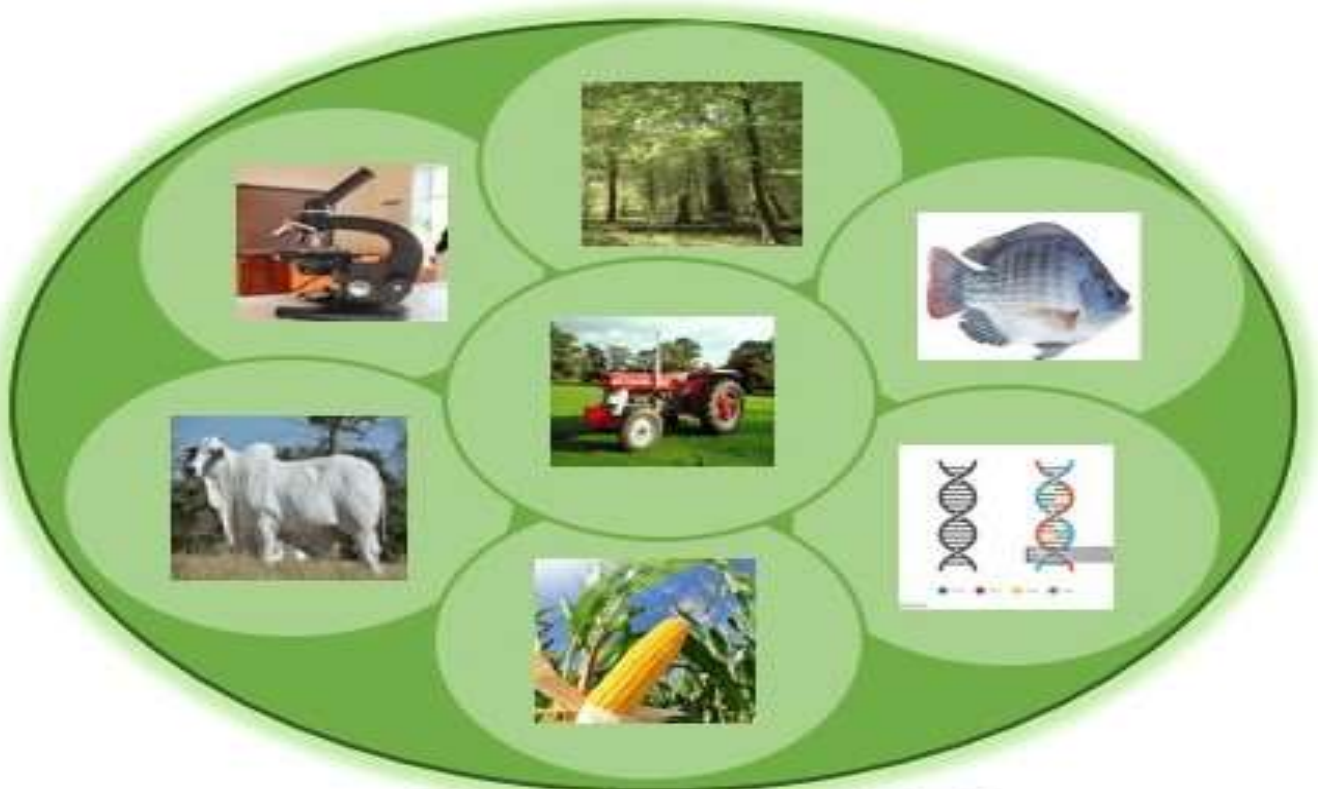




(KEJAANS)

KEBBI JOURNAL OF AGRICULTURE AND NATURAL SCIENCES

January, 2025 Vol. 1, issue 1



KEJAANS

CONTACT:

The Editor-in-Chief,
Kebbi Journal of Agriculture and Natural Sciences,
Faculty of Agriculture,
Kebbi State University of Science and Technology Aliero,
PMB 1144, Birnin kebbi, Nigeria.
Email: kejaanseditor@ksusta.edu.ng, kejaans.foa@gmail.com.
Phone: +234 8039370546

ISSN: 1595-5776



KEBBI JOURNAL OF AGRICULTURE AND NATURAL SCIENCES
(KEJAANS)

JANUARY, 2025: Volume 1, Issue 1

OFFICIAL JOURNAL OF THE
FACULTY OF AGRICULTURE
KEBBI STATE UNIVERSITY OF SCIENCE AND TECHNOLOGY, ALIERO

Editors

**I.S. Jega
M.I. Ribah
I. Sani
M. Atiku
M.N. Kwaifa**

KEJAANS

ISSN: 1595-5776

(c) 2025



About the Journal

This official scientific publication of the Faculty of Agriculture, Abdullahi Fodiyo University of Science and Technology Aliero, is a non-profit, open access, double-blind peer-reviewed Journal publishing four issues (January, April, July and October) per annum. The Journal is a platform open to collaborations with researchers, authors, institutions, research agencies and private companies related to Agriculture. The Mission of the Journal is to disseminate scientific knowledge through the publication of original research articles, research notes, book reviews, letters to the editor and reviews of Literature, representing a contribution to scientific and technological knowledge in respective areas covered by the Journal. The Kebbi Journal of Agriculture and Natural Sciences seeks to validate and disseminate new knowledge, making it public in order to strengthen the human capacity, constitute a link in the scientific community to the society and encouraging the expansion of University and academic researches.

Scope of Kebbi Journal of Agriculture and Natural Sciences (KEJAANS)

The Kebbi Journal of Agriculture and Natural Sciences has the sole aim of providing an intellectual platform and ideas for scholars, by promoting interdisciplinary studies related to agriculture and natural science through publishing the latest scientific research findings that are of direct policy implications and beneficial to the research community. Consequently, the journal covers all aspects of Crop Science, Animal Science, Agricultural Economics, Agricultural Extension and Rural Development, Food Science, Fisheries and Aquaculture, Biotechnology, Soil Science and Agricultural Engineering, Forestry and Environment, Wildlife, Agricultural Education, Agro-allied Industries as well as all Natural Science researches related to Agriculture.

KEJAANS

INSTRUCTIONS FOR AUTHORS

Submission of Manuscript

Submission of manuscript to JAANS shall be on an online platform. Papers could also be submitted as e-mail attachment to the Editor-in-Chief using the kejaanseditor@ksusta.edu.ng or kejaans.foa@gmail.com. The paper should be submitted as a single file in Microsoft Word Format (no other formats will be accepted) and the file shall not be more than 5 Megabytes so that it can be e-mailed to reviewers. The first author, month and year of submission shall be the file name (e.g Ibrahim *et al.* Aug 2010 doc). Once the Editorial Board receives the submission, acknowledgement shall be sent to the corresponding author. If acknowledgement of submission is not received within a week, the author shall remind the Editor-in-Chief through the official email.

Preparation of Manuscript

General presentation: The manuscript should be presented clearly and concisely in English Language. Manuscripts must be prepared (preferably with MS word package) using 12-point New Times Roman (TNR) font, double line-spaced on A4 size paper (210 — 297mm) with at least 3cm margins on all sides. All typing should be justified. Pages including figures and Tables, should be numbered consecutively in the bottom middle with the title page as page 1. Manuscript should contain the following sections (except for review and commentary articles): **Title page; Abstract; Introduction; Materials and Methods; Results, Discussion (Results and Discussion could be combined); Conclusion and References.**

Title page

The first page of the manuscript should contain the title of the article, which should be concise and explicit, typed with upper-case, bold, 14 font size, TNR and not more than 21 words. The surname and forenames (in full) of authors, affiliation of each author should be provided. Phone number and email address of the corresponding author (identified by an asterisk) should be provided. Superscripts should be used to relate authors to their affiliations.

Abstract

The next page should contain abstract in English. Abstract should not be more than 250 words and should provide sufficient information to give the reader a full understanding of the content of the article. Paragraphs, footnotes, references and undefined abbreviations should be avoided.

Keywords

Up to five keywords in normal fonts, separated by semi-column, should be provided to assist the reader and facilitate information retrieval.

Body of Text

The title of the article should be typed in upper-case letters and bold. All other headings should be typed in upper-case letters and bolded while sub-headings should be in lower-case and bolded. The main headings should not be indented. The SI unit system must be used. Standard abbreviations may be used without definition, and specialized abbreviations should be used only after they are defined when they first appear. Use capital 'T' for Table and 'F' for figure. Mathematical formulae should be carefully typed with symbols, correct alignment and must be adequately spaced. Statistical evaluation of results should be described briefly and if necessary, supported by references.

Introduction

A conscience introduction of the background to the subject is required and should include a brief statement of the problem, significance and purpose of the research and relationship to earlier works with well acknowledged references.

Materials and Methods

This section must be presented with adequate clarity and provide sufficient details to permit the repetition of the experimental work. The techniques and the methodologies adopted should be supported with standard references. Subheadings under this section should be in lower case except the first letter.

Results and Discussion

Results should be presented concisely. Only in exceptional cases will it be permissible to present the same set of results in both Table and figure. In discussion, point out the significance of the results and place the results in the context of other work and theoretical background. Results and Discussion part could be written separately if author so wish.

References

Only published articles (Journals and Proceedings) or Books may be cited. In addition, articles with evidence of Journal acceptance are considered as "in press" and are also citable. The reference list should be arranged alphabetically. Authors should be referred to in text by name and year (Harvard system). Examples:

For Journals, list as:

Jega, I.S. and Kwaifa, M.N. (2017). Statistics of Cassava Yield Trials with the Additive Main Effects and Multiplicative Interaction (AMMI) model. *African Journal of Root and Tuber Crops*, 3 (1), 46-50.

Within the text, references should be given as: Meaza *et al.* (2007), or similar results have been obtained (Meaza *et al.*, 2007).

For proceedings, list as:

Aina, O.O., Dixon, A.G.O. and Akinrinde, E.A. (2021). Influence of shoot and root characteristics of cassava genotypes on yields in Nigeria. *African Crop Science Conference Proceedings*, Vol. 5. pp. 1119-1125.

For Books, list as:

DeVries, J. and Toenniessen, G. (2001). *Securing the Han/est Biotechnology, Breeding and Seed Systems for African Crops*. The Cromwell Press, Trowbridge, Wiltshire, UK. 208pp.

For electronic resource materials (online publications) list as:

Zachary, G.P. *Africa plays the rice card*. Foreign Policy. May/June 2008 (web-exclusive story). http://WWW.foreignpolicy.com/stogg/cms.php?story_id=4306. Accessed 26 August 2008.

Tables and Figures

Tables and Figures should be labelled serially using Arabic numerals (e.g Table 1, Table 2, etc; Figure 1, Figure 2, etc.)

Abbreviations

Avoid the use of abbreviations at the beginning of the title, heading or sentence. The following abbreviations with numerals can be used without spelling out at first use. H, min, s, yr, mo, mm, kg, g, DNA, RNA, cpDNA, dNTP.

Numbers

Avoid the use of figures /numbers at the beginning of a sentence. Write out one through nine unless a measurement, a designator, or a range (e.g five seeds, 8cm, 3yr, 5-11 flowers)

Ethical matters

The author using experimental animals must seek permission and include a statement that the investigation was approved by the Ethics Clearance Committee of the researchers' institution.

Copyright

Submission of a copyright to **KEJAANS** implies that the study presented has not been published before or under consideration for publication elsewhere. Once an article has been accepted for publication, author concedes the copyright to **KEJAANS**. However, authors are responsible for the content that appeared in their manuscripts.

Plagiarism Check on Submitted Papers

Since academic Journals must strictly audit the quality of the papers, prevent plagiarism, fraud and other phenomena, ensure that the papers are scientific, original and standardized; cultivate the author's research integrity and consciousness in the process, create a healthy and fair academic environment. It is advisable that each author first conduct a plagiarism check on their paper before submission. Every submitted paper undergoes a plagiarism check by the editors. The editors of **KEJAANS** shall liaise with the Academic Librarians of the organization/institution to do this. Any paper that is more than 20% (or less as determined by the editors) in its plagiarism check shall be sent back to the author for reworking and resubmission.

Blind Peer-review of Submitted Papers

Submitted papers that passed a plagiarism check by **KEJAANS** shall be sent to at least two reviewers that are expert in the field, after every piece of information that can reveal the identity or the affiliation of authors has been concealed for fair, blind peer review. The reviewers shall give a comprehensive report of their review. The editors shall design a Form to be completed by the reviewers after the review. The reports and completed forms shall guide the editors in their further decisions on the reviewed article. The reviewers shall recommend the paper for publication or otherwise.

Publication of Papers

This shall be done after the acceptance of articles for publication in line with the next publication time of the Journal. Prior to publication, a galley proof copy shall be sent to the corresponding author who shall immediately effect correction (if any), and return to the editors. The number of articles to be published in a given issue of the Journal shall be at least 15. It is not compulsory for **KEJAANS** to produce an issue of the Journal if there are no accepted articles ready for publication at a given time of publication.

EFFECT OF MYCORRHIZAL INFECTION ON *CALAPOGONIUM MUCUNOIDES* IN NKO COMMUNITY, YAKURR LOCAL GOVERNMENT AREA, CROSS RIVER STATE, NIGERIA

¹Eteng, Edet Emmanuel and ²Eteng, George Emmanuel

¹Department of Geography and Environmental Science, Faculty of Environmental Science, University of Calabar, Cross Rivers State, Nigeria.

²Department of Soil Science Faculty of Agriculture, Kebbi State University of Science and Technology, Aliero, Kebbi State, Nigeria

Corresponding author: eddyeteng05@gmail.com +234 8065912500

ABSTRACT

Fertilizer, herbicides, and pesticides application has been the major options for sustainability in global food and tree crop production, the danger is felt in rising acidity of the soil, with low crop yield. Mycorrhizae and nodulation bacteria are necessary for tree seedlings establishment, arable, pasture, and fodder crops production in over-cultivated, degraded, and low to moderate soil fertility areas. *Calapogonium mucunoides* is cover crop, and leguminous plant. The paper seek to carry out preliminary assessment of mycorrhizal infection on *Calapogonium mucunoides* in Nko community soils. The study is to; examine if there are mycorrhizal spores associated with the *Calapogonium mucunoides* environment; examine if there are roots infection by mycorrhiza; estimate the percentage infection of the roots. Result of analysis explained that, *Calapogonium mucunoides* is infested with Vesicular Arbuscular Mycorrhizae type of endomycorrhizae with a percentage root colonization of 69% and Rhizosphere spore count of 52 spore per 5gm of soil. Significant linear relationship existed between plant "P", the bulk soil "P" and rhizosphere soil "P". There was no significant difference between the bulk soil "P" and the rhizosphere soil "P" at 5% and 1% probability levels. Thus, the mycorrhizal infection may not have been the major contributing factor to "P" in the plant, as clearly seen in *Calapogonium m.* with plant "P" (0.071%), bulk soil "P" (35.5ppm) and rhizosphere soil "P" (39,8ppm). Further research on *Calapogonium m.* to ascertain the possible effect in its synergism is recommended, if addressed, its rich potentials shall be harness for sustainable crop production.

Keywords; mycorrhizae; nodulation bacteria; rhizosphere soil; VAM; endomycorrhizae,

Introduction

Fertilizer application has been the most outstanding option for sustainability in global food and tree crop production, but the input is often limited by economic constraints and short falls in supply. Tropical Africa agricultural production is associated with peasantry, and land tenure agriculture. Hence, these approach

is exposed to continuous cultivation, bush burning, and deforestation with it attendant soil erosion (Michelsen, 2014)). Whereas, those within the savanna region, who engaged in animal rearing experienced overgrazing. The consequences arising from these systems of agriculture, leads to soil nutrient loss and land degradation. To activation the soil nutrient

efficiency for effective crop production, inorganic fertilizer became necessary tool and the only simple and fast alternative to soil nutrient replenishment. The continuous use of inorganic fertilizers on the soil acidify the soil, resulting to low crop output.

In tree seedlings establishment and arable, pasture, and fodder crops production in degraded, and low to moderate soil fertility areas of the Temperate and Tropics, Mycorrhizal fungi and nodulating bacteria are of immense significance. These importance are indicated by concomitant setback in the recent degradation of soil and natural forest ecosystem, woodlands and shrub-lands, and increased yield in arable, pasture and fodder crops in the tropics (Hayman, 2016).

The paper, effect of Mycorrhizal Infection on *Calapogonium m*; is designed to give a clear analysis of mycorrhizae vis-à-vis the nodulating plant, but not the bacteria. It also provide synergy in promoting and providing alternative method of replenishing lost soil nutrient without negative effect on the soil and crop yield.

The term mycorrhizae has a myriad of definitions as there are many researchers. (Amijee, Tinker, & Stribley, 2009)., defined mycorrhiza as an association of fungus and root where a considerable number of plants are regularly associated with the filaments or hyphae of soil fungi. Mycorrhiza was also referred to as “*My’kes rhiza*” a Greek word meaning “fungus-root” which involves the intimate association of plant roots with specialized soil fungi (Gamalero, Trotta, Massa, Copetta, Martinotti, & Berta, 2004); Broadly defined, mycorrhizae refer to root-like organs formed as a result of the symbiotic association of certain fungi with the roots of higher plants (Smith Smith, & Jakobsen 2004). However, defining mycorrhizae, specific terms are common to all such as; symbiosis,

association, soil fungi and plant roots. Hence mycorrhizae could be taken to mean a fungi-roots interplay or interaction with corresponding benefits. The benefits of mycorrhizae to soil and plants are as follows:

- Uptake of phosphate especially phosphorus which enhances plant growth (Smith, Smith, & Jaobsen I. 2004).
 - Uptake of nutrients and water for increased drought resistance (Hayman, 2016).
 - Detoxify certain soil toxins or enable seedlings to withstand high temperatures or extreme acidity (Molina & Trappe, 2001).
 - Protect roots against certain pathogen eg. *Laccaria laccata scop. Ex Fr.* Protects feeders roots from *Fusarium* infection (Offre, Pivato, Siblot, Gamalero, Corberand, Lamanceau, & Mouget, 2007)
 - Provide the host plant with growth hormones eg. Auxins, Cytokinin, Gibbrelins and growth regulating B vitamin (Kironomos, McCoune, Hart, & Neville, 2000).
 - Contribute to organic matter turn over and nutrient cycling in forest ecosystem (Offre, Pivato, Mazurier, Siblot, Berta, Lemanceau, & Mouget. 2008).
- Infection according to advance learners dictionary by (Hornby, Gatenby, & wakefield, 2014), means infecting or being infested; communication of disease or contagion; disease, influence that infects. From the foregoing, therefore mycorrhizal infection refers to the ability of beneficial soil fungi to contaminate or infect a host-plant in other to enhance some mutual relationship and not disease communication per se.

Calapogonium Mucunoides is a species out of 18,000 in 650 genera that constitute the family leguminosae (Polhill and Ravin, 2012). It belongs to the subfamily papilionoideae that is made up of several species of agricultural

importance in both the tropical and temperate regions (Kironomos, McCoune, Hart, Neville. 2000). Generally, leguminosae is ranked second to Gramineae in terms of economic importance, among other legumes of economic importance in the subfamily (Hayman 2016), Calapogonium is a highly nutritional source of food for animals as pasture legume, useful as ground cover in plantation and eroded soils and as green manure (Hayman, opcit) as well as fixing atmospheric nitrogen in the soil.

Calapogonium m. as a pasture legume together with others like; stylosanthes, Desmodium, Centrosema, Glycin, Leucaena, Pueraria were introduced to Nigeria in the 1940's, and investigation into its performance was done in Shika – Ibadan. The beneficial effects of Calapogonium and other legumes on the soil as nitrogen fixers was long recognized by the Greeks and Romans, but was not explained until towards the end of the 19th Century. The experiment was carried out by growing wheat and legumes in Nitrate deficient soil. Result revealed that while the wheat showed unhealthy growth, the legumes flourished very well on such soil. This concluded that the legumes were able to utilize atmospheric nitrogen in their growth, because of their close association with certain soil bacterium known as Rhizobium (or Bacillus radicola) (Abbott, & Robson, 2016). Michelsen, (2014) and other Co-workers like Hayman, (2016); Molina & Trappe, (2001) among others. have reported the preponderance of Mycotrophy (ie, greater strength and influence of mycorrhizal activity) in the tropical environment.

Besides the numerous forest trees and arable crops that have been tested and proved mycorrhizal, most legumes in the tropics have equally been experimentally tested in the field and confirmed mycorrhizal, Legumes generally are unique and thought to exhibit a

"Tripartite" kind of symbiosis that involve mycorrhiza and Rhizobium relationship in the roots of leguminous plants (Offre, Pivato, Siblot, Gamalero, Corberand, Lamanceau, Mouget, 2007). *Tested tripartite legumes include: Arachis hypogaea; Centrosema Pubescens; Glycin Max; Phaseolus spp.; Pueraria SP.; Trifolium repens; Vigna unguiculata among others.*

The paper, preliminary investigation of mycorrhizal infection on Calapogonium m. explore and exploit the richness of past works on mycorrhizae in determining " what is " to Calapogonium with respect to mycorrhizae , relative to " what was " to other crops or plants with mycorrhizae.

However, the theory of symbiosis as postulated by Russian botanist Konstantin Mereschkowski in 1905 (Cornish-Browden, 2017). The theory presupposes an active relationship and association where two contending organism, or species interact within the ecosystem with mutual benefits from each other, without harm. Mycorrhizae are symbiotic relationship between specialized soil fungi and roots of higher plants. In this association, each mycosymbiont derive some form of benefit from one another. While there is a nutrient gain to the host plant symbiont from the fungus, there is a carbon cost in the form of carbohydrate to the plant, serving as gain to the fungus symbiont (Hayman, 2016) . Even though, some contending views tend to disagree that mycorrhiza is a symbiotic relationship as was seen in non - photosynthetic plant mycotrophy (bird's nest orchis), being totally dependent on the fungus associated with it for both nitrate and carbohydrate supply (Lumini, Bianciotto, Jargeat, Novero, Salvioli, Faccio, Becard, & Bonfante, 2007), and carbon drain by Vesicular Arbuscular Mycorrhizae (VAM) from the host plant (Acacia), resulting

to growth depression (Sanders,2002; & Hayman,2016). The balance of evidence, however, suggests that mycorrhiza is in fact, a symbiotic relationship. That is the bird's-nest Orchis (*Neottia Midus-avis*), though with no chlorophyll, provides the fungus with a suitable habitat, and on the other hand, the carbon drain by Vesicular Arbuscular Mycorrhizae (VAM) from the host plant is equally compensated by increased nutrients up-take mostly phosphorus and Nitrogen (P and N) which increased leaf size, hence photosynthesis is increased and the carbon drained is replenished back to the host symbiont.

Mycorrhizal fungi-plant symbiosis, enables classification of mycorrhizae into two main groups: Ectomycorrhizae, in which the fungal hyphae (propagules) usually form a compact mantle around the short roots and also occur in the intercellular spaces of the cortical cells. According to Hayman,(2016) ectomycorrhizae are also called sheathing mycorrhizae because they produce a mass of mycelium which forms a sheath around lateral roots;

Endomycorrhizae, in which the hypae ramify intracellularly and extracellularly through the root cortex, but do not form a fungal mantle around the roots. Besides the two main groups classified by Frank,the third type called ectendomycorrhizae was also recognised (Molina, & Trappe, 2001); (Sanders,2002), Ectendomycorrhizae possess some of the features or characteristics of both ectomycorrhizae and endomycorrhizae.

Endomycorrhizal fungi form large conspicuous thick-walled spores in the rhizosphere, on the root surface, and sometimes in the feeder root cortical tissue. Upon the formation of spores, fungal hyphae penetrate epidermal cell walls of the root and then grow into the cortical cells, where they develop specialized absorbing structures called "Arbuscules" in the

cytoplasmic matrix. Thin-walled spherical vesicles may also be produced in cortical cells, hence the term vesicular-arbuscular (VA) denotes a type of endomycorrhizae (Gamalero, Trotta, Massa, Copetta, Martinotti, Berta, 2004; & Hayman, 2016). In some instances, the fungus completely colonizes the cortical region of the cell without invading the epidermis, stele, or meristem, (Cornish-Browden, 2017). Therefore this study seek to carry out a preliminary investigation of mycorrhizal infection on *Calapogonium* in Nko community soils. Specifically the study sought to; effect of mycorrhizal infection on *calapogonium mucunoides* in Nko community, Yakurr local government area, Cross River state, Nigeria

Materials and Methods

The experimental site falls within the bounds of the Mkpani communities to the south in Yakurr Local Government Area, and to the North by Oyadama community in Obubra L.G.A of Cross River State, Nigeria. Sample location consisting such areas as;

1. Government primary school garden, Nko
2. Behind Abanamkpai health center, Nko
3. Comprehensive secondary school garden, Nko
4. Behind Unity primary school, Nko
5. Global college school garden, Nko
6. Community secondary school garden, Nko
7. St. Pius primary school garden, Nko
8. Agoi Ekpo road by general Frank house, Nko
9. Ekpekama Farm Road by oil mill, Nko
10. Nko Rubber estate Staff quarters, Nko

Nko community is located between latitude 05° 52' 32" North of the Equator, and longitude 8°11'23". East of the Greenwich Meridian. The average annual rainfall is about 2500mm with a higher spell between April and October, giving an average fall of 38.85mm per week. The mean annual temperature falls within the range of 67°F and 88°F. Nko consists of soils made up of sands of alluvial soils. The texture of the soil ranges from sand to clay but predominantly loamy. The soils are characterized by a deep well drained and well weathered profile with porous weak structures that are susceptible to erosion.

Collection and preparation of samples

Plants samples:

The plant used for the experiment was calapogonium mucunoides. The plant sample consisted of two categories of samples used in the experiment; Root and leaf samples. The root samples were used for quantification of infection while leaf samples were for chemical analysis.

(a) Root Sample:

Collection of root sample for determination of mycorrhizal infection and quantification of infection or level of root colonization was done in the early morning prior to analysis in the laboratory to avoid excessive dehydration of the roots due to high sun intensity. Shelving was avoided as much as possible until the processes of fixing and staining were completed. In sampling of the roots, matured plants of about 4 months or 4 months old were obtained from two of the ten sample areas listed above. Fine terminal feeder roots of at least 1 cm. Length were collected from the entire root system and enclosed in two marcarthney bottles, with each containing about twenty 1 cm. cut feeder roots.

In the laboratory, the root samples were washed and immediately preserved (fixed) and store in 50% ethanol. The fixed roots were cleared in 2.5% KOH (Potassium Hydroxide) and heated in a water bath for 20 min. After heating, the roots were rinsed in several changes of water. Bleaching of the roots was done at room temperature in a freshly prepared solution of alkaline hydrogen peroxide (H₂O₂) comprising of 3 ml. of 20% Ammonium hydroxide (NH₄ OH) in 30 ml. of 3% H₂CO₂ for 10-45 min. However, the roots did not bleach to the naked eyes as recommended by Michelsen, (1992), thus the bleaching time was extended to 24 hours. The roots after bleaching were washed off the alkaline hydrogen peroxide thoroughly in several changes of water, and acidified overnight in 1% HCl.

After acidifying overnight, the roots were stained in acidic glycerol solution (500 ml. of glycerol, 450ml.of H₂O, 50ml of 1% HCl) containing 0.05% Aniline blue and allowed for 20 minutes. The stain was poured-off and roots dis-stained in acidified glycerol at room temperature. Roots were then retained in the acidified glycerol until when root colonization and infection will be assayed.

(b) Leaf Samples:

The entire plant was collected from the Ten various locations listed above, where the plant population was found to be in abundance. From each plant collected, leaves were sampled while the vines were discarded. The leaves were enveloped in a large paper envelop to ease drying and then oven-dried (in the make "ELE") for about 24 hours at 75°C.

After drying, the leaf samples were blended using the christison Scientific laboratory blender. 2g of the blended sample were used for determination of N and 1g for P and K.

(c) Soil Samples:

Like the plant samples, the soil used for this experiment consisted of two categories:

(i) The Rhizosphere Soil

(ii) The Bulk Soil/Non-Rhizosphere soil sample.

Both the Rhizosphere soil and Bulk/Non-Rhizosphere soils were obtained from ten (10) fields located within the NKO community. The Rhizosphere soil was used for spore count and P determination whereas the Bulk soil was for chemical analysis only.

(i) Rhizosphere Soil:

The Rhizosphere soil is a mass of soil usually under the influence of plant roots, is the interface of the soil-root system. The Rhizosphere soil is often noticed to get attached to the base and root hairs of a plant when uprooted from the soil. During sample collection, the Rhizosphere soils from individual plants obtained from the various location earlier mentioned were shaken into a polythene bag and bulked together by mixing thoroughly.

After collection, the Rhizo. soil was air-dried in the laboratory, gently homogenized to avoid damaging the spores and then sieved using a 2mm sieve.

(ii) Bulk / Non-Rhizosphere Soil:

The Bulk or Non-Rhizosphere soils were randomly sampled by collection from the fields, each to a depth of 15cm from the surface using a spade measured to 15cm mark. The samples were thoroughly mixed and bulked together to form a composite sample in which a representative sample would be taken or sub sampled for analysis of desired parameters. The system of collecting the Non-Rhizosphere soil was to make a V-shaped groove to a stated depth and from one side of the groove, a thin slice of soil was taken and divided into three equal parts. The two outer most parts were discarded while only the middle Part was retained in the sampling container.

Like the Rhizosphere soil, the Bulk/Non-Rhizo soil after collection was air-dried in the laboratory, homogenized and sieved with a 2mm sieve type.

Laboratory analysis of samples

The invitro experiment was conducted in the soil Chemistry and Soil microbiology laboratories of the department of soil science, Faculty of Agriculture, University of Calabar.

1) Root Infection and quantification of infection

Root infection investigation for the presence of VAM and ectomycorrhizal fungi was carried out under a compound microscope. The presence of arbuscules (with or without vesicles), hyphae resulted to a positive count of VAM fungi, whereas the presence of a fungal mantis or sheat around root and the presence of a Harting net were the diagnostic characters of ectomycorrhizal fungi. The level of mycorrhizal colonization or quantification of infection was assayed (assessed) with the Grid-line intersect method examining 84 intersects for the sample. In this method, horizontal and vertical grid-lines of fixed distances same as that of the colony counter were ruled at the bottom of a plastic petri dish and the stored roots were evenly spread in the petri dish. The counting or quantification of infection was done under a dissecting microscope at 14x magnification. On this basis the infection was expressed as a percentage of the root infected. Calculation of the percentage root colonization was as follows:

$$\frac{\text{No. of root/grid line intersects with colonization} \times 101}{\text{Total No. of root/grid line intersect counted}}$$

1

2) Spore Count

The number of spores in the Rhizosphere soil determined by the wet sieving and decanting technique, 5g of the homogenized Rhizosphere soil we suspended in distilled water for 10-20 seconds for sedimentation of the coarse sand. Decanting of the suspension was carried out over a series of sieves arranged on top of one another in a descending mesh order of 500 UM,

250 UM, 106 UM and 53UM. After repeated suspension and decanting, the medium and finest sieve contents were collected with distilled water into a 100ml centrifuge tube. Content was suspended in 70% sucrose solution and centrifuged at 3000 rpm for 4 minutes. The gradient of the mixture was extracted with a syringe and transferred to a plastic petri dish a determination of the number of spores per 5g of soil was done by counting under a dissecting microscope at 14x magnification.

Chemical analysis

(a) Rhizosphere Soil Analysis:

A quantity of air-dried Rhizosphere soil passed through 2mm sieve was measured using electronic weighing balance for determination of available phosphorus (P).

(i) Available Phosphorus (P):

Available phosphorus was extracted using the Bray P-1 method (Bray and Kurtz, 2015). The phosphorus was determined colorimetrically from the soil extract by the ascorbic acid method and examined in a spectrophotometer at a wavelength of 882 nm (Murphy and Riley, 2012).

(b) Bulk / Non-Rhizosphere Soil Analysis:

Similarly, in the Non-Rhizosphere soil, air-dried weight of soil passed through 2mm sieve was measured for determination of the following parameters;

(i) Available Phosphorus:

The Bray P-1 extraction method, and the Murphy and Riley, (2012) method of P determination using a spectrophotometer was employed.

(ii) Exchangeable Potassium:

Exchangeable k was extracted by the Ammonium acetate method and K determined in a flame photometer.

(iii) Organic Carbon:

Organic carbon was determined by the wet oxidation method using concentrated sulphuric acid and potassium dichromate.

(iv) Organic Matter:

Soil organic matter was estimated from the determination of Carbon which was made by oxidation under standardized conditions with potassium dichromate in sulphuric acid medium. The % organic matter was calculated by multiplying % organic carbon by a factor (1.729).

(v) Total Nitrogen:

Nitrogen in soil was calculated by multiplying % organic matter by a factor (0.05).

(C) Plant Analysis

(i) Total Nitrogen

Nitrogen in plant was determined by the Macro Kjeldahl digestion method.

(ii) Total Phosphorus

Total P was extracted by the wet digestion method using a mixture of Nitric acid (HNO_3) and perchloric acid (H_2O_2): Determination of phosphorus was done colorimetrically by the Vanado-Molybdate (Yellow) method and a spectrophotometer at a wavelength of 400nm.

(iii) Total Potassium

Total K in plant was extracted by the wet digestion method as in plant P. K. was determined using standard reagents (K-stock solution of 1000 ppm and K-working solution of 0 - 100ppm) and examined in a flame photometer.

Mechanical analysis for bulk soil

The hydrometer method using Calgon (Sodium hexametaphosphate) as a dispersing agent was employed in the particle size analysis of % sand, silt and clay. The textural class of the Bulk soil was determined using a textural triangle.

Results and Discussion

Table 1: Result of Chemical Analysis for Bulk Soil (Non-Rhizosphere, Rhizosphere Soil and Plant

	Bulk Soil/Non-Rhizosphere Soil			Rhizo/Soil		Plant
Total N %	Available P ppm	Exchange-Able K Meg/100g	Organic C %	Organic Matter %	Available P ppm	Total N %
0.10	35.50	0.30	1.18	2.04	39.80	4.7

Table 2: Result of Mechanical and Biological Analysis

Bulk Soil/Non Rhizosphere Soil			Plant Roots	Rhizosphere Soil
% SAND	% SILT	% CLAY	%ROOTS COLONIZATION	SPORE COUNT
75.6	7.7	16.7	69.42	10

Table 3 correlation of bulk soil p and plant p; rhizosphere soil and plant p in different crops

Crop	Bulk Soil P In Ppm X1	Rhizosphere Soil P In Ppm X2	Plant P In % Y
Calapogonium	35.50	39.80	0.071
Water Leaf	44.13	42.70	0.241
Telferia	61.50	50.10	0.358
Maize	78.62	33.90	0.143
Cocoyam	28.25	41.30	0.258
Peper	12.87	33.10	0.252
Okro	62.87	45.30	0.375
Centrosema	36.50	46.60	0.190
Cassava	17.37	22.20	0.258
Cowpea	38.25	37.10	0.205
Ipomea	30.50	18.80	0.238

Table 4 Correlation coefficient (r) for Bulk soil/Plant P and Rhizosphere soil/plant P.

	Correl. Coefficient(r)	Significant Level	
		5%	1%
$r_{X1Y(n-2)}$	0.8707 *	0.6021	0.5214
	0.05		
	0.01		
$r_{X2Y(n-2)}$	0.9253**	0.6021	0.5214
	0.05		
	0.01		

Discussion

Chemical analysis that was carried out for Non-Rhizosphere (Bulk) soil, Rhizosphere soil and plant tissue (leaf) shows result in Table 1 with Total plant N of 4.27% higher (above the optimum level of 3.3% for normal growth in plants) than the total N in the Bulk soil that gave 0.10%.

Available P in Bulk soil and Rhizosphere soil of 35.50PPM (35500Ug1g) and 39.80PPM (39800Ug1g) above the critical soil level of 25-36 $\mu\text{g g}^{-1}$ was higher than the total P in plant with 0.071% below the optimum level of 0.3% for normal plant growth. Though a correlation of Bulk soil P and plant P, and Rhizosphere P and plant P for *Calapogonium* and Ten (10) other crops that were experimented, showed a significant difference at both 5% and 1% levels, a T-test through mean separation of the Bulk soil/plant P and Rhizosphere soil/plant P revealed that the two means were not significantly different. The different plants' P with their respective Bulk soil and Rhizosphere soil P used in the correlation test are shown in table 3.

As in N, the total K content was higher in the plant with 4.30% more than optimum level of 0.80% than the Bulk soil Exchangeable K of 0.30 Meg/100g less or within the critical limit of 0.2-0.3 Meg/100g in the soil. The organic matter content of the Bulk soil stood at 2.04% indicating a moderate level base on standard given as; <2%, 2-3% and >3% for deficiency, moderate and excess respectively.

Result on particle size determination of the Bulk soil, percentage root colonization of *Calapogonium* and the spore count/g of soil in the Rhizosphere soil as contain in Table 2; shows the result of % Sand, silt and clay proportion in the Bulk soil which gave 75.6%, 7.7% and 16.7% respectively with sand being the highest. The % root colonization was 69% approximately. This therefore fell within the range of 50-100% colonized root length

considered as high infection or high level of colonization Michelsen, (2001). The high level of colonization indicates that there was a host-fungi compatibility in the relationship, thus degree of colonization is a function of the plant's response to the inoculum (Offre, Pivato, Mazurier, Siblot, Berta, Lemanceau & Mouget 2008).

The spore count per 5g of soil used in the analysis was 52/5g of Rhizosphere soil, thus estimating approximately 10 spores per 1g of soil. This shows high sporulation, indicating also a soil – endophyte compatibility. The high sporulation was a function of K content of the soil, since high soil P depresses sporulation and K enhances sporulation (Xavier & Germida, 2003).

The study confirms previous report that almost all plants are mycorrhizal especially Agronomic and Horticultural Crops (Kironomos, McCoune, Hart, & Neville, 2000). The study also supports the view that VAM are the most ubiquitous and economically important group since they infect majority of grass and herbaceous species (Ross, 1989; Hayman, 2016;). The study equally confirms that legumes exhibit and display a dual relationship between its rhizobial and mycorrhizal symbionts, otherwise referred to as tripartite relationship (Offre, et al 2008; Hayman 2016); Lumini et'al (2007).

The high N. content in the plant could support the opinion that mycorrhizae enhance N-up-take or confirm the view that mycorrhizal legumes are N_2 -fixers. K content in the plant is another strong affirmative that mycorrhizae help in nutrient mobilization. The organic matter level of the Bulk soil with respect to the high sporulation and percentage root colonization confirms previous reports that organic matter enhance mycorrhizal infection and developments.

Conclusion

This study has shown that mycorrhizal spores are located or associated with the rhizosphere-the immediate vicinity or environment of the feeder roots of calapogonium, termed the "nutrient depletion zones". From analysis, the study also found that, there is a high percentage root colonization of calapogonium by vesicular-Arbuscular Mycorrhizae. This shows that roots of Calapogonium are in association with endomycorrhizae (VAM). It has been found that mycorrhizal plants extract more soil P and are richer in plant P than non-mycorrhizal plants especially legumes. Mycorrhizal Calapogonium in the study was very low in plant P meaning that mycorrhizae had no effect in plant P uptake. However, there is no correct measure to assess a significant or non-significant effect since a control experiment was not carried out.

In conclusion, Calapogonium a legume crop has been found to be highly mycorrhizal than ten other crops that were tested. This shows that mycorrhizal fungi are present in the roots of all well known Agronomic and Horticultural crops, and the Vesicular Arbuscular type of endomycorrhizae is the most ubiquitous in these crops and plants. Calapogonium by implication stands to be synergistically boosted in nutrient uptake especially P and N by its micro-symbionts (Mycorrhizae and rhizobia). However, in this study, mycorrhizae did not improve P uptake in the plant. This work demonstrates the need to further explore the direct effects of mycorrhizae - Rhizobia relationship that affect P uptake in Calapogonium and the full exploitation of its yield potential. It is hoped that when this research work is properly under-taken and the potentials of calapogonium fully harnessed, the call for sustainability in global food supply and environmental protection will be enormous in Nko community, Yakurr Local Government Area and beyond.

References

- Abbott, L., K., and Robson, A.D. (2016). Formation of external hyphae in soil of four species of VA mycorrhizal fungi. *New phytologist* 99: 245-255.
- Bray, R.H., and Kurtz, L.T. (2015). Determination of Total organic and Available forms of phosphorus in Soil. *Soil. Sci.* 59: 39-49.
- Cornish-Browden, Athel (2017) "Lynn Margulis and the origin of the eukaryotes." The origin of mitosing cells: 50th anniversary of a classic paper by Lynn Sagan(mrgulis) 7 December 2017. 434 (1) Bibcode:2017JThBi.434....1C doi.10.1016/j.jtbi2017.09.027 PMID 28992902
- Gamalero E, Trotta A, Massa N, Copetta A, Martinotti MG, Berta G, (2004) Impact of fluorescent pseudomonads and an arbuscular mycorrhizal fungus on tomato plant growth, root architecture and P acquisition. *Mycorrhiza* 14:185-192
- Hayman, D.S. (2016). *My Mycorrhizae of Nitrogen-fixing legumes*, Miroen J. *App1. Microbiol. Biotech.* 2:I2I-I45. Oxford University press, Rothamsted Experimental Station, Harpenden, Herifordshire U.K.
- Hornby, A. S., Gatenby, E, V., and wakefield, H. (2014) *The Advance Learners Dictionary of Current English*. Oxford University Press, London.
- Kironomos JN, McCoune J, Hart M, Neville J. (2000) The influence of arbuscular mycorrhizae on the relationship between plant diversity and productivity. *Ecol lett* 3137-141
- Lumini E, Bianciotto V, Jargeat P, Novero M, Salvioli A, Faccio A, Becard G,

- Bonfante P (2007) Presymbiotic growth and sporal morphology are affected in the arbuscular mycorrhizal fungus *Gigaspora margarita* cure of its endobacteria. *Cell microbiol* (9)1716-1729
- Michelsen, A. (2014). Mycorrhiza and Root nodulation in Tree seedlings from five Nurseries in Ethiopia and Somalia. *For. Ecol. and Management* 48:335-344. Elsevier Sc. Publ. B.V., Amsterdam.
- Molina, R. and Trappe, J. M.(2001). Mycorrhiza management in Bare root Nurseries. In *Forest Nursery Manuel* pp. 221-221. Martinus Nijhoff/Dr. W.Junk pub. The Hague/Boston/Lancaster.
- Murphy, J., and Riley, J.P. (2012). A modified single solution method for Determination of phosphate in Natural Water. *Anal. Chem. Acta* 27: 31-36.
- Offre P, Pivato B, Mazurier S, Siblot S, Berta G, Lemanceau P, Mouget P. (2008) Microdiversity of Burkholderiales associated with mycorrhizal and non-mycorrhizal roots of *Medicago truncatula* *FEMS Microbiol Ecol* (65) 180-192
- Offre P, Pivato B, Siblot S, Gamalero E, Corberand T, Lamanceau P, Mouget C, (2007) Identification of bacterial groups preferentially associated with mycorrhizal roots of *Medicago truncatula*. *Appl Environ Microbiol* (73) 913-921
- Polhill, R.M., and Reven, P.H.(2012). *Advances in legumes systematic*. Royal Botanic Garden, Kew. P.I049.
- Sanders, I. R. (2002) Specificity in the abuscular mycorrhizal Symbiosis. IN Van der Heijden MGA, Sanders IE (eds) *Mycorrhial Ecology*, Springer, Heidenberg, Germany, PP 415 437
- Smith SE, Smith FA, Jakobsen I. (2004) Functional diversity in abuscular mycorrhizal (AM) symbioses: the contribution of the mycorrhizal P uptake pathway is not correlated with mycorrhizal responses in growth or total P uptake. *New Phytol* 162:511-524
- Xavier LJC, Germida J.J (2003) Bacteria associated with *Glomus clarum* spores influence mycorrhizal activity. *Soil Biol Biochem* 35:471-478.