EQUIJOST

Available online at www.equijost.com
Equity Journal of Science and Technology, 2021 8(1): 100 - 104
ISSN 2354-1814; E-ISSN 2682-5961

An Official Publication of Kebbi State University of Science and Technology, Aliero, Nigeria

Antibacterial Potential of Pod Extracts of Gum Arabic Tree (*Acacia nilotica*)

[1]Albert O. Fasogbon, *[2]Joseph O. Odewade [3]Ruth T. Omojoyegbe
[1]Department of Microbiology, Bayero University Kano, Kano State, Nigeria.
[2]Department of Microbiology, Federal University, Dutsin-Ma, Katsina, Katsina State, Nigeria.
[3]Department of Biological Sciences, Samuel Adegboyega University, Ogwa, Edo State, Nigeria
*Corresponding author's email: odewadejoseph@yahoo.com

Received: May 23, 2021: Accepted: June 26, 2021; Published Online: July 30, 2021

Abstract

This study was designed to determine the antibacterial potential of Gum Arabic Tree (*Acacia nilotica*) pod extract on some pathogenic bacteria which could have health implications. Powdered pods of *Acacia nilotica* were extracted with ethanol and distilled water separately. The extracts were tested for antibacterial activities against the test isolates using Agar Well Diffusion method. The extracts were further subjected to qualitative phytochemical screening to detect secondary metabolites present using standard procedures. The minimum inhibitory and minimum bactericidal concentrations of the extracts were also detected using standard procedures. The phytochemical screening revealed certain metabolites (steroids, saponins, terpenoids, tannins, flavonoids, and alkaloids) in the ethanol and aqueous extracts, except for steroids, which were present only in ethanol extract. The result of the sensitivity test showed that ethanol pod extract produced the highest and least zone of inhibition against *Pseudomonas aeruginosa* (22 mm) and *S. typhi* (15 mm) respectively at 50mg/ml while the aqueous extract of the pod exhibited activity against *S. typhi* (20 mm) and *E. coli* (12 mm) at 50 mg/ml. This showed that the *A. nilotica* pod extracts could serve as potential antibacterial agents against pathogenic bacteria.

Keywords: Antibacterial potential, *Acacia nilotica*, Pathogenic bacteria, Phytochemical, Screening

1. Introduction

In tropical countries, the major cause of 50% of death cases is an infectious disease. Death resulting from infectious diseases was ranked 5th in 1981, and became the 3rd leading cause of death with about 58% increase in 1992 [1]. The potency of the available antibiotics is increasingly becoming threatened by the emergence of MDR pathogens [2]. This calls for urgent attention for the discovery of newer, effective, and safer antimicrobial agents with various chemical structures and novel mechanisms of action due to the increased rate in the incidence of new (re-emerging) infections [3]. The potentials of higher plants as sources of new antimicrobial agents remain largely unexplored. Only a minute portion of the plant population has been well investigated phytochemically, biologically or pharmacologically. Natural or synthetic compounds serve as the source of numerous therapeutic agents [4]. Medicinal plants are rich sources of antimicrobial agents. Plants serve as the source of potential drugs and are utilized in folk medicine in various countries. Different phytochemicals possessing different medicinal properties against pathogenic microbes are sourced from medicinal plants around us. Among several plant species investigated for antimicrobial properties. Majority of them are yet to be thoroughly evaluated. The problems associated with infectious microorganisms thought to be controlled by antibiotics have resulted in the re-emergence of resistant strains called superbug [5, 6].

Plants explored for treating infections are as old as civilization [7]. Tradio-medicine still serves as habitual treatments [8]. This plant-based tradio-medicine continues to play a vital role in the health delivery system by accounting for about eighty percent of the world human population that depends solely on tradio-medicines for their health survival [9]. Despite synthetic chemistry as a means of drug discovery, bioactive plant extracts still have the potential to produce new novel drugs for the treatment and prevention of diseases [10]. Sourcing for newer and more potent drugs with little or safe side effects, completely reversible, self-administrable, and cost- effective is one of the most challenging pursuits in the area of medical and pharmaceutical sciences. Most of these attributes are observed in medicinal plants of natural origin. Medicinal plants (the bedrock of traditional medicine), were the subject of pharmacological research in combating pathogenic microbes during the last few decades [11]. A good example of a plant used in treating infections caused by pathogens is Gum Arabic Tree also known as *Vichelia nilotica* or *Acacia nilotica* which belongs to the family Fabaceae. It is a native species of Acacia in Africa and other continents. Different parts of the tree are widely utilized in folk medicine. Most of the acacias are of medicinal benefits to the human being. *Acacia nilotica* plant is used as antiscorbutic, nerve stimulant, cold, natriuretic, diarrhea, congestion, dysentery, leucorrhea, sclerosis, anti-oxidant, ophthalmia, etc. [12].

https://dx.doi.org/10.4314/equijost.v8i1.17

Antibacterial drugs are becoming less effective resulting in global health insecurity and challenge that is rapidly outpacing the available treatment options [13]. Therefore, this study was aimed at evaluating the antibacterial potential of pod extracts of *Acacia nilotica* against some pathogenic bacteria.

2. Materials and Methods

2.1 Collection / Authentication of Plant Samples

The pods of *Acacia nilotica* were collected at farmland in Bichi town, Kano state, located on Latitude 12º23'39''N and Longitude 8º27'94''E. The pods were taken to Herbarium, Plant Biology Department for identification and authentication.

2.2 *Extraction of plant samples*

The pods used in this study were thoroughly rinsed with distilled water. The samples were air-dried, grounded into a fine powder using a blender and stored in an air tight bottle for future use. The powdered pods (100 g each) of *A. nilotica* were soaked in 500ml ethanol and distilled water separately and kept for three days with intermittent shaking. The solutions were separately filtered using Whatman filter paper. The ethanol and aqueous extracts were separately concentrated *in-vacuo* and lyophilized.

2.3 Phytochemical screening of Acacia nilotica

Phytochemical analysis was carried out on the ethanol and aqueous pod extracts to detect the presence of certain secondary metabolites [14].

2.4 Source of the test isolates

Escherichia coli, *Salmonella typhimurium*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* were obtained from Microbiology Laboratory, Bayero University, Kano, Kano State, Nigeria. The isolates were sub-cultured on Eosin Methylene Blue, Salmonella-Shigella Agar, and MacConkey Agar to reconfirm the isolates after which they were stored on nutrient agar slants and kept in the refrigerator at 4 ºC.

2.5 Preparation of inoculum

A loopful of the test isolate was picked using a sterile wire 1oop and emulsified in 10 ml of sterile physiological saline. Exactly 0.5 McFarland standard was used as the standard for preparing the inoculum used [15].

2.6 Sensitivity testing

The antibacterial screening was achieved using Agar Well Diffusion Method in which a standardized inoculum equivalent to 0.5 McFarland standard was swabbed on each plate. Then 6 mm diameter wells were made on the agar using a sterile cork borer. The wells were filled with different concentrations of the extracts and were left for 1 hr at 37 ºC for the antimicrobial agents present in the extract to diffuse properly. Standard antibiotic (Ciprofloxacin) was used as control. The plates were incubated at 37ºC for 24 hrs. The zone of inhibition was measured in millimeters using a ruler [16].

2.7 Determination of Minimum Inhibitory Concentration (MIC)

A two-fold serial dilution of the reconstituted extract was prepared by transferring 2 ml of each dilution into 18 ml molten Mueller-Hinton agar and thoroughly mixed. The MIC was determined after 24hrs of incubation at 37ºC [17].

2.8 Determination of Minimum Bactericidal Concentration (MBC)

Nutrient agar plates were streaked with the sample from the MIC plates that produced no physical growth and were incubated at 37 ºC for 24 hrs to determine the minimum bactericidal concentration [18].

2.9 Statistical analysis

The data were expressed as mean ±SD (standard deviation) of three replicates and were statistically analyzed using one way analysis of variance (ANOVA). Values were considered significant at $p<0.05$.

3. Results and Discussion

3.1 The Yield

The weight of each pod samples before extraction was 100 g. After extraction, the aqueous extract yielded 14g while the ethanol extract was 28.6 g. The extracts were dark brown and gummy as shown in Table 3.1 below.

Table 3.1: The physical properties of *Acacia nilotica* (Pod) extracts and the yields

Solvent	Samples	Weight of Powdered samples (g)	Yield(g)	Texture	Colour
Aqueous	Pod	100	14	Gummy	Dark brown
Ethanol	Pod	100	28.6	Gummy	Dark brown

3.2 Phytochemical screening

Acacia nilotica pod extracts were found to possess secondary metabolites. Alkaloids, flavonoids, steroids, saponins, tannins, and terpenoids were all present in ethanol extract while the same secondary metabolites were also found in aqueous extract excluding steroid as stated in Table 3.2.

Table 3.2: Phytochemical Composition of *Acacia nilotica*

Extract	Alkaloids	Flavonoids	Steroid	Saponins	Tannins	Terpenoid
AE	+	+	+	+	+	+
EE	+	+	-	+	+	+

Key: + = present, - = absent, EE = Ethanol extract, AE = Aqueous extract

3.3 Antibacterial Activity of the Pod Extracts

Tables 3 and 4 below showed the antibacterial activity of aqueous and ethanol pod extracts of *Acacia nilotica* against the test isolates. The antibacterial activity of the ethanol and aqueous pod extracts showed that the extracts were active against the test bacteria.

According to Table 3.3, the aqueous pod extract of *A. nilotica*, produced the highest zone of inhibition (20 mm) against *Salmonella typhi* while the least zone of inhibition (12 mm) was produced against *Escherichia coli* at the same highest dose (50 mg/ml). The highest zone of inhibition exhibited by the least concentration (6.25 mg/ml) was observed in *S. typhi* (10 mm) while the extract was inactive against *E. coli* (6 mm) at an equal concentration of 6.25 mg/ml.

Table 3.4 below showed that at 50 mg/ml, the highest zone of inhibition was observed in *P. aeruginosa* (22 mm) while the least zone of inhibition was observed in *S. typhi* (15 mm). At the lowest concentration (6.25 mg/ml), the highest zone of inhibition of 11 mm was observed in *P. aeruginosa* while the least zone of inhibition of 8 mm was recorded against *Salmonella typhi*

.

Table 3.3: The Antibacterial Activity of Aqueous Pod Extract of *Acacia Nilotica*

	ZONE OF INHIBITION (mm) **				
Isolates	50mg/ml	25mg/ml	12.5mg/ml	6.25mg/ml	CPR (1mg/ml)
E. coli	12±0.82	10±0.00	08±0.00	0	19±1.63
S. typhi	20±1.41	15±2.16	12±0.00	10±0.82	28±2.16
*K. pneumonia*e	14±0.82	12±1.41	10±0.82	08±0.82	23±0.00
P. aeruginosa	16±0.82	14±1.41	11±0.82	08±1.63	26±0.82

Key: mm**=Mean of Three Replicates, CPR = Ciprofloxacin, 0=Not sensitive

Table 3.4: The Antibacterial Activity of Ethanol Pod Extract of *Acacia nilotica*

	ZONE OF INHIBITION (mm)				
Isolates	50mg/ml	25mg/ml	12.5mg/ml	6.25mg/ml	CPR (1mg/ml)
E. coli	16±0.82	12±0.82	10±0.82	09±0.82	28±0.00
S. typhi	15±0.00	13±1.63	11±0.00	08±0.82	26±0.82
K. pneumoniae	20±0.82	17±0.00	13±0.82	10±0.00	31±0.82
P. aeruginosa	22±0.00	18±1.63	14±1.63	11±0.82	35±1.63

Key: mm**=Mean of Three Replicates, CPR = Ciprofloxacin, 0=Not sensitive

3.4 Minimum Inhibitory and Minimum Bactericidal Concentrations of Acacia Nilotica Pod Extracts

The minimum inhibitory concentration of the aqueous and ethanol extracts was determined between 12.5 to 50 mg/ml and 6.25 to 12.5 mg/ml respectively while the minimum bactericidal concentration was found to be 12.5 to 50 mg/ml and 6.25 to 25 mg/ml respectively as shown in Table 3.5 below.

Table 3.5: The Minimum Inhibitory and Minimum Bactericidal Concentrations of The *A. Nilotica* Pod Extracts.

Bacterial isolates	Aqueous extract (mg/ml)		Ethanol extract (mg/ml)	
	MIC	MBC	MIC	MBC
E. coli	50	50	12.5	12.5
Salmonella typhi	12.5	12.5	12.5	25
Klebsiella pneumonia	25	50	6.25	12.5
Pseudomonas aeruginosa	12.5	25	6.25	6.25

The bioactive compounds found in plants are known as phytochemicals. Several bioactive compounds detected in *Acacia nilotica* plant extracts are known to have medicinal significance, health benefits, and physiological activities [19]. Table 3.2 showed the phytochemical constituents of both the aqueous and ethanol pod extracts of *Acacia nilotica*. These results supported previous studies conducted by Mohammed *et al.* and Atiku *et al*. [20, 21].

Screening for antibacterial activity indicated that *Acacia nilotica* pod extracts possessed antibacterial activities against the test isolates. The result of the sensitivity test

showed that ethanol pod extract produced the highest and least zone of inhibition against *Pseudomonas aeruginosa* (22 mm) and *S. typhi* (15 mm) respectively at 50mg/ml while the aqueous extract of the pod exhibited activity against *S. typhi* (20 mm) and *E. coli* (12 mm) at 50 mg/ml (Table 3 and Table 4). In this study, the ethanol extract was found to have a higher inhibitory effect than the aqueous extract. This agrees with the finding of Shuai *et al*. [23].

The antibacterial activity of the extracts was also evaluated by determining their minimum inhibitory and minimum bactericidal concentrations. The results of the MIC suggested that the pod extracts of *Acacia nilotica* could be a good source of bacteriostatic activity against the isolates. The MIC of the aqueous and ethanol extracts was determined between 12.5 to 50 mg/mL and 6.25 to 12.5 mg/mL respectively while the MBC was found to be 12.5 to 50 mg/mL and 6.25 to 25 mg/mL respectively (Table 5). The study showed that the ethanol pod extract was more potent compared with the aqueous pod extract. Therefore, the lowest MIC and MBC value of the pod extracts against the test isolates indicated that the extract was a good one. The potency may be a result of phytochemical constituents present in the pod extracts.

4. Conclusions

The extracts of *Acacia nilotica*, particularly the ethanol pod extract used in this study displayed a good antibacterial activity. Thus, the literature review so far tends to support the fact that the plant (*Acacia nilotica*) could find application towards treating ailments. This finding also suggests good antibacterial potential of the active components (phytochemical constituents) present in the extracts, which might be responsible for its biological activity.

References

1. Venkataswamy R, Doss A, Muhammed MH, Sukumar M. Phytochemical, HPTLC Finger Printing and Antibacterial Activity of *Acacia nilotica* (D.) Delile. Hygeia. *Journal of Dental Medicine*. 2010; 2(2): 38-42.
2. Doss A, Muhamed H, Dhanabalan R. Antibacterial Activity of Tanins from *Solanum Trilobatum* Linn. Leaves. *Indian Journal of Science and Technology*. 2009; 2(2): 41–43.
3. Anand SP, Doss A, Nandagopalan V. Antibacterial Studies of *Clitoria ernatea* Linn. A high Potential Medicinal Plant. *International Journal of Applied Biology and Pharmaceutical Technology*. 2011; 2(3): 453–456.
4. Mahesh B, Satish S. Antimicrobial Activity of Some Important Medicinal Plan against Plant and Human Pathogens. *World Journal of Agricultural Sciences*. 2008; 4: 839–843.
5. Udobi CE, Onaolapo JA, Agunu A. Antibacterial Activities and Bioactive Components of the Aqueous Fraction of the Stem Bark of *Parkia biglobosa* (Jacq) (Mimosaceae*). Nigerian Journal of Pharmaceutical Science*. 2008; 7(1):49–55.
6. Levy SB, Marshal B. Antibacterial resistance worldwide: Causes, Challenges and Responses. *Natural Medicine*. 2004; 10: S122–S129.
7. Fabricant D, Farnsworth N. The Value of Plants used in Traditional Medicine for drug Discovery. *Medicine: Environmental Health Perspectives.* 2001; 1091(11):69.
8. Alviano D, Alviano A. Plant extracts: search for new alternative to treat microbial Diseases. *Current Pharmaceutical Biotechnology*. 2009; 10:106-121.
9. Christiana JD, Ishaku LE, Nkechi VO, Jurbe GG, Olumola OO, Micah SM, Sunday M, David S. Antidiarrheal Evaluation of Aqueous and Ethanolic Leaf Extracts of *Acacia sieberiana* D.C. (Fabaceae) in Albino Rats. *Asian Journal of experimental biological Sciences*. 2012; 3(4): 79 - 803.
10. Raskin I, Ribnicky D, Komarnytsky S, Ilic N, Poulev A, Borisjuk N, Brinker A, Moreno A, Ripoll C, Yakoby N, Cornwell T, Pastor I, Fridlender B. Plants and Human Health in the Twenty-first Century. *Trends in Biotechnology*. 2002; 20 (12): 522 - 531.
11. Unny R, Chauhan AK, Joshi YC, Dobhal, MP, Gupta RS. A Review on Potentiality of Medicinal Plants as the Source of New Contraceptive Principles. Phytomedicine. 2002;10: 233 - 260.
12. Saini ML. Comparative Pharmacognostical and antimicrobial studies of Acacia Species (Mimosaceae). *Journal of Medicinal Plants Research*. 2008; 2(12): 378–386.
13. World Health Organization (WHO). Antimicrobial Resistance Global Report on Surveillance. 2014, pg. 23 of 256.
14. AOAC. Official Methods of Analysis of Association of Official Analytical Chemists. 2010, 18th Edition, Washington, D. C.
15. Cheesbrough M. *District Laboratory Practice Manual in Tropical Countries*. Part 2. Cambridge: Cambridge University Press; 2000.
16. Clinical and Laboratory Standards Institute. *Performance standards for Antimicrobial Susceptibility Testing. Twenty Second Informational Supplement Update, CLSI Document, M100.* Clinical and Laboratory Standards Institute, Wayne, PA; 2017.
17. Akinpelu DA, Kolawole DO. Phytochemical and Antimicrobial Activity of Leaf Extract of *Piliostigma Thonningii* (Schum.). *Science Focus*. 2004; 7: 64–70.

18. Ashraf A, Mostafa AA, Al-Askar Khalid S, Almaary Turki MD, Essam NS, Marwah MB. Antimicrobial Activity of Some Plant Extracts Against Bacterial Strains Causing Food Poisoning Diseases. *Saudi Journal of Biological Sciences*. 2017; 25(2): 361–366.
19. Cheik YA, Summers RA, Kahaka G. Qualitative and Quantitative Analysis of Phytochemical Compounds in Namibian *Myrothamnus Flabellifolius*. *International Science and Technology Journal of Namibia*. 2015; 5:71–83.
20. Mohammed AM, Adamu MW, Ali AA, Isa AG. Antimicrobial Activities of Aqueous and Ethanolic Leaves Extracts of Ficus Platyphylla Del. *Archives of Applied Science Research.* 2015; 7 (3):37–42.
21. Atiku A, Oladipo OO, Forcados EG, Usman SA, Mancha DM. Anti-nutritional and Phytochemical profile of Some plants grazed upon by ruminants in North Central 62 Nigeria During the Dry season (January to April). *International Journal of Livestock of Livestock Production*. 2016; 7(4): 19–23
22. Zellagui A, Gherraf N, Elkhateeb A, Hegazy MEF, Mohamed TA, Touil A, Shahat AA, Rhouati S. Chemical Constituents from Algerian *Foeniculum Vulgare* Aerial Parts and Evaluation of Antimicrobial Activity. *Journal of the Chilean Chemical Society*. 2011; 56:759–763.
23. Shuai P, Weichang D, Hansong Y, Yuhua WS, Xuelin, W, Shumin S. Antibacterial Activity of Aqueous and Ethanolic Extracts of Portulaca oleracea L and Taraxacum Mongolicum Against Pathogenic Bacteria of Cow Mastitis, *International Journal of Applied Research in Veterinary Medicine*. 2015; 49 (6):827–829.