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Potential of *Moringa oleifera* Leaf Powder and Beneficial Microorganisms (Mycorrhizal Fungi and Rhizobia) In Modulating Plant Symbiosis and Yield

Djouhou Fowe M.C[1,2], Fokou E[1], Nwaga D[2]*
[1]Department of Biochemistry, Laboratory of Food Science and Metabolism, Biotechnology Centre, The University of Yaoundé I, P. O. Box 812 Yaoundé, Cameroon
[2]Department of Microbiology, Laboratory of Soil Microbiology, Biotechnology Centre, The University of Yaoundé I, P. O. Box 812 Yaoundé, Cameroon

Original Research Article

***Corresponding author**
Nwaga D

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Abstract: In recent decades, preventing crop losses due to biotic and abiotic stresses and producing more food and feed to meet the demands of ever-increasing human populations have gained unprecedented importance. In the botanical garden of the University of Yaoundé I, trials were carried out to evaluate the effect of *Moringa oleifera* leaf powder and arbuscular mycorrhizae fungi on the growth of *Vigna unguiculata* and *Sorghum bicolor* plants. Twelve treatments for each plant were used; These treatments were: T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa 2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g). *Vigna unguiculata* and *Sorghum bicolor* seeds were planted at a rate of four seeds per plastic bag. From the germination date, agronomic parameters were recorded and four months after planting, leaves, pods and roots were collected for the determination of biomass. Data collected were analysed using ANOVA at $p < 0.05$. Results showed that low rate of *Moringa oleifera* leaf powder and mycorrhizal fungi increased the agronomic parameters and biomass production (20% and 94% respectively for cowpea and sorghum). *Moringa oleifera* leaf powder and inoculation increased root system development, biomass production and yield cowpea. The substrate inoculation increased nodulation and decreased mycorrhizal sporulation. The best results obtained were a low rate of leaf powder application.
Keywords: *Moringa oleifera*, arbuscular mycorrhizae fungi, bio-stimulant, biomass and growth.

INTRODUCTION

The dependency on the use of inorganic fertilizers as a source of plant nutrients by farmers and their high cost is further associated with land and soil degradation and environmental pollution. Thus, the innovative view of farm production attracts the growing demand of biological based organic fertilizers exclusive of alternative to agro-chemicals [1,2]. Organic farming is one of such strategies that not only ensures food safety but also adds to the biodiversity of soil [3]. The additional advantages of bio-fertilizers include longer shelf life causing no adverse effects to ecosystem [4].

Actually, there is continuous need to search for alternative safe natural sources of plant nutrients. *Moringa oleifera* (Moringaceae family) and Arbuscular Mycorrhizae Fungi (AMF) are some of such alternatives, being investigated to ascertain their effect on growth and yield of crops and thus can be promoted among farmers as a possible supplement or substitute to inorganic fertilizers [5]. Since leaves of *Moringa oleifera* are rich in zeatin, it can be used as natural source of cytokinin [6]. Several studies have demonstrated the ability of Moringa leaf extracts to improve plant growth. However, the treatments leading to extracts are long and sometimes difficult to put in place; it is therefore important to explore new application alternatives. In view of all these reports, it is then hypothesized that *Moringa oleifera* leaves, having a number of plant growth promoters, mineral nutrients and vitamins in a naturally balanced composition, may promote plant growth. Therefore, the objective of this study was to evaluate the potential of *Moringa oleifera* leaf powder and AMF in the stimulation of growth of *Vigna unguiculata* and *Sorghum bicolor*.

MATERIALS AND METHODS

Experimental substrate

Soil that provided support for the experiment was a forest soil collected in Yaoundé in the Centre region of Cameroon. It consisted of a mixture of soil and sand (proportions 3-1) that was sieved (using a mason sieve of 5 mm) and introduced into perforated plastic bags (30 × 20 cm).

Experimental site

The experiment was conducted at the Botanical Garden of the University of Yaoundé I from March to October 2016. The area lies between latitude North 3 degrees 52 minutes 0 seconds (3°52'0'') and longitude East 11 degrees 31 minutes 0 seconds (11°31'0'').

Arbuscular mycorrhizal fungi (AMF) and rhizobia

Inoculum, provided by the Laboratory of Soil Microbiology, University of Yaoundé I consisted of a local selected strains of AMF and rhizobia isolated from cowpea and groundnut. The selected stains of AMF have been defined, described and used for experiments under field conditions.

Moringa oleifera leaf powder

The *Moringa oleifera* leaves used as fertilizer have been harvested in Yaoundé on over 5 years old trees. These leaves have been dried at 40°C (to a relative humidity of less than 10%) and ground with mortar; then sieved (using 0.5 mm sieve).

Experimental design

Five liters contain plastic bags according to the treatments were ranged on shelves in the botanical garden of the University of Yaounde I. A factorial design consisting of twelve treatments replicated twenty times for each plant was used. These treatments were: T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa 2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g).

Fertilizers application

AMF, rhizobia and Moringa leaf powder were applied in pots before sowing. Na_2SO_4 was first dissolved in water and applied two weeks after planting.

Planting operation

Vigna unguiculata and *Sorghum bicolor* seeds were planted at a rate of four seeds per plastic bag.

Watering and weeding

Watering of plants was done twice a day with tap water in the absence of rain. Weeding was done according to the grass thrust.

Treatment against insect attacks

Plant insect attacks were solved by using *Azadirachta indica* (Neem) oil. This was dissolved in water-detergent mixture (1/100 v/v).

Soil analysis

Soil sample was passed through a 0.5 cm mesh to remove any plant or grass fragment at the field. It was air-dried and ground to pass through a 2 mm sieve. Soil pH was determined in a 1:2.5 (w/v) soil: water suspension. Organic carbon was determined by chromic acid digestion [7] and spectrophotometric analysis [8]. Soil N (%) was determined by wet acid digests [9] and analysed colorimetrically [10]. CEC was determined by ammonium acetate extraction and quantified colorimetrically.

DATA COLLECTION

From the germination date, agronomic parameters were recorded. These growth parameters were: disease occurrence, leaf yellowing and frequency, flowering date, plant height. Four months after planting, leaves, pods and roots of plants were collected for the determination of biomass.

Number of nodules and nodule weight

Nodules were collected from five randomly selected plants of both legumes at flowering stages. The excavated roots and nodules were washed under running tap water to remove soil and inert particles. Exposed root nodules were collected from the root systems using forceps and amassed. The number of collected nodules of each legume plants were counted and then dried in an oven at 72°C for 24 hours to determine nodule dry weight per plant.

STATISTICAL ANALYSIS

Data from the quantitative study was entered into Microsoft Excel 2007 and exported to SPSS 20.0 statistical program for analysis to verify and evaluate the difference between the levels of biomass production of the treatments. Data were analysed following analysis of variance procedure and expressed as mean ± S.E. Treatment means were separated using least significant difference. The difference was considered significant if $P < 0.05$. Results of parameters were presented in form of Tables and figures.

RESULTS AND DISCUSSION

Physico-chemical characteristics of the experimental soil

The physico-chemical characteristics of the soil used for the experiment are presented in the table 1.

From this table the pH of the soil is 5.12; this value, lower than 5.20 is an indicator of acidic soils. This soil showed low nitrogen content (1.32) and a C/N value of 13.71; this low ratio can suggest a high activity of soil microorganisms. Soil acidity is a major constraint in soil fertility maintenance, particularly, in the humid tropics. Sustainable production of crops on acidic soils depends on soil amendment to remediate acidity and fertility status. Actually the ability of fertilizers, especially organic fertilizers to mediate this acidic character of soil can be attributed to their mineral content. They can enrich soil through mineralization of cations particularly in Calcium [11].

Table-1: Physico-chemical properties of the experimental soil mixture before planting

Soil characteristic	Value
Total N (g/kg)	1.32
Org C (g/kg)	18.10
pH (H_2O)	5.12
C/N	13.71
CEC (mmol/kg)	94.30
Total P (g/kg)	0.4
Sand (%)	52
Silt (%)	25
Clay (%)	23

Influence of bio-fertilizers on the protection of plant against attacks

Results presented in Figure 1 show that *Moringa oleifera* leaf powder and AMF increased ($p < 0.05$) the insect attack frequency of *Vigna unguculata* leaves compare to control treatment.

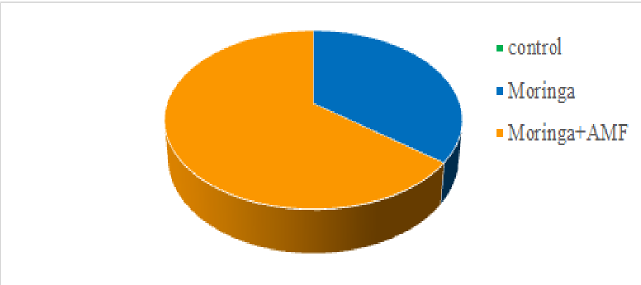

Fig-1: Influence of *Moringa oleifera* leaf powder and beneficial microorganisms (AMF + rhizobia) application on the frequency of insect attack of *Vigna unguiculata* plants

The application of Moringa leaf powder shows a frequency of insect attack of 35% (Moringa treatments) and 65% (Moringa + AMF treatments) as compared to untreated control plants. This result can be an indicator of the nutritional potential of attacked plants. Insects are heterotrophs organisms; thus, they must consume other organisms (plants and animals) in order to acquire energy-rich molecules needed for survival, growth, and reproduction. A balanced diet including proteins, lipids, sugars, minerals, vitamins shall be provided by their feed to meet the metabolic needs of the animal.

The fact that pests have a preference for certain plants may be an indicator of their nutritional composition. Thus the plants of the treatments combining both Moringa leaf powder and microorganisms would be nutritionally richer than others; followed by those receiving only Moringa oleifera leaf powder as fertilizer.

Influence of *Moringa oleifera* leaf powder on biomass production of *Vigna unguiculata* and *Sorghum bicolor*

Vigna unguiculata root nodulation

Nodules are special plant organs that house nitrogen-fixing bacteria called Rhizobium. Root nodule symbiosis enables nitrogen-fixing bacteria to convert atmospheric nitrogen into a form that is directly available for plant growth [12]. At the flowering stage of *Vigna unguiculata*, plant roots showed large number of nodules as presented by the figure 2 and 3.

Fig-3: Nodules on cowpea roots

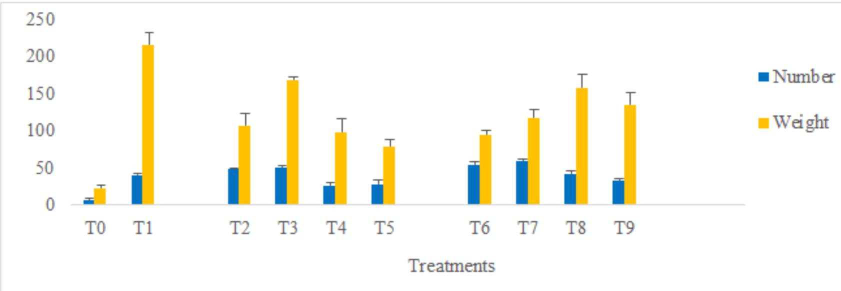

Fig-3: shows the influence of *Moringa oleifera* leaf powder and AMF on the nodule number and weight developed on cowpea (*Vigna unguiculata*) roots.

T0 (Control), T1 (Na2SO4), T2 (10 g AMF), T3 (10 g AMF + Na2SO4), T4 (Moringa 1g), T5 Moringa 2g), T6 (Moringa 4g), T7 (Moringa 8g), T8 (10 g AMF + Moringa 1g), T9 (10 g AMF + Moringa 2g), T10 (10 g AMF + Moringa 4g), T11 (10 g AMF + Moringa 8g)

Figure 3: Influence of *Moringa oleifera* leaf powder and beneficial microorganisms (AMF + rhizobia) application on root nodulation of Cowpea (*Vigna unguiculata* L. Walp.) two months after planting

This figure reveals that the highest number of nodules (61) was recorded by AMF + Moringa 2g; followed by AMF + Moringa 1g (55). The highest nodule weight was recorded by the AMF treatment plant (217.00 g). Either for nodule number or weight, the control treatment is the one with the lowest values (7 and 23.33 g respectively). These results showed that although AMF treatment does not produce as many nodules as some others, theirs are quite large and dense and gave the greatest weight. Although the number of nodules of the Na_2SO_4 treatment remained higher than the control treatment, the application of this chemical product greatly reduces the production of nodules on *Vigna unguiculata* roots compared to treatments receiving bio-fertilizers.

Nodules are very complex organs, containing several interacting processes that operate at distinct levels, including, at least, nodule formation, carbon metabolism, oxygen supply, cellular redox and transmembrane transport.

Biomass production

The influence of Moringa leaf powder and AMF on the production of leaf biomass of cowpea and sorghum is presented by the figure 4.

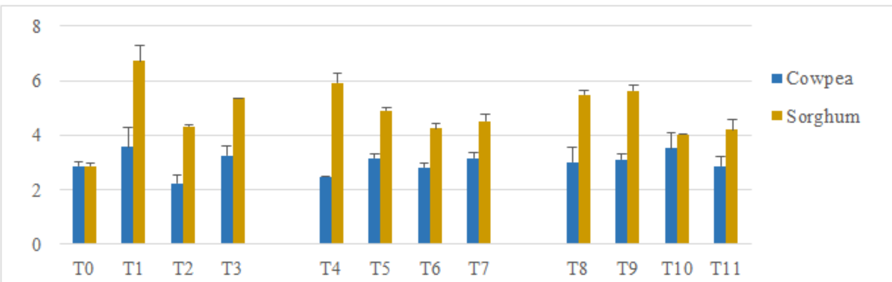

Fig-4: Effect of *Moringa oleifera* leaf powder and beneficial microorganisms (AMF + rhizobia) application on dry biomass of cowpea (*Vigna unguiculata*) and sorghum (*Sorghum bicolor*) two months after planting

T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa 2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g)

Leaf productivity of *S. bicolor* and *V. unguiculata* was influenced by the application of Moringa leaf powder and AMF. This result could be very interesting for local development since farmers could be encouraged to adopt for more sustainable agriculture, replacing the use of synthetic agrochemical and derivate fertilizers [13]. The treatment Na_2SO_4 obtained the highest values (with an increase level of 24, 04% and 134,15% respectively for cowpea and sorghum compare to control) of dry leaf weight for both cowpea (*V. unguculata*) and sorghum (*S. bicolor*). This was followed by AMF + Moringa 4g (23% of augmentation compared to control) for cowpea and Moringa 1g (106,27% of augmentation compared to control) and AMF + Moringa 2g (95,82% of augmentation compared to control) treatments for sorghum. The positive effect of Na_2SO_4 can be attributed to sulphur that it contains. Sulphur is one of essential macronutrients for plants. It occurs predominantly in soil in the form of sulphate. Sulphur is a necessary element in forming protein, enzymes, vitamins, and chlorophyll in plants, and nodule development in legumes. *Moringa oleifera* leaves are rich in sulphur and other minerals [14]. This can be responsible for plant growth stimulation and yield enhancement. Soil beneficial microorganisms such as Arbuscular mycorrhizae fungi and rhizobia have been investigated for their agricultural role in sub Saharan Africa [15;16]. These microorganisms establish a symbiotic relation with host plants to augment nutrient uptake by roots, which deplete the rhizosphere at a rate dependent on water availability, as well as the concentration and the cycling and mobilization of nutrients [17].

Root system development

In this study, crops grown with fertilizer successfully developed their root system and as consequence, obtained the highest root size and weight. Moringa leaf powder and AMF enhanced the development of the plant root system. AMF, Moringa 2g, Moringa 4g, AMF + Moringa 2g and AMF + Moringa 4g treatments are those with the biggest root system size (table 2). Roots are organs ensuring the absorption of nutrients (minerals) by the plant; well-developed root explore a larger area and a better absorption of essential nutrient for plant growth.

It has been demonstrated that root system is one of most important organs for plant [18]. It absorbs water and nutrients and synthesizes organic compounds. The growth and physiological activities of the root system directly affect the growth and physiology of the whole plant. The application of *Moringa oleifera* leaf powder and AMF helps plants to develop their root system by promoting the formation of adventitious roots and affecting cell differentiation and root morphogenesis.

Table-2: Influence of *Moringa oleifera* leaf powder and beneficial microorganisms (AMF + rhizobia) application on root development of *Vigna unguiculata* and *Sorghum bicolor* two months after planting

Treatments	Cowpea root weight (g)	Sorghum root weight (g)
T0	10,82 ± 0,52^{a}	45,00 ± 0,00^{a}
T1	18,99 ± 0,17^{de}	87,00 ± 4,04^{bc}
T2	19,59 ± 0,24^{ef}	88,00 ± 5,20^{bc}
T3	17,32 ± 0,24^{c}	86,50 ±7,79^{bc}
T4	17,00 ± 0,02^{c}	92,00 ± 9,81^{bc}
T5	18,96 ± 0,09^{de}	92,50 ± 3,75^{bc}
T6	19,73 ± 0,17^{ef}	80,50 ± 11,26^{ab}
T7	14,36 ± 0,20^{b}	104,50 ± 12,41^{bc}
T8	17,29 ± 0,37^{c}	123,00 ± 5,77^{c}
T9	19,88 ± 0,39^{ef}	107,00 ± 4,62^{bc}
T10	20,62 ± 0,22^{f}	121,50 ± 2,02^{c}
T11	17,69 ± 0,45^{cd}	122,50 ± 8,37^{c}

Means followed by the same letters are not significantly different for each treatment means ($P< 0.05$) by LSD

T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa 2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g)

Pod and seed production by *V. unguculata*

M. oleifera leaf powder and AMF significantly increased the availability of micro and macro nutrients in the soil for plant uptake. This can be justified by the production level of pods and seeds by *V. unguculata* plants (table 3).

Table-3: Influence of *Moringa oleifera* leaf powder and beneficial microorganisms (AMF + rhizobia) application on cowpea (*Vigna unguiculata* L. Walp.) pod and seed production four months after planting

	Pod weight (g/plant)	100 seed weight (g)
T0	1,29 ± 0,15[abcd]	05,1 ± 0,1[a]
T1	1,74 ± 0,12[abcd]	11,0 ± 0,7[e]
T2	1,40 ± 0,16[abcd]	07,9 ± 0,4[bc]
T3	1,83 ± 0,15[bcd]	10,9 ± 0,6[de]
T4	1,60 ± 0,07[abcd]	10,1 ± 1,1[cde]
T5	1,52 ± 0,11[abcd]	07,2 ± 0,3[ab]
T6	1,27 ± 0,40[abc]	10,5 ± 0,1[cde]
T7	1,32 ± 0,07[abcd]	08,6 ± 0,9[bcde]
T8	2,09 ± 0,14[d]	08,0 ± 0,2[bc]
T9	2,05 ± 0,06[cd]	10,7 ± 0,3[de]
T10	1,18 ± 0,12[ab]	08,3 ± 0,2[bcd]
T11	1,00 ± 0,06[a]	08,2 ± 0,1[bcd]

Means followed by the same letters are not significantly different for each treatment means ($P< 0.05$) by LSD T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa 2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g).

The result on pod and seed production as influenced by the various treatments indicated that the highest weight of pods was obtained from the treatment that received the fertilizer supplemented with *Moringa oleifera* leaf powder and AMF at the rates of 2 g and 4 g.

The highest result obtained by Moringa leaf powder application might be due to the action of zeatin which is a natural growth promoting substance belonging to the class of cytokinins [19]. Fertilizers are any organic or inorganic material of natural or synthetic origin that is added to soil to supply one or more plant nutrients essential to the growth of the plants. It has been stated that plant leaf and fruit peel increase fertility of soil. They contain potassium, vitamins, minerals and some essential elements which enhance the growth of plants [20]. *Moringa oleifera* is capable to increase the availability of micro and macro nutrients in the soil for plant uptake; apart from that, it can also act as a scavenger of certain nutrients such as calcium, potassium and sodium [21].

AMF sporulation

Plant-growth-promoting rhizobacteria (PGPR) and plant-growth-promoting fungi are further components of rhizosphere microflora that can also play a relevant role in plant growth and phytopathogen suppression, mainly due to their synergistic interaction with mycorrhizae [22; 23]. The application of *Moringa oleifera* leaf powder and beneficial microorganisms (mycorrhizal fungi and rhizobia) showed a variability of the colonisation rate (not shown) and sporulation level (table 4) in soil after culture of both cowpea and sorghum.

Table-4: Influence of Moringa oleifera leaf powder and beneficial microorganisms (AMF + rhizobia) application on sporulation of cowpea (Vigna unguiculata L. Walp.) and sorghum (Sorghum bicolor) four months after planting

	Cowpea (number/g)	Sorghum (number/g)
T0	18,2	14,7
T1	11,1	10,1
T2	18,1	9,6
T3	14,2	13,6
T4	12,3	9,3
T5	11,9	10,7
T6	15,1	7,1
T7	9,3	10,6
T8	14,5	10
T9	19,6	8,5
T10	11,3	8,8
T11	20,4	11,7

T_0 (Control), T_1 (Na_2SO_4), T_2 (10 g AMF), T_3 (10 g AMF + Na_2SO_4), T_4 (Moringa 1g), T_5 Moringa

2g), T_6 (Moringa 4g), T_7 (Moringa 8g), T_8 (10 g AMF + Moringa 1g), T_9 (10 g AMF + Moringa 2g), T_{10} (10 g AMF + Moringa 4g), T_{11} (10 g AMF + Moringa 8g)

A spore is a unit of reproduction that is adapted for survival for extended periods of time, under unfavorable conditions, in unfavourable conditions. The dispersion and variability of the obtained results could be due on the one hand to the non-sterilization of substrates with risk of the contamination; and on the other hand to the fertilization provided. Indeed, fertilization is a technique to improve plant nutrition. So if plants are able to draw all the nutrients they need from the soil, the activity of microorganisms decreases. Higher sporulation of AMF was observed in soils where cowpea plants were grown. This can be due to the fact that strains used were isolated from cowpea and groundnut.

CONCLUSION

The regulation of plant growth and development and alleviation of the negative effects of environmental stresses during ontogenesis, are important factors determining the productivity of cultivated plants. While it is well recognized that biotic and abiotic stresses prevent essentially all crop systems from achieving their yield potential, current understanding of the mechanisms involved, and the strategies to mitigate these effects are more and more developed. Abiotic stresses may be prevented by optimizing plant growth conditions and through provision of water and nutrients and plant growth regulators. In addition to these traditional approaches, bio stimulants are increasingly being integrated into production systems with the goal of modifying physiological processes in plants to optimize productivity. The influence of *Moringa oleifera* leaf powder and arbuscular mycorrhizae fungi (AMF) on the growth improvement and biomass production of *V. unguiculata* and *S. bicolor* was investigated. It can be concluded that organic fertilization at different levels has a significant effect on biomass production of *V. unguiculata* and *S. bicolor* which gives moreproductivity and better yields productivity and better yields. The result of this study will give hope to farmers who spend important parts of their income to purchase inorganic fertilizers. *M. oleifera* which is seen growing commonly around homes and homestead gardens will present a suitable substitute to the chemical fertilizers. When *Moringa oleifera* leaf powder increases soil fertility, AMF activity enhances the nutrient uptake and transfer for stimulating plant functioning.

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Abbreviations

AMF: arbuscular mycorrhizae fungi; CEC: Cation Exchange Capacity; S.E: standard error; SPSS: Statistical Package for the Social Sciences; LSD: Least Significant Difference.

REFERENCES

1. Phiri C. Influence of Moringa oleifera leaf extracts on germination and early seedling development of major cereals. Agriculture and Biology Journal of North America. 2010;1(5):774-7.
2. Raja N. Biopesticides and biofertilizers: ecofriendly sources for sustainable agriculture. J Biofertil Biopestici. 2013;4(1).
3. Megali L, Glauser G, Rasmann S. Fertilization with beneficial microorganisms decreases tomato defenses against insect pests. Agronomy for sustainable development. 2014 Jul 1;34(3):649-56.
4. Sahoo RK, Ansari MW, Dangar TK, Mohanty S, Tuteja N. Phenotypic and molecular characterisation of efficient nitrogen-fixing Azotobacter strains from rice fields for crop improvement. Protoplasma. 2014 May 1;251(3):511-23.
5. Abdalla MM. The potential of Moringa oleifera extract as a biostimulant in enhancing the growth, biochemical and hormonal contents in rocket (Eruca vesicaria subsp. sativa) plants. International journal of plant physiology and biochemistry. 2013 Sep 30;5(3):42-9.
6. Fuglie LJ. The miracle tree: Moringa oleifera, natural nutrition for the tropics.1999.
7. Black CA. Methods of Soil Analysis. Agronomy, No. 9, Part 2, American Society of Agronomy & Soil Science Society of America, Madison, Wisconsin;1982 Vol; 9, pp. 539-579.
8. Heanes DL. Determination of total organic-C in soils by an improved chromic acid digestion and spectrophotometric procedure. Communications in soil science and plant analysis. 1984 Sep 1;15(10):1191-213.
9. Buondonno A, Rashad AA, Coppola E. Comparing tests for soil fertility. II. The hydrogen peroxide/sulfuric acid treatment as an alternative to the copper/selenium catalyzed digestion process for routine determination of soil nitrogen-kjeldahl. Communications in soil science and plant analysis. 1995 May 1;26(9-10):1607-19.
10. Robinson JB. Tropical Soil Biology and Fertility: A Handbook of Methods. Edited by Anderson JM and Ingram JSI, with 13 appendices by various authors. Wallingford, Oxfordshire: CAB International (1993), pp. 221,£ 19.95. ISBN 0-85198-821-0. Experimental Agriculture. 1994 Oct; 30(4):487-.
11. Undie UL, Kekong MA, Ojikpong T. Moringa (Moringa oleifera Lam.) leaves effect on soil pH and garden egg (Solanum aethiopicum L.) yield in

two Nigeria agro-ecologies. Eur J Agric For Res. 2013 Sep;1(1):17-25.
12. Brewin NJ. Root nodules (legume–rhizobium symbiosis). eLS. 2010.
13. Campos M, Uliana M, Montero DV, Lima G, Ming LC. Effect of Organic Fertilization on Biomass Production and Bioactive Compounds in Passiflora incarnata L. International Journal of Phytocosmetics and Natural Ingredients. 2015 Aug 21;2(1):11-.
14. Foidl N, Makkar HPS and Becker K. The potential of Moringa oleifera for agricultural and industrial uses. In: Fuglie L J (editor). The miracle tree: the multiple attributes of Moringa. Centre Technique de Coopération Agricole et Rurale / Church World Service. Dakar, Sénégal. 2001; pp. 45 - 76.
15. Nwaga D, Jansa J, Angue MA, Frossard E. The potential of soil beneficial micro-organisms for slash-and-burn agriculture in the humid forest zone of sub-Saharan Africa. InSoil Biology and Agriculture in the Tropics 2010 (pp. 81-107). Springer, Berlin, Heidelberg.
16. Abdalla MM. Boosting the growth of rocket plants in response to the application of Moringa oleifera extracts as a biostimulant. Life Science Journal. 2014;11(11):1113-21.
17. Wang ZY, Kelly JM, Kovar JL. In situ dynamics of phosphorus in the rhizosphere solution of five species. Journal of environmental quality. 2004 Jul 1;33(4):1387-92.
18. Zhang C, Li Q, Zhang M, Zhang N, Li M. Effects of rare earth elements on growth and metabolism of medicinal plants. Acta Pharmaceutica Sinica B. 2013 Feb 1;3(1):20-4.
19. Foidl N, Makkar HPS and Becker K. The potential of Moringa oleifera for agricultural and industrial uses. In: Fuglie LJ. (eds), The Miracle Tree: The Multiple Attributes of Moringa, 2001; pp: 45–76. Wageningen, the Netherlands.
20. Mercy S, Jenifer I. Application of different fruit peels formulations as a natural fertilizer for plant growth. International Journal of Scientific & Technology Research. 2014 Jan 25;3(1):300-7.
21. Ozobia AP. Comparative assessment of effect of Moringa extracts, NPK fertilizer and poultry manure on soil properties and growth performance of Solanium menlongina in Abuja, North Central Region of Nigeria. Journal of Agricultural and Crop Research. 2014 May;2(5):88-93.
22. Compant S, Duffy B, Nowak J, Clément C, Barka EA. Use of plant growth-promoting bacteria for biocontrol of plant diseases: principles, mechanisms of action, and future prospects. Applied and environmental microbiology. 2005 Sep 1;71(9):4951-9.
23. Jeffries P, Gianinazzi S, Perotto S, Turnau K, Barea JM. The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. Biology and fertility of soils. 2003 Jan 1;37(1):1-6.