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Yield Performance of Wheat Genotypes (*Triticum aestivum* L.) and Occurrence of Native Arbuscular Mycorrhiza Fungi in Contrasted Conditions of the Bimodal Humid Forest Zone of Cameroon

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Abstract Increased temperature and degradation of soil fertility are the principal constraints affecting wheat production in Cameroon. As such, this study aimed to determine the potential of native Arbuscular Mycorrhiza (AM) fungi in yield performance and variability of wheat genotypes cultivated in contrasted conditions of the bimodal humid forest zone of Cameroon. For this, 34 wheat genotypes were sown following an incomplete alpha-lattice design. The genotypes were evaluated on the basis of their yield potential (grain weight) and their affinity to mycorrhiza symbiosis. The efficiency of the occurred mycorrhiza was verified on two wheat genotypes and compared to the standard chemical fertilization used and a control test. Wheat grain weight, shoot dry mass, number of leaf, shoot length and leaf area were collected for this purpose. Biplot analysis revealed positive significant correlations ($r=0.83$ $P<0.01$; $r=0.77$, $P<0.01$) between the grain weight of wheat genotypes and mycorrhiza parameters (percentage of AM colonization and number of AM spores respectively) in Mbankolo (high altitude) and positive significant correlations ($r=0.56$, $P<0.01$; $r=0.57$, $P<0.01$) between the same parameters in Nkolbisson (low altitude). The grain weight and mycorrhiza parameters explained 90.1% and 71.3% of variance among all the wheat genotypes in high and low altitudes respectively. The following genotypes SST015, SST087, SST866 and SST88 in high altitude and the following genotypes Nd643-5, Kenya2, Babax8 and SST895 in low altitude were identified to have the highest mycorrhiza symbiotic affinity and yield performances. The mycorrhiza spores isolation and morphological identification revealed the presence of three species in wheat rhizosphere in both study sites: *Scutellospora sp*, *Gigaspora sp*, and *Septoglomus sp*. The species *Scutellospora sp*, was identified to be present and dominant in both study sites. These latter were observed to have a high growth and yield enhancing ability on wheat varieties compared to the conventional chemical method of fertilization hence could be use as efficient bio-fertilizer. Nonetheless, it will be necessary to perform molecular phylogeny to identify each AM specie in order to individually evaluate their efficiency on wheat growth and yield. Also, the targeted wheat genotypes can be used to promote large scale production in both sites and vulgarize to local farmers. Additionally, the determination of the mechanism underpinning their symbiotic preferences will be essential in targeting the genes implicated to be used for breeding purposes.

Keywords Bio-fertilizer, Wheat symbiotic affinity, Genotype variability, Yield

1. Introduction

Wheat is the second most consume cereal after maize and rice with a world annual production of approximately 766 million ton (FAO, 2019). Wheat represents one of the main sources of food security in Africa (Negassa et *al*., 2013). In Cameroon, its demand has increase along the years with an increase in wheat consumption of 98% in urban strata and 90-91% in non-urban strata (Engle-Stone and Brown, 2015). However, 100% of the total domestic consumption is satisfied by import translating Cameroon vulnerability to food security risk (PAM, 2011). In Cameroon, the production which is almost non-existent is nonetheless estimated at 900 tons annually (FAO, 2019), Highly inferior to the national importation estimated at 725,000 tons in 2015

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(Mocauley et Ramadjita, 2015). To satisfy this population demand, the government allocated an amount of 103 billion of FCFA to import approximately 518, 000 tons in 2012 (Yamdjeu, 2012). Facing this situation, the Cameroon government is urge to implement appropriate strategies to reduce the wheat dependency, to minimize price fluctuation and attain a minimum threshold for self-sufficiency for this commodity.

Increase temperature has been the major constraint responsible for 58-92% decrease in wheat yield in this sub region. Evaluations to adaptation and breeding programs have been conducted by local researchers to obviate this constraint (Ayuk-takem, 1983). However, these trials have seriously decreased the soil productivity capacity along the years due to the use of conventional practices. An alternative solution might be the application of natural method of fertilization, which will maintain at the same time a good soil health and enhance tolerance of wheat plant to high temperature. But none of such has yet been done in Cameroon.

Arbuscular mycorrhizae also called endomycorrhizae are the most ancient, widespread and common soil denizens, that establish an association with the root of 80% of vascular plant species (Wang and Qiu, 2006). Arbuscular Mycorrhizal (AM) fungi are endophytes that constitute the phylum of Glomeromycota. They are ubiquitous in soil around the globe and have been associated with improve plant growth for over 100 years (Hamel, 1996; Singh and Adholeya, 2013). These symbiotic fungi are the main components of the soil micro-biota in most of the agroecosystems and account for 25% of the biomass of soil micro flora and micro fauna combined (Singh and Adholeya, 2013). Mycorrhizae are mutualistic symbiotic associations based on bidirectional nutrients transfer between soil fungi and the roots of vascular plants, where the plant supplies the fungi with sugars produced by photosynthesis, while the hyphae network improves the plant capacity to absorb water and nutrients (Roy-Bolduc and Hijri, 2011). AM fungi are obligate biotrophs since the rely on their host plant to proliferate and survive (Pozo and Azcón-Aguilar, 2007). They are also known to provide several ecosystem services including promoting plant growth, nutrient uptake and enhancing soil stability (Velázquez *et al*., 2016), improvement of water retention through mycorrhizal symbiosis (Bernardo *et al*., 2019) and confer to crops resistance to biotic and abiotic stress (Augé and Moore, 2005; (Filho *et al.*, 2017).

This enhancing ability of Arbuscular Mycorrhizal fungi on plant growth and adaptation is generally determined by the degree of symbiotic compatibility, which is affected by the host plant and fungi species (Silva *et al*., 2018). This evidence was also proven by (Munkvold *et al.*, 2004; Stover *et al.*, 2012; Monier *et al.*, 2017), who emphasized on the large functional variability among distinct AM fungi species and fungi isolates of the same species. Hence the continuous study of Arbuscular mycorrhizal diversity is a necessity in order to target the predominant species and their corresponding host. This has justify the extensive studies of AM fungi diversity in natural ecosystems. However, few studies have been conducted in order to appreciate the diversity and influence of these indigenous mycorrhizal fungi in plant growth and yield in agricultural lands (Singh and Adholeya, 2013; Loit et al., 2018). Indeed, agricultural lands are constantly subjected to human intervention which tends generally to reduce AM fungi diversity, their colonization ability thereby reducing their impact in crop adaptation and yield (Singh and Adholeya, 2013; Loit et al., 2018). Therefore, there is an urgent need in exploring these indigenous mycorrhiza diversities, their influences in increasing crops adaptations and yield, which might be highly informative on their presence, host specificity and symbiotic compatibility with most of the cultivated crops in agricultural lands in view of promoting good fertilization processes.

In Cameroon, Kamko et *al*. (2020) showed that *Gigaspora margarita* is an efficient mycorrhiza species, which can be used as an effective bio-fertilizer for the domestic propagation of *Prunus africana*. Mbogne *et al.* (2015), studied the diversity of AM fungi of pumpkins, and isolated two AM fungi species *Glomus* and *Acaulospora*, with *Acaulospora* identified to be the abundant species. Nzweundji *et al.* (2015), identified three families associated to *Prunus africana,* which included *Gigasporaceae* (*Gigaspora margarita*), *Acaulosporaceae* (*Acaulospora tuberculate, A. longula* and *Enthrophosphora colombiana*) and *Glomeraceae* (*Glomus manihotis and G. etunicatum*). Bechem and Alexander, (2012), showed that *Gnetum Spp*. had a high preference to ectomycorrhizal colonization of genus *Scleroderma* (*Scleroderma sinnamariense*), which may highly contribute to domestication efforts. Therefore, undergoing this same purpose in contrasted wheat cultivated areas might be highly informative in highlighting not only the mycorrhizal diversity and their influence in wheat yield capacity but also inform of the degree of wheat symbiotic need in constraint environments. Since plant mostly adhere to symbiotic association when facing high level of abiotic stress. This study might be a promising step for the restoration of soil productivity capacity in wheat agricultural areas and also for the improvement of wheat productivity in sub-Saharan Africa and Cameroon in particular.

Furthermore, a recent report made by (Sawers *et al.*, 2008), mention the fact that, in a study comparing the performance of wheat varieties developed before and after 1900, varieties developed before 1900 were more responsive to AM colonization than those developed later and it has been suggested that plant breeding has selected against AM association. However, in their report, it was mention that the observed differences in responsiveness were largely determined by an increased ability of modern lines to take up phosphate without AM fungi (i.e. a reduction in dependence, not a loss of compatibility with the fungus). This signifies cultivating wheat with a high level of chemical input. But high level of inputs decreases soil fertility along the years. Thus investigating the influence of

mycorrhizal symbiosis on wheat yield and symbiotic preference in cultivated land will be a promising introduction to encourage plant breeders to put in place new wheat lines with increase in dependency to AM colonization. This justifies the aim of this study, whose purpose was to determine the potential of native Arbuscular Mycorrhiza (AM) fungi in yield performance and variability of wheat genotypes cultivated in contrasted conditions of the bimodal humid forest zone of Cameroon.

2. Materials and Methods

Field experiment

Table 1. Wheat genotypes (*Triticum aestivum* L.) selected for this study

No	Lines	Origins	No	Lines	Origins
1	Atilla4	South Africa	18	Nd643-2	South Africa
2	Babax1	South Africa	19	Nd643-5	South Africa
3	Babax10	South Africa	20	Pfunye1	South Africa
4	Babax11	South Africa	21	Premio1	South Africa
5	Babax12	South Africa	22	Premio3	South Africa
6	Babax13	South Africa	23	SST015	CIMMYT
7	Babax14	South Africa	24	SST027	CIMMYT
8	Babax15	South Africa	25	SST056	CIMMYT
9	Babax17	South Africa	26	SST087	CIMMYT
10	Babax2	South Africa	27	SST806	CIMMYT
11	Babax7	South Africa	28	SST835	CIMMYT
12	Babax8	South Africa	29	SST843	CIMMYT
13	Croc_1	South Africa	30	SST866	CIMMYT
14	Kenya2	South Africa	31	SST88	CIMMYT
15	Kenya4	South Africa	32	SST895	CIMMYT
16	Kronstad F2004-1	South Africa	33	Waxwing1	South Africa
17	Nd643-1	South Africa	34	Wbll3	South Africa

The experiment was conducted on two sites of different altitudes of the bimodal humid forest zone of Cameroon. The high altitude site of 1057m above the sea level, with average temperature and rainfall of 17℃ and 1592.2 mm respectively, was located in Mbankolo mountainous zone. This site had a relative humidity of 77.53%. The low altitude site standing at 650m above the sea level with average temperature and rainfall of 23.5℃ and 1560 mm respectively was located in lowland of Nkolbisson zone, with a relative humidity of 62%. A total of thirty-four (34) wheat genotypes were evaluated during a period of March-August 2016. Accessions were collected in Africa, Mexico and some from *international Maize and wheat improvement center (*CIMMYT) (Table.1). These genotypes were evaluated in an incomplete alpha-lattice design with 3 repetitions. The wheat genotypes were scored on the basis of the grain weight (Wgr), which was collected after harvest using a four digits' unit weight balance (KERN, Germany).

Samples collection

The rhizosphere of each wheat genotype was sampled per block in each repetition following the experimental design. With the help of a core sampler, samples of 500g of soil (rhizosphere and roots) from a depth of 0-30cm were collected in the rhizosphere of four wheat plant randomly selected for each wheat genotype at the time of harvest and mixed up to form a composite sample. That gave a total of 34 samples obtained from each study sites. The samples were dried in a shade with no moisture and placed in zip plastic bags. These samples (rhizosphere and root) were stored at 4℃ and were subsequently used to undergo AM fungal isolation and quantification. Some of the collected wheat root were washed with distilled water and stored in 70% ethanol and were later on used for observation of the percentage fungi colonization.

Soil analysis

A portion of soil samples from each plot were mixed, ground and sieved through a filter of 2mm. Soil organic carbon (C total), was determined by calorimetric method after oxidation with H_2SO_4 and $K_2Cr_2O_7$; total nitrogen (total N) by Kedjahl digestion, total phosphorus (total P) by dosage in continuous flow and available phosphorus by the Olsen method (Olsen, 1954). Soil pH was measured in 1/2.5 ratio of soil to distilled water. Soil analyses were performed at LASPEE (Laboratoire d'analyse sol, plante et engrais / Soil, plant, water and fertilizer laboratory) at the Institute of Agricultural Research for development (IRAD) Nkolbisson, Cameroon.

AMF spore isolation, quantification and identification

AM spores were isolated from 100g of soil obtained from the conserved plastic bags. The soil was wet sieved in filters of different dimensions and decanted, the supernatant was then introduced in a petri dish where the spores were observed under a dissecting microscope (HUVITZ, Korea) following the method described by Gerdemann and Nicolson, (1963). The spores were counted in a squared petri dish following the formula: number of spores in each residue of 100g = 69.4/2.5 × n (where n is the number of spores observed in the squared area of the squared petri dish) described by (Ngonkeu, 2003). For proper identification of different spores parts (spore membrane, hyphae, suspensory bulb), each spore type was mounted in polyvinyl-lactic acid glycerine (PVLG) (Brundrett et *al.*, 1994) and the polyvinyl mixed with Melzer's reagent (Koske and Tessier, 1983). Spore morphology was compared with on-line description of the International Culture Collection of Vesicular Arbuscular Mycorrhizal Fungi (INVAM: http//www.invam.caf.wdu.edu/), and genus deduce following Morton, (1988) genus description. The identified genus were also compared to the Oehl, de Souza & Sieverding, (2008), revision of *Scutellospora* and description of five new genera and three new families in the arbuscular mycorrhiza-forming Glomeromycetes.

Intraradical colonization by AM Fungi

The harvested root of the wheat lines collected in both study area, were used to undergo intraradical colonization study. The staining technique used was that preconized by (Phillips and Hayman, 1970); (Brundrett *et al*., 1996), which consisted of staining the root with 0.05% tryptan blue for 30min, after clearing of the root with 10% of KOH for 15min at 90℃. The percentage of colonization was calculated using the principle of colonization divided by the total number of segments examined as described by Bhuvaneswari *et al*., (2014). Pi = ni/n, gives the relative abundance of the specie on the site, where (ni) is the proportion of individual of one particular specie divided by the total number of individuals found (n) (Legendre and Legendre, 1984).

Evaluation of the efficiency of the occurred mycorrhiza

A pot experiment was conducted in the Regional Biological control and Applied Microbiology laboratory at the Institute of Agricultural Research for Development (IRAD). Sterilized substrate was introduced in polythene bags (29×23) cm of length and width respectively. The occurred mycorrhiza spores were purified through a purification and pre-germination process; which consisted briefly of introducing the identified spores into Eppendorf tubes for sterilization with 3ml of 2% chloramine T. 3ml of streptomycin 0.025% was introduced in the tube and the mixture was stirred for 20min after removal of chloramine T and rinsed with distilled water thrice for 15min. The disinfected spores were sown on the basis of their morphotype in petri dishes containing agar medium (0.7%), at the rate of 4 spores per petri dishes with 3 replicates. The whole mixture was sealed and allowed in the dark for at least 4 days at 30℃ (Ngonkeu, 2003). These pre-germinated spores were later multiply on the roots of mycotrophic plant (*Sorghum bicolor*) on sterilized sand. For that purpose, AM species were inoculated (inoculum consisted of a mixture of the identified AM species) at 1cm above the substrate in order to verify their effectiveness on growth and yield of the highest performant wheat genotypes (SST88 and Nd643-2) identified after field evaluation in Mbankolo and Nkolbisson respectively. This AM species treatment was compared to the standard NPK fertilization (i.e 50 kg N. ha^{-1} as 34% of ammonium nitrate, 80 kg P_2O_5. ha^{-1} as granular triple superphosphate, 100 kg K_2O. ha^{-1} as potassium salt before sowing and 40 kg N. ha^{-1} as 34% ammonium nitrate at elongation phase) and a control test (no treatment + sterilized substrate). Parameters like shoot length, number of leaf and leaf area were collected at 45 and 65 days after sowing (DAS) and the grain weight and shoot dry mass were collected after harvesting. The root percentage colonization of the two wheat genotypes was determined following the method described previously.

Data Analysis

Collected parameter (grains weight), was used for analysis of variance (ANOVA). The grain weight of each genotype repetition was mean. The mean values of the wheat genotypes were compared using the Student Newman keul (SNK) test at 5% level of significance (Hsu, 1996). AM fungi parameters (number of spores and percentage colonization) collected for each wheat genotype were mean. The mean values were used to perform correlation analysis (pearson correlation), principal component analysis, grouping of the genotypes following their affinity to microbes all combined in a Biplot for principal components using ggplot2 package in R software. The wheat grain weight and root dry mass parameters collected on the two wheat genotypes in laboratory test were mean. The mean values were compared using the Duncan test at 5% level of significance following the Felipe de Mendiburu method (Hsu, 1996). The analysis of variance, comparisons of the mean values using the SNK test and Duncan test and the Biplot analysis were performed with R software of version 3.5.3.

3. Results

Soil characterization of both study sites

The soil analysis presented different characteristics on both experimental study sites (Table.2). In Nkolbisson (low altitude) study site, the soil had a pH value of 5.12, whereas in Mbankolo (high altitude) study site the soil had a pH value of 5.7 (Table.2). Nkolbisson presented low mean value of available phosphorus (4.65µg/g) compared to Mbankolo (421.051µg/g). The available nitrogen and carbon ratio (C/N) was also evaluated for both sites and the ratio of 9.07 was obtained at Nkolbisson compared to a C/N ratio value of 10.407 obtained at Mbankolo (Table.2).

Table 2. Soil characteristics of both study sites

Soil elements	Nkolbisson	Mbankolo
P (ppm or µg/g)	4.65	421.051
N total (%)	0.13	1.980
C total (%)	1.18	20.606
C/N	9.07	10.407
pH(H20)	5.12	5.70

Relation between occurred Abuscular mycorrhizal fungi, yield and wheat genotypes

Case of Mbankolo study site

A biplot was constructed to appreciate the relationship between the occurred microbes (Arbuscular mycorrhizal fungi), yield performances and wheat genotypes (Fig.1). Hence, the analysis revealed positive significant correlations (r= 0.83 and r= 0.78) between the yield performances of wheat genotypes cultivated in Mbankolo, percentage of Arbuscular Mycorrhiza fungi root colonization (CAM) and number of Arbuscular Mycorrhizal fungi (NAM) spore present in their rhizosphere respectively. It was observed that the yield performance of wheat genotypes and mycorrhiza parameters (CAM and NAM) were highly loaded in the first

principal component (PC1) which explained 90.1% variance among all the wheat genotypes. Also the clustering of wheat genotypes following their affinity to mycorrhiza parameters permitted to observe that wheat genotypes numbered 23, 26, 30 and 31, corresponding to wheat genotypes SST015, SST087, SST866 and SST88 (Table.1), of the same cluster, showed high affinity to mycorrhiza colonization. These wheat genotypes also showed high yield performances among the cultivated wheat genotypes in Mbankolo study site (Fig.2).

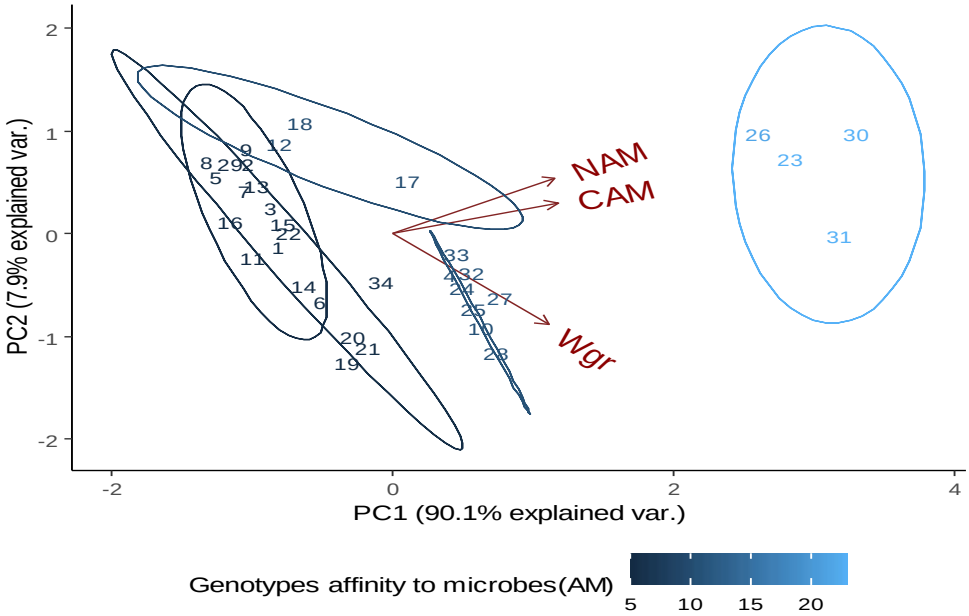

Figure 1. Biplot of yield performance of wheat genotypes related to occurred Arbuscular mycorrhiza (AM) fungi parameters in Mbankolo study site, CAM: AM colonization percentage, NAM: Number of AM spores

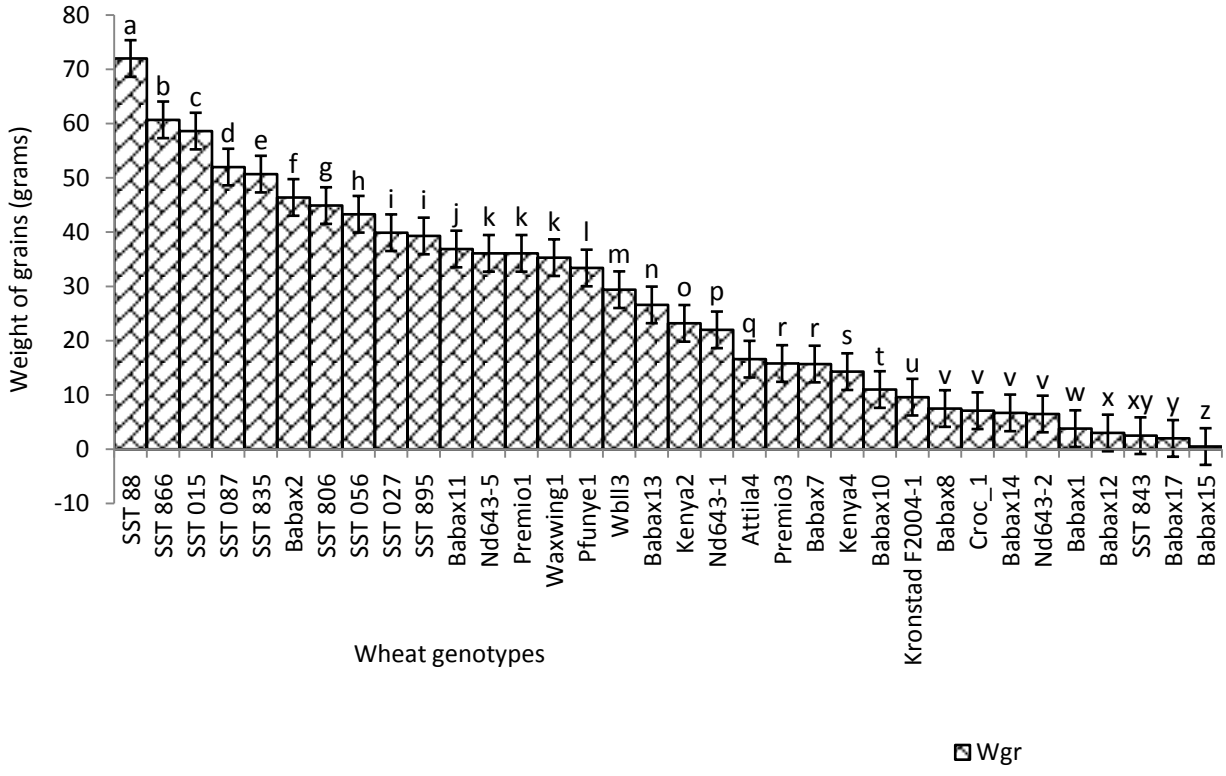

Figure 2. Yield performance of wheat genotypes in Mbankolo

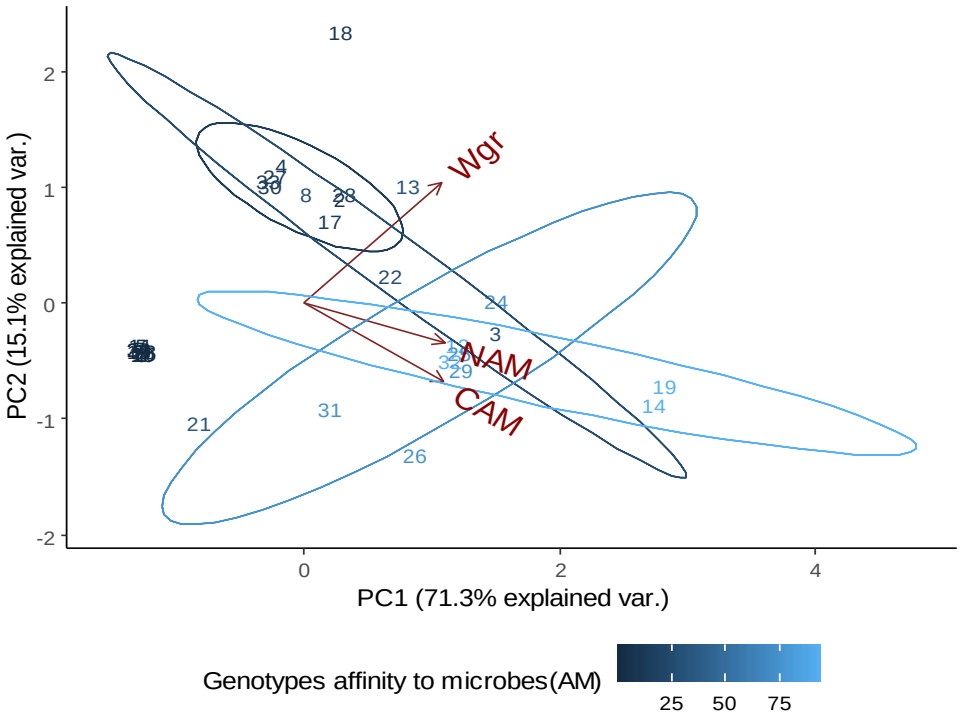

Figure 3. Biplot of yield performance of wheat genotypes related to occurred Arbuscular mycorrhiza (AM) fungi parameters in Nkolbisson study site, CAM: AM colonization percentage, NAM: Number of AM spores

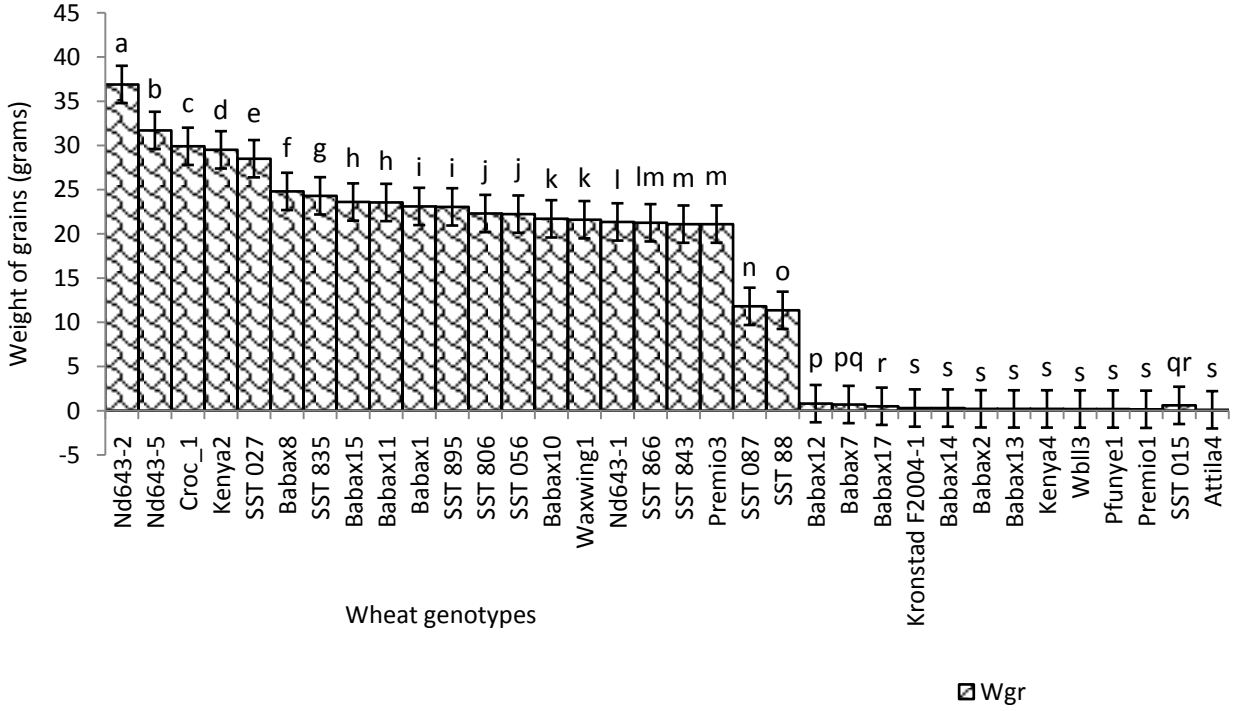

Figure 4. Yield performance of wheat genotypes in Nkolbisson

Case of Nkolbisson study site

In Nkolbisson study site, after the biplot analysis, it was observed positive significant correlations (r= 0.55 and r = 0.56) between the yield performance of wheat genotypes, percentage of the occurred Arbuscular Mycorrhizal fungi root colonization (CAM) and number of Arbuscular Mycorrhizal fungi spores in wheat rhizosphere (NAM) respectively (Fig. 3). Also, the grains weight and the mycorrhiza parameters (CAM and NAM) were observed to be highly loaded in the first principal component which

explained 71.3% of variance among the wheat genotypes. Two clusters were identified with high affinity to mycorrhiza colonization. The first cluster was composed of wheat genotypes numbered 12, 14, 19 and 32 corresponding to Babax8, Kenya2, Nd643-5 and SST895 respectively (Table.1). The second cluster was composed of wheat genotypes numbered 13, 24, 25, 26, 29 and 31 corresponding to Croc-1, SST027, SST056, SST086, SST843 and SST88 (Table.1). These genotypes in both clusters were observed to have high yield performance after evaluation in Nkolbisson study site (Fig.4).

Occurrence of AMF species in both study sites

Arbuscular Mycorrhiza (AM) fungi spores were isolated from the rhizosphere of wheat genotypes cultivated in both study sites (Fig.5). A single species (*Scutellospora sp*.) was present in the rhizosphere of wheat genotypes in Mbankolo (high altitude) with a total of 917 AM spores isolated. Main while, three species (*Gigaspora sp.*, *Scutellospora sp*. and *Septoglomus sp*.,), were observed in the rhizosphere of wheat genotypes cultivated in Nkolbisson (low altitude) with a total of 15254 AM spores isolated. Also, a single AM species (*Scutellospora sp*.) was observed to be present and dominant in the rhizosphere of wheat genotypes cultivated in both sites (Fig.4), with a relative abundance of 63.1%.

Influence of occurred mycorrhiza on wheat yield and biomass

The occurred Arbuscular Mycorrhiza (AM) fungi from both sites (Scutellospora sp and Gigaspora sp) were purified and used as bio-fertilizer. The enhancing ability of this latter on the wheat grain weight of two wheat varieties was evaluated and compared to that of chemical fertilizer (Table.3). The result obtained showed that the mycorrhiza treatment had a significant effect on grain weight on Var1 (SST88) compared to the chemical treatment, which showed non-significant difference with the control test (Table.3). This observation differs on Var2 (Nd643-2), where mycorrhiza and the chemical fertilizer showed non-significant difference on wheat grain weight, but instead, showed a significant difference with the control test which had the lowest grain weight value (Table.3).

Table 3. Influence of occurred mycorrhiza treatments on wheat grain weight

Treatments / Varieties	Mycorrhiza treatment (M2)	NPK treatment (M1)	Control test (M0)
Var1	0.85± 0.18 a	0.49±0.19 b	0.37±0.16 b
Var2	0.76± 0.45 a	0.766±0.34 a	0.12 ± 0.11 b

Values with the same letters are not significantly different at 5% level of significance

The evaluation of the influence of different treatments showed that mycorrhiza used as bio-fertilizer had a high enhancing ability on shoot dry mass of Var1 compared to the chemical fertilizer and control test, which instead showed non-significant difference (Table.4). On the other hand, non-significant difference was observed between mycorrhiza and chemical fertilizer on shoot dry mass of Var2, with the control test having the lowest effect (Table.4).

Table 4. Influence of occurred mycorrhiza treatments on shoot dry mass of wheat

Treatments / Varieties	*Mycorrhiza treatment* (M1)	NPK treatment (M2)	Control test (M0)
Var1	1.82±1.04 a	0.68±0.33 b	0.30±0.05 b
Var2	0.91±0.55 a	1.24±0.49 a	0.31±0.07 b

Values with the same letters are not significantly different at 5% level of significance

At 45 days after sowing, the mycorrhiza and chemical treatments showed non-significant difference on shoot length variable of Var1. Main while, at 65 days after sowing, the mycorrhiza treatment was observed to be significantly high compared to the chemical fertilizer, which in turn showed a high significant value compared to the control test (Table.5). This observation varied for Var2, where mycorrhiza treatment showed the highest enhancing value followed by the chemical treatment that in turn showed a significant value of shoot length compared to the control test at DAS 45. Instead at DAS65, the mycorrhiza and chemical treatments showed the highest significant values of shoot length compared to the control test (Table.5).

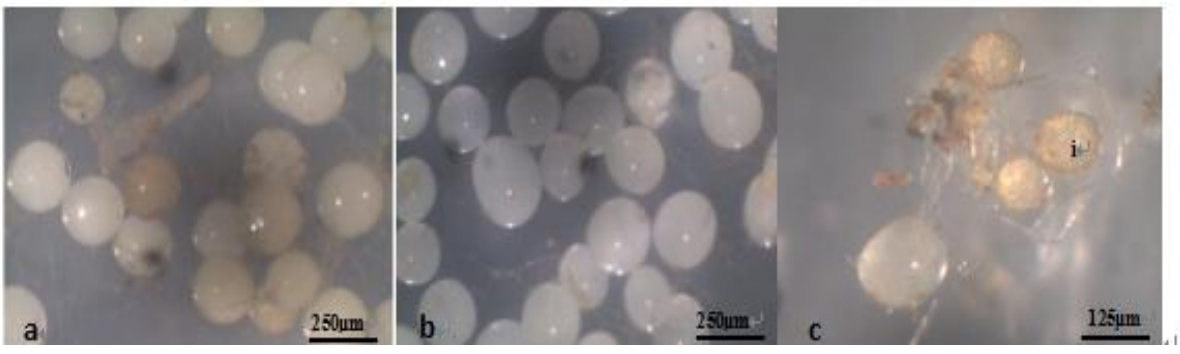

Figure 5. Some Arbuscular Mycorrhiza (AM) fungi spores isolated in wheat rhizosphere from both study sites in Cameroon, observed through a dissecting microscope; a: *Gigaspora sp.* (250µm) b: *Scutellospora sp.* (250µm) c: i.Septoglomus sp. (125µm)

Table 5. Influence of mycorrhiza treatments on growth parameters of wheat lines

DAS	Varieties	Treatments	Shoot length	No. of leaf	Leaf area
45	var1	Control	4.75 b	5.33 b	1.99 b
		Mycorrhiza	28.93 a	13.33 a	12.58 a
		NPK treatment	23.45 a	9.66 ab	8.91 a
	var2	Control	5.11 c	4.0 b	1.99 b
		Mycorrhiza	26.6 a	17.33 a	7.12 a
		NPK treatment	15.83 b	9.33a b	5.55 a
65	var1	Control	6.66 c	6.83 c	5.15 b
		Mycorrhiza	49.03 a	43.83 a	17.97 a
		NPK treatment	36.48 b	32.16 b	14.98 a
	var2	Control	5.283 b	7.5 b	7.07 a
		Mycorrhiza	40.73 a	36.16 a	9.51 a
		NPK treatment	34.53 a	25.66 a	9.83 a

Values with the same letters are not significantly different at 5% level of significance

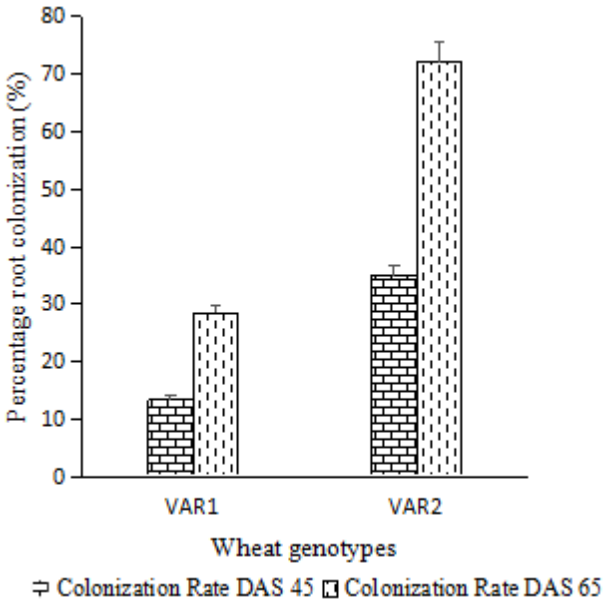

Figure 6. Root percentage colonization of wheat genotypes

A least significant difference was observed between the mycorrhiza treatment and chemical fertilizer, and between the chemical fertilizer and the control test for the variable leaf number at DAS 45 on VAR1 (Table.5). This observation differs at DAS 65 with the mycorrhiza showing the highest significant value followed by the chemical fertilizer treatment and lastly by the control test. On the other hand, on VAR2 the mycorrhiza treatment showed the highest leaf number value at DAS 45 and the chemical treatment showed non-significant difference with the control test. At DAS 65 the chemical fertilizer and mycorrhiza treatment showed non-significant difference (Table.5).

For the leaf area variable, the mycorrhiza treatment and chemical fertilizer showed non-significant difference at DAS 45 on VAR1, the same observation was verified t DAS 65. Also, the mycorrhiza treatment and chemical fertilizer showed non-significant difference, with the control test showing the lowest value for VAR2 at DAS 45. This observation differs at DAS 65, where a non-significant difference as observed among all the treatments (Table.5).

Root colonization of the wheat genotypes

The root colonization percentage evaluated for the selected wheat varieties showed a high variation among them (Fig.6). It was observed that, VAR1 (SST88), had a lowest root colonization percentage compared to VAR2 (Nd643-2) at 45 and 65 days after sowing (Fig.6).

4. Discussion

Soil characteristics of both study sites

The contrasted soil characteristics observed in both study sites, was due to the difference in altitude, since according to the result obtained by (Saeed *et al.*, 2014), the soil physico-chemical properties showed a positive correlation with elevation gradient (altitude). In Nkolbisson (low altitude), the soil pH was 5.12, lower than the critical soil pH (5.29) for wheat preconized by (Baquy *et al.*, 2016). Whereas in Mbankolo (high altitude), the soil pH (5.7) was higher than the critical and tolerable soil pH, but lower than the adequate pH (6.0-7.0) for wheat production (Brann *et al.*, 2009). Phosphorus content in low altitude was observed to be lower compared to high altitude and this can be explained by the low soil pH that affected the availability of important element in the soil. A similar result was observed in the same site by Tekeu et *al*. (2015).

Relation between occurred Abuscular mycorrhizal fungi, yield and wheat genotypes

Positive significant correlations revealed between the grain weight, number of AM spores and percentage colonization in Mbankolo (high altitude) and Nkolbisson (low altitude), indicate a positive influence of mycorrhizal symbiosis on yield performance of wheat. A similar result was observed by (Essiane-Ondo *et al.*, 2019), in a study of wheat landraces with low mycorrhizing ability at field respond differently to inoculation with introduced or indigenous arbuscular mycorrhizal fungal communities.

It was observed in this study that the community of arbuscular mycorrhizal fungi can have an effect on yield, even in wheat landraces with low mycorrhizogenous ability. This also justifies the positive influence of the few AM species identified in both study sites on wheat grain weight. Nonetheless, a high correlation was observed between mycorrhiza parameters and wheat grain weight in Mbankolo (r= 0.83 and r= 0.78) compared to Nkolbisson (r= 0.55 and r = 0.56). This result could be justified by the observations made by (Cardoso Filho, Sobrinho, & Pascholati, 2017; Francis and Read, 1995), where they mention a spectrum of fungal impacts in which some species respond mutualistically, while others, putative host or nonhosts, are antagonized or antagonized the action of other species. Since three species were identified in Nkolbisson and the efficiency of a particular species of mycorrhiza might sometimes be influenced by the presence of another mycorrhizal species that is not specific to the host. It was also observed that AM parameters highly contributed to the variation in wheat genotypes in both study sites, as these parameters were highly loaded in PC1 that respectively explained 90.1% and 71.3% of variance in Mbankolo (high altitude) and Nkolbisson (low altitude) respectively. This result justifies the allegation made by (Silva *et al*., 2018) mentioning the fact that the enhancing ability of arbuscular Mycorrhizal fungi species on plant growth and adaptation is generally determined by the degree of symbiotic compatibility, which is affected by the host plant and fungi genotypes. Hence the symbiotic preference of the wheat genotypes might have affected the mycorrhiza parameters and thus has caused this variation. Increase number of genotypes (Nd643-5, Kenya2, Babax8, Croc-1, SST027, SST056, SST086, SST843 and SST88) showed high affinity to mycorrhiza colonization in Nkolbisson compared to Mbankolo (SST015, SST087, SST866 and SST88). This result could be justified by the low soil fertility observed in Nkolbisson site compared to Mbankolo characterized by reduced available phosphorus, which is the principal element mycorrhiza supply to plants. Hence increasing their mycorrhiza affinity permitted them to easily acquire essential elements for their growth and yield. Also, the increase in temperature in Nkolbisson (low altitude) study site might have reduced the water availability in the soil due to increase in evapo-transpiration, hence disturbing the translocation of nutrients from the soil to the plant (Altuhaish et *al*., 2014). Thus these symbiotic associations in this midst could be extremely beneficial to the wheat plant. These allegations were confirmed through a study conducted by Al-Karaki, McMichael, and Zak, (2004), when working on the field response of wheat to Arbuscular Mycorrhizal symbiosis in drought stress, observed that mycorrhizal symbiosis enhances wheat biomass and yield.

Occurred of AMF species in both study sites

Morphological characterization of the occurred AM permitted to identify three Arbuscular Mycorrhizal species that is *Septoglomus sp, Gigaspora sp, Scutellospora sp,* with dominant species in the rhizosphere of wheat cultivated in both altitudes study field identified to belong to the genus *Scutellospora* and family of *Scutellosporaceae* following the revision of Scutellospora and description of five new genera and three new families in the arbuscular mycorrhiza-forming Glomeromycetes (Oehl, de Souza & Sieverding, 2008). A different result was obtained by, Velázquez *et al.*, (2016) who obtained as result a dominant AM fungi community in wheat rhizosphere belonging to the family of *Acaulosporacea.* This difference in result could be explained by the difference in studied environment and wheat genotypes. A high number of AM spores were present in the rhizosphere of wheat genotypes in Nkolbisson (low altitude) study site, compared to Mbankolo (high altitude) study site. This result indicates the increase in dependency of wheat plant to AM symbiosis in low soil fertility areas (Campos *et al.*, 2018; Rubio *et al.*, 2003), and high temperature areas as mycorrhizal trigger the metabolic response of wheat plant in order to increase their tolerance to drought (Al-Karaki *et al.*, 2004; Bernardo *et al*., (2019); Llorens *et al.*, 2019).

Influence of occurred mycorrhiza on wheat yield and biomass

Overall, the mycorrhiza treatment showed a high enhancing ability on wheat grain weight and biomass compared to the other treatments. This enhancing growth ability of mycorrhiza was also observed by (Pérez et *al*., 2016), in studying the effect of inoculation with Arbuscular Mycorrhizal on selected spring wheat lines, obtained as result an enhancing yield potential of spring wheat by Arbuscular Mycorrhizal symbiosis. On the other hand, the responding ability to mycorrhiza colonization varied in function of the wheat genotype. These results simply corroborate the observation made by (Munkvold *et al.*, 2004; Stover et al., 2012b); Smith *et al.*, 2004; Monier *et al.*, 2017; Silva *et al.*, 2018), mentioning the fact that there exist a variation in the degree of specific symbiotic compatibility of the wheat plant. This might justify the variation in response in yield and growth of the two wheat genotypes.

5. Conclusions

The purpose of this study was to determine the potential of native Arbuscular Mycorrhiza (AM) fungi in yield performance and variability of wheat genotypes cultivated in contrasted conditions of the bimodal humid forest zone of Cameroon. The results permitted us to conclude that, the following wheat genotypes SST015, SST087, SST866 and SST88 in Mbankolo (high altitude) and the following genotypes Nd643-5, Kenya2 and Babax8 in Nkolbisson (low altitude) had a high yield potential in each of these areas and showed a high symbiotic affinity with AM fungi. Thus determining the mechanism underpinning these symbiotic preferences will be essential in targeting the genes implicated and important for breeding purposes. The Mycorrhiza parameters were observed to highly influence the variability of wheat genotypes in both study sites, hence

could be essential criterion for plant breeders in wheat improvement. *Scutellospora sp.*, *Gigaspora sp*., and *Septoglomus sp*., were morphologically identified as present in the rhizosphere of wheat lines in both altitudes and were observed to have a high growth and yield enhancing ability on wheat varieties compared to the conventional chemical method of fertilization, hence could be use as efficient bio-fertilizer. Nonetheless, it will be necessary to perform molecular identification (molecular phylogeny) to identify each AM specie in order to individually evaluate their efficiency on wheat growth and yield.

Conflicts of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

Data Availability

The data analysis used to support the findings of this study and other supplementary images related to this study are available from the corresponding author upon request.

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