https://dx.doi.org/10.4314/dujopas.v10i3b.24
ISSN (Print): 2476-8316
ISSN (Online): 2635-3490
Dutse Journal of Pure and Applied Sciences (DUJOPAS), *Vol. 10 No. 3b September 2024*

The Influence of selected Arbuscular Mycorrhizal Fungi on growth of *Curcuma longa* and *Cymbopogon citratus* grown on Biochar Amended Soil

[*1] Archibong, A. E.; [1]Okon, O. G.; [1]Udoh, L. I. and [2]Nyong, I. A.

[1]Department of Botany,
Akwa Ibom State University,
Ikot Akpaden,
Nigeria.

Email: augustinearchibong01@gmail.com

Abstract
The composition of active compounds in medicinal plants is mostly influenced by their growth period and factors that either facilitate or mar their growth. This study assessed the growth of Curcuma longa (Tumeric) and Cymbopogon citratus (Lemon grass) inoculated with two species of arbuscular mycorrhizal fungi (AMF) (Funneliformis mosseae and Rhizophagus irregularis) and grown in soil augmented with biochar. The physicochemical properties of the soil were determined using standard methods. Standard methods were used to measure the growth parameters of both plants and mycorrhizal colonization assessment was examined using standard methods. The physicochemical properties of the soil used were: light intensity (5.00) and pH (7.00). For growth parameters, C. longa responded better to AMF inoculation and biochar application for shoot length and leaf area (17.70±0.56cm; 230.00±6.56 cm^2) than C. citratus (16.30±0.00cm; 53.23±4.28 cm^2). The number of leaves on C. citratus was higher (68.00±0.00) when compared to C. longa (9.33±0.67). For total photosynthetic Pigment (TPP), C. longa inoculated with F. mosseae recorded (62.60±18.58mg/kg) and C. citratus inoculated with a combination of R. irregularis + biochar recorded (64.57±1.87mg/kg). For total fresh weight; C. longa inoculated with F. mosseae yielded 259.03±1.64g compared to C. citratus, treated with R. irregularis + F. mosseae + biochar (189.63±0.49g). For mycorrhizal colonization percentage, C. longa inoculated with a combination of R. irregularis, F. mosseae and biochar recorded the highest AMF colonization (56.60%) when compared to C. citratus inoculated with F. mosseae (4.85%). The increase in growth observed in both plants could influence their significance and use in pharmaceutical, herbal and food industries. Inoculation of aromatic and medicinal plants with AMF species and soil amended with biochar can contribute positively to increase its medicinal value and potency for human use.

Keywords: Biochar, *Cymbopogon citratus, Curcuma longa, Funneliformis mosseae*, *Rhizophagus irregularis*

INTRODUCTION
Medicinal plants have been highly regarded since ancient times for their healing properties and various other benefits (Yasser *et al*., 2008). They provide a reliable source for preparation of new drugs as well as combating diseases from the dawn of civilization. *Curcuma longa* L. (Turmeric) from the family of Zingiberaceae is greatly known for their significance in the herbal industry due to its wide spectrum of pharmacological activities. It is widely cultivated in many Asian countries such as China and India and also distributed throughout the tropical and subtropical regions of the world. (Qaderi *et al*., 2023). *Cymbopogon citratus* (Lemon grass)

Author for Correspondence

is composed of several bioactive compounds that impart medicinal value to it. It is from the Poaceae family and belongs to the genus *Cymbopogon*. The genus *Cymbopogon* constitutes approximately 140 species greatly distributed across the semi-temperate and tropical regions of Asian, African and American continents. Plants in this genus synthesize volatile oils and thus are also known as aromatic grasses (Kumar *et al*., 2009: Alhikari *et al*., 2013).

In nature, plants are in association with a vast number of beneficial microorganisms (e.g. endophytic or symbiotic bacteria and fungi) that play a significant role in growth, development, productivity of plants and in the regulation of metabolite synthesis (Compant *et al*., 2021). Among these are the arbuscular mycorrhizal fungi (AMF), an ubiquitous group of soil microorganisms, that forms a symbiotic association with majority of vascular plants (Brundrett and Tedersoo, 2018). Arbuscular mycorrhizas are characterized by the formation of finely branched structures called arbuscules within the root cortical cells of host plants (Coleman *et al*. 2004), which are the site of bidirectional transport, i.e., minerals from the fungal cell to the plant cell and carbon compounds in the opposite direction.

Arbuscular mycorrhizal fungi, including *Rhizophagus irregularis* and *Funneliformis mosseae* have been shown to increase the production of diverse group of secondary metabolites in plants such as terpenoids or terpenes, steroids, phenolics, and alkaloids. Although they are non-essential to life, they ensure the continued existence of plants in their ecosystems. They play key roles in plant performance as signaling molecules, chemical defense mechanism and adaptation, pollination and seed dispersal, protection from herbivores, predators, pathogens and allelopathic agents.

Nutrient deficiency is prevalent in many crop production systems of the tropics. Hence more sustainable practices, are needed to improve and sustain the productivity of their crops especially as cost of chemical fertilizers and their associated risks on the environmental safety is becoming unaffordable (Mahajan *et al*., 2008). Organic soil amendment is among the sustainable management practices that is used to enhance soil fertility and have been proven to positively improve poor soils' nutrient content and other soil chemical properties (Guo *et al*., 2020; Siedt *et al*., 2021). Organic soil amendments such as biochar, the by-product of biomass and organic waste through thermal degradation (Hansen *et al*., 2016), have shown to enhance soil function (Drinkwater *et al*., 1995) and increases the total soil organic carbon content (Fidel *et al*., 2017). When applied to soil, biochar can help reduce the bulk density of the soil and increase its overall porosity, water retention capacity, and cation exchange capacity hence increase crop productivity (Doan *et al*., 2015). The goal of this research is to access the influence of *Rhizophagus irregularis*, *Funeliformis mosseae* and biochar on the growth/biomass yield and photosynthetic pigment, of *Curcuma longa* and *Cymbopogon citratus.*

MATERIALS AND METHODS

Sample collection

Vegetative Propagules of *C. longa* and *C. citratus* was obtained from a local farm at Effiat offot. The Arbuscular mycorrhizal fungi (*Rhizophagus irregularis* and *Funneliformis mosseae*) inoculum were obtained from The International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State. The biochar obtained from Akpan-andem market in Uyo was pulverized using local method. The sandy-loam soil used in the research was obtained from the Department of Botany research garden, Akwa Ibom State University. The soil was sterilized

for two (2) hours and passed through a 2 mm mesh sieve to remove indigenous AM propagules, soil microorganisms, weeds, seeds, pebbles etc.

Determination of Soil Properties
The soil pH, temperature, moisture and light intensity was determined using a rain trip 4 – in – 1 soil tester (p403 model).

Experimental design
The experiment was set up in a complete block design (CBD) with 8 treatments per plant = 16 treatments for both plates. The experiment was setup in triplicates. The treatment codes and labels is explained in Table 1.

Table 1: Experimental Design

Treatment	Meaning
C	+ Control
+Ri	+ *R. irregularis*
+Fm	+ *F. mosseae*
+Ri+Fm	+ *R. irregularis* + *F. mosseae*
+B	+ Biochar
+Ri+B	+ *R. irregularis* +Biochar
+Fm+B	+ *F. mosseae* + Biochar
+Ri+Fm+B	+ *R. irregularis* + *F. mosseae* +Biochar

Planting/Mycorrhizal Innoculation
Curcuma longa and *Cymbopogon citratus* were sown in pots filled with about 10kg of sterilized soils. The mycorrhizal inoculation was carried out by placing 25g of the AM inoculum per pot and placed at 15cm depth before planting the seeds and biochar was applied at a ratio of 2g – 7g of soil. Inoculum consisted of external mycelium, spores and colonized roots mixed with soil. Water was supplied to individual every two days.

Determination of Growth Parameters
The shoot length of plants was measured using a meter ruler. The leaf area (LA) was determined every two weeks after sprouting. Measurements were taken using measuring tape; the area (A) of the leaf was determined by tracing the outline of the activities on the leaf. The area covered by the outline was then calculated. The correlation factor (K) was determined by dividing the area (A) by product of length x breadth of the leaf. Therefore, the leaf area for each plant was determined using the formula:
$A = L \times B \times K$.

The number of leaves was determined by carefully counting the number of leaves on each plant. The Photosynthetic pigment of the leaves was determined with the aid of electronic digital chlorophyll meter atLEAF with the model number (PN: 0131 USA). This was carried out in-situ (Okon *et al.*, 2021).

Quantification of Arbuscular Mycorrhizal Colonization in Plant Roots
Feeder roots of about 2-4cm of *C. longa* and *C. citratus* were separately collected, fixed in 50% ethanol and stored for colonization assessment. The fixed roots were rinsed in tap water before clearing in 10% KOHw/v and autoclaved for about 15 minutes at 1210 C. Cleared roots were collected on a fine sieve and rinsed with water several times before being transferred into the staining solution. Staining of the plants roots was carried out 5% acetic acid. The roots segments were soaked in the acetic acid and left in staining solution at room temperature for one day. Stained roots were later destained in 50% glycerol for 1 hour (Walker, 2005).

Stained roots were randomly dispersed in a 9cm diameter petri plate with grid lines. Vertical and horizontal gridlines were scanned at x40 magnification with a dissecting microscope. The proportion of root length that is mycorrhizal and total root length can then be calculated from a conversion factor derived from the total length of grid lines and the area of the dish. A minimum of 100 intersections was used to assess the stained root samples; the samples were rerandomized and counted several times. Mycorrhizal root colonization was thus determined by estimation of percentage of root segments containing hyphae, arbuscules and vesicles (Giovannetti and Mosse, 1980).

$$\text{Mycorrhizal colonization} = \frac{\text{Total number of roots infected intersecting gridlines}}{\text{Total number of roots intersecting gridlines}} \times 100$$

RESULTS

Properties of the Soil

Light intensity, moisture content, nutrient content and pH value of the experimental soils was evaluated. Soils on which *Curcuma longa* and *Cymbopogon citratus* were planted had same value for light intensity (5.00±0.00) and pH (7.00±0.00). However moisture ranked from 2.50±0.29 - 3.37±0.67 and nutrient content 2.00±0.00 - 4.67±1.33 (Table 2).

Table 2: Properties of the Soil

Light Intensity	Moisture content	Nutrient	pH Value
5.00±0.00 -5.00±0.00	2.50±0.29 - 4.67±1.33	2.00±0.00 - 3.37±0.67	7.00±0.00 - 7.00±0.00

Morphological parameters of *Curcuma longa* and *Cymbopogon citratus* inoculated with arbuscular mycorrhizal fungi on biochar amended soil.

At 4 and 7 weeks after planting (WAP), *C. longa* and *C. citratus* responded equally to inoculation of AMF and biochar application as observed in shoot length and were significantly (p=0.05) increased when compared to control. Five treatments applications were observed to influence the increase in shoot length in *C. longa* and *C. citratus* (+Fm, +Ri+Fm, +B, +Ri+B, +Ri+Fm+B). At 10 WAP all treatments increased the shoot length of both plants significantly (p = 0.05) when compared to control. At 13 WAP, *C. longa* inoculated with *F. mosseae* (+Fm) had the highest value for shoot length (16.17±3.51 cm) but was not significantly increased when compared to some other AMF inoculated and biochar plants (*R. irregularis* (16.20±0.00) and *R. irregularis* + Biochar (16.30±0.00)) of *C. citratus* (Figure 1).

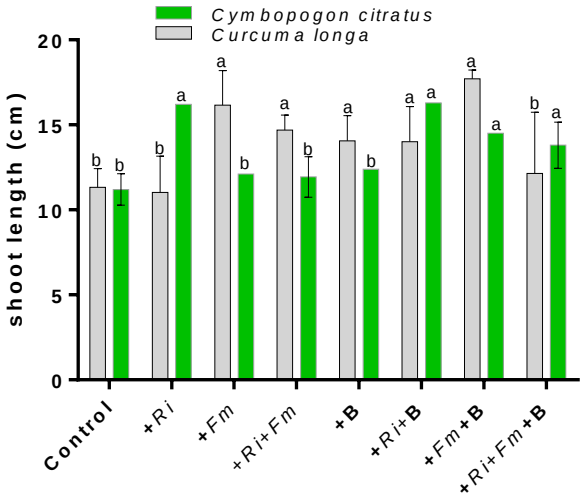

Figure 1: Shoot length of *Curcuma longa* and *Cymbopogon citratus* inoculated with arbuscular mycorrhizal fungi with biochar soil amendment at 13 WAP

The Leaf area of all AMF inoculated and biochar soil amended treatments of *C. longa* and *C. citratus* increased with number of weeks and most were significantly (p=0.05) increased when compared to the control. At 4 WAP, four treatments had better influence on the leaf area of *C. longa* (+Ri, +Fm, +Ri+B, +Ri+Fm+B) when compared to the other three (+Ri, +Ri+Fm, +Ri+B) on *C. citratus* (Figure 4.5). At 10 WAP, all treatments significantly (P = 0.05) increased the leaf area of both plants when compared to the control with *C. longa* inoculated with *F. mosseae* (+Fm) with the highest value (230.00±6.26 cm²). However at 13 WAP, all AMF inoculated and biochar soil amended plants of *C. longa* maintained the same level of performance as observed at 10 WAP, while for *C. citratus*, plants leaf area were not significantly (p = 0.05) increased when compared to control (Figure 2).

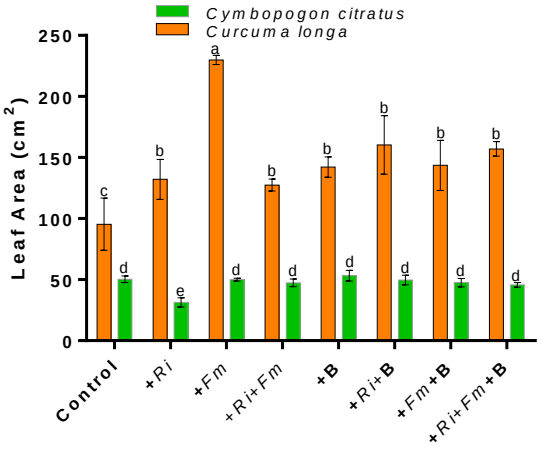

Figure 2: Leaf area of *Curcuma longa* and *Cymbopogon citratus* inoculated with arbuscular mycorrhizal with biochar soil amendments at 13 WAP

AMF inoculation and biochar application greatly influenced increase in the number of leaves of *C. longa* at 4 WAP when compared to *C. citratus*. *C. longa* inoculated with *R. irregularis* + *F. mosseae* + Biochar (+Ri+Fm+B) had the highest number of leaves (7.00+2.83). At 7 – 13 WAP, the number of leaves of *C. longa* were not significantly (p = 0.05) increased when compared to control and had less number of plants that performed better than the control; *C. citratus* (68.00±0.00) had more (Figure 3).

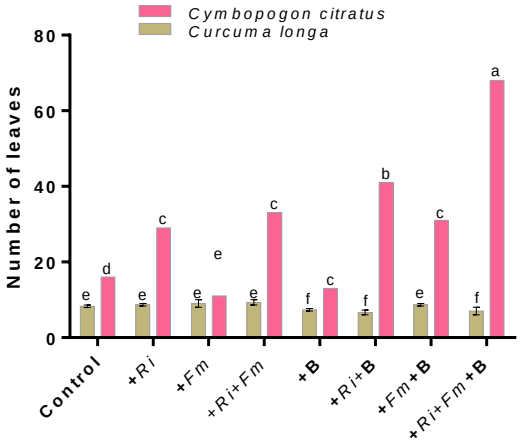

Figure 3: Number of leaves of *Curcuma longa* and *Cymbopogon citratus* inoculated with arbuscular mycorrhizal fungi with biochar soil amendments at 13 WAP

Inoculation of AMF and biochar application had no significant (p = 0.05) effect on the total photosynthetic pigment of *C. longa* and *C. citratus* at 10 WAP. At 13 WAP, *C. citratus* plants treated with *F. mosseae* (+Fm) (62.00±18.58 mg/kg) and *R. irregularis* + Biochar (+Ri+B) (57.00±1.61 mg/kg); then *C. longa* treated with *R. irregularis* + Biochar + *F. mosseae* (+Ri+Fm+B) (54.47±2.04 mg/kg) had significant influence in their total photosynthetic pigment when compared to the controls and other treatments. However, at 4 and 7 WAP, inoculation with *R. irregularis* alone and *R. irregularis* + Biochar application significantly (p = 0.05) increased the total photosynthetic pigment of *C. citratus* when compared to control (Figure 4).

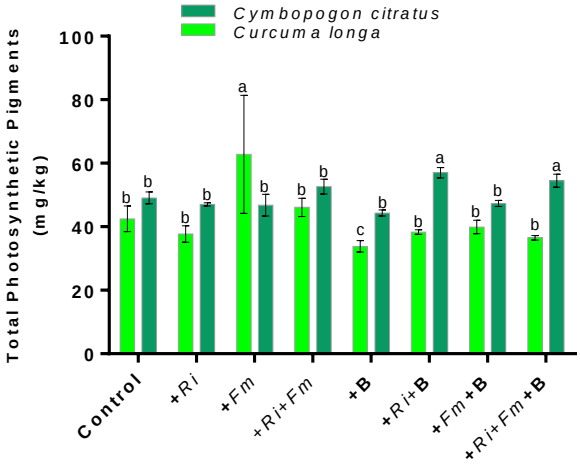

Figure 4: Total photosynthetic pigment of *Curcuma longa* and *Cymbopogon citratus* inoculated with arbuscular mycorrhizal fungi with biochar soil amendments at 13 WAP

All treatments except *R. irregularis* inoculated plant (+Ri) significantly (p=0.05) recorded and increase in the total fresh weight of *C. longa* when compared to control. *C. longa* inoculated with *F. mosseae* (+Fm) alone recorded the highest (348.47±0.07g) fresh weight. For *C. citratus* only four (+Ri, +Ri+Fm, +Fm+B, +Ri+Fm+B) treatments significantly (p=0.05) increase in the total fresh weight when compared to control (Figure 4.17). For rhizome weight of *C. longa*, *F. mosseae* (+Fm) had the highest rhizome weight with 145.30g. All other treatments had influence on the rhizome weight as all had higher values above the control (Figure 5 and 6).

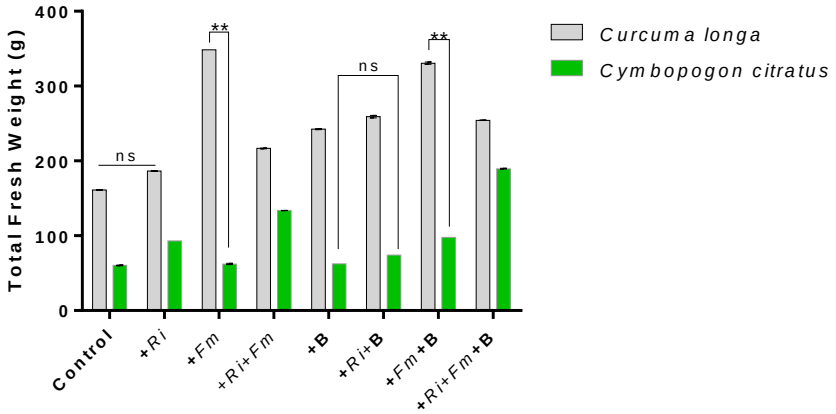

Figure 5: Total fresh weight of *Curcuma longa* and C*ymbopogon citrtaus* inoculated with arbuscular mycorrhizal fungi with biochar soil amendments at 13 WAP

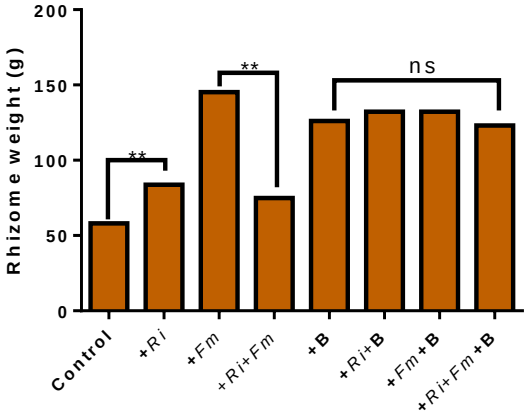

Figure 6: Rhizome weight of *Curcuma longa* inoculated with arbuscular mycorrhizal fungi with biochar soil amendments at 13 WAP

The rate of mycorrhizal root colonization varied in both plants. *C. longa* AMF root colonization was significantly (p=0.05) better when compared to *C. citratus*. *C. longa* inoculated with +Ri+Fm+B and +Ri+B had better rate of mycorrhizal colonization 56.60% and 54.55% respectively. *C. citratus* inoculated with +Fm alone had a mycorrhizal colonization rate of 4.85% (Table 4.1).

Table 3: Total root infection and AMF percentage of *Curcuma longa* and *Cymbopogon citratus*

Treatment	*Curcuma longa*		*Cymbopogon citratus*	
	Total Root infection	**% AMF of Infection**	**Total Root infection**	**% AMF of Infection**
+Ri	50	48.54	0	0.00
+Fm	48	46.60	5	4.85
+Ri+Fm	38	36.89	4	3.92
+Ri+B	48	54.55	0	0.00
+Fm+B	57	53.77	2	1.92
+Ri+Fm+B	60	56.60	3	2.86

DISCUSSION

The results from this study indicated that the growth of both plants was influenced by the inoculation of AMF and biochar application. *Curcuma longa* and *Cymbopogon citratus* growth parameters were increased above the control with AMF inoculation and biochar application. The increase in shoot length might be due to the availability of nutrients through the presence of biochar which is rich in carbon, nitrogen and phosphorus which are critical for plant growth, and the role of AMF which exploits these nutrients, ensure their availability and assimilation in plants. These results were similar to those obtained by Virginie *et al*. (2022) in turmeric (*C. longa*).

This work agreed with the findings of Asma *et al*. (2018) and Virginie *et al*. (2022) who reported the positive influence of organic amendments and biofertilizers on growth of Beetroot (*Beta vulgaris*) and Tumeric (*Curcuma longa*) respectively. The strong observation in the value of leaf area of *C. citratus* from week 10 to week 13 could be an indication that the plant may have reached its peak of growth at week 10 and while still maintaining its growth at week 13.

The number of leaves of *C. longa* and *C. citratus* at week 4 was increased when compared to the control. However, from week 7 through week 13, there was no significant effect of AMF and biochar on the number of leaves of *C. longa* when compared to the control, but increased the number of leaves of *C. citratus* when compared to control. The reduction in the number of leaves of *C. longa* from week 4 could be due to environmental factors, biotic or abiotic, as reported by Ma and Zhang (2010), plant growth and development are usually elicited or inhibited by different environmental conditions, hence plant morphology, anatomy, and physiology reacts to the changes that occurs in the environment as influenced by biotic and abiotic factors.

At week 4 and 7, there was a significant increase in the total photosynthetic pigment of AMF inoculation and biochar applied plants of *C. citratus* when compared to control while AMF inoculated and biochar application on plants of *C. longa* showed no significant increase in their total photosynthetic pigment when compared to control. However, at week 13 only few AMF inoculated and biochar amended treatments of *C. citratus* and *C. longa* showed increased total photosynthetic pigment when compared to control. This study corroborates the result of Virginie *et al*. (2022), who reported that mycorrhizal inoculation and organic amendment had no significant ($p = 0.05$) effect on the chlorophyll content of turmeric (*C. longa*). Also Liang *et al*. (2019), reported that the presence of AMF significantly decreased concentrations of chlorophyll in *P. australis*.

All treatment had a positive significant effect on the total fresh weight of *C. longa* while only four treatments significantly ($p = 0.05$) influenced the total fresh weight of *C. citratus* when compared to control. For rhizome weight of *C. longa*, all treatment significantly ($p = 0.05$) increased the rhizome weight of plants. The significant increase in the total fresh weight of *C. longa* and *C. citratus* and in the rhizome weight of *C. longa* can be attributed to the ability of biochar which does not only serve as a source of nutrients to the plants but also retain and accumulate them and the role played by AMF in enabling plant through its hyphae to access and take up those nutrients, which consequently increases the above and below-ground biomass of the plants. This result is similar to those of Ndonda (2018), who obtained increased production of cassava inoculated with mycorrhizae combined with organic amendment and Virginie *et al*. (2022), who reported that mycorrhizae and organic amendment significantly increased the fresh weight of rhizomes of *Curcuma longa*.

AMF root colonization of *C. longa* was significantly increased when compared to *C. citratus*. Consequently, *C. longa* had high percentage of AMF infection when compared to *C. citratus*. The rate of mycorrhizal colonization between a fungus and the host is influenced by various factors, both biotic and abiotic. According to Halder *et al*. (2015) the colonization of plant root by AMF is affected by effects of climate, soil, host relationship, and species diversity, hence they play a critical role in the interaction between soil microbes and host plants.

Therefore, improved nutrients and water uptake caused by the availability of these nutrients through biochar application and inoculation/colonization of AMF in plants facilitates and increases the growth of plants. The AMF symbiosis have been reported to increase shoot biomass, shoot length, number of nodes in *Ocimum basilicum* (Rasouli-Sadaghiani *et al*., 2010) and increased leaf biomass which resulted in improved photosynthetic capacity (Dave *et al*., 2011).

CONCLUSION

Our results indicates that the use of mycorrhizal inoculation and biochar application is a feasible approach to increase growth in plants, especially for *C. longa* and *C. citratus. C. longa* and *C. citratus* are aromatic plants with essential oils and metabolites which are influenced during their growth period, hence increases their value and could be used for similar purposes in herbal medicine and pharmaceutical industries. The AMF and biochar, separately have been reported in this study to improve the growth of *C. longa* and *C. citratus*. It is important to take into consideration the species of mycorrhizal fungi used, the plant, the soil and environment, as all these might influence the effectiveness of AMF and biochar. Consequently, this study has revealed that the inoculation of arbuscular mycorrhizal fungi (*R. irregularis* and *F. mosseae*) and biochar soil amendment improves the growth/yield in the study plants, therefore increasing the value and use of *C. longa* and *C. citratus* in herbal medicine.

REFERENCES

Asma, J., Sumati, N., Khursheed, H., Shakeel, A. M. and Farooq, A. K. (2018). Effect of Organic Manures and Biofertilizers on Quality of Beetroot (*Beta vulgaris* var. bengalensis). *International Journal of Current Microbiology and Applied Sciences*, **7**(9):1312-1317.

Brundrett, M. C.and Tedersoo, L. (2018) Evolutionary history of mycorrhizal symbioses and global host plant diversity. *New Phytologist*, **220**:1108–1115.

Coleman, D. C., Crossley, D. A. and Hendrix, P. F. (2004). Primary production processes in soils: roots and rhizosphere associates. *In*: Coleman, D. C, Crossley, D. A, Hendrix, P. F (eds). *Fundamentals of Soil Ecology* (2nd Edition). Academic Press. pp. 23–46.

Compant, S., Cambon, M. C., Vacher,, C., Mitter, B., Samad, A., *et al*. (2021). The plant endosphere world – bacterial life within plants. *Environmental Microbiology*, **23**(4):1812–1829.

Doan, T. T., Henry-Des-Tureaux, T., Rumpel, C., Janeau, J. L. and Jouquet, P. (2015). Impact of compost, vermicompost and biochar on soil fertility, maize yield and soil erosion in Northern Vietnam: A three year mesocosm experiment. *Science, Total and Environ*ment, **514**: 147–154.

Drinkwater, L. E. Letourneau, D. K., Workneh, F., van Bruggen, A. H. C. and Shennan, C. (1995). Fundamental Differences between Conventional and Organic Tomato Agroecosystems in California. *Ecological Applications*, **5**:1098–1112.

Fidel, R. B., Laird, D. A., Parkin, T. B. (2017). Impact of Biochar Organic and Inorganic Carbon on Soil CO2 and N2O Emissions. *Journal of Environmental Quality,* **46**: 505–513.

Giovannetti, M. and Mosse B. (1980). An evaluation of techniques for measuring vesicular arbuscular mycorrhizal infection in roots. *New Phytologist*, **84**(3):489–500.

Guo, Y., Niu, G., Starman, T. and Gu, M. (2020). Growth and Development of Easter Lily in Response to Container Substrate with Biochar. *Journal of Horticultural Science and Biotechnology*, **94**(21): 80–86.

Halder, M., Karim, R., Islam, S., and Akhter, S. (2015). Seasonal variation of arbuscular mycorrhiza fungi colonization with some medicinal plant species of chittagong BCSIR forest. *Plant Science Today*, **2**: 87–92.

Hansen, V., Hauggaard-Nielsen, H., Petersen, C. T., Mikkelsen, T. N. and Müller-Stöver, D. (2016). Effects of gasification biochar on plant available water capacity and plant growth in two contrasting soil types. *Soil Tillage Res*earch, **161**: 1–9.

Kumar, J., Verma, V, Goyal, A., Shahi, A. K., Sparoo, R., Sangwan, R. S. and Qazi, G. N. (2009). Genetic diversity analysis in *Cymbopogon* species using DNA markers, *Plant Omics Journal*, **2**: 20-29.

Kumar, J., Verma, V, Goyal, A., Shahi, A. K., Sparoo, R., *et al*. (2009). Genetic diversity analysis in *Cymbopogon* species using DNA markers, *Plant Omics Journal*, **2**: 20-29.

Liang, J., Li, Q., Gao, J., Feng, J., Zhang, X., *et al*. (2021). Biochar rhizosphere addition promoted *Phragmites australis* growth and changed soil properties in the Yellow River Delta. *Science of The Total Environment,* **761**: 143291.

Ma, Z. Q. and Zhang, S. S. (2010). Light intensity affects growth, photosynthetic capability, and total flavonoid accumulation of *Anoectochilus* plants. *HortScience*, **45**: 863–867.

Ndonda, M. (2018). Evaluation agronomiques des champignons mycorhiziens locaux sur la productivité du manioc (*Manicot esculenta* crantz) en sols dégradés des jachères herbeuses à Kisangani (R. D Congo). *Agronomie*. p. 209.

Okon, O. G., Okon, J. E., Sam, S. M., Udoh, L. I and Udoh, F. E. (2021). Checklist Conservsation status and health status assessment via total photosynthetic pigment content of plants found at the Akwa Ibom State University Botanical Garden, Nigeria. *Journal of Biodiversity and Environmental Sciences*, **19**(6): 20-29.

Qaderi, M. M., Martel, A. B. and Strugnell, C. A. (2023). Environmental Factors Regulate Plant Secondary Metabolites. *Plants*, **12**: 447.

Rasouli-Sadaghiani, M., Hassani, A., Barin, M., Danesh, Y. R. and Sefdkon, F. (2010) Effects of arbuscular mycorrhizal (AM) fungi on growth, essential oil production and nutrients uptake in basil. *Journal of Medicinal Plants Research*, **4**(21):2222–2228.

Siedt, M., Schäffer, A., Smith, K. E., Nabel, M., Roß-Nickoll, M., *et al*. (2021)**.** Comparing straw, compost, and biochar regarding their suitability as agricultural soil amendments to affect soil structure, nutrient leaching, microbial communities, and the fate of pesticides. *Science of the Total Environment*, **751**: 141607.

Virginie, T. A., Fridolin. C., Fabien, D. F., Doriane1, K. M., Fotso A., *et al*. (2022). Growth, rhizome yield and biochemical components of turmeric (Curcuma longa L.) as influenced by mycorrhizae, poultry droppings and chemical fertilizers. *International Journal of Agronomy and Agricultural Research*, **21**: 35-49.

Walker, C. A. (2005). Simple blue staining technique for arbuscular mycorrhizal and other root-inhibiting fungi. *Inoculum*, **56**(4):68–69.

Yasser, K., Abdallah, M. and Abdelmadjid, B. (2018**).** Étude ethnobotanique de quelques plantes médicinales dans une région hyper aride du Sud-ouest Algérien Cas du Touat dans la wilaya d'Adrar. *Journal of Animal and Plant Science*, **36**: 5844–5857.