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EVALUATION OF PHYTOCHEMICAL AND ANTIBACTERIAL PROPERTIES OF TERMINALIA AVICENNIOIDES CRUDE EXTRACT AGAINST SELECTED BACTERIA FROM DIARRHOEIC PATIENTS

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ABSTRACT
Phytochemical screening of aqueous and ethanol crude extracts of the different plant parts of Terminalia avicennioides was carried out using standard chemical evaluation methods. The antibacterial effects of aqueous and ethanol crude extracts of Terminalia avicennioides against E.coli and S.typhimurium clinical and reference isolates from diarrhoeic patients were also evaluated using agar-well diffusion method. The minimum inhibitory concentrations (MICs) and minimum bactericidal concentrations (MBCs) of aqueous and ethanol crude extracts were evaluated by broth dilution techniques. The result revealed the presence of carbohydrates, alkaloids, tannins, flavonoids saponins, triterpens and glycosides. All bacteria were found to be susceptible to the extracts which were indicated by the various zones of inhibition. The activity of extracts was concentration dependent. The reference strains were less susceptible to all extracts at low concentrations of 12.5mg/ml, but highly susceptible to extracts at varied concentrations of 25, 50 and 100mg/ml. However, all test bacteria were more susceptible to the ethanol extracts compared to the aqueous extracts with mean zones of inhibition ranging between 0.68 $\pm$ 2.54 mm to 22.08 $\pm$ 1.75 mm on E. coli clinical isolates, 0.0 $\pm$ 0.0 mm to 20.00$\pm$0.mm on E. coli reference isolate, 3.08 $\pm$ 6.0 mm to 21.50 $\pm$ 0.00 mm on S. typhimurium clinical isolates and 0.00 $\pm$ 0.mm to 20.00 $\pm$ 0.0 mm on S. typhimurium reference isolate. The ethanol crude extracts exhibited lower MICs (12.5 to 25mg/ml) and MBCs (25 to 50mg/ml) values indicating higher efficacy of ethanol extracts, with the leaf extract demonstrating the highest activity against all the bacterial isolates. The important bioactive compounds present in the plant may be responsible for the observed antibacterial activity of the plant and hence its potential use as an antibacterial agent.
Keywords: Phytochemical, Antibacterial effect, Terminalia avicennioides, diarrhoeaic patients.

INTRODUTION

Enteric bacteria are important pathogenic group because of their involvement in a number of diarrhoeal diseases that account for a significant number of death among infants and adults living in developing countries. Some strains, of *Salmonella typhimurium* and *Escherichia coli* are emerging as significant agents of diarrhoea world wide and have also become endemic in many parts of developing countries including Nigeria. Some *Escherichia coli* strains can cause malnutrition and growth defect in children (Iruka *et al*., 2003). *Salmonella typhimurium* and *Salmonella enteritidis* are common causative agents of bacteremia in young children living in developing countries. Similarly, Salmonellosis remain an important public health problem (Rotimi *et al;* 2008) and is more frequent with people who consume foods from contaminated fresh, poultry, water, fruits and vegetables (Momoh *et al.,* 2013).

Moreover, diarrhoea is self-limiting but when it is as a result of bacterial infection antibiotic therapy may be needed. Despite the numerous number of antibiotics used for effective treatment of diarrhoea and other ailments, there is a need to search for alternative due to the growing incidence of multi drug resistance among bacterial pathogens, coupled with the rising costs and low therapeutic index of many synthetic drugs especially in developing countries with weak economic indices (Bulus *et al.,* 2011; Adebolu, 2012). Species of plants belonging to the family combretaceae have been tested for their antimicrobial activities against some pathogenic microorganisms that are prone to drug resistance (Mann *et al*., 2008). In the light of this, an update information on the properties and uses of any medicinal plant belonging to this group needs to be investigated.

Terminalia avicennioides (Guill and Perr), has been discovered to have some medicinal values. It is used in the treatment of different types of ailments. The plant grow abundantly in the Savanna region of Africa as a shrub or small tree. It is popularly found growing in the northwest vegetations of Nigeria. The common name of the plant; *T.avicennioides* is 'Indian laurel' and in Nigeria, it is locally called 'baushe' in Hausa 'Idi' in Yoruba, 'Edo' in Ibo, 'Kpace' in Nupe; 'Kpayi' in Gwari and 'Bodeyi' in Fulfulde (Mann *et al.,* 2008).

However, there are few reported literatures, on the antidirrhoeal activity of *Terminalia avicennioides* against the endemic bacterial pathogens associated with diarrhoea. Therefore, screening the plant for its activity against some selected endemic bacterial pathogens associated with diarrhoea will provide valuable source of new anti diarrhoeal agent that may offer safe and effective prevention and treatment of diarrhoeal diseases.

MATERIALS AND METHODS

Collection and Identification of Plant Material

Terminalia avicennioides plant (Guil and Perr) was collected from Karaukaraw village in Zaria Local Government in Kaduna state, Nigeria in October 2011. The plant was identified and confirmed by a botanist from Biological Science Department in Ahmadu Bello University Zaria Nigeria with a voucher number 104, deposited at the herbarium section.

Preparation of Plant Material

The fresh plant material was separated into three potions (roots, stems and leaves) and air dried at room temperature under shade for two weeks. Each dried plant part was separately ground to coarse powder using a motar and pistle, and put in separate containers.

Extraction of Plant Material

To each powdered plant part (800g leaf, stem and root) was macerated in two different solvents, (1 liter of distilled water and 70% ethanol) in conical flasks. The flasks were left to stand for 72hours with constant shaking. At the end of 3 days, each extract was filtered with a muslin cloth and then with filter paper. The filtrate was concentrated in a water bath at 40^0C for two (2) days. Each dried crude extract was kept in a sterile container and kept in a refrigerator for further use. The percentage yield of each aqueous and ethanol crude extracts of *T. avicennioides* plant part was calculated (Temidayo, 2013).

Phytochemical screening.

The crude aqueous and ethanol extracts of *T. avicennioides* leaf, stem and root barks were subjected to standard reagent-based phytochemical tests for ten major constituents, namely: carbohydrates, cardiac glycosides, saponins, flavonoides, Tannins, alkaloids anthroquinones, phenols, steroids, and triterpenes. (Trease and Evans, 1983; Tiwari *et al.,* 2011).

Collection of Test Bacteria

A total of 58 clinical isolates of *E.coli* and 20 clinical isolates of *S. typhimurium* isolated from patients suffering from diarrhoea were obtained from Yusuf Dantsoho Memorial Hospital Tudun Wada (YDH) Kaduna, Gwamnaawan General Hospital (GAH) Kakuri Kaduna and Ahmadu Bello University Teaching Hospital (ABU) Zaria Kaduna state. The standard strain, *E .coli* ATCC 25922 was sourced from the Department of Pharmaceutical Microbiology ABU, Zaria while *S. typhimurium* ATCC 14028 reference strain was sourced from the National Veterinary Research Institute (NVRI), VOM, Jos. Plateau state. All Samples were collected in nutrient agar slants, labeled, placed in a cold box, transported to the Post graduate Laboratory of the Department of Microbiology ABU Zaria and incubated at 37^OC for 24 hours

Identification of Test Bacteria

Inocula from overnight growth broth culture of *E.coli* were streaked on freshly prepared plates of eosin methylen blue (EMB) and on MacConkey agar plates. Similarly, inocula from overnight growth broth culture of *S. typhimurum* were streaked on freshly prepared plates of xylose lysine deoxycholate (XLD) and Brilliant green agar BGA (pro-lab diagnostic USA). All plates were incubated for 24hours at 37^OC to obtain fresh cultures of test isolates. The identity of each isolate was confirmed by conventional biochemical test, microgen identification kit and PCR assay**.**

Antibacterial Activity of Crude Extracts

Agar well diffusion method, described in the National Committee for Clinical Laboratory Standards manual, (2OO3) with slight modification, was used to determine the antibacterial activity of the crude aqueous and ethanol extracts of leaves, stems and roots of *Terminalia avicennioides* against the clinical and reference isolates of *E.coli* and *S. typhimurium*. One gram each of aqueous and ethanol crude extracts of leaf, stem and root of *T. avicennioides* was weighed and added to 10ml each of 10% dimethyl sulfoxide (DMSO) to obtain 100mg/ml stock solutions of each extract. Using two-fold serial dilution, concentrations of 50mg/ml 25mg/ml and 12.5mg/m were prepared from each stock solution. The different concentrations, were labeled and kept in bijou bottles for subsequent use. Some quantity of each test bacteria from an overnight growth culture was added to 2ml of sterile physiological saline as a suspension medium and compared with 0.5 McFarland standard (1.5×10^8 CFU ml). Using a micropipette about 100µl of standardized inoculum of a bacterial suspenssion was inoculated into Mueller Hinton agar plate and spread evenly over the entire surface of the plates using a sterile cotton swab stick. The plates were left for 10 minutes before wells were dug in the agar using a 6mm sterile cork borer. The wells were each filled with 100µl volume of the prepared concentrations of extracts. Additional wells were filled with dimethyl sulfoxide (DMSO) to serve as negative controls. Each extract test was replicated three times. The plates were left for 10 minutes at room temperature for diffusion of extracts into the agar to take place and then incubated at 37^OC for 24 hours. For all bacteria tested, zones of inhibition of growth were examined, and the diameter of each zone was recorded in millimeters with a meter ruler. The means were calculated to the nearest whole number.

Determination of Minimum Inhibitory Concentration (MIC)

The MICs of crude extracts against the clinical and reference isolates of *E .coli* and *S. typhimurium* were evaluated using broth dilution method. The following concentrations of extracts; 25mg/ml, 12.5mg/ml and 6.25mg/ml, were prepared by two - fold serial dilutions. 1ml of extract concentration was added to a test tube containing 9ml of Mueller Hinton broth.

About 100µl each of a standardized inoculum of a test bacterium was added to mixtures of different concentrations of extracts with Mueller Hinton broth. The test tubes were incubated at 37^{O}C for 24 hours. The growth of bacteria in the broth were examined which were indicated by the turbidity of the broth. However, the lowest concentration of the extract which inhibited the growth of a test organism was recorded as the minimum inhibitory concentration (MIC). Negative controls were Mueller Hinton broth only and Mueller Hinton broth with extract. While positive control comprised of Mueller Hinton broth with test bacteria. (Sule and Agbabiaka 2008).

Determination of Minimum Bactericidal Concentration (MBC)

The minimum bactericidal concentration (MBC) was determined from the positive MIC tubes. An inoculum from a positive tube was sub cultured on to nutrient agar plate and incubated at 37^{0}C for 24 hours. The lowest concentration of extract that yielded no growth was the minimum bactericidal concentration (MBC) (Andrew, 2001). The negative controls were nutrient agar only and nutrient agar with extract only.

Statistical analysis

Data was analyzed by ANOVA and student t-test using Statistical Package for Social Sciences (SPSS) computer package. For all evaluations, the level of statistical significance was fixed at $P \leq 0.05$.

RESULTS.

After the extraction process, the aqueous crude extracts of the leaf, stem and root barks of *T. avicennioides* showed higher yields of 20.2%, 19.4% and 18.7%, while the yields from ethanol crude extracts were 18.8%, 17.6% and 17.1% respectively (Table 1).

The photochemical screening carried out on the leaves, stems and roots of *T. avicennioides* using qualitative methods, revealed the presence of the phytoconstituents shown in Table 2. Carbohydrates, cardiac glycosides, saponins, flavonoides, tannins, alkaloids, phenols and triterpenes were detected in all the aqueous and ethanol crude extracts of leaf, stem and root barks of *Terminalia avicennioides*. However, anthroquinones and steroids were not detected in all the crude extracts tested.

The results of susceptibility tests of clinical isolates of *Escherichia coli, Escherichia coli* ATCC 2592, clinical isolates of *Salmonella typhimurium* and *Salmonella typhimurium* ATCC 14028 to different concentrations of aqueous and ethanol extracts of leaf, stem and root of *Terminalia avicennioides* showed that there were no inhibitory activities at 12.5mg/ml concentrations of the aqueous leaf extract on *E.coli* and *S.typhimurium* clinical islates. All the reference strains were resistant to the aqueous leaf extract at low concentrations of 12.5mg/ml. Higher inhibitory activities were exibited on all test isolates at 25, 50 and 100mg/ml concentrations of extract with mean zones of inhibition ranging between 0.34 ± 1.82mm to 19.81 ± 1.24 on *E. coli* clinical isolates, 0.0 to 19.0 ± 0.0 on *E. coli* reference isolate, 1.43±3.63mm to 19.85+1.35mm on *S. typhimurium* clinical isolate and 0.0 to 20.00 ± 0.0mm on *S.typhimurium* reference strain respectively (Figure 1). However, all test bacteria were highly susceptible to the ethanol extracts compared to the aqueous extracts (Figure2) with mean zones of inhibition ranging between 0.68 ± 2.54mm to 22.08 ± 1.75mm on *E. coli* clinical isolates, 0.0 ± 0.0mm to 20.00±0.0mm on *E. coli* reference isolate, 3.08 ± 6.08mm to 21.50 ± 0.00 on *S. typhimurium* clinical isolates and 0.00 ± 0.0 to 20.00 ± 0.0 on *S. typhimurium* reference isolate. Similarly there were low inhibitory activities at 12.5 mg/ml concentration of the stem extract on clinical isolates of *E.coli* (0.52± 2.23 mm) and no inhibitory activities at same concentration on the remaining bacteria tested. But, there were significant inhibitory activities at 25, 50 and 100 mg/ml concentrations with mean zones of inhibition ranging between 0.52 ± 2.23mm to 17.98 ± 2.03mm on clinical isolates of *E.coli*, 0.00 to 19.00 ± 0.00mm on *E.coli* reference isolate, 0.00 to 17.95±1.99mm on clinical isolates of *S.typhimurium* and 0.00 ± 0.00 to 19.00 ± 0.00 on *S.typhimurium* reference isolate respectively (Figure3). The ethanol stem extract also demonstrated higher inhibitory activities compared to the aqueous stem extract with mean zones of inhibition ranging between 0.00 to 18.95 ± 2.07mm, 0.00 to 20.00 ± 0.00 on clinical isolates of *E.coli* and *E.coli* reference isolates, 10.50 ± 9.99mm to 19.85 ± 0.99mm and 0.00mm to 20.00±0.00mm on clinical isolates of *S. typhimurium* and *S .typhimurium* reference isolates (Figure4).

All the bacteria tested also exhibited high degree of resistance to the aqueous root extracts at 12.5mg/ml concentrations but were highly susceptible to the aqueous root extracts at varied concentrations of 25, 50 and 100mg/ml concentration (Figure 5) with mean zones of inhibition ranging between 0.0mm to 18.12 ± 2.40mm on clinical isolates of *E. coli,* 0.00 to 19.00 ± 0.00 on *E. coli* reference isolates, 0.0 to 19.85 ± 1.39 on clinical isolates of *S. typhimurium* and 0.00 to 19.00 ± 0.00 on *S. typhimurium* referece isolate. There were no inhibitory activities observed among the two reference strains of bacteria at 12.5mg/ml concentrations of the ethanol root extracts but all bacteria showed higher susceptibilities to the ethanol extracts compared to the aqueous extracts at varied concentrations of 25, 50 and 100 mg/ml with mean zones of inhibition ranging between 0.17± 1.31.00 to 20.19 ± 1.58 on *E. coli* clinical isolates, 0.00 to 20.00 ± 0.00 on *E. coli* reference isolate, 1.61 ± 3.41 to 20.73 ± 1.29 on *S. typhimurium* clinical isolates, and 0.00±0.41 to 21.00 ± 0.00 on *S. typhimurium* referece isolate respectively (Figure 6).

The MICs of aqueous extracts of the different plant parts against *E. coli* clinical isolates and *E. coli* reference strain were all found to be 25mg/ml while the MICs of extracts against *S. typhimurium* clinical isolates and *S. typhimurium* reference strain ranged

between 12.5 and 25mg/ml. Similarly the MBC values of all aqueous extracts against *E. coli* clinical isolates and *E. coli* referece strain were found to be 50mg/ml. The MBC of extracts against *S. typhimurium* clinical isolates and *S. typhimurium* reference strain ranged between 25 to 50mg/ml respectively. However the aqueous leaf extract exhibited high antibacterial activity on *S.typhimurium* reference strain showing low MIC of 12.5mg/ml and MBC of 25.0mg/ml (Table 4). Similarly, the MICs of ethanol extracts of the different plant parts against *E.coli* clinical isolates and *E.coli* reference strain were in the range of 12.5 to 25mg/ml with the ethanol leaf extract demonstrating the highest activity with lower MIC of 12.5 mg/ml. This was followed by the root and stem extracts, both showing MIC values of 25mg/ml against *E. coli* clinical isolates and *E. coli* reference strain respectively. Furthermore the MBCs of the various ethanol extracts against the test *E. coli* clinical isolates and *E .coli* referece strain ranged between 25 to 50mg/ml with the stem and root extracts demonstrating the least activities with higher MBC values of 50mg/ml. Similarly the MIC of various extracts tested against clinical isolates of *S.typhimurium* and *S. typhimurium* referece strain were all found to be 12.5mg/ml with the corresponding MBC values 25.0mg/ml.

Table 1: percentage yields of aqueous and ethanol crude extracts of leaf, stem and roots of *T. avicennioides* per 800g of plant part.

Plant Part	Aqueous extract yield (g)(%)	Ethanol Extract yield (g)(%)
Leaf	161.4(20.2)	150.2(18.8)
Stem	155.3(19.4)	140.7(17.6)
Root	149.3(18.7)	137.1(17.1)

Table 2: Phytochemical constituents of aqueous and ethanol crude extracts of *T. avicennioides*.

Constituents	**Aqueous extract**			**Ethanol extracts**		
	Leaf	**Stem**	**Root**	**Leaf**	**Stem**	**Root**
Carbohydrates	+	+	+	+	+	+
glycocides	+	+	+	+	+	+
Saponins	+	+	+	+	+	+
Flavonoides	+	+	+	+	+	+
Tannins	+	+	+	+	+	+
Alkaloids	+	+	+	+	+	+
Anthroquinone	-	-	-	-	-	-
Phenol	+	+	+	+	+	+
Steroides	-	-	-	-	-	-
Triterpenes	+	+	+	+	+	+

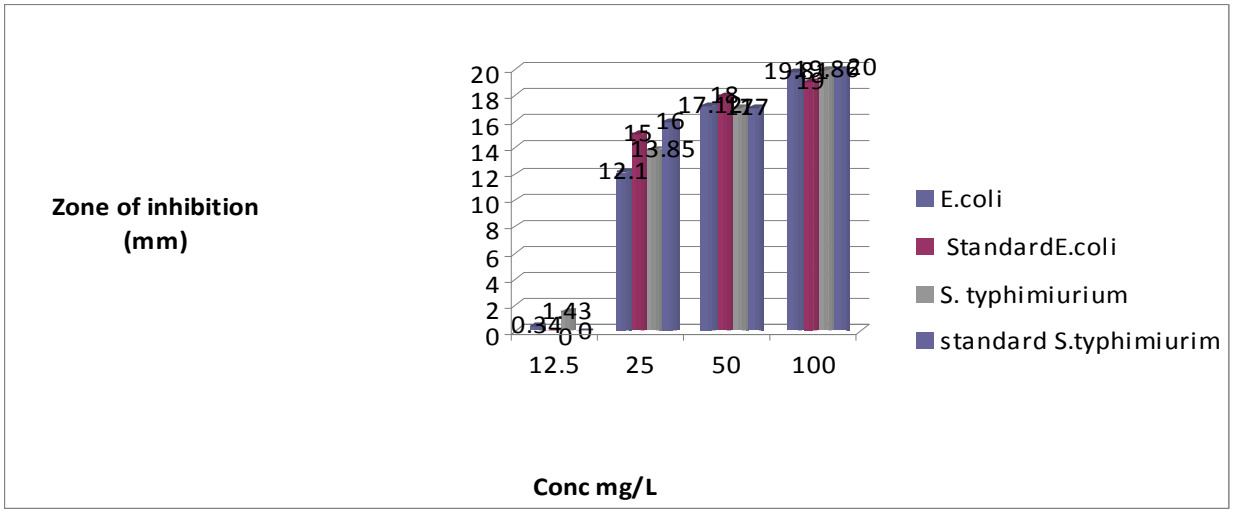

Figure 1: Mean zone inhibition of aqueous leaf extracts of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolates

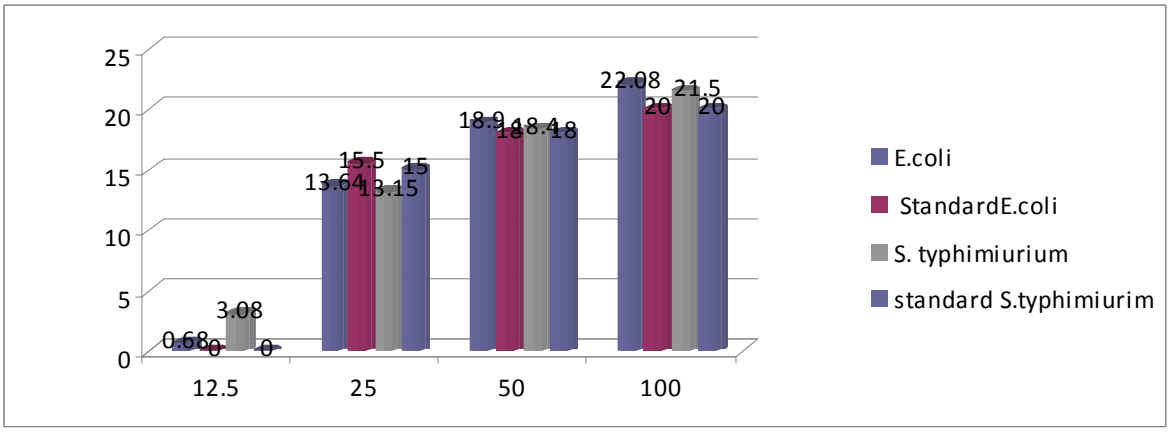

Figure 2: Mean zone inhibition of ethanol leaf extract of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolates

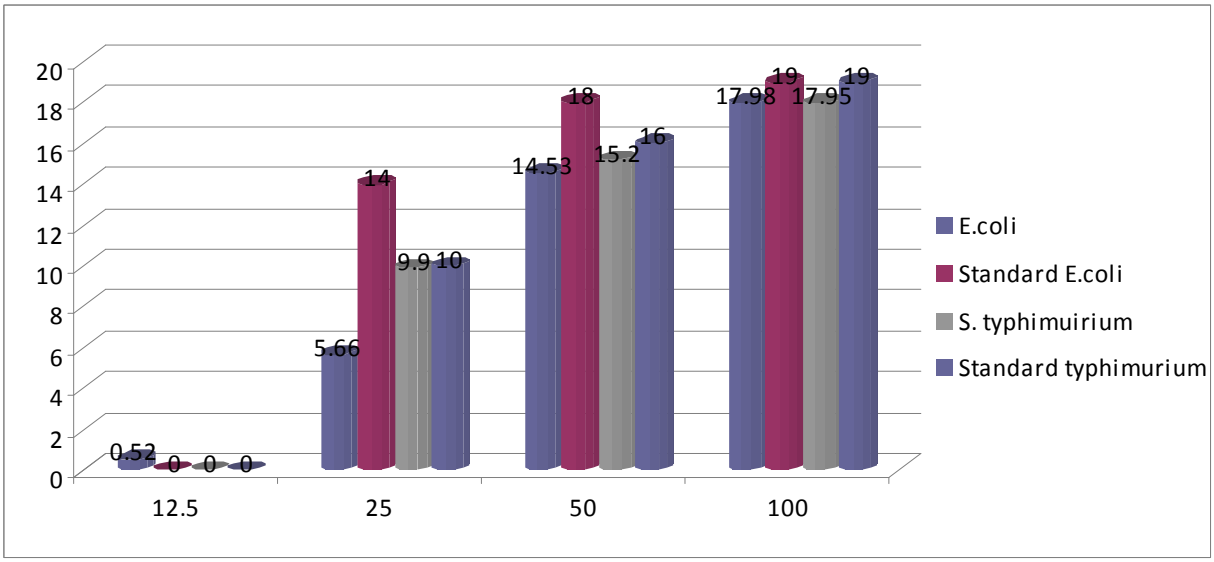

Figure 3: Mean zone inhibition of aqueous stem extract of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolates

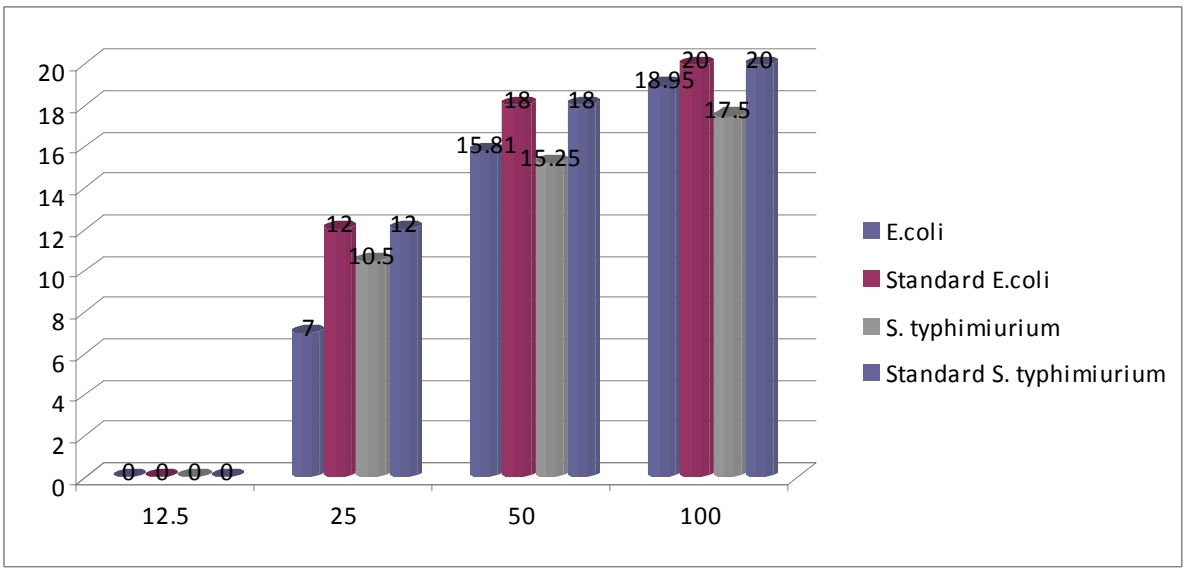

Figure 4: Mean zone inhibition of ethanol stem extract of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolate

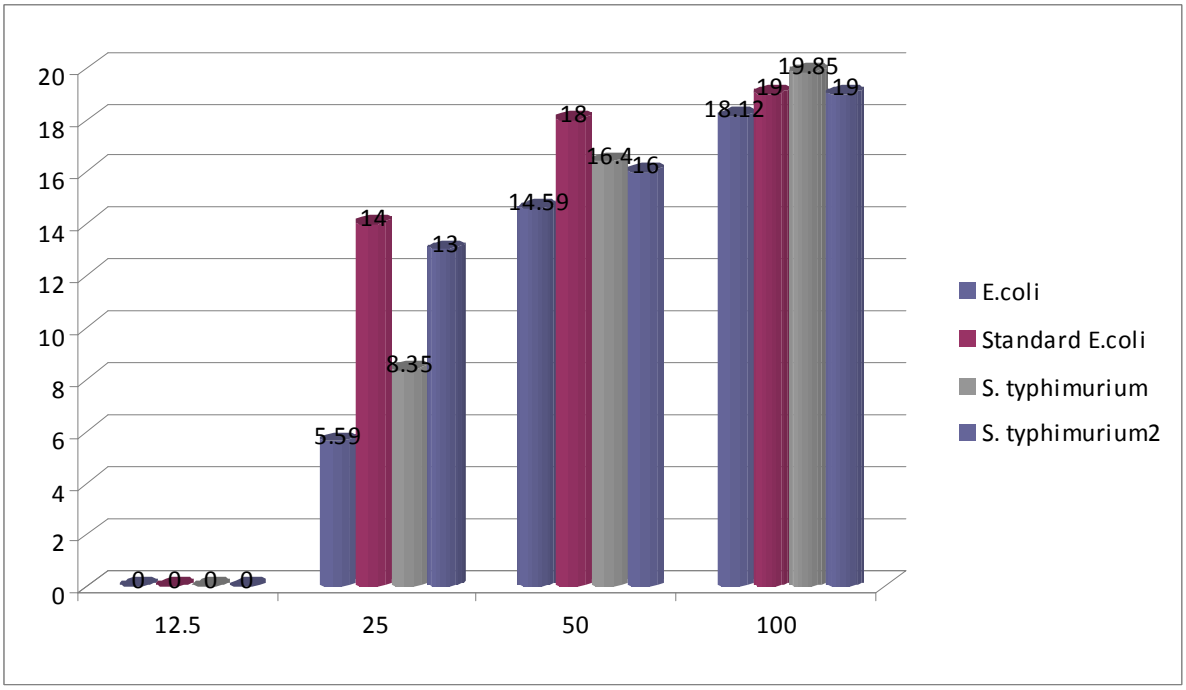

Figure 5: Mean zone inhibition of aqueous root extract of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolates

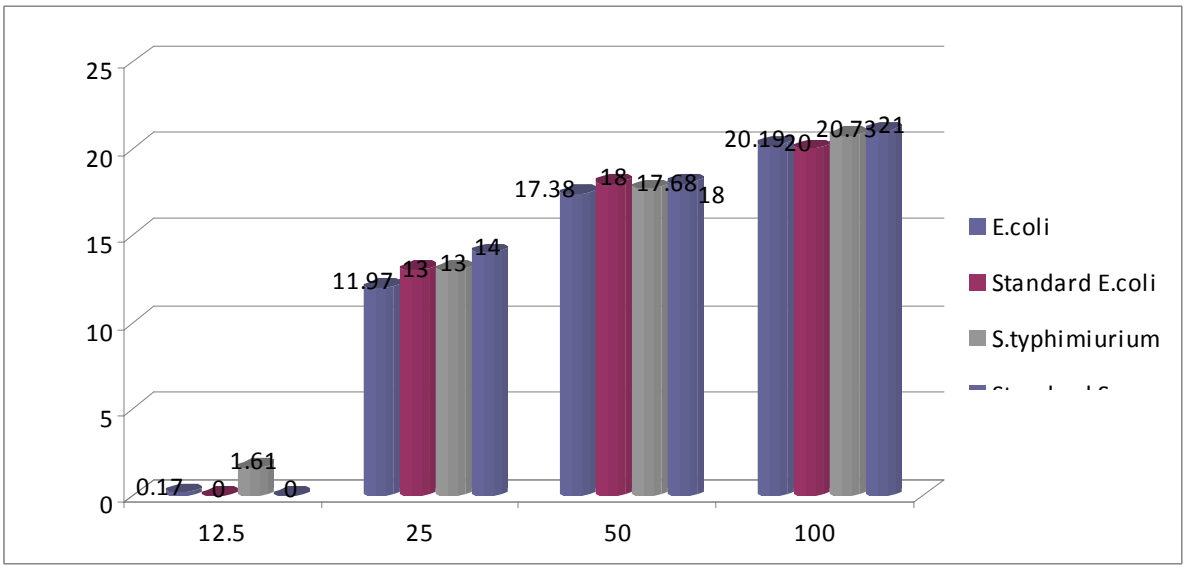

Figure 6: Mean zone inhibition of ethanol root extract of *T. avicennioides* against *E. coli* and *S. tyhpimurium* isolates

Table 3: Minimum Inhibitory Concentration (MIC) and Minimum Bacterial Concentration (MBC) of aqueous crude extracts of *T. avicennioides* on *E.coli* and *S. typhimurium* isolates.

BACTERIA	MIC(mg/ml)			MBC(mg/ml)		
	L	S	R	L	S	R
E. coli(CI)	25	25	25	50	50	50
E. coli(RI)	25	25	25	50	50	50
S. typhimurium(CI)	25	25	25	50	50	50
S. typhimurium(RI)	12.5	25	25	25	50	50

Key: L=Leaf, S=Stem, R=Root, CI=Clinical isolate, RI=Reference isolate.

Table 4: Minimum Inhibitory Concentrations (MIC) and Minimum Bactericidal Concentrations (MBC) of ethanol crude extracts of *T.avicennioides* on *E.coli* and *S.typhimurium* isolates.

BACTERIA	MIC(mg/ml)			MBC(mg/ml)		
	L	S	R	L	S	R
E. coli(CI)	12.5	25	25	25	50	50
E. coli(RI)	12.5	25	25	25	50	50
*S. typhimurium(*CI)	12.5	12.5	12.5	25	25	25
*S. typhimurium(*RI)	12.5	12.5	12.5	25	25	25

Key: L=Leaf, S=Stem, R=Root, CI=Clinical isolate, RI=Reference isolate.

DISCUSSION

The differences in the crude extract yields obtained from the aqueous and ethanol extracts of leaf stem and root barks of *T. avicennioides* might be ascribed to the polarity of solvents used. Water has high partial charges with increasing elution strength that would solubilize more compounds to form complex mixtures. The availability of mixtures of these compounds extracted from the various parts of the plant material may vary in their proportions (Hsu *et al.,* 2006). However, these mixtures can be separated using appropriate selective solvents (Menstrum) (Tiwari *et al.,* 2011).

Our investigation in the preliminary phytochemical screening revealed the presence of many phytocompounds in both aqueous and ethanol crude extracts of *T. avicennoiodes* plant parts studied. The compounds include alkaloids, flavonoides, tannins , saponins , phenols , triterpenes, cardiac glycosides and carbohydrates. However, steroids and anthroquinones were not detected in all the crude extracts of the different plant parts. Related findings on phytochemical constituents of *T. avicennioides* plant parts obtained from Bida , Niger State was reported by Mann *et al.* (2008). Their result was found to slightly contradict our findings. Alkaloids were not detected in the root bark and saponins were reported to be absent in the leaves. The variation in the composition of secondary metabolites extracted from plant tissues can be attributed to several factors including geographical source ,soil condition , harvest processing time and post harvest processing time, moisture content, drying method and storage condition. Additionally, the high temperature generated during tissue grinding can denature chemical constituents and this may invariably affect the composition of compounds and the level of biological activity of a plant material (Wendakoon *et al.,* 2012).

Phytocompounds are known to be biologically active and can aid the antibacterial activities of *T, avicenniodes* through different mechanisms. Alkaloids for example, being one of the compounds present in the plant are one of the largest groups of phytochemicals in plants, with amazing effects in humans leading to the development of a powerful pain killer medication (Akinpelu, 2009). One of the most common biological properties of alkaloids is their toxicity against cells of foreign organisms which have been widely studied for their potential use in the elimination and reduction of human cancer cells. (Dharmananda, 2003). These observations corroborate the use of *T. avicennioides* in folklore remedies. Tannins is another phytochemical compound observed in the extracts of *T. avicenniods*. It has antiviral effect and is used to pull out poisons from poison oak or from bee stings causing instant relief (Dharmananda, 2003). The presence of Tannins in *T. avicennioides* supports the traditional medicinal use of plant in the treatment of different ailments. Li *et al*, (2003) reviewed the biological activities of tannins and observed that tannins (whether total or pure compounds) have remarkable activity in cancer prevention which implies that *T. avicennioides* can serve to aid prevention of cancer. Flavonoids are also one of the constituents of *T. avicennioides* plant extracts. They are natural plant pigments, responsible for large variety of colours in flowers (Temidayo, 2013). . Saponins which are natural surfactants, also known as soap plants, detected in *T.ancennioides* are also found in many plants and act as active immune system. They are also used as cough remedies and for diuretics. They act as natural antibiotics and also bind cholesterol which interfere with cell growth and division but are found to be highly toxic to fish (Temidayo, 2013). This also supports the numerous pharmacological properties of *T. avicennindes*. Therefore, the cited observations on the phytochemical compounds support our findings on the usefulness of *Tavcennioides* in traditional medicine (Han *et al.,* 2007). Lastly phenols is another important constituent discovered in the crude extracts of *T. avicennioides* Primarily phenolioc compounds are of great importance as cellular support material because they form the integral part of cell wall structure (Gupta *et al.,* 2010). All these facts could be one of the reasons why *T. avicenniodes* is widely used for the treatment of many ailments among many tribes in Nigeria hence supporting its usefullness in folklore remedies.

In the present study the aqueous and ethanol crude extracts of the different parts of *T. avicennioides* plant used against the test bacterial isolates demonstrated significant antibacterial activities. The activity of each extract was shown to be concentration dependent, as the concentrations increased from 12.5 to 100mg/ml. Additionally the level of inhibition of bacterial growth exhibited by the active extracts could be due to the initial population density of the organisms, there growth rate and the rate of diffusion of the extracts. This is in line with the report made by Prescott *et al*. (2002) where he stated that the activity of antimicrobial agent is concentration dependent and a related report previously made by Mann *et al.* (2008). But the activities of aqueous stem and ethanol stem extracts against all bacteria at concentrations of 12.5 and 25mg/ml did not differ significantly ($p > 0.05$).

The same applied to ethanol extracts at concentrations of 50 and 100mg/ml. However the ethanol crude extracts of the different parts of *T. avicennioides* demonstrated higher activities against the test bacterial isolates compared to the aqueous extracts.The use of ethanol must have accounted for increased extraction of the biologically active constituents thus displaying wider zones of inhibition. This could probably be why ethanol extraction is widely used by many researhers and herbal medicine industries to obtain crude extracts of phytochemicals from plant materials, for therapeutic application. This observation may be attributed to some reasons. Firstly, the polarity of ethanol used in the initial extraction process, (that is adding 30ml of water to 70ml of absolute ethanol to prepare 70% ethanol), makes it easier to penetrate the cellular membrane to extract the intracellular ingredients from the plant material. Therefore the nature and composition of some biologically active components (saponins, tannins, alkaloids, flavonoids and phenols) could be enhanced in the presence of ethanol due to the stronger extraction capacity of ethanol which may be responsible for the antibacterial activity. Additionally, the higher activity of ethanol extract compared to aqueous extract can be attributed to the presence of higher amounts of polyphenols in ethanol extracts compared to the amount present in water extracts. They are more efficient in cell walls and seed degradation which have un polar character and cause polyphenols to be released from cells. More useful explanation can be ascribed to the enzyme polyphenol oxidase which degrade polyphenols in aqueous extracts. Where as in ethanol or methanol they are inactive. Moreover, water is a better medium for the occurrence of micro organisms compared to ethanol (Lapornik *et al*., 2005). Traditionally, leaves and barks of *T. avicennioides* are soaked in water for days. Large quantities of these extracts which lack specific concentrations are usually administered to patients: Our result therefore showed that crude extracts from the investigated plant material should preferably be extracted with ethanol. This supports the findings of Tiwari *et al.* (2011) that plant extracts from organic solvents give more consistent antimicrobial activity compared to aqueous extracts. Nearly all of the identified plant components active against microorganism are obtained through initial ethanol or methanol extraction because they are aromatic or saturated organic coupounds. Though methanol is also widely used and it is more polar, but due to its cytotoxic effect, it is unsuitable for extraction in certain kinds of studies (Das and Tiwari 2010).

Among the ethanol extracts of leaves, stem and root barks, the leaf extract of *Terminalia avcennioides was* found to be the most active demonstrating the lowest MIC and MBC values. The differences in susceptibities between *Escherichia coli* and *Salmonella tyhphinurium* isolates to different concentrations of *T. avicennioides* crude extracts could be attributed to species differences or differences in the chemical compositions of these extracts as revealed by the phytochemical analyses (Escalante *et al.,*2002;Yimta *et al.,* 2014).The values of MIC of extracts against the test isolates were lower than the MBC values showing that the extract of *T.avicennioides* plant is bactericidal in action and this corresponds with the findings of Yimta *et al.*(2014) and Mann *et al.* (2008).

CONCLUSION AND RECOMMENDATIONS

Phytochemical analysis of aqueous and ethanol crude extracts of leaf stem and root barks revealed the presence of compounds like alkaloids, flavonoids, tannins, saponins , cardiac glycosides, phenols, triterpenes, steroids and carbohydrates. These important bioactive compounds may be responsible for the observed antibacterial activities of crude extracts of the investigated plant material. Among the ethanol extracts of leaf, stem and root barks, higher antibacterial activity was observed with the leaf extract of *Terminalia avcennioides* showing lower MIC of 12.5 mg/ml, followed by the root and stem extracts, both showing MIC values of 25mg/ml against *E. coli* clinical isolates and *E. coli* reference strain respectively. Furthermore the MBC of the various ethanol extracts against the test *E. coli* clinical and reference isolates ranged between 25 to 50mg/ml with the stem and root extracts demonstrating the least activities with higher MBC values of 50mg/ml. Similarly, the MICs of various ethanol extracts tested against clinical and reference isolates of *S. typhimurium* were all found to be 12.5mg/ml with the corresponding MBC values of 25.0 mg/ml. The values of MIC of extracts against the test isolates were lower than the MBC values showing that the extract of *T. avicennioides* plant is bactericidal in action.

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