None
56	Peter Okia	SELKB	1.37978	34.15097	Cowpeas	None
57	Gilbert Ilomu	SELKB	1.33623	34.05870	Sorghum	None
58	Omerisa	SELKB	1.49332	34.01407	Sorghum	None
59	John Ojakolo	SELKB	1.34400	34.18990	Sorghum	None
60	Gilbert Ilomu	SELKB	1.38537	34.05912	Sweet potato	None
61	Esugut Daniel	SELKB	1.49795	34.01535	Sweet potato	None
62	Peter Igala	SELKB	1.25462	34.13432	Sweet potato	None
63	Mary Atimong	SELKB	1.26866	34.15145	Fallow	None
64	Hellen Agoti	SELKB	1.35250	34.16271	Fallow	None
65	Joseph Akol	SELKB	1.47300	34.02448	Fallow	None
66	Anastancia Among'	SELKB	1.37253	34.05537	Fallow	None

compound microscope at magnification x40 with spore germination characteristics, spore wall characteristics, type of spore wall, number of layers and reaction to PVLG- Polyvinyl lacto glycerin (1.66g polyvinyl alcohol 20-25 cP, 10 ml lactic acid, glycerin 1 ml and 10 ml distilled H2O) and Melzer's reagent (chloral hydrate, 1.5 g iodine, 5.0 g potassium iodide and 100 ml distilled H2O + PVLG). The spores were matched with genera described by International Culture Collection of VA Mycorrhizal Fungi (INVAM) West Virginia University Morgantown, WV, USA Website and Schenck and Perez (55, 56).

2.5 Data analysis

Analysis of Variance (ANOVA) using statistical analysis software (SAS), version 9.4 generalized linear model was used to determine the effects of AEZ and CT on AMF spore abundance. Spore abundance data were transformed in excel using square root pi (SQRTPI) the most preferred type of transformation for count data, to obtain symmetric distribution. Agro-ecological zone and CT were treated as fixed effects while fields were the random effects using the generalized linear model. Treatment means were separated using the least significant difference (LSD). Redundancy analysis (RDA) (57), the canonical version of principal component analysis (PCA), was used to examine the multiple correlations between soil properties (pH, OM and total N, available P, exchangeable Ca, K, Mg, and Na, sand, silt, and clay content) and genera of AMF in the sampling sites. The abundance values of AMF genera were centered and standardized in RDA using square root transformation. Soil properties were also standardized as explanatory variables using arcsine square root transformation before performing RDA. The AMF abundance data were pre-

analyzed by Detrended Correspondence Analysis (DCA) using CANOCO software 4.5 (Micro-computer Power, Ithaca, NY) to choose a linear or unimodal ordination model for analysis. As the length of the gradient (first axis) was 0.668 below 3 by DCA, the RDA (canonical correlation analysis) was applied to the data obtained in this study.

3 Results

3.1 Arbuscular mycorrhizal fungi genera and spore abundance

In this study, six AMF genera were distinguished based on morphological features. Spore features that were used to classify the AMF genera included color of spore, spore size, hyphal attachments on the spore, and surface appearance of spore (INVAM and 55). *Gigaspora* species were identified as large spores with a bulbous hyphal attachment. *Scutellospora* species were identified as large spores with bulbous hyphal attachment, germination shield, and flexible/separating walls. *Acaulospora* species were identified as spores having ornamented spore walls, presence of cicatrix/cicatrices, and spores forming on the side of the hypha. *Entrophospora* species had almost similar characteristics to those of *Acaulospora* spp. But their spores form into the neck of the hypha, and *Glomus* spp. Have either curved, straight, or gourd-like hyphal attachment (Figure 2).

Exploratory data analysis using box plots showed a variable range of distribution of spores within AEZs and CTs. The box represents the spore abundance range, the rhombus inside the box represents the mean of the spore abundance data, and the middle line represents the median, the lower and the upper bar mean the minimum and the maximum values of the data, respectively. Mean spore abundance across AEZs ranged between 48 and 58 spores while across CTs it ranged between 47 and 60 spores as shown by the medians of the box plots. Spore abundance was positively skewed across AEZs particularly in Mt. Elgon High Farmlands AEZ and in sweet potato fields because their medians are closer to the lower quartile, signifying non-normal distribution. The highest spore abundance was recorded in Southern and Eastern Lake Kyoga Basin AEZ, and in fallow fields since they had the highest mean values. The widest range of spore abundance was observed in Lake Victoria Crescent AEZ, and in legume fields since they had the longest whiskers (Figures 3A, B).

Using a generalized linear model for the presence and absence of AMF, the occurrence of AMF showed a slight variation in each AEZ and CT with two AMF genera significantly affected. The AEZs differently affected the mean spore abundance of *Entrophospora* spp. ($p < 0.05$) with SEKLB having a significantly ($p < 0.05$) higher number of spores than MEHF. The mean spore abundance of *Gigaspora* spp. ($p < 0.05$) was variable across the CTs with AMF spores dominating in fallow fields than the rest of the CTs (Table 3). T-test accepted the null hypothesis that total spore abundance between CTs in all AEZs was equal except for sorghum and sweet potato (t value=31.28; p=0.0010), sorghum and fallow (t value=0.00; p<.0001) and sweet potato and fallow (t value=9.28; p=0.0114) in MEHF and, sorghum and fallow (t value=0.00; p<.0001) in LVC.

The frequency of observation of the identified AMF genera was 100% except that of *Archaeospora* which was 86%. The dominant AMF genera across the different AEZs were Glomus spp., *Acaulospora* spp., *Scutellospora* spp., and *Entrophospora* spp., each constituting ≥ 10% of the spore abundance of the identified AMF. They accounted for 87% of the spores of the identified AMF in each AEZ while *Gigaspora* spp. And *Archaeospora* spp. Contributed only 13% of the spores. The total number of the AMF genera spores across AEZs was highest in *Glomus* followed by *Acaulospora*, *Scutellospora*, *Entrophospora*, *Archaeospora*, and *Gigaspora* except for MEHF AEZ where *Gigaspora* was more dominant than *Archaeospora* (Table 4).

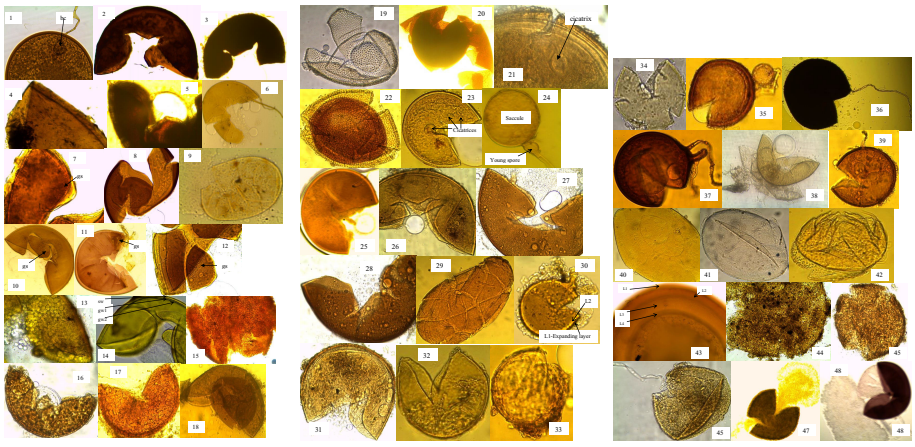

FIGURE 2
Some of the Glomeromycotan species identified from field soils across the AEZs and CTs. Magnification x40, bc, bulbous cell; gs, germination shield; sw, spore wall; gw, germination wall, l,layer, **1-6**=*Gigaspora* spp., **7-12**=*Scutellospora* spp., **13-20**=*Acaulospora* spp., **21-25**=*Entrophospora* spp., **26-39**=*Glomus* spp., **40-48**=Undefined spp.

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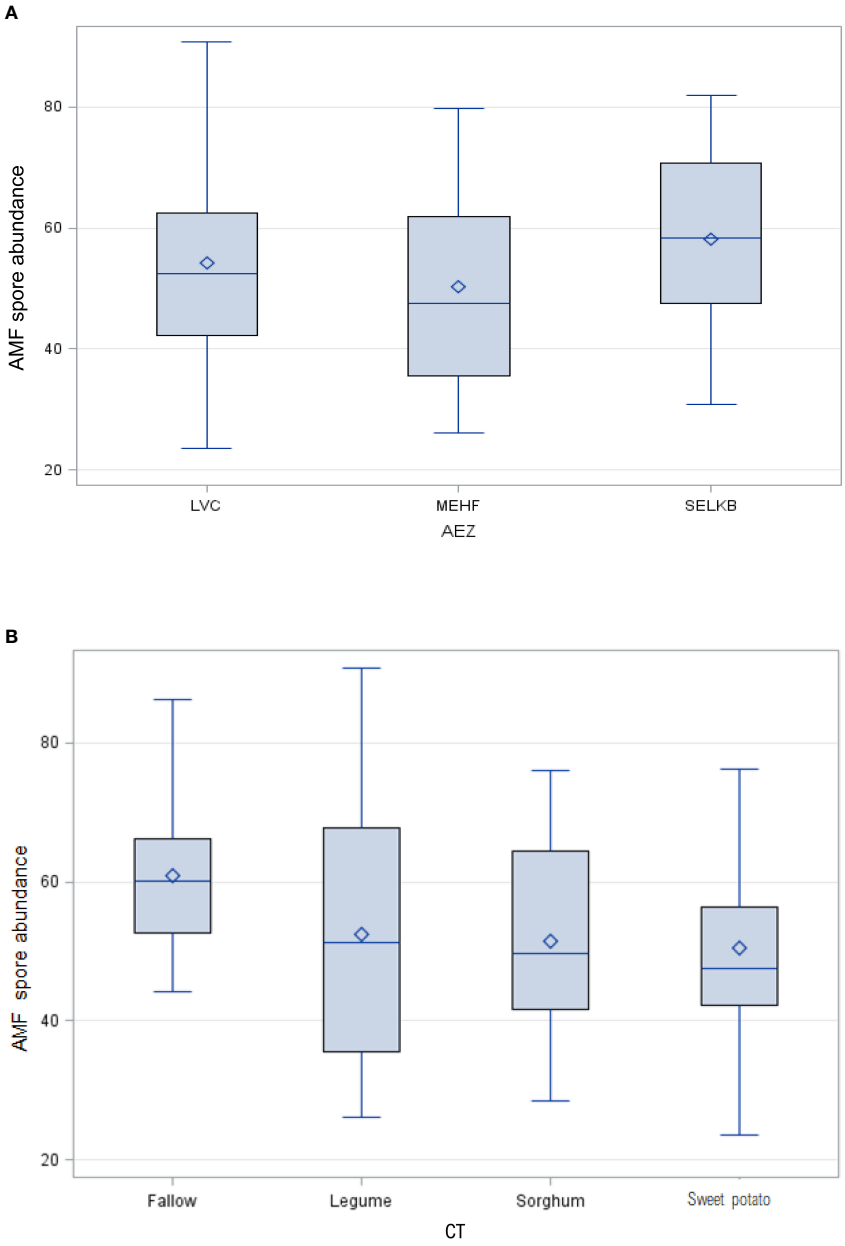

FIGURE 3
(A, B) Range of distribution of AMF spores within AEZs and CTs.

3.2 Correlation of soil properties and distribution of arbuscular mycorrhizal fungi

The soils in this study were weakly acidic but with low to moderate nutrient and OM levels. The mean values of measured soil properties were significantly ($p \leq 0.05$) different across AEZs except for P and Na. Crop types significantly ($p \leq 0.05$) influenced all soil properties except for pH, Ca, and Mg. The effect of the interaction of AEZs and CTs was only significant ($p \leq 0.05$) on P, Na, Ca, and Mg (Table 5).

The first and the second RDA axis explained variance for 71 and 18%, respectively (Figure 4). According to the lengths of the arrows and the angles among them, OM, N, and pH had a strong positive correlation with the sporulation of *Archaeospora*, *Entrophospora*, *Glomus* and *Scutellospora*, and strong negative correlation with the sporulation of *Acaulosopra* and *Gigaspora*. Clay, K, Ca, and P had slight positive effects on the sporulation of *Archaeospora*, *Entrophospora*, *Glomus* and *Scutellospora*, and slight negative effects on the sporulation of *Acaulospora* and *Gigaspora*, because the arrows representing them are relatively short. Sand also had slight positive effects on the sporulation of *Acaulospora*, *Archaeospora* and *Entrophospora*, and slight negative effects on the sporulation of *Glomus*, *Scutellospora* and *Gigaspora*. Based upon the direction of the arrows, silt and Mg had a strong positive correlation with the sporulation of *Acaulospora*, *Gigaspora*, *Scutellospora* and *Glomus*, and a strong negative correlation with the sporulation of

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TABLE 3 Mean spore abundance of arbuscular mycorrhizal fungi in 50 g^{-1} air-dried soil across AEZs and CTs.

AEZ	CT	AMF genera					
		Glomus spp.	*Acaulospora* spp.	*Scutellospora* spp.	*Archaeospora* spp.	*Entrophospora* spp.	*Gigaspora* spp.
Southern and Eastern Lake Kyoga Basin (SELKB)	Fallow	58	45	31	23	35	15
	Legumes	104	51	43	17	33	8
	Sorghum	47	35	21	14	24	5
	Sweet potato	77	51	32	25	34	11
	p-value	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*
Mt. Elgon High Farmlands (MEHF)	Fallow	72	39	46	12	22	22
	Legumes	55	29	26	3	11	11
	Sorghum	117	38	47	1	20	15
	Sweet potato	33	25	20	14	15	5
	p-value	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*
Lake Victoria Crescent (LVC)	Fallow	81	46	47	22	32	26
	Legumes	82	39	36	13	22	9
	Sorghum	71	36	37	12	22	16
	Sweet potato	80	31	28	8	15	8
	p-value	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*
		F-test (*p-value*)					
AEZ		*Ns*	*Ns*	*Ns*	*Ns*	*0.0057*	*Ns*
CT		*Ns*	*Ns*	*Ns*	*Ns*	*Ns*	*0.0122*
*AEZ*CT*		*Ns*	*Ns*	*Ns*	*Ns*	*Ns*	*Ns*

Ns, not significant (p>0.05).

Archaeospora and *Entrophospora*. Sodium had a strong positive correlation with the sporulation of *Glomus*, *Scutellospora* and *Gigaspora*, and a strong negative correlation with the sporulation of *Acaulospora*, *Archaeospora* and *Entrophospora*.

4 Discussion

The high spore abundance of *Glomus* spp. And *Acaulospora* spp. Across AEZs and CTs may be attributed to their high frequency of hyphal fusions that plug into compatible extraradical networks and hence immediate access to host plants and subsequent spore formation (58). Glomus and *Acaulospora* produce more spores in a shorter time than *Scutellospora* and *Gigaspora* in the same environment (23, 35, 36). Gigasporaceae species produce large spores (260 to 440 µm for *Gigaspora margarita*) that require a longer developmental period than small spores (59). Hence, their occurrence can be further ascertained through the study of AMF diversity in the root systems since spore extraction alone may miss species that may have not sporulated. The insignificant response of AMF genera except for Entrophospora to changes in AEZs could be due to quite similar soil properties attributed to similar agronomic practices carried out in the fields (Table 3). The total and mean spore abundance in this study (involving annual crops) were high compared to soils dominated by perennial crops and frequently supplied with inorganic fertilizers and pesticides (23, 34). The high spore abundance can be attributed to increased sporulation caused by frequent soil disturbance in annual crops fields. However, trap cultures were necessary to observe the composition of AMF as evidenced by several authors who reported fewer species from direct isolation from local soil and more species after trapping (26).

Crop type did not significantly affect AMF due to the similar conventional tillage carried out in the fields with the different crops (Table 3). The slight significant difference between fallow and sweet potato in MEHF and LVC was because fallows had been rested for an average of 6 months allowing for the recolonization and sporulation of AMF. Results showed that *Gigaspora* spp. Abundance was highest in the fallow fields (Table 3) which confirmed the results of Gai et al. (60) that *Gigasporaceae* spp. Are more often associated with wild plants than open fields. It also

TABLE 4 Rank of total number of arbuscular mycorrhizal fungi spores per genera and their proportions in 50 g^{-1} air dried soil.

Agro-ecological zone	AMF genera	Rank	Total number of spores	Proportion (%)
Southern and Eastern Lake Kyoga Basin (SELKB)	*Glomus* spp.	1	1332	36
	Acaulospora spp.	2	791	21
	Scutellospora spp.	3	581	16
	Entrophospora spp.	4	541	14
	Archaeospora spp.	5	327	9
	Gigaspora spp.	6	162	4
	Total		3734	100
Mt. Elgon High Farmlands (MEHF)	*Glomus* spp.	1	1134	38
	Scutellospora spp.	2	612	20
	Acaulospora spp.	3	571	19
	Entrophospora spp.	4	291	10
	Gigaspora spp.	5	263	9
	Archaeospora spp.	6	130	4
	Total		3001	100
Lake Victoria Crescent (LVC)	*Glomus* spp.	1	2398	39
	Acaulospora spp.	2	1167	19
	Scutellospora spp.	3	1118	18
	Entrophospora spp.	4	673	11
	Archaeospora spp.	5	406	7
	Gigaspora spp.	6	380	6
	Total		6142	100

Relative proportion (%) of the AMF across agroecological zones depended on the spore abundance of the identified genera.

TABLE 5 Selected soil physical and chemical properties across three AEZs and four CTs.

AEZ	CT	Soil properties										
		pH (H_2O)	OM (%)	Total N (%)	Extractable P (mg kg^{-1})	K^+	Na^+	Ca^+	Mg^+	Sand (%)	Clay (%)	Silt (%)
						cmol(+) kg^{-1}						
SELKB	Fallow	6.14± 0.17	1.35± 0.49	0.13± 0.04	21.21± 20.00	0.46± 0.12	0.46± 0.14^{a}	7.46± 3.66	2.05± 0.57	70.00 ± 6.93	14.00 ± 7.12	16.00 ± 1.63
	Legumes	6.07± 0.58	3.24± 2.44	0.12± 0.04	11.83± 7.11	0.64± 0.36	0.33± 0.10^{ab}	6.46± 3.98	1.99± 0.59	68.00 ± 10.30	19.00 ± 7.55	13.00 ± 5.16
	Sorghum	5.89± 0.46	4.12± 2.51	0.12± 0.03	7.18± 1.45	0.54± 0.28	0.26± 0.06^{b}	4.02± 1.02	1.46± 0.54	69.75 ± 7.22	14.50 ± 5.97	15.75 ± 5.56
	Sweet potato	5.93± 0.41	2.98± 2.00	0.12± 0.03	15.11± 12.09	0.61± 0.31	0.37± 0.10^{ab}	5.44± 2.06	1.92± 0.62	70.83 ± 5.31	16.00 ± 5.83	13.17 ± 4.19
	Mean	*6.03^{A}*	*3.03^{B}*	*0.12^{B}*	*13.26^{A}*	*0.59^{B}*	*0.35^{A}*	*6.01^{AB}*	*1.90^{B}*	*69.19^{A}*	*16.93^{B}*	*13.89^{B}*
	p-value	*NS*	*NS*	*NS*	*NS*	*NS*	*0.0308*	*NS*	*NS*	*NS*	*NS*	*0.0158*

(Continued)

TABLE 5 Continued

AEZ	CT	Soil properties										
		pH (H_2O)	OM (%)	Total N (%)	Extractable P (mg kg^{-1})	K^+	Na^+	Ca^+	Mg^+	Sand (%)	Clay (%)	Silt (%)
MEHF	Fallow	6.14± 0.25	1.40± 0.40^{b}	0.19± 0.14	18.25± 10.36	0.44± 0.25	0.40± 0.16	6.32± 2.44	2.05± 0.61	69.43 ± 8.06	14.86 ± 7.20	15.71 ± 2.43
	Legumes	5.85± 0.36	3.31± 1.40^{a}	0.11± 0.03	19.51± 16.03	0.45± 0.24	0.22± 0.11	9.91± 7.30	3.38± 2.37	72.62 ± 7.23	12.54 ± 2.85	14.85 ± 6.16
	Sorghum	6.23± 0.83	2.65± 1.47^{ab}	0.08± 0.01	12.76± 9.57	0.56± 0.30	0.27± 0.17	8.89± 5.97	3.27± 1.48	69.33 ± 1.15	12.67 ± 5.77	18.00 ± 6.93
	Sweet potato	6.11± 0.35	2.73± 1.48^{a}	0.10± 0.03	20.42± 17.39	0.38± 0.15	0.24± 0.10	7.88 ± 4.12	2.74± 1.26	70.40 ± 8.17	12.80 ± 3.63	16.80 ± 5.22
	Mean	*6.01^{A}*	*2.66^{B}*	*0.13^{B}*	*18.64^{A}*	*0.45^{B}*	*0.27^{A}*	*8.54^{A}*	*2.93^{A}*	*71.07^{A}*	*13.14^{B}*	*15.75^{B}*
	p-value	*NS*	*0.0016*	*NS*	*NS*	*NS*	*NS*	*NS*	*NS*	*NS*	*NS*	*NS*
LVC	Fallow	6.64± 1.71	1.88± 0.78^{b}	0.33± 0.16^{a}	42.36± 41.61	0.51± 0.14	2.29± 2.18^{a}	13.01 ± 7.07^{a}	2.97± 0.84^{a}	52.00 ± 11.40	26.80 ± 10.45	21.20 ± 7.82
	Legumes	6.34± 0.50	7.13± 1.27^{a}	0.16± 0.03^{b}	11.09± 7.53	1.08± 0.34	0.19± 0.06^{b}	4.22± 1.98^{b}	1.38± 0.65^{bc}	50.31 ± 7.02	21.38 ± 5.11	28.69 ± 6.55
	Sorghum	6.32± 0.45	6.57± 3.06^{a}	0.14± 0.03^{b}	11.56± 6.64	0.96± 0.36	0.18± 0.15^{b}	2.59± 0.85^{b}	0.89± 0.34^{c}	53.83 ± 10.13	21.83 ± 7.00	24.33 ± 7.71
	Sweet potato	6.36± 0.32	4.28± 3.34^{ab}	0.18 0.02^{b}	29.55± 27.91	0.72± 0.34	0.30± 0.14^{b}	6.21± 3.80^{b}	2.06± 0.92^{ab}	51.83 ± 14.51	23.67 ± 14.05	24.50 ± 11.20
	Mean	*6.39^{A}*	*5.57^{A}*	*0.19^{A}*	*19.76^{A}*	*0.89^{A}*	*0.54^{A}*	*5.76^{B}*	*1.68^{B}*	*51.60^{B}*	*22.83^{A}*	*25.73^{A}*
	p-value	*NS*	*0.0041*	*0.0146*	*NS*	*NS*	*0.0208*	*0.0044*	*0.0008*	*NS*	*NS*	*NS*
		F-test (p-value)										
AEZ		0.0421	<.0001	0.0002	NS	<.0001	NS	0.0309	0.0003	<.0001	<.0001	<.0001
CT		NS	<.0001	<.0001	0.0028	0.0070	0.0005	NS	NS	NS	NS	NS
AEZ* CT		NS	NS	NS	0.0198	NS	0.0005	0.0154	0.0429	NS	NS	NS

Values=Mean ± standard deviation; LSD= Least significant difference; means followed by the same lower-case letters in the same column are not significantly different (p>0.05); means followed by the same upper-case letters in the same column are not significantly different (p>0.05); NS= not significant (p>0.05). The soils sampled from SELKB were sandy loam (14 fields), sandy clay loam (4 fields), and loamy fine sand (1 field); those sampled from MEHF were sandy loam (15 fields), sandy clay loam (4 fields), and loamy fine sand (1 field); while those from LVC were sandy clay loam (8 fields), loam (9 fields), sandy loam (3 fields), sandy clay (2 fields), clay (2 fields), and loamy fine sand (1 field). Soil textural classes for SELKB, MEHF and LVC were generally sandy-loam, sandy-loam and sandy-clay-loam, respectively across CTs.

confirmed that the 6 months of fallowing was enough time for the development of their large spores (59).

The varying effects of sodium and magnesium on AMF genera in the present study (Figure 4) may be attributed to varying adaptability of AMF genera to varying levels of exchangeable bases (61). The positive effect of nitrogen on the spore abundance of *Glomus*, *Scutellospora*, *Entrophospora*, and *Archaeospora* might have been indirect through the increased supply of photosynthates from the host-crop to the fungi. Availability of soil N increases mycorrhizal activity (62) and hence its positive effect on the AMF genera.

It was expected that sporulation of all AMF genera is positively affected by organic matter since it is considered a source of energy for the growth and functioning of the fungi. However, the effect of organic matter on AMF growth and sporulation depends on the efficiency of the individual species in acquiring resources from the organic matter (63). Ng et al. (64) also reported that the chemical nature of soil carbon drives the structure and functioning of soil microbial communities which may vary from one AMF type to another. For example, pure cellulose obtained after proper decomposition increases asymbiotic AMF extraradical hyphae growth and root colonization (18, 65).

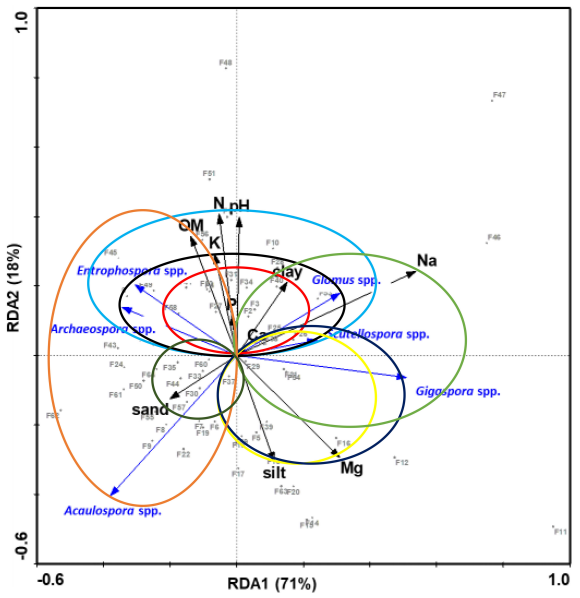

FIGURE 4
RDA (canonical correlation analysis) biplot of the 6 arbuscular mycorrhizal fungi genera (*Glomus, Scutellospora, Gigaspora, Acaulospora, Archaeospora* and *Entrophospora*) and soil properties from the sampled fields. The circle's diameter is equal to the length of the soil property's arrow. Genera lines that end in that circle have a positive regression for that soil property. The length of the arrow indicates increasing influence and the smaller the angle between the two arrows indicates closer relationships.

Soil texture has been reported to alter colonization of AMF (8, 66) depending on the sporulation patterns. The general soil textural classes in the present study were sandy clay loam and sandy loam which are reported to favor mycorrhizal development (67). Gigasporaceae (*Gigaspora* spp. And Scutellospora spp.) dominate in sandy soils (68) and they are also indicators of soils with lower clay content (69). *Glomaraceae* and other AMF families with small spores do not show any strong dependence on soil characteristics (70). However, Lekberg et al. (68) also reported that *Scutellospora cerradensis (Sc. Rtl)* was the only member of Gigasporaceae that occurred predominantly in clayey soil (containing 41:11:48 of clay, silt, and sand). From the same study they reported that Glomaraceae colonized roots well in sandy soil (containing 3:7:90 of clay, silt, and sand), clayey soil (containing 41:11:48 of clay, silt, and sand) and sand/clay mixture (containing 4:1 v/v). There's a possibility that the difference in biomass allocation and the growth patterns of extraradical hyphae of *Gigasporaceae* and *Glomaraceae* are affected by soil texture (71, 72).

Most importantly, the soil pH was within a favorable range (5.85-6.64) for fungi and supported sporulation of especially, *Glomus, Scutellospora, Entrophospora*, and *Archaeospora*, and negatively affected the sporulation of *Acaulospora* and *Gigaspora*. Earlier, Muchane et al. (28) reported that AMF were favored by pH 5.51 to 6.67, while Dobo et al. (73) reported pH 6.18 to 6.28 to increase AMF sporulation in agricultural soils. However, it was expected that *Acaulospora* spp. Would be positively affected by the present pH since Acaulosporaceae are tolerant to acidic tropical soils (26, 74) and they highly sporulate under conventional tillage (a common practice in the sampled sites) as compared to other AMF families (31). For example, *Acaulospora laevis* is predominat in low pH soils and germinates well at pH 4-5 while *Gigaspora* heterogama isolated from warm climates and maintained in tropical areas showed varying germination rates (8 to 78%) in the same environmental conditions (75).

The six AMF genera identified in this study were present in all the AEZs and CTs but differences in their spore production were not significant. The AMF genera observed in this study were lower than 12 isolated in Sudan (76), 15 in Ethiopia (24), 15 in Southern China (77), 9 in Ethiopia (73) but higher than the 4 to 5 genera isolated in Kenya (23, 28, 34) and 4 in Rwanda (78). This may be due to variances in edaphic conditions and the cropping systems of the study sites. However, *Glomus*, *Acaulospora*, *Scutellospora*, and *Gigaspora* reported in this study were also observed in the studies carried out in the Sub-Saharan Africa (SSA) countries mentioned above. Incidentally, there were no new AMF genera identified, which alludes to low composition in these AEZs with the mean spore abundance of only *Entrophospora* spp. In SEKLB being significantly higher than in MEHF.

Most of the commercial mycorrhizal bioinoculants available in SSA are imported, expensive, and majorly contain *Glomus* species. During efficacy testing of these strains, the environmental conditions of the origin of the strains and where the bioinoculant is to be used may not be compared (79) yet it is important for the adaptability of AMF in the different local SSA edaphic and climatic conditions (80). The population of the introduced strains must build up for their increased competitiveness and effectiveness. In Tchabi et al. (81), AMF from Tropical Africa (*Glomus hoi*, *Acaulospora spinosa*, *Glomus mosseae*, *Glomus etunicatum*, and *Acaulospora scrobiculata*) averagely led to increased yam tuber growth by 51, 49, 38, 38 and 31%, respectively. Whereas exotic species from Europe were less efficient except for the three isolates of *G. clarum* isolates which increased tuber yield by 8.8, 15.6 and 55.6%, respectively compared to non-mycorrhizal control. Additionally, soil (from yam field) increased tuber yield by 40, 33, and 20% compared to exotic *G. constrictum*, the non-mycorrhizal control, and the exotic *G. luteum* confirming the existence of superior native AMF species in yam producing regions. Therefore, to overcome the problem of local adaptability of imported species and cost implications, mycorrhizal bioinoculants can be produced locally from the native species observed across different locations in SSA. The physiological characteristics of the species determine to a greater extent their survival and activity in the soil. Hence, different species will show varying responses, in terms of survival and activity. The ability of AMF to enhance root surface area by hyphal growth and provide an extra route for uptake as mycorrhizal pathway (82) depends on the AMF strains colonizing the plant roots. The efficiency of AMF strains is influenced differently by their development and activity of the external hyphae, hyphal transport rates, and solute interchange at the arbuscule-host root cell interface (83, 84). Therefore, the mixing of different compatible AMF strains during bioinoculant production will promote the complementary benefits of AMF strains in crop production.

Plant-host generalist AMF strains should be considered for bioinoculant production as compared to plant-host specialist AMF strains since during fallowing the latter can colonize weeds or new

crops which eventually can serve as a source of additional spores (85). The compatibility of the sporogenous *Glomus* and *Acaulospora* species with different crops and environmental conditions require further testing for suitable local bioinoculant production. However, *Acaulospora* spp. Would greatly influence crop production in acidic tropical soils since the *Acaulosporaceae* species are tolerant to acidic soils (26, 74); highly effective in P-uptake and transfer to the host plant compared to *Glomeraceae* species (86); and they highly sporulate under conventional tillage (a common practice in sweet potato producing areas) as compared to other AMF families (31).

Glomus and *Acaulospora* species can be further identified using molecular techniques and their compatibility with different crops and environmental conditions in varying dosage tested for suitable local bioinoculant production. To reduce the cost of soil fertility amending inputs in crop production, bioinoculants containing the most effective species can be integrated with reduced rates of inorganic fertilizers.

5 Conclusion

A total of six AMF genera comprising of *Glomus, Acaulospora*, *Scutellospora*, *Entrophospora*, *Archaeospora*, and *Gigaspora* were isolated from the study sites. The composition and spore abundance of AMF recorded in the AEZs and CTs in the major sweet potato growing areas had limited significant differences due to the similar agricultural practices employed by farmers. The effect of soil parameters on AMF spore abundance varied from genera to genera, however, the strongest influence was by OC, N, pH, silt, Mg and Na. The most dominant AMF i.e., *Glomus* and *Acaulospora* species can be isolated for local bioinoculant production.

Data availability statement

The original contributions presented in the study are included in the article/supplementary materials. Further inquiries can be directed to the corresponding author.

Author contributions

RM conceived the research idea and conducted the research, collected, and analyzed the data, and co-wrote the manuscript; JT, PE, and CM assisted with conceiving the research and critically reviewed the manuscript. All authors contributed to the article and approved the submitter version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential competing interest.

Publisher's note

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