Phytochemical Evaluation and Antimicrobial Activity of Leaf Extracts of *Cymbopogon Citratus* (Lemon Grass)

Abbas Ibrahim[*1], Umar Dan'azumi[2], Suleiman Ali[2], Mohammed Audu[3], Saratu Yakubu[1], Amina Rabi'u Umar[1], Mohammed Sani Dankoly[4], M. Ibrahim Misau[5], Ibrahim Sa'id Isma'il[5], Mohammed Aminu Usman[5]

[1]*Department of Chemistry, Bauchi state University, P.M.B 65, Itas/Gadau, Bauchi state, Nigeria*
[2]*Department of Basic Science, College of Agriculture Gujba, P.M.B 1104, Damaturu Yobe State, Nigeria*
[3] *Department of Science Laboratory Technology, Mai Idris Alooma Polytechnic Geidam, P.M.B 1020, Geidam Yobe State, Nigeria*
[4]*Department of Biological Sciences, University of Maiduguri P.M.B 1069, Borno State, Nigeria*
[5]*School of Remedial and Continuing education, A. D. Rufa'I College for Legal and Islamic Studies Misau, P.M.B 004, Misau Bauchi State, Nigeria*

***Abstract*: - Plants are important source of drugs especially in traditional medicine. Fifty gram (50g) of *Cymbopogon citratus* was extracted with different solvent such as ethanol, acetone chloroform, petroleum ether, diethyl ether and n-hexane using merceration (cold infusion method) and phytochemically screened which revealed the presence of some important secondary metabolites such as alkaloids, flavonoids, tannins, saponins and steroids which is found presence in all the extracts. The extracts were tested against both negative and positive gram bacteria which include ; *Staphalococcus aureus* and *Escherichia coliby* using disc diffusion assay and the activity of the plant extract against the listed microorganism were recorded as result and was assessed by measuring of zone diameter around the disc paper and it showed susceptibility at the zone of inhibition of 10µg/ml and 8µg/ml while it showed resistance at 6 µg/ml and 4 µg/ml although this plant can be use in the treatment of diseases caused by the microorganism and as a source of useful phytochemical in drugs.**

***Keywords:* *Cymbopogon citrates, Staphalococcus aureus, Escherichia coliby,* Phytochemical**

I. INTRODUCTION

Plant are important source of drugs especially in traditional medicine(1). It is a common practice In Nigeria and other parts of the world to used plant as crude extracts, decoction, infusion, to treat common infection and chronic conditions. According to WHO, over 70% of the world population rely on medicinal plants for primary health care and there are report from various researches on natural substances of plant origin which are biologically active, with desirable and antimicrobial activity (1).A medicinal is used to attempt to maintain health, to be administered for a specific condition, or both, whether in modern medicine or in traditional medicine. Plant can cause adverse effects and even death, whether by side-effects of their active substances, by adulteration or contamination, by overdose, or by inappropriate prescription. During the last two decades, the development of drug resistance as well as the appearance of undesirable side effects of certain antibiotics has lead the search for new antimicrobial agents mainly among plant extract with goal to discover new chemical structures, which overcome the above disadvantages (2). It has been reported that there are over 8000 species of known medicinal plants in African considered as essential part of traditional health care systems. More than 80 percent of African population is dependent on this cheap and effective traditional medicines used against many diseases and infections (3). The aim of this research is to identify the phytochemical components present in the leaves of Lemon grass and the antimicrobial effect of the plant through the following objectives

II. EXPERIMENTATION

Sample collection and identification

Lemon grass leaf was collected from Jos plateau state and their environment such as state lowcost and anguwan doki bukuru. The sample was healthy and uninfected and was identified by appropriate voucher in the Herbarium unit Botany Department at Bauchi State University Gadau, Bauchi State.

Sample Preparation and Extraction

The lemon grass leaf sample was air dried under laboratory condition and the sample was grinded into powder using a well cleaned wooden mortar and pestle, 50g of the powdered material was extracted using solvent such as ethanol, chloroform and diethyl ether using merceration (cold Infusion method) for 24hours , the extracts were concentrated under reduce temperature .

Phytochemical Screening

The ethanol extract, chloroform extract and diethyl ether extract were phytochemically screened for alkaloids, flavonoids, saponins, sreroids and polyphenols using guided protocols (4,5,6)

Test Organisms

The test organisms used namely Staphlocuccus aureus, Esherichia coli, were obtained from stuck cultures in the microbiology laboratory for Bauchi state university Gadau Nigeria. They were sub cultured and identified based on their colonial morphology, microscopic appearance. The test organisms were sub cultured in 10ml broth each and incubated at 37C for 24 hours. After 24 hours, the organisms were sub cultured into fresh Mueller Broth and incubated for 3 hours which was used for analyses.

Antimicrobial Susceptibility Test

The antimicrobial test was carried out using disc diffusion method using guided protocol.

Preparation of sensitivity discs

Discs of about 6mm in diameter was made from Whatmans No.1 filter paper, then transferred into Bijour bottle and sterilized at 121Cº for 15 minutes using standard procedure.

Preparation of culture media

Nutrient agar was prepared according to manufacturer's instruction and sterilized by Autoclaving at 121ºC for 15 minutes and then cooled to room temperature. The molten nutrient agar was poured in to Petric dish and allowed to solidify.

Standardization of inoculums

The standardization of inoculums was carried out using inoculation wire loop. Enough material from an over nigh culture of the test organisms were transferred in to a test tube containing normal saline until the turbidity of the suspension matched the turbidity of the 0.5Mcfarland standards as described by national community for clinical laboratory standards.

Bioassay procedure

Standard inoculums of the isolates were swabbed on the surface of prepared and solidified nutrient agar in separate Petri dishes. The discs of the extracts (3000µg/ml, 2000µg/ml 1000µg/ml, 100µg/ml) and the standard antibiotic discs (ofloxacin and Erythromycin) were placed on the surface of the inoculated media at intervals. The plates were then incubated at 37ºC for 24 hours, observation and measurements of zones of inhibition (in millimeters) were made.

Determination of minimum inhibitory concentration (MIC) and Minimum Bacterial concentration growth (MBC)

Minimum inhibitory concentration of the extract and fractions were prepared by serial doubling dilution using distilled water to obtain concentrations of 2000µg/ml, 1000µg/ml and 100µg/ml. Equal volume (2ml) of extract and Muller-Hinton broth were mixed. Specifically 0.1ml of standardized inoculate (3.310^6 CFU/ml) was added to each of the test tube above. The tubes were incubated aerobically at 35ºC for 24 hours. Tubes containing broth and leaf extract without inocula which served as positive control while tubes containing broth and inocula served as negative control. The tubes were observed after 24 hours incubations to determine minimum inhibitory concentration (MIC). That is the lowest concentration that showed no evidence of growth. Sterile Muller-Hinton agar plates were separately inoculated with sample from each of the test tubes that showed no evidence of growth. The plates were further incubated at 35ºC for 24 hours and observed. The highest dilution that yielded no bacterial growth was regarded as MBC.

III. RESULT

Table 1: Phytochemical Screening of ethanol extract, acetone extract, chloroform extract, Petroleum ether extract, ethyl ether extract and n-hexane extract of the leaf of *Cymbopogon citratus*

Phytochemicals	Extracts					
	Ethanol extract	Acetone extract	Chloroform extract	Petroleum ether extract	Ethyl ether extract	n-Hexane
Alkaloids	+	+	-	+	+	+
Flavonoids	+	-	+	+	+	+
Saponins	-	+	+	-	+	+
Tannins	+	+	-	+	+	-
Steroids	+	+	+	+	+	+

Key: + = Present, - = Absent

Table 2: Result of the Antibacterial activity of ethanolic extract of lemon grass on *Escherichia coli*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	17	S
8	15	S
6	12	R
4	11	R

Key: S= Susceptibility R= Resistance

Table 3: Antibacterial activity of Acetone extract of lemon grass leaves *Staphylococcus Aureus*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	14	S
8	16	S
6	12	R
4	11	R

Table 4: Results of the Antibacterial activity of chloroform extract of lemon grass on *Escherichia coli (E. coli)*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	14	S
8	16	S
6	12	R
4	11	R

Table 5: Results of Antibacterial Activity of Petroleum ether of lemon grass extract on *Staplococcus Aureus*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	17	S
8	15	S
6	12	R
4	11	R

Table 6: Results of the Antibacterial Activity of Diethyl ether extract of lemon grass on *Escherichia Coli*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	17	S
8	15	S
6	12	R
4	11	R

Table 7: Results of Antimicrobial Activity of N-hexane extract of lemon grass on *Staphylococcus Aureus*

Conc. Of the extract (µg/ml)	Zone of Inhibition (mm)	Susceptibility pattern
10	14	S
8	15	S
6	12	R
4	11	R

Table 8: Antibacterial Activity Shows MIC and MBC of *Cymbopogon Citrastus*

Isolate	Ethanol extract	Chloroform extract	Acetone extract	Petroleum ether extract	n-Hexane extract	Diethyl ether extract
	MIC MBC	MIC MBC	MIC MBC	MIC MBC	MIC MBC	MIC MBC
E. Coli	(4, 10)	(4, 10)	(4, 8)	(4, 10)	(4, 10)	(4, 8)
S. Aureus	(4, 10)	(6, 10)	(4, 8)	(4, 10)	(4, 10)	(4, 8)

Key: (MIC) = Minimum Inhibitory Concentration (MBC) = Minimum Bacterial Concentration

IV. DISCUSSION

Extraction and Phytochemical screening of bioactive component from medicinal plants permits the demonstration of their physiological activities. The phytochemical analyses of the extracts ethanol, acetone, chloroform, petroleum ether, diethyl ether and n-hexane showed the presence of some important secondary metabolites such as steroids, tannins, flavonoids, alkaloid and saponins. Research have shown that tannins have been found to inhibit bacterial growth and also capable of protecting certain plants against infection (7). The presence of this phytochemical implicates the medicinal value as well as the antibacterial effect of this plant. The leaf of *Cymbopogon citratus* showed more antimicrobial activity despite the presence of bioactive agents in the leaf extracts, which showed that there are more different types of active ingredient in different plant species and different plant parts. The antimicrobial activity of the plant extract against tested organisms increase in the concentration of the extracts and this could be attributed due to presence of phytochemicals presence in this study. The antimicrobial Screening of the ethanol, Acetone, chloroform, Diethyl ether, n-hexane and Petroleum ether showed that the Acetone of the isolate *Esherichia coli* and *Staphylococcus aureus* has more inhibitory effect than Ethanol extract.

V. CONCLUSION

Cymbopogon *citratus* have led to the following conclusions: The extracts of *Cymbopogon citratus* leaf (chloroform extracts) possessed intermediate antimicrobial activity against *Staphylococcus aureus*, and *E. coliand*, the MIC of *Cymbopogon citratus* reveals that a higher dose of the plant extract is required to bring about a significant activity in the body and bioactive components were identified in the plant which include flavonoids, tannins, steriods, alkaloids and saponin.

REFERENCES

[1]. Hamid, A. A., Aiyelaagbe, O. O., Usman, L. A., Ameen, O. M. & Lawal, A. (2010). "Antioxidants: Its medicinal and pharmacological applications". African J. of Pure and Applied Chemistry. 4(1):00710

[2]. Harbone, J. B. (1973) "Phytochemical methods". A guide to modern technique of plant Analysis London: Chapinanand Hall; 33–185

[3]. Neuwinger, H. D. (2000). "African traditional medicine: a dictionary of plant use and applications". Med. Pharm.Scientific, Stuttgart, Germany, 589.

[4]. Silva, L. G. Lee, I. S. & Afkinnghorn, D. A. (1998). "Special Problem with Extraction of Plant In Natural Products Isolation (cannel R. JD)". Human Press Inc. 999, Review Drive, Suite 208 Totowa, New Jersey, USA 072512., 343-364

[5]. Trease, G. E. & Evans, W. C. (2002). "Textbook of Pharmacogonosy, 14th Edition". W.B Saundors Company LTD. 24-25 Oval, London. NW7DX, UK and printed by Harcourt Brace and Company. 13-53, 117-139,227,293-334,471-551.

[6]. Brain, K. R & Turner. (1995). "The Practical Evaluation of Mophormaceutical". *J. Wright Scientechnica*., 190-191.

[7]. Chung, K. T., Lu, Z. & Chou, M. W (1998). "Mechanism of inhibition of tannic acid and related compounds on the growth of intestinal bacteria". Food and chemical Toxicology 36,1053-60