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ABUNDANCE OF SOIL MICROORGANISMS UNDER THE INFLUENCE OF

Acacia senegal (L. Benth.) *Balanites aegyptiaca and Faidherbia albida* IN MAIDUGURI, BORNO STATE, NIGERIA

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ABSTRACT

This study was carried out to assess the abundance of soil microorganism under the influence of tree species of Acacia senegal, *Balanites aegyptiaca* and *Faidherbia albida*. The experimental design used was 3x2 factorial in CRD (Complete Randomized Design). The two factors were three tree species of Acacia senegal, *Balanites aegyptiaca* and *Faidherbia albida*; two soil depth levels (20cm and 40cm) and control (0cm) were studied. The result had shown that *Balanites aegyptiaca* had the highest number of bacteria and fungi with means of 1.97E+18a and 2.43E+14a respectively. Acacia senegal had the highest number of nematodes with a mean of 1062.5a but lowest in terms of number of bacteria and fungi with a mean of 1.25E+18b and 2.20E+14a respectively. *Balanites aegyptiaca* gave the lowest in terms of number of nematodes. Soil depth of 20cm performed better in the number of nematodes and fungi found in the soil with a mean 1068.8a and 2.16E+14a respectively while 40cm depth had the highest in the number of bacteria with a mean of 1,56E+18a. In conclusion *Balanites aegyptiaca* was observed to have the highest impact on the abundance of these microorganisms followed by *Acacia senegal* with prevalence of the microorganisms in the 20cm soil depth. It is recommended that *Balanites aegyptiaca* be used in research requiring the study of fungi, bacteria and nematodes in soils and samples to be collected at the depth of 20cm.

Keywords: Abundace, Acacia Senegal, Micooganism, Soil

INTRODUCTION

Acacia senegal, Balanites aegyptiaca and *Faidherbia albida* grow commonly in tropical area, they were imported into Nigeria in the early 19th century and later given a trial in the north eastern state of the country, in Maiduguri in order to sustain deforestation and desertification that have been occurring in the area for long time [1]. *Acacia senegal, Balanites aegyptiaca* and *Faidherbia albida,* have the ability to adapt to semi-arid condition which make them well known amongst the tree species. They contain many natural substances in their leaves, seeds, barks, roots, etc., they also have impact on biological activities against disease causing organism as well as containing about 140 chemical compounds [1, 2].

Soil microorganisms play important roles in the cycle of matter in nature, soil formation, and soil fertility. Soil microorganisms can develop directly in soil, as well as in decomposing plant residues [3]. Microorganisms are beneficial in increasing the soil fertility and plant growth as they are involved in several biochemical transformation and mineralization activities in soil, continuous use of chemical fertilizer over a period may cause imbalance in soil micro flora and indirectly affecting biological properties of the soil leading to soil degradation. Some pathogenic microbes and aquatic microorganisms may accidentally enter soil during the decomposition of dead bodies, from the gastrointestinal tract of animals and man, with irrigation water, or by other routes, but they generally die quickly [4]. Most soil microorganisms are found ubiquitous and their introduction to a new environment is a haphazard occurrence. Soil factors such as moisture, temperature, aeration, aridity, nutrient and energy supply as well as the characteristic of the organisms determine how well the organisms thrive in new surroundings; some factors greatly influence the spatial distribution of organisms in the soil [5]. A large number of bacteria exist in the soil, but because of their small size, they have a smaller biomass. Fungus population numbers are smaller but they dominate the soil biomass [6]. There are more microbes in the soil than the people on earth, soil contains about 8 to 15 tons of bacteria, fungi protozoa, nematodes, earthworms, and arthropods. At some level within the litter layer and underlying soil horizons are the most favorable conditions for most organisms, the surface litter of humus layers, where space and light conditions fit their particular needs is where most soil animals make their homes [7].

There are billions to hundreds of billions of soil microorganisms in mere handful of a typical garden soil. That single handful

might well contain thousands of different species of bacteria most of which are yet to be classified, hundreds of different species of fungi and protozoa, dozens of different species of nematodes. Almost all of these countless soil organisms are not only beneficial, but essential to the life giving properties of soil [8]. Soil microorganisms are responsible for transforming raw materials from one chemical form to another. Important nutrients in the soil are released by microbial activities, these nutrients include nitrogen, phosphorus, sulfur, iron and others; breaking down soil organic matter into a form useful to plants. This increases soil fertility by making nutrients available and other chemicals found in the soil, suppression of pathogenic microorganisms that cause diseases. Pathogens are also part of this group, but are highly outnumbered by beneficial microbes.

Plant microorganism system is a set-up which undergoes short and long term fluctuation depending on plant development stage as well as agro-ecological condition. Microbial composition and survival in the soil is solely affected by plant species, differences in canopy cover, in that the shortage of canopy cover to an extent exposes the microbial communities to intensive rays of sunlight thereby reducing their population. Microbial composition in the soil is also affected or altered by plant species differences in rooting depth, and litter quality and quantity or even to secondary effects on soil pH, moisture and nutrient levels which in turn affect biogeochemical cycling and ecosystem functioning [9].

It is very important to study the relationship between various plant species and various microorganisms, how the plant species affect the population of the microorganisms in the soil and the factors affecting their growth, so as to apply optimum management practices in the enhancement of the growth and population of microorganisms, and also to know the particular microorganisms that contribute to the proper growth and development of certain plant species. Soil microorganisms perform an important planetary function of participating in the cycle of matter and in the conversion process of important biogenic elements – O, C, N, P, S, and Fe [10, 11].

Soil microorganisms are capable of breaking down all natural organic compounds and some organic compounds not found in nature. They perform an important role in freeing the biosphere from pollutants, chiefly by decomposing pesticides and oxidizing carbon monoxide. The properties of different soil groups and their variations in their fertility are largely determined by the nature of the soil microorganisms and by their activity. Many

different microorganisms inhabit the soil; therefore, this research focuses on their abundance, identification and interactions with these three plant species.

Aim and objectives of the Study

The aim of the study is to determine the abundance of some microorganisms (Bacteria, Fungi, and Nematodes) in soil under the influence of *Acacia senegal, Balanites aegyptiaca and Faidherbia albida*. The specific objectives are to:

i. determine the abundance and prevalence of bacteria, fungi and nematodes at different soil depths under *Acacia senegal, Balanites aegyptiaca and Faidherbia albida* tree species and

ii. identify the species of bacteria, fungi and nematodes found in the study.

MATERIALS AND METHODS

Study Area

The study was carried out in the Laboratory of the Department of Forestry and Wildlife nursery, Faculty of Agriculture University of Maiduguri, Borno State, Nigeria. The nursery is situated within the University campus; it covers a total of $400m^2$. It is located at longitude 14^0 45E and latitude 11^030N, behind Geography Department, maximum temperatures can reach up to 40^0C in April and minimum temperature can be as low as 18^0C between December and January [12]. Major crops grown in the area include groundnut and cowpea,poultry

are reared for domestic consumption and commercial purposes.

Methodology

Sample Collection and Preparation

Soil samples were collected around Forestry and wildlife Nursery, while others around the science complex, with the use of soil auger under the canopies of standing tree species of *Acacia senegal, Balanites aegyptiaca and Faidherbia albida,* Samples were collected at different depths of 20cm and 40cm each and replicated 3 times for each species. Control samples were collected in areas with no vegetation cover, also replicated 3 times. Soil sample was prepared by weighing 1g of the soil sample into 10mls of sterile distilled water, designated as stock and used for serial dilution.

Media Preparation

The media used were potato dextrose Agar (PDA) and Nutrient Agar (NA). These were weighed according to the manufacturer's instruction dissolved by boiling and sterilized using the autoclave at 121^0C for 15mins. Lactic acid was added to the PDA at the appropriate quantity and the media were allowed to cool at 45^0C using the water bath, before pouring into Petri dishes.

Serial Dilution

900ul of sterile water was introduced into each of the sterile Eppendorf tube arranged on the Eppendorf rack. The 100ul of

sample was introduced into the Eppendorf tube containing 900ul of sterile water. This was mixed properly using the vortex mixer. After this, 10 fold serial dilutions were made until dilution was completed.

Total Viable Count for Bacteria

1g each of the soil samples was collected and dissolved in 10mls of distilled water and labelled accordingly. Serial dilution was carried out for each of the samples using test tubes. The test tubes numbered 10^{-3} and 10^{-5} were collected and kept for each of the samples. Test tubes number 10^{-5} was used for inoculation to avoid getting numerous colonies. Serial dilution was done from 10^{-1} to 10^{-10} to enable counting. Using Xiong [13] method of TVCB.

Total Viable Count for Fungi

1g each of the soil samples was collected, weighted and labeled accordingly. Potato dextrose agar (PDA) was plated. 1ml of diluted samples in the test tube number 10^{-5} for each was collected using a pipette and inoculated into the sterile Petri dishes containing media (PDA) and then placed inside an incubator for 24 hours. Fungal growths were observed and mold count was taken for each of the samples using a colony counter. After which a slide was prepared and fungi were identified using a compound microscope.

Inoculation

100ul of diluted sample at dilution 10^{-3} and 10^{-5} were pipette respectively into sterile Petri dishes, and added about 15ml to 20ml of cooled media from water bath at 45^{0}C into each of the inoculated plate. Rock/swirl the plate clockwise and anticlockwise for homogenization, and then the plate was allowed to solidify.

Incubation Period

Incubation period refers to the time interval between the inoculation of sample and appearance of the disease symptom.

- For bacterial incubated at 37^{0}C for 18-24 hours

- For Fungi incubated at 28^{0}C for 48-72 hours.

After incubation the plate count and the colony forming unit were calculated as follows.

$$UFU/ML = \frac{Number\ of\ colony\ \times\ dilution}{Volume\ inoculate}$$

Preparation of Microscope Slide

A piece of Fungi was transferred to a cleaned microscope slide with a cleaned sterilized needle and a drop of water, subsequently, mounting fluid was added and a cleaned cover-slip placed over the slide. During preparation the followings were observed, sporulating structures were taken including some agar, from young areas of the colonies. The Agar was gently melted above aflame and the slide was examined under the microscope. This technique is very helpful for Deuteromycetes to observe the

development of dry chain, or wet conidial heads.

Nematode Extraction Method and Procedure

100g each of the soil sample were collected, weighed and labeled accordingly for the sample. Nematode was extracted from each of the soil samples using **[14]** tray method.

Experimental Design

The experiment was laid out in a 4 x 3 x 2 factorial design in CRD, replicated 3 times.

Statistical Analysis

Data obtained from all the variables in the experiment were subjected to statistical analysis of variance (ANOVA) using SPSS 16.0 package. Significant difference was further subjected to Least Significant Difference (LSD) for the separation of treatment means.

RESULTS AND DISCUSSION

Abundance of Fungi, Bacteria and Nematode

Number of Fungi

The results on numbers of fungi in **Table 1** shows that at alpha 0.05, *Acacia albida, Balanites aegyptiaca and Faidherbia albida,* and had no significant difference on their influence on the abundance of fungi in the soil beneath them respectively, i.e. there was not much difference between *Acacia senegal, Balanites aegyptiaca and Faidherbia albida* and in their influence on the abundance of fungi in the soil, but there

was significant difference between *Balanites aegyptiaca* and Control, *Faidherbia albida* and Control, *Acacia senegal* and control on the abundance of fungi, i.e. each of the plant species had much effect on the population of fungi in the soil compared to the control, i.e. the soil that is not influenced by any vegetation. This may be as a result of the effects of the species canopy on the soil, which perform certain functions such as the regulation of the soil temperature, provision of litters which adds nutrients to the soil there by making the soil favorable and habitable for the microorganisms. This agrees with the work of Qadir **[6]**. The highest number of fungi was recorded in the under *Balanites aegyptiaca* with a mean of 2.43E+14, the least was observed in the soil not influenced by vegetation i.e. Control with a mean of 9.86E+13.

The greater abundance of soil meso-fauna under vegetation compared to bare soil is associated with the availability of plant material, exemplified by the abundance of roots under the tree species and their absence in the bare fallow soil. Similarly, the larger microbial community in the tree species soil may be because substrates are more readily available to microorganisms as well as more plentiful than the bare fallow soil i.e. control. The difference in the resident plant species has long been thought to be a driving force in structure of

bacterial community within the rhizosphere [15]. Even though grasses have diffuse root systems that permeate the soil, some investigators showed the effects of plants on soil bacteria community to be different in the rhizosphere and in the bulk soil. Plants exhibit a wide variety of interactions with microorganisms in both physical and chemical aspects [16]. These interactions explain the relative abundance of microorganisms in the soil.

Number of Bacteria

The results on the numbers of bacteria in **Table 2** shows that at alpha 0.05, there was no significant difference between *Balanites aegyptiaca* and *Faidherbia albida*, i.e. there was not much difference between the influence of *Balanites aegyptiaca* and *Faidherbia albida* on the population of Bacteria, but there was a significant difference between *Balanites aegyptiaca, Acacia senegal* and Control, also between *Faidherbia albida*, *Acacia senegal* and Control and also between *Acacia senegal* and Control on the abundance of bacteria in the soil (P<0.05). The highest number of bacteria was recorded in the soil under *Balanites aegyptiaca* with a mean of 1.97E+18. The least was observed in the soil not under the tree species canopy i.e. Control with a mean of 6.78E17. This result is in accordance to the work of **[17, 18]** who stated that the abundance of

bacteria in the soil is greatly influenced by plant species.

Number of Nematodes

The results on numbers of Nematodes in **Table 2a** shows that no significant difference was found between *Acacia senegal, Balanites aegyptiaca and Faidherbia albida*on their effects on the abundance of nematodes in the soil under the species [i.e. there was not much significant difference between *Acacia senegal, Balanites aegyptiaca* and *Faidherbia albida* on their influence on the population of Nematodes. Also there was no significant difference between the effect of *Faidherbia albida, Balanites aegyptiaca* and the control on the abundance of nematodes, that is to say that the population of Nematodes found in the soil that was not influenced by vegetation had not much difference with that influenced by *Faidherbia albida* and *Balanites aegyptiaca*. But *Acacia senegal* had significant difference from control on its effects on the abundance of nematodes, indicating that the soil influenced by *Acacia senegal* had much effect on the population of nematode compared to the soil that has no treatment. (P<0.05).

However, the highest number of nematodes was recorded in the soil under *Acacia senegal* with a mean of 1062.5. This might be as a result of the canopies of the specie providing an adequate temperature for their

growth, and also provides litter which adds nutrients to the soil through decomposition making it suitable for growth of Nematode. And also the roots of the tree species in the soil make it habitable for the Nematodes as agreed by Wang *et al*. [19]. The least was observed in soil not under the canopy of the studied species i.e. control, with a mean of 679.2. This result corresponds with the work of Wallace [20]; Steer and Harris [15], who stated that the rhizosphere soil around small plant roots and root hairs is a particularly rich habitat for many kinds of nematodes.

Influence of Soil Depth on Fungi, Bacteria and Nematodes Abundance

Fungi

The results in influence on soil depth on Fungi in **Table 2b** shows that there was a significant difference between depth 20cm, and 40cm, ($P<0.05$). The highest number of fungi was observed and recorded at depth 20cm with a mean of 2.16E+14. This might be as a result of the fungi having more access to those favorable factors and the least was recorded at depth 40cm with a mean of 1.84E+14.

Bacteria

The results on influence of Soil Depth on Bacteria in **Table 2c** shows that there was a significant difference between depth 20cm and 40cm on their influence on the abundance of bacteria in the soil ($P<0.05$). However, the depth with the highest abundance of bacteria was depth 40cm with a mean of 1.56E+18 and the least was recorded at depth 20cm with a mean of 1.28E+18. This agrees with the work of Garman, [21] who worked on the influence of depth and sampling time on bacterial community structure in an upland grassland soil.

Nematode

The results on the influence of soil Depth on Nematodes in **Table 3** shows that there was significant difference between the two depths (20cm and 40cm), $P<0.05$. However, the highest number of nematode was recorded at 20cm depth, with a mean of 1068.8. This might be as a result of the depth being closer or more accessible to the nutrients and benefits provided by the plant species and also due to the availability of certain factors such as; good soil aeration, and water percolation. While at depth 40cm a mean of 614.6 was recorded, owing to the shortage of these factors.

Table 1: Pair wise Comparisons Test for Total Viable Count for Fungi TVCF Species

Species	Mean Homogeneous Groups
Control	**6.78E+17[c]**
Acacia senegal	**1.25E+18[b]**
Balanites aegyptiaca	**1.97E+18[a]**
Faidherbia albida	**1.79E+18[a]**

Table 2: Pair wise Comparisons Test for Total Viable Count for Bacteria TVCB Species

Species	Mean Homogeneous Groups
Control	9.86E+13[b]
Acacia senegal	2.20E+14[a]
Balanites aegyptiaca	2.43E+14[a]
Faidherbia albida	2.37E+14[a]

Table 2a: Pair wise Comparisons Test for Total Number of Nematodes TNN Species

Species	Mean Homogeneous Groups
Control	679.2[b]
Acacia senegal	1062.5[a]
Balanites aegyptiaca	750.0[ab]
Faidherbia albida	875.0[ab]

Table 2b: Pair wise Comparisons Test for Total Viable Count of Fungi TVCF Depth

Depth (cm)	Mean Homogeneous Groups
20	2.16E+14[a]
40	1.84E+14[b]

Table 2c: Pair wise Comparisons Test for Total Viable Count of Bacteria TVCB Depth

Depth (cm)	Mean Homogeneous Groups
20	1.56E+18[a]
40	1.28E+18[b]

Table 2d: Pair wise Comparisons Test for Total Number of Nematodes TNN Depth

Depth (cm)	Mean Homogeneous Groups
20	1068.8[a]
40	614.6[b]

Table 3: Species identification of Fungi, Bacteria and Nematodes

Species	Depth (cm)	Species Identification		
		Fungi	Bacteria	Nematodes
Control	20/40	*Aspergilus flavus* *Saccharomyces cerevisiae* *Aspergilusniger* *Candidia tropicalis*	*Clostridium botulinium* *Drechlariasorgicola* *Staphylococcus aurous.*	*Meloidogyne spp* *Longidorus menthasolanum* *Paratylenchus spp* *Heplolaimus galeatus* *Meloidogyne spp.*
A.senegal	20/40	*Aspergilus niger* *Aspergilus parasiticus* *Saccharomyces cerevisiae* *Trichoderna harzianum* *Aspergilus flavus*	*Eweniacharactovora* *Streptococcus faecalis* *Staphylococcus aurous* *Drechlariasorgicolas*	*Tylenechulus mangenoti* *Xiphinema americanum* *Longidorusmentha solanum* *Aphelenchoides sacchari* *Sphaeronema californicum* *Meloidogyne spp*
B. aegyptiaca	20/40	*Candidia tropicalis* *Aspergilus niger* *Aspergilus flavus* *Trichoderma harzianum.*	*Staphilococcus aurous* *Streptococcus faecalis* *Escherichia coli* *Drechlaria sorgicola* *Clostridium botulinium.*	*Psilenchus hilarulus* *Trichodorus christiei* *Aphelenchoides sacchari* *Longidorusmentha solanum*
F.albida	20/40	*Saccharomyces cerevisiae* *Aspergilus niger* *Aspergilus flavus.* *Aspergilus. parasiticus.*	*Staphilococcus aurous* *Ewenia charactavora.* *Drechlaria sorgicola*	*Longidorusmentha solanum* *Tylenchorhynchus F.* *Heteroderas chachtii* *Trichodorus christiei*

CONCLUSION & RECOMMENDATIONS
Conclusion

The results of this research showed that soil under the plant species of (*Acacia senegal,Balanites aegyptiaca and Faidherbia albida,*) had influenced the abundance of microorganisms (Fungi, Bacteria, and Nematodes). *Balanites aegyptiaca* was seen to have the highest impact on the abundance of these

microorganisms followed by *Acacia senegal.* Also, the result of this study showed that microorganisms were found in abundance in the soil at 20cm depth.

The results of the study further showed the interaction between the microorganisms and the plant species in achieving the optimal population of the microorganisms in the soil.

Recommendations

Since *Balanites aegyptiaca* had been observed to have the highest impact on the abundance of these microorganisms, soils under the influence of *Balanites aegyptiaca* should be used in research requiring the study of Fungi, Bacteria and Nematodes in the soil, and it is recommended that collection of soil for identification should be at a depth of 20cm as the abundance of microorganisms were found at 20cm depth.

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