Published by
NIGERIAN SOCIETY OF PHYSICAL SCIENCES
Available online @ https://journal.nsps.org.ng/index.php/jnsps
J. Nig. Soc. Phys. Sci. 1 (2019) 6–11

Journal of the Nigerian Society of Physical Sciences

Original Research

Antimicrobial Assessment of Different Solvent Extracts of the Root-bark, Stem-bark and Leaves of *Acacia ataxacantha* Linn.

O. S. Oguntoye[a], A. M. Okooboh[b], O. M. Bello[c,d,*], A. H. Usman[a], S. S. Abdussalam[a], G. V. Awolola[d]

[a]*Dept. of Applied Chemistry, faculty of Science, Federal University Dutsin-Ma, Katsina State, Nigeria.*
[b]*Federal College of Education, Pankshin Plateau State-Nigeria*
[c]*Dept. of Chemistry, Faculty of Science, University of Ilorin, Kwara State, Nigeria.*
[d]*Dept. of Industrial Chemistry, faculty of Science, University of Ilorin, Kwara State, Nigeria.*

Abstract

Acacia ataxacantha DC. (Fabaceae) is one of the most important medicinal plant used traditionally in managing many ailments in Nigeria particularly infectious diseases. In this study, the stem-bark, roots and leaves of *A. ataxacantha* were extracted using hexane, ethyl acetate, methanol and water (from non-polar to polar solvents). These extracts were initially screened to identify the secondary metabolites present. They were then examined against eight bacteria (*Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Streptococcus pneumonia, Proteus mirabilis, Salmonella typhi-murium*) and seven fungi (*Candida albicans, Penicillium spp, Aspergillus fumigatus, Aspergillus flavus, Trichophyton megninii, Trichophyton. rubrum, Salmonella typhi-murium*). Disc diffusion method was employed to establish the antimicrobial activity of the plant parts. Serial dilution method was used to determine the minimum inhibitory concentration (MIC), minimum bactericidal (MBC) and fungicidal concentration (MFC). Methanol and hexane fractions of the root bark exhibited the highest inhibition zone ranging from 19-21 mm against the tested microorganism hence these were further assessed by determining their MIC, MBC and MFC. MIC value for the aqueous extract of the plant was 25 mg/cm^3 against the fungi used while the methanol fraction was 15 mg/cm^3 against the bacteria used. This work suggests that *Acacia ataxacantha* may possess compounds which can be template for acquiring novel drugs which could act against infectious diseases.

Keywords: *Acacia ataxacantha*, infectious diseases, MIC, antimicrobial drugs

Article History :
Received: 13-03-2019
Received in revised form: 06-04-2019
Accepted for publication: 06-04-2019
Published: 09-04-2019

Communicated by: B. J. Falaye

1. Introduction

Over a million and half of deaths associated with human diseases in the tropical countries are from infectious diseases. The cause of death and illness amongst inhabitants in developing countries like West Africa can be traced to these infectious diseases [1]. Bacteria, viruses, parasites or fungi are some of these deadly microorganism, they are pathogen in nature and they are responsible for the alarming and high incidence of infectious diseases [2]. The recent discovery of developing resistance of pathogens to prevailing and current antibiotics is obvious [3]. There is a pressing demand to discover novel compounds with excellent antimicrobial activities because there has been a growing and disturbing increase in the occurrence of new and re-emerging infectious diseases [4, 5].

*Corresponding Author Tel. No: +2348062320327
Email address: obello@fudutsinma.edu.ng (O. M. Bello)

Many bacterial species already have the genetic make-up to gain and transmit resistance against the current available antibiotics. Many clinical challenges experienced around the world are as a result of the resistance posed by pathogens. The growth of this incidence is so alarming that, it has provided access to a new wave of widespread consumption of antibiotics [6-9]. The search for new antimicrobial agents is a serious burden due to the constant emergence of micro-organisms resistant to conventional antimicrobials, associated and adverse side effects of some antimicrobial drugs and the suddenly emergence of some uncommon infections [10, 11]. Pathogen resistance to currently used antibiotics increases death incidence, there is serious likelihood of increase in patients visitation and stay in the clinics and hospitals [12].

Plant-based therapy has been used against array of diseases for generations hence looking to them for solution against microbes may not be an exception [5,13]. Many secondary metabolites of excellent biological activities have been isolated from mother "Nature". Nature wherein medicinal plants are found, provides a serious hope a better template in the discovery of new antimicrobial drugs [5, 14]. It is obvious from the literature gathered that little work has been done on establishing the phytochemistry of *Acacia ataxacantha*. So far only five compounds have been isolated from the root-bark of this medicinal plant. Most biological evaluations were done on the root-bark, very few research was done on the stem bark while none on the leaves [15]. The medicinal plant is renowned traditionally for its antimicrobial uses, so there is a need to further explore, compare and justify using all the different parts i.e. stem bark, leaves and root-bark for antimicrobial use. This will serve as basis for further studies on the phytochemistry of this plant employing spectroscopic techniques to elucidate the active and non-active constituents in the fractions.

2. Materials and Methods

2.1. Plant material

Different parts of *A. ataxacantha* were obtained from Jos, Nigeria in November, 2016. The plant was properly identified by the curator of the Herbarium, Department of Biological Science, University of Jos, Nigeria.

2.2. Extraction

The root-barks, stem-barks and leaves were air-dried and grounded using wooden mortar and pestle. The powdered sample was subjected to cold extraction (maceration) using hexane and then methanol. The crude methanol extract was suspended in water and washed with ethyl acetate, in that order, to yield water (WAF), methanol (MEF), ethyl acetate fraction (EAF) and hexane (HEF) respectively.

2.3. Phytochemical Screening

Standard techniques of screening and detecting secondary metabolites in plants was used [16, 17]. The metabolites tested for were alkaloids, anthraquinones, cardiac glycosides, carbohydrates, flavonoids, saponins, steroids, tannins and terpenes.

2.4. Determination of Zone of Inhibition.

The zone of inhibition of *A. ataxacantha* plant extracts were determined by using selected pathogenic micro-organisms obtained from the National Veterinary Research Institute, (NVRI) Vom, near Jos in Plateau State. Purity of the clinical isolates employed were verified and they were preserved in nutrient agar. The weight of the plant extracts (0.2 g) were taken, dimethyl sulphoxide (DMSO) (10 mls) was used to dissolve the plant extracts to have a fixed concentration of 20 mg/ml. This was the preliminary concentration of the extracts that was employed to test the antimicrobial properties of the plant extracts. The growth medium for the microbes was Mueller Hinton agar. The medium was prepared according to the Manufacturers guidelines were followed in preparing the medium used. Furthermore, zone of inhibition of *A. ataxacantha* plant extracts were evaluated according to the methods used by Lalnundanga *et al.* and Mathur [18, 19]. Then these plates of the medium were observed for the zone of inhibition of growth of the microbes, the diameter of the zones were taken and recorded in millimetres (mm).

2.5. Determination of Minimum Inhibitory Concentration (MIC)

Resistance is a growing menace, MICs are often used by laboratories to check resistance, though it is also employed as a means to evaluate the antimicrobial activity (in vitro) of new agents [20]. The MIC of the methanol and water crude extracts of the root bark of *A. ataxacantha* was verified using Broth Dilution method [21]. The broth labelled "Mueller Hinton Broth" was made by strictly following the manufacturers instructions. The MIC of the various solvents fractions of the parts of *A. ataxacantha* was determined using the modified agar well diffusion method [16, 18, 19, 22].

2.6. Determination of Minimum Bactericidal Concentration (MBC).

The minimum bactericidal concentrations (MBCs) of the water and methanol extracts were ascertained by selecting and sub-culturing the tubes that did not show growth during MIC determination, into fresh medium which contained no extract. Wire loop was used to transfer from these tubes, these were sub-cultured over the surface of extract free nutrient agar, in petri dishes. These were incubated for 24 h at 37 °C. The lowest extract concentration from which the organism did not recover and grow on the nutrient agar was documented as MBC.

2.7. Determination of the Minimum Fungicidal Concentration (MFC).

Sabour and Dextrose Agar (SDA) plates were made following the manufacturers instructions. The lowest dilution that did not show growth during MIC was inoculated onto the SDA plates. These plates were incubated at ambient temperature for 24 hours. The plates were then observed for growth. The lowest extract concentration from which the fungi did not recover and grow on the agar was noted as the minimum fungicidal concentration (MFC). The result is recorded in table 2.0.

2.8. Test Organisms

The biological activities of the extracts were carried out at the National Veterinary Research Institute (NVRI), Vom, Plateau State, Nigeria. All the microorganisms used are of clinical strains from the same institute. The choice of these pathogens is based on their implication in human diseases such as typhoid, enteric fever, pneumonia, urinary tract infections, dysentery, wound infections and others.

3. Result and Discussion

3.1. Phytochemical Screening

Phytochemical constituents extracts of *A. ataxacantha* are shown in Table 1. On the whole, coumarins, alkaloids, saponins, tannins, flavonoids, tannins, anthraquinones, cardiac glycosides and triterpenes were identified in all extracts. The ethyl acetate of the root-bark extract gave a poor result for most groups of secondary metabolites investigated. The phytochemical screening reveals that flavonoids and carbohydrates are present in the various extracts.

3.2. Diameters of Zones of Inhibition

The bacteria and fungi inhibition produced by *A. ataxacantha* extracts varied in relation to the type of extract and to the micro-organism strain used. Though most of the crude extracts exhibited varying degree of inhibition zones. Methanol and hexane extracts of the roots of *A. ataxacantha* had the highest inhibition zone ranging from 10-19 mm against the tested bacteria used while the ethyl acetate extract of the stem bark and the hexane extract of the roots showed the highest zone of inhibition ranging 05 10 mm against the tested fungi. Implying that it is the more likely active fraction. The antibacterial activity of the exhibited by the roots extracts is good though from this study the antifungal activity is not too commendable. The antibacterial activity of the plant must be due to the phytochemicals identified in the extracts.

3.3. MIC, MBC and MFCs Result

3.3.1. MIC

This was carried out to determine the minimum concentration of the extracts that can inhibit the growth of the microbes. The MIC results showed for only aqueous and methanol extract of the root-bark for the plant, these were chosen because of their interesting activity by the zone of inhibition as Table 4 reveal. The extracts showed moderate activity against the bacteria and fungi used. The water extract of the roots exhibited a moderate activity against the fungi with MIC of 25 mg/ml while the methanol extract of the same plant showed moderate activity against the bacterial with MIC ranging from 25-50 mg/ml. The former showed MIC of 25 mg/ml against P. aeruginosa which a gram negative bacteria but the latter showed a better MIC against all the tested bacterial.

3.3.2. MBC and MFC

Determination of MBC and MFC was done to check if microbes evaluated were actually killed by the extracts or their growth was impeded. The results from MBC and MFC showed that none of the extract killed the bacteria after they were sub-cultured onto agar plate. That is when the results from MIC test tubes were sub-cultured, they still showed growth. This means none of the extract was bactericidal to the eight (8) bacteria employed for the study. In the same vein, the results showed no fungicidal effects on the three (3) fungi used in the study.

In this study, the preliminary phytochemical screening tests were done for three different parts of the *A. ataxacantha* (leaves, stem bark and roots), each of the parts into four different polarities (water, methanol, ethyl acetate, hexane). The tests showed that alkaloids, anthraquinones, cardiac glycosides, coumarins, flavonoids, saponins, tannins and triterpenes were present in all extracts as shown in Table I. This screening may be useful in the discovery of the bioactive compounds and subsequently may lead to drug discovery and development. In antibacterial and antifungal studies carried out earlier, Akapa *et al.* (2015) investigated the antibacterial and antifungal of the root-bark of *A. ataxacantha* using different polarities [23], it was reported that ethyl acetate fraction showed the most significant result with MIC of 2.5 mg/mL towards *B. subtilis, E. coli, S. Typhi and K. pneumonia* [24, 25]. The antifungal activity of stembark of *A. ataxacantha* extracts against six strains of *Aspergillus* was determined by Amoussa *et al.* [14]

The ethyl acetate extract of the stem bark was reported to significantly inhibits the growth of mycelial and sporulation of Aspergillus strains. This study compares favourably with the literature where the stem bark extract (ethyl acetate fraction) of the plant was the most active against the fungi used. Looking at the results in table II and III, it was found that the stem bark extract (ethyl acetate fraction) showed the most interesting zone of inhibition (10 mm) against *Candida albican* in comparison with the control (0.5 mm), the same extract showed a better antibacterial activity against *S. pneumoniae* with an inhibition zone of 15 mm, which is better than to the control (10 mm).

The ethyl acetate extract of the stem-bark showed an inhibition zone of 5 mm against *T. rubrum* while the control showed no zone of inhibition. Looking at the results in the table III for antibacterial activity, it was found that the hexane and methanol extracts of the root bark showed the most interesting zone of inhibition (19 mm) against *Pseudomonas aeruginosa* in comparison with the control "Cyprofloxacin" (19 mm), the hexane extract of the root-bark showed also an interesting antibacterial activity against S. pneumoniae with an inhibition zone which is equal to 15 mm, which is better than to the control (10 mm).

Though the ethyl acetate extract of the leaves showed an inhibition zone of 20 mm against S. aureus as same with the control (20 mm). Figure 1 showed that methanol extract of the root-bark gave the highest zone of inhibition against *S. typhi* when compared with the other extracts. Ethyl acetate extract of the leaves of *A. ataxacantha*, methanol and ethyl acetate extracts of the stem-bark displayed similar zone of inhibition against the fungi used. Figure 2 revealed the antibacterial activities of

Table 1. The phytochemical screening of different parts of *A. ataxacantha* with solvents of different polarities. H_2O : Water, MeOH : Methanol, E.A : Ethyl acetate, Hex : Hexane, +++ = Very Good, ++ = Good, + = Fair, - = Not present

S/N	Constituents	Leaf Extract				Stem bark Extract				Root-bark Extracts			
		H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex
1	*Alkaloids*	+	-	-	-	-	-	-	-	-	-	-	-
2	*Saponins*	+	+	-	-	+	+++	-	-	+++	++	-	+
3	*Tannins*	++	+++	++	+	++	+	-	-	-	+++	-	-
4	*Flavonoids*	+++	+++	++	+	++	++	-	-	++	++	-	-
5	*Carbohydrates*	++	++	++	+	++	+++	++	+	+++	++	-	+++
6	*Steroids*	+	++	++	++	-	+	++	+++	-	+	+	++
7	*Anthraquinones*	-	-			-	-	++	+	-	+	++	+
8	*Cardiac glycosides*	-	+	+	+	-	+	+	+	-	-	-	-
9	*Terpenes*	-	-	++	-	-	-	-	-	-	-	+	-
	% Yield	8.2	17	3	2	2.4	8.8	1	0.68	3	10.4		

Table 2. Diameter of Zones of Inhibition of Fungi (*mm*). H_20: Water, MeOH: Methanol, E.A: Ethyl acetate, Hex: Hexane, Values are means of three replicates ($N = 3 \pm SD$).

S/N	Fungi	Ctrl	Leaf Extract				Stem-bark Extract				Root-bark Extracts			
			H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex
1	*Candida albican*	0.5	0	0	0	08	0	0	10	0	0	06	0	0
2	*Penicillium spp*	0.8	0	0	0	0	0	0	0	0	0	0	0	05
3	*A. flavus*	10	0	05	08	05	03	03	08	05	03	0	0	05
4	*A. fumigatus*	12	0	05	05	0	05	05	05	05	05	0	0	05
5	*T. megninii*	0.3	0	0	0	0	0	0	0	0	0	0	0	05
6	*T. rubrum*	0	0	0	0	0	04	0	05	0	0	0	0	0
7	*S. typhi*	12	08	09	10	05	0	10	10	03	0	11	0	0

Table 3. Diameter of Zones of Inhibition of Bacteria (*mm*). H_2O = water, MeOH = methanol, E.A = Ethyl acetate, Hex = Hexane, ketoconazole (An antifungal conventional drug), cyprofloxacin with (an antibacterial conventional drug). Values are means of three replicates ($N = 3 \pm SD$).

S/N	Bacteria	Ctrl	Leaf Extract				Stem-bark Extract				Root-bark Extracts			
			H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex	H_2O	MeOH	E.A	Hex
1	*P. aeruginosa*	19	0	0	0	0	0	0	0	0	0	19	0	19
2	*E. coli*	13	0	09	11	10	09	0	11	10	0	09	0	11
3	*S. aureus*	20	0	10	20	12	0	0	13	0	15	10	0	11
4	*Bacillus spp*	20	10	0	09	10	0	08	07	0	0	08	10	10
5	*K. pneumoniae*	30	10	15	15	10	10	10	10	05	11	10	0	10
6	*S. pneumoniae*	10	07	0	0	0	10	07	0	0	0	10	0	15
7	*P. mirabilis*	13	10	0	09	10	0	0	10	0	0	10	0	07
8	*S. typhi*	15	10	11	10	10	0	10	10	09	0	10	0	11

the various parts of this medicinal plant. Ethyl acetate extract of the leaves gave the highest zone of inhibition against S. aureus

Table 4. MIC result of the crude methanol and water extracts of the root-bark of *Acacia ataxacantha*. MER: methanol extract of the root, WER: water extract of the root, values are means of three replicates ($N = 3 \pm SD$).

S/N	Test Organism Bacteria	MER (mg/ml)	WER (mg/ml)	Control +ve	Control - ve
1	*Pseudomonas aeruginosa*	200	25	+	-
2	*Escherichia coli*	50	400	+	-
3	*Staphylococcus aureus*	50	400	+	-
4	*Bacillus spp*	100	400	+	-
5	*Klebsiella pneumoniae*	100	400	+	-
6	*Streptococcus pneumonia*	25	200	+	-
7	*Proteus mirabilis*	400	400	+	-
8	*Citrobacter spp*	50	100	+	-
	Fungi				
1	*Candida abaccum*	25	25	+	-
2	*Aspagirus flavus*	100	25	+	-
3	*Aspagirus funigatus*	100	25	+	-

while methanol extract of the root-bark showed a promising activity too. The presence the phytochemical compounds in these active extracts are responsible for the antimicrobial activity.

The MIC results showed in Table IV are for aqueous and methanol extract of the root bark of the plant, these were chosen because of their interesting activity by the zone of inhibition. The MIC values for methanol extract of the root was better than the aqueous extracts for bacteria while aqueous extract was better for the fungi tested. Aqueous extract showed the MIC values of 25 mg/ml against the fungi i.e. *Candida abaccum, Aspagirus flavus, Aspagirus funigatus* while methanol extract displayed a MIC value of 25 mg/ml against only *Candida abaccum* and displayed MIC values of 100 mg/ml and 200 mg/ml against both *Aspagirus flavus and Aspagirus funigatus* respectively. The methanol extract of the root exhibited a moderate activity against *Streptococcus pneumonia* with MIC of 25 mg/ml while the aqueous extract of the same plant showed a very low activity against the same bacterial with MIC ranging from 200 mg/ml. MIC values for the aqueous extract are relatively low compared to methanol extract. This confirm the reported work of Akapa *et al.* [23] that methanol and chloroform fractions of root-bark had MIC values of 5 mg/ml and petroleum ether had 10 mg/ml for all the test organisms [23], though ethyl acetate gave the most significant result with 2.5 mg/ml for *B. subtilis, E. coli, S. typhi and K. pneumonia* [23, 24]. The result for MBC and MFC tests for both the methanol and water extracts on bacteria and fungi are not significant.

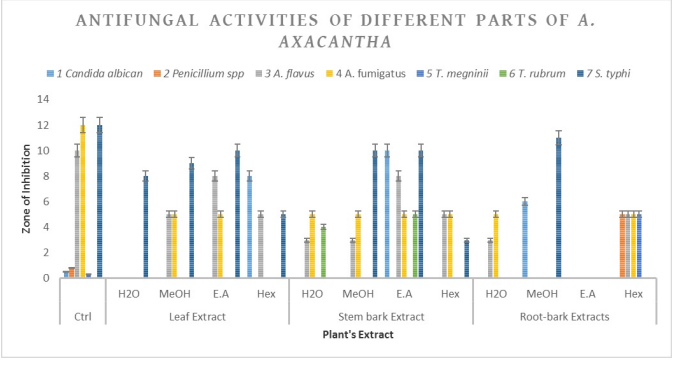

Figure 1. Antifungal activities of different parts of *A. ataxacantha*

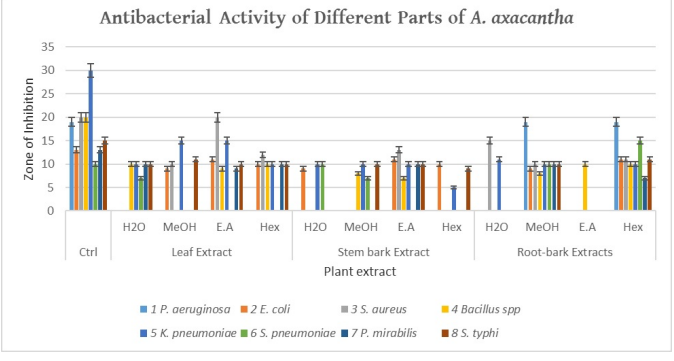

Figure 2. Antibacterial activity of different parts of *A. ataxacantha*

4. Conclusion

This study on the antimicrobial activity of different fractions of the roots, stembark and leaves of *Acacia ataxacantha*

Linn were carried out employing disc diffusion method. The extracts were examined against eight bacteria (which consist of both + Gram and Gram bacteria) (*P. aeruginosa, E. coli, S. aureus, Bacillus spp, K. pneumonia, S. pneumonia, P. mirabilis, S. typhi*) and seven fungi (*C. albican Penicillium spp, A. flavus, A. fumigatus, T. megninii, T. rubrum, S. typhi*). Within the use of traditional Nigeria medicine, our studies are in agreement with the reputed potency of parts of *A. ataxacantha* against common bacterial and fungal complaints. The facts in this study confirms the use and potency of *A. ataxacantha* as an effective antimicrobial plant whose active principles could serve as a potential chemotherapeutic agent.

Acknowledgments

We thank the referees for the positive enlightening comments and suggestions, which have greatly helped us in making improvements to this paper.

Conflict of interest

No conflict of interest

Funding

No funding was received for this study

References

[1] A. O. Amoussa, A. L. Lagnik & A. Sanni, "*Acacia ataxacantha* (Bark): Chemical composition and antibacterial activity of the extracts", International Journal of Pharmacy and Pharmaceutical Science **6** (2014) 11.

[2] www.who.int/gho/publications/world_health_statistics/2016/Annex_B/en/(Date accessed: May 2018).

[3] A. Doss, M. H. Muhammed & R. Dhanabalan, "Antibacterial activity of tannins from Solanum trilobatum Linn. Leaves", Indian Journal of Science and Technology **2** (2009) 41.

[4] V. P. R., Anand Bhatt J. K., J. M. Varghese & K. R. Das Sushanta, "Supplementation of Vitamin E improves cognitive status and oxidative stress in type 2 diabetes mellitus", International Research Journal of Pharmacy **2** (2011) 169.

[5] O. M. Bello, T. Ibitoye & C. Adetunji, "Assessing antimicrobial agents of Nigeria flora", Journal of King Saud University Science, (2018). *doi.org/10.1016/j.jksus.2018.04.017*.

[6] J. Harvey & A. Gilmour, "Characterization of recurrent and sporadic Listeria monocytogenes isolates from raw milk and nondairy foods by pulsed field gel electrophoresis, monocin typing, plasmid profiling, and cadmium and antibiotic resistance determination", Applied Environment Microbiology **67** (2001) 840.

[7] Z. Ruiz-Bolivar, A. K. Carrascal-Camacho, M. C. Neuque-Rico, C. Gutierrez-Trivino, M. X. Rodriguez-Bocanegra, R. A Poutou-Pinales & M. Salim, "Enterobacterial repetitive intergenic consensuspolymerase chain reaction (ERIC-PCR) fingerprinting reveals intra-serotype variations among circulating Listeria monocytogenes strains", African Journal of Microbiology Research **5** (2008) 1586.

[8] G. Sakoulas & R. C. Moellering, Jnr. "Increasing antibiotic resistance among Methicillin-Resistance Staphylococcus Aureus Strains", Clinical Infectious Diseases 46 (2008) 360.

[9] B. P. Howden, J. K. Davies, P. D. R. Johnson, T. P. Stinear & M. L. Grayson, "Reduced vancomycin susceptibility in Staphylococcus aureus, including vancomycin-intermediate and heterogeneous vancomycin-intermediate strains: resistance mechanisms, laboratory detection, and clinical implications", Clinical Microbiology Reviews **23** (2010) 99.

[10] B. L. Stuart & M. Bonnie, "Antibacterial resistance worldwide: causes, challenges and responses", Nature Medicine Supplement **10** (2004) 134.

[11] M. Sourav, H. Bibhabasu, S. Rhitajit, B. Santanu & M. Nripendranath, "Hemidesmus indicus, an age-old Plant: Study of its in vitro antioxidant and free radical scavenging potentials", Pharmacology-Online **1** (2009) 604.

[12] T. G. Winstanley, D. I. Limb, R. Eggington & F. Hancock, "10-year survey of the antimicrobial susceptibility of urinary tract isolates in the UK: The Microbe Base project. Journal Antimicrobiology Chemotherapy **40** (1997) 591.

[13] O. M. Bello, A. A. Zaki, I. S. Khan, P. S. Fasinu, Z. Ali, I. A. Khan, L. A. Usman & O. S. Oguntoye," Assessment of selected medicinal plants indigenous to West Africa for antiprotozoal activity", South African Journal of Botany **113** (2017) 200.

[14] A. M. O. Amoussa, L. Latifou, B. Malanie, C. Vonthron-Senecheau & S. Ambaliou, "Triterpenoids from *Acacia ataxacantha* DC: antimicrobial and antioxidant activities", BMC Complementary and Alternative Medicine **16** (2016) 284.

[15] O. M. Bello, S. M. Jagaba, S. M. Abubakar, "Recent perception of the ethnomedicinal importance of *Acacia Ataxacantha* (DC) its pharmacology and phytochemistry: a review", FUDMA Journal of Sciences **3** (2019) 91.

[16] E. A. Sofowora, *Medicinal Plants and Traditional Medicine in Africa (2nd ed.)*, Spectrum Books limited Ibadan, Nigeria **9** (1993) 289.

[17] G. E. Trease & W. C. Evans, *A Textbook of Pharmacognosy, (13th ed.)* Bailliere Tindall Ltd, London (1989) 134.

[18] N. Lalnundanga, N. Lalchawimawii & K. Thanzami, "Antimicrobial activity of methanol extract of root bark of hiptage benghalensis (L) Kurz", Journal of Pharmacognosy and Phytochemistry **3** (2015) 119.

[19] R. Mathur, "Phytochemical and antimicrobial evaluation of plant extracts of enicostemma hyssopifolium", Journal of Pharmacognosy and Phytochemistry **2** (2013) 30.

[20] A. P., Ravathie, R., Sevvel, N. Nirmala & H. Kularajany, "Alternative culture media for bacterial growth using different formulation of protein sources", Journal of Natural Product and Plant Resources **2** (2012) 697-700.

[21] C. Thornsberry, & R. S. Donald, "Broth-Dilution method for determining the antibiotic susceptibility of anaerobic bacteria", Antimicrobial Agents and Chemotherapy **7** (1975) 15.

[22] D. Umaiyambigai & K. Saravanakumar, "Screening of phytochemical and antimicrobial activity from the leaves of pleiospermium alatum (Wall. & Arn.) Swingle", Journal of Chemical and Pharmaceutical Research **7** (2015) 498.

[23] T. C. Akapa, R. O. Arise, O. J. Olajide & I. T. Ikusemoro, "Ulceroprotective potentials of methanolic extract of *Acacia Ataxacantha* leaves in indomethacin and stress induced gastric ulcer models", International Journal of Biochemistry Research **4** (2015) 312.

[24] R., Venkataswamy, H., Mohamed Mubarack, A., Doss, S. Lakshmi Devi, & M. Sukumar, 2010. Antimicrobial activity of some ethnomedicinal plants used by the Malasar tribe of Tamilnadu, South India. Research Journal of Biological Sciences **2** (2010) 25.

[25] K. Seo, H. Akiyoshi & Y. Ohnishi, "Alteration of cell wall composition leads to amphotericin B resistance in Aspergillus flavus", Microbiology and Immunology **43** (1993) 1017.