IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS)
e-ISSN:2278-3008, p-ISSN:2319-7676. Volume 13, Issue 5 Ver. III (Sep – Oct 2018), PP 72-81
www.iosrjournals.org

Antimicrobial, Antioxidant Properties And Phytochemical Analysis Of *Opilia Amentaceae*

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Abstract: *Plants produce thousands of specialized metabolites, many of which have medicinal uses. Over the next few years, the study of medicinal plants as antimicrobial agents should be focused on ascertaining specific information about the plant's antimicrobial activity. In the present investigation the phytochemical, antimicrobial and DPPH radial scavenging activities were screened by using two plants namely Opilia amentaceaea and Anogeissus latifolia. Different solvent extracts were used in the phytochemical analysis to elucidate the tannins, Saponins, Flavonoids, Catachins and Sugars from these plants. Significantly Anthroquinones are present only in Anogeissus latifolia. Both of these plants inhibit the growth of Escherichia coli ATCC 25922, Salmonella entericatyphimurium, Shigella dysentriae, Klebseilla pneumoniae. In DPPH assay maximum inhibition (70.800) was observed at 200 µg/ml concentration of Anogeissus latifolia and maximum percentage of inhibition (66.426) was observed at 500 µg/ml concentration of Opilia amentaceae.*
Key words: *Anogeissus latifolia ,Opilia amentaceae, phytochemical, antimicrobial and DPPH.*

Date of Submission: 06-10-2018 Date of acceptance: 21-10-2018

I. Introduction

A vast knowledge of medicinal plants usage against various diseases and illness may be expected and accumulated in the areas of traditional knowledge and ethno medicinal practice. Medicinal plants have been used for the treatment of a large number of human diseases in different parts of the world throughout the history of human kind. About 80% of rural population and communities of many developing countries were still dependent on traditional medicines both in health care and practice. (Palombo, 2006).Traditional healers pass the plant knowledge to their families or relatives orally or practically. Traditional knowledge is secured for future by showing the geographic habitation with vernacular/local names and with various methods drug preparations for a particular disease (Kokwaro, 2009). Around 20% of people in rural areas consult herbalists before seeking treatment in government clinics and hospitals. Traditional medicine is a major source of treatment for African patients (Dejong, 1991). The most important of these bioactive compounds of plants are alkaloids, flavanoids, tannins and phenolic compounds. Rural communities, in particular the tribal people depend on plant resources for herbal medicines, food, forage, construction of dwellings, making household implements, sleeping mats, and for fire and shade. The use of medicinal plants as traditional medicines is well known in rural areas of many developing countries.

*Opilia amentaceae*is a climbing shrub, also had tree like behavior. Leaves are ovate, tip pointed, leathery with leaf stalk to 8 mm. Flowers are born in racemes at leaf axils, 1-5 together, up to 3 cm long. Fragratopilia is found in Peninsular India through Srilanka to Tropical Australia and Africa.

Anogeissus latifolia is a small to medium sized tree, grows up to 20 m and closely related to the Button tree. The species name is latifolia with reference to its wider leaves. Leaves are oppositely arranged, simple, entire, with greyish-yellow or whitish hairs at nodes and internodes. Flowers are small and have part in fives with sepals are joined together to form a stalk like tube. There are no petals and the Fruit is a 2-winged Pseudoachene, packed into a dense head. In the Present study the antibacterial activity of *Opilia amentaceae* Roxb and *Anogeissius latifolia* were screened against human pathogens.

II. Materials And Methods

Collection of plant samples

The leaves of *Opilia amentaceae* Roxb and *Anogeissius latifolia* were collected in the month of January 2018 from Siriyakalvarayan hills of Eastern Ghats present in Villupuram district of Tamilnadu. Samples were shade dried and pulverised under the room temperature for 48 hrs.

Preparation of the extract

The dried leaves of *Opilia amentaceae* Roxb and *Anogeissius latifolia* were powdered and defatted with methanol (60-80ºc) in a Soxhlet Apparatus by continuous hot percolation. The solvent was removed by keeping the extracts in the incubator for 24 hrs. The resultant dried extracts were used for further study.

Microorganisms

Three bacterial species like *Escherichia coli ATCC 25922 (E.coli), Salmonella entericatyphimurium* (*S.enterica), Shigella dysentriae and Klebseilla pneumonia* are used in the present study. These bacterial cultures were obtained from K.A.P. Viwsanathan Government Medical College, Trichy, India. And the bacterial species were subcultured on nutrient broth and incubated at 37°C for 18-24 h. The bacterial strains were cultured on nutrient agar slants and the cultures were maintained by sub culturing, periodically and preserved at 4° C for further use.

Phytochemical analysis

The extracts were then tested for their respective phytochemical properties and compositions, as per the analytical method of Brinda *et al.*1981.(Table 1).

Table 1: Preliminary qualitative analysis (*Brinda et al., 1981*) method.

S.no	Test	Observation	Inference
1.	2ml of test solution and a minimum amount of chloroform, 3 to 4 drops of acetic anhydride, followed by one drop of conc. H_2SO_4 was added.	Purple colour changes to blue or green.	Steroid present.
2.	2ml of test solution was reacted with a piece of tin and two drops of thionyl chloride.	Violet or purple colour developed.	Triterpenoids present.
3.	2ml of test solution was mixed with a very small quantity of Anthrone reagent and a few drops of conc. H_2So_4 and then heated.	Green or purple colour developed.	Sugar present.
4.	2ml of test solution was mixed with 2ml of Fehlings reagent and 3 ml of water.	Red-orange colour formed.	Presence of reducing sugar.
5.	2 ml of the test solution was taken with 2N HCl, and the aqueous layer formed was decanted, to which, one to few drops of Mayers reagent was added.	Formation of white precipitate or turbidity.	Alkaloids present.
6.	2 ml of test solution alcohol was taken along with a drop of neutral Ferric chloride (5%) solution.	Intense blue colour developed	Phenolic compound present.
7.	2 ml of the solution in alcohol was mixed with a bit of magnesium and 1 or 2 drops of concentrated HCl and then heated.	Red or orange-red colour formed.	Presence of flavonoid.
8.	2 ml of test solution in alcohol was taken along with a few drops of Ehrlichs reagent and a few drops of conc. HCl.	Pink colour formed.	Presence of catachins
9.	2 ml of test solution was mixed with water and shaken well.	Foamy lather formed.	Saponins present.
10.	2 ml of test solution was mixed with water and then with lead acetate solution.	White precipitate was developed	Tannins present.
11.	2 ml of test solution was mixed with magnesium acetate solution.	Pink colour developed.	Anthroquinones present.
12.	2 ml of test solution was mixed with 1% Ninhydrin in alcohol.	Blue or violet colour obtained.	Presence of amino acids.

Determination of antibacterial activity using standard antibiotics by disc diffusion method

The disc diffusion method was used with few modifications to evaluate anti-microbial activities. Sterile Disc (Whatman, 6 mm) were impregnated with 50 µl of reconstituted crude extracts (1 mg mL-1) in the solvent used for extraction (ethanol). And then placed on the surface of Muller-Hinton agar dispersion plates inoculated with bacterial cultures. Each extract was tested in triplicate. Control disc containing 50 µl DMSO (100 %) was used as negative control. Standard antibiotics such as Vancomycin, Gentamicin, Bacitracin and Amoxicillin-clavulanate (HIMEDIA) were used as reference or positive control. Agar plates containing bacteria were incubated at 37°C for 24 h. Inhibition zones were recorded as the diameter of growth-free zones that includes the diameter of the disc in mm at the end of the incubation period.

Determination of antibacterial activity by agar well diffusion method

Antimicrobial activity was determined by the well diffusion method according to National Committee for Clinical Laboratory Standards, 1993 (NCCLS). Petri plates containing 20 ml of Nutrient Agar medium were seeded with 1-3 day bacterial cultures of microbial inoculums (standardized inoculums 1-2 X 107 cfu/ml 0.5 Mcfarland standards). Wells (6 mm in diameter) were cut off into agar and 50 µl of plant extracts were tested in

a concentration of 100 mg/ml and then incubated at 37ºC (bacterial strains) and at 25ºC (fungal strains) for 24-48 h. The assessment of antimicrobial activity was based on measurement of the diameter of the inhibition zone formed around the well.

DPPH Radical Scavenging Assay:

The antioxidant activity of the plant extracts were measured on the base of the scavenge activity of the stable 2, 2- diphenyl 1-picryl hyorazyl (DPPH), free radical with small modifications. 1ml of 0.1mM DPPH solution in methanol was mixed with 1ml of plant extract solution of different concentrations (50, 100, 150, 200,250,300, 350, 400, 450 and 500 µg/ml). Equivalent blank sample was prepared and L-Ascorbic acid (1-100 µg/ml) was used as reference standard. Mixer of 1ml methanol and 1ml DPPH solution was used as control. The reaction was done in triplicate and the decrease in absorbance was measured at 520 nm after 30 minutes in dark using Colorimeter. The inhibition % was calculated using the following formula. Inhibition % = (Ac-As/Ac) ×100 Where Ac is the absorbance of the control as is the absorbance of the sample.

III. Results And Discussion

The present study was conducted to investigate antibacterial properties of *Opiliaa mentaceaea* and *Anogeissus latifolia.* Powdered samples of plant leaves were extracted in different solvents.

Phytochemical analysis

The plant material was subjected to soxhlation and the dried paste was weighed and dissolved appropriately in the original solvent used for extraction. This mode of reconstitution of the plant material allows for convenient dissolution of the material, without precipitation. The yields of extraction from originally used 15-20 g of material were around 2 g. Before proceeding with ethanol as the solvent of choice for soxhlation, different solvents are used quickly by taking 2g of the plant powder. Then grinding smoothly with a mortar and pestle and finally filtered by using a Whatman no.1 filter paper. The extracts were assessed qualitatively for phytochemical composition. Based on the abundant presence of the phytochemicals (in terms of qualitative analysis), we decided, then most phytochemical classes were found in the ethanolic extract. Then we used different solvents such as water, methanol, acetone, chloroform, ethylacetate and hexane for extraction. (In order to decrease polarities).

In the phytochemical analysis different types of solvent extracts were used. Tannins, Saponins, Flavonoids, Catachins and Sugar were present in the *Opilia amentaceae and Anogeissus latifolia, that y*ielded positive results for Anthroquinones, Tannins, Saponins, Flavonoids, Catachins and Sugars. Anthroquinones were completely absent in tested solvents and the results were categorically tabulated.

Among the different extracts used, we identified the acetone extract tested negative for all the phytochemical classes. Water extract of *O. amentaceae* tested positive for tannins, saponins, flavonoids and catachins, however, it did not contain sugars or anthraquinones. Among the several solvents used, only the EtOH extract contained, almost all the classes of phytochemicals (save, tannins). EtOH often extracts tannins, polyphenols, polyacetylenes, flavonols, terpenoids, sterols and alkaloids. In this case, we did not obtain tannins presumably because the tannins may be utterly absent in the plant. The results obtained during phytochemical analyses were photographed (Eg. Colour change/ Effervescence) immediately. (Figure 1, 2 and 3; Table 2&3).

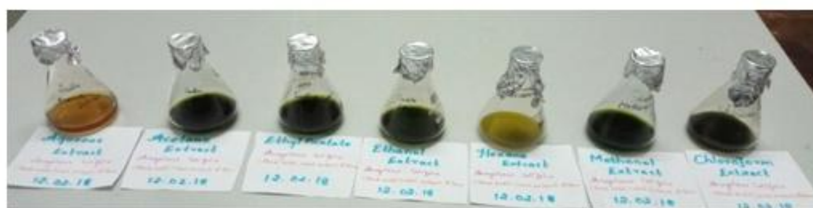


Figure 1: Photograph of the various solvent extracts of A. latifolia obtained by grinding 5g powder with mortar and pestle.

Table 2. Preliminary phytochemical analysis of *Opilia amentaceae* (Roxb.)

S.NO	*TEST*	*Distilled water*	*Ethanol*	*Methanol*	*Chloroform*	*Hexane*
1.	**Anthroquinone**	-	-	-	-	-
2.	**Tannins**	-	+	-	-	-
3.	**Saponins**	+	+	+	-	+
4.	**Flavonoids**	-	+	+	+	-
5.	**Catachins**	-	+	-	-	-
6.	**Sugar**	-	+	-	-	+

***Figure 2: Phytochemical analysis of Opilia amentaceae* (Roxb.).** *5g of plant powder was weighed in butter paper and extracts of different solvents, ranging from water to n-hexane (in order of polarity), were prepared. Qualitative phytochemical analysis was carried out as per the method of Brindha et al. From left to right: Catechins,sugars, tannins, anthraquinones, flavonoids and saponin tests.*

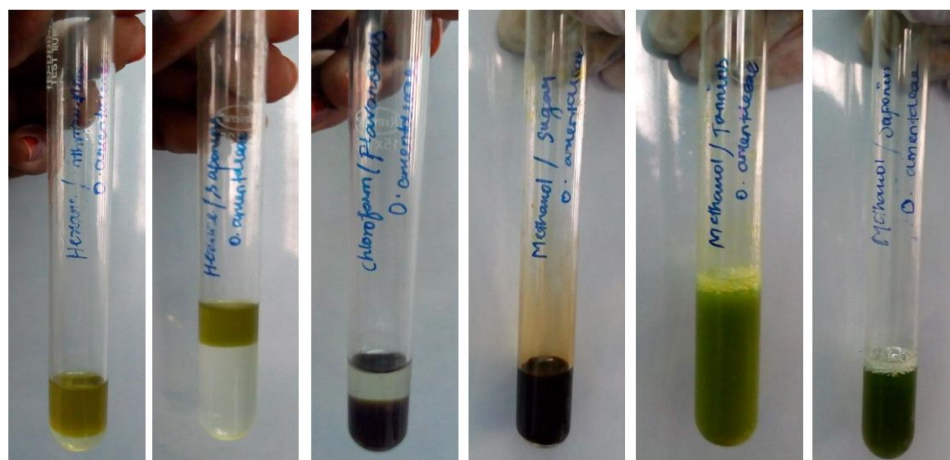
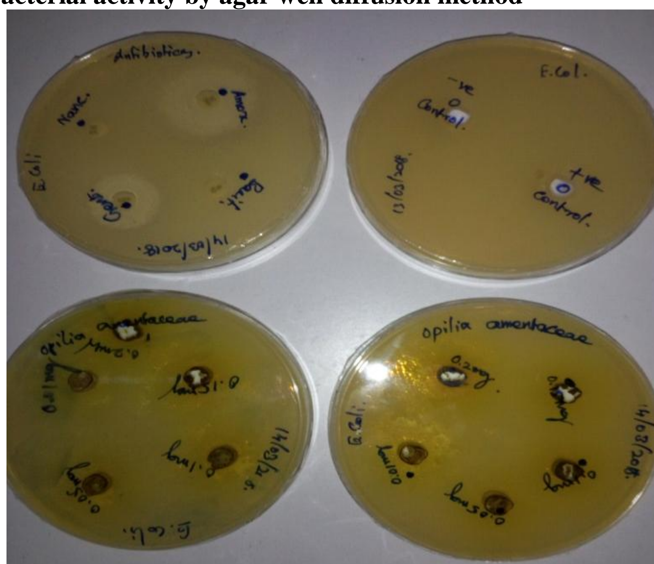


Table 3: Phytochemical analysis of *Anogeissius latifolia* (Brindha*et al*. method).

S.No.	***Test***	***Aqueous***	***Ethanol***	***Methanol***	***Chloroform***	***Acetone***
1	Anthroquinone	-	-	-	+	-
2	Tannins	+	+	+	-	-
3	Saponins	+	-	+	-	-
4	Flavonoids	+	-	+	-	-
5	Catachins	+	+	+	+	-
6	Sugar	+	-	+	+	-

***Figure 3: Phytochemical analysis ofA. latifolia* (Roxb.)** *5g plant powder was weighed in butter paper and extracts of different solvents, ranging from water to n-hexane (in order of polarity), were prepared. Qualitative phytochemical analysis was carried out as per the method of Brindha et al. From left to right: Catechins, sugars, tannins, anthraquinones, flavonoids and saponin tests.*

Determination of antibacterial activity by agar well diffusion method



Antibacterial activity of ethanolic extract of *O.amentaceae* was tested against *E.coli, S. typhi* and *S. dysentriae.* Maximum zone of inhibition was observed in 0.2mg concentration against *E. coli* and *S. dysentriae.* And *S. yphi* inhibited by 0.15mg concentration. Gentamicin, Bacitracin, Amoxyclav and Vancomycin discs were used as controls (Fig 4,5 and 6 and Table 4).

Figure 4: Well diffusion assay to measure the activity of the ethanolic extract of O. amentaceae against Escherichia coli ATCC25922. *The zones of inhibition were measured by taking five different concentrations of the ethanolic extract – 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate positive and negative controls (ethanolic disc) as well as antibiotic discs were maintained*

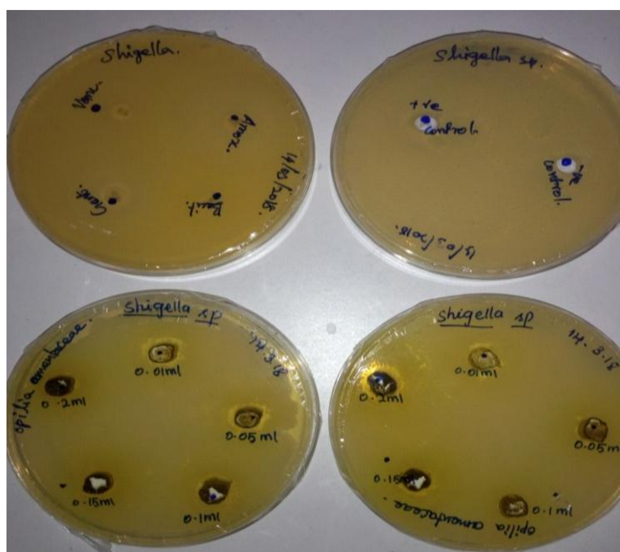


Figure 5 Well diffusion assay to measure the activity of the ethanolic extract of O. amentaceae against Shigella dysentriae. *The zones of inhibition were measured by taking five different concentrations of the ethanolic extract – 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate positive and negative controls (ethanolic disc) as well as antibiotic discs were maintained*

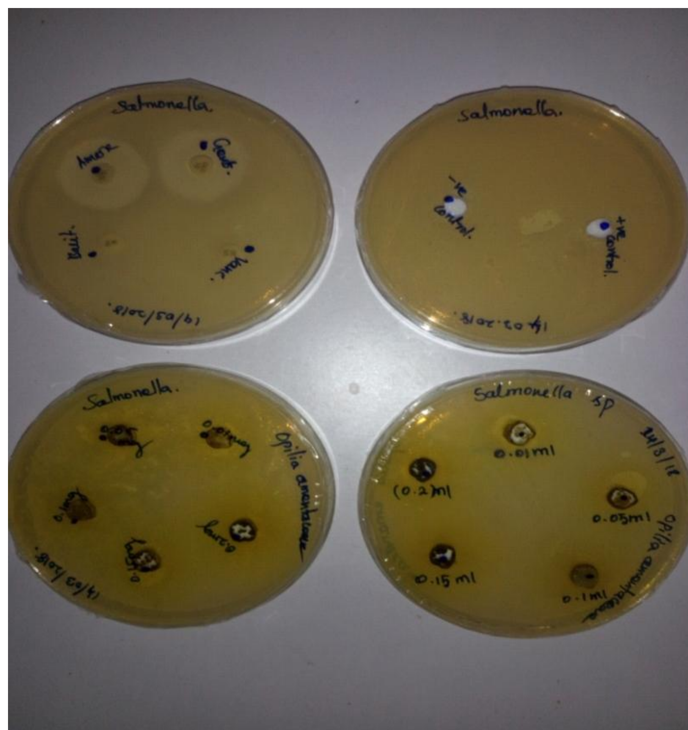


Figure 6: Well diffusion assay to measure the activity of the ethanolic extract of O. amentaceae against Salmonella typhi. *The zones of inhibition were measured by taking five different concentrations of the ethanolic extract; 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate positive and negative controls (ethanolic disc) as well as antibiotic discs were maintained.*

Table 4: Zones of inhibition for Ethanolic extract of *O. amentaceae.*

Organism	*0.01 mg*	*0.05 mg*	*0.1 mg*	*0.15 mg*	*0.2 mg*	*Gentamicin (10 µg)*	*Bacitracin (10 µg)*	*Amoxyclav (30 µg)*	*Vancomycin (10 µg)*
E. coli	0.0	0.0	0.0	0.3	0.5	2.0 cm	1.5 cm	2.7 cm	1.8 cm
S. typhi	0.1	0.3	0.4	0.7	0.6	2.9 cm	None	2.5 cm	None
S. dysentriae	0.0	0.0	0.0	0.4	0.5	2.5 cm	None	2.5 cm	None

The antibacterial affects at different concentrations of the Methanolic extract of *O. amentaceae*

The data of antibacterial activity acquired in duplicates has been plotted in the graph, which has the most potent concentrations of the methanolic extract against the test organisms. For *E. coli*, the most potent concentration was 0.1 mg and *S. dysentriae*, has 0.15 mg. For *S. aureus*, it was 0.2 mg and for *S. typhi*, the most potent concentration was 0.15 mg (Fig

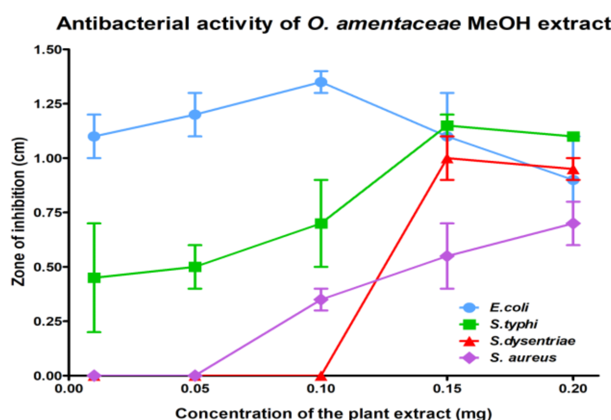


Figure 7: Summary of the antibacterial effects of different concentrations of the extract of O. amentaceae against clinically relevant human pathogens

Antibacterial activity of the methanolic extract of *A. latifolia*

The zones of inhibition were measured by taking five different concentrations of the methanolic extract, 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate negative controls (methanolic disc) as well as positive controls (antibiotic discs) were maintained, to cross-check the activity of the plant extracts. Maximum zone of inhibition was observed in 0.15 mg/ml against *E.coli,* 0.15mg/ml against *S.typhi,* 0.20 mg/ml against *S.dysentriae and* 0.20mg/ml against *K.pneumoniae* (Fig 8, 9, 10, 11 and Table 5).

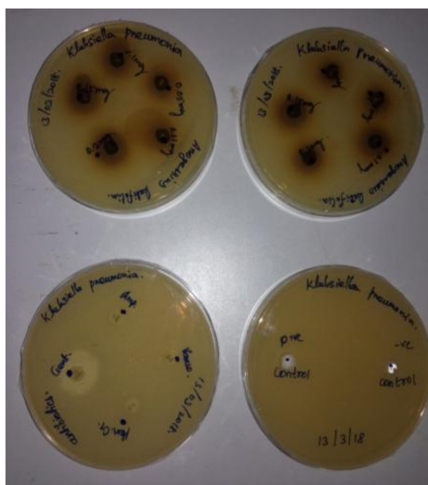


Figure 8: Well diffusion assay to measure the activity of the methanolic extract of A. latifolia against Klebsiella pneumoniae. The zones of inhibition were measured by taking five different concentrations of the methanolic extract, 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate negative controls (methanolic disc) as well as positive controls (antibiotic discs) were maintained .

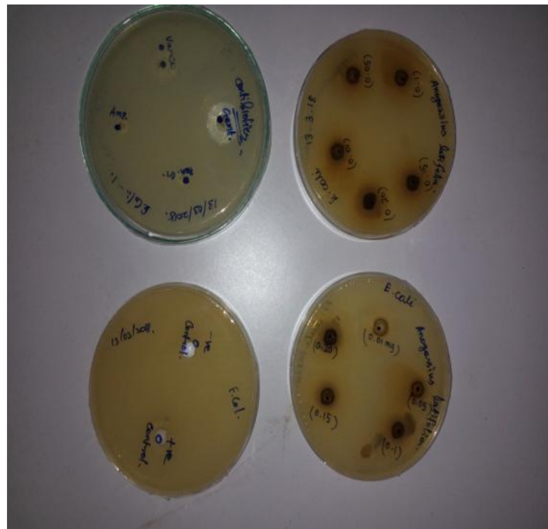


Figure 9: Well diffusion assay to measure the activity of the methanolic extract of A. latifolia against E.coli ATCC25922. *The zones of inhibition were measured by taking five different concentrations of the methanolic extract, 0.01, 0.05, 0.1, 0.15 and 0.2 mg/ml into wells punched with a sterile, autoclaved gel puncher. Appropriate negative controls (methanolic disc) as well as positive controls (antibiotic discs) were maintained in order to cross-check the activity of the plant extract.*

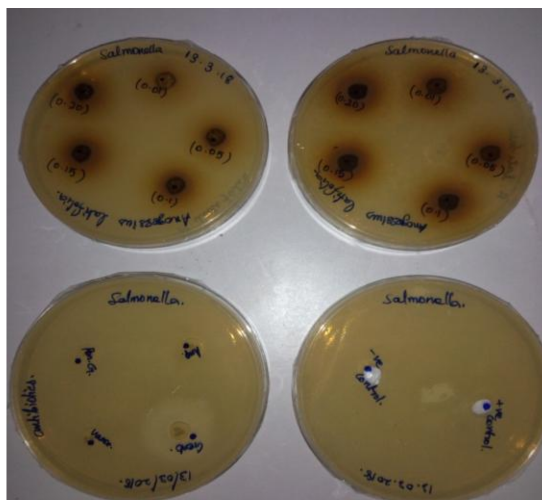


Figure 10: Well diffusion assay to measure the activity of the methanolic extract of A. latifolia against Salmonella typhi.

Table 5: Zones of inhibition obtained for methanolic extract of A. latifolia.

Concentration of extract (mg/ml)	*E. coli*		*S. typhi*		*S. dysentriae*		*K. pneumoniae*	
0.01	1.6	1.2	0.8	1.1	1.1	1.2	1.0	1.3
0.05	0.5	1.0	1.1	1.2	1.0	1.1	1.1	1.1
0.10	1.0	1.1	1.1	1.6	1.1	1.4	1.4	1.1
0.15	1.7	2.1	1.3	1.4	1.2	1.3	1.5	1.4
0.20	1.2	1.2	1.2	1.3	1.4	1.3	1.6	1.2

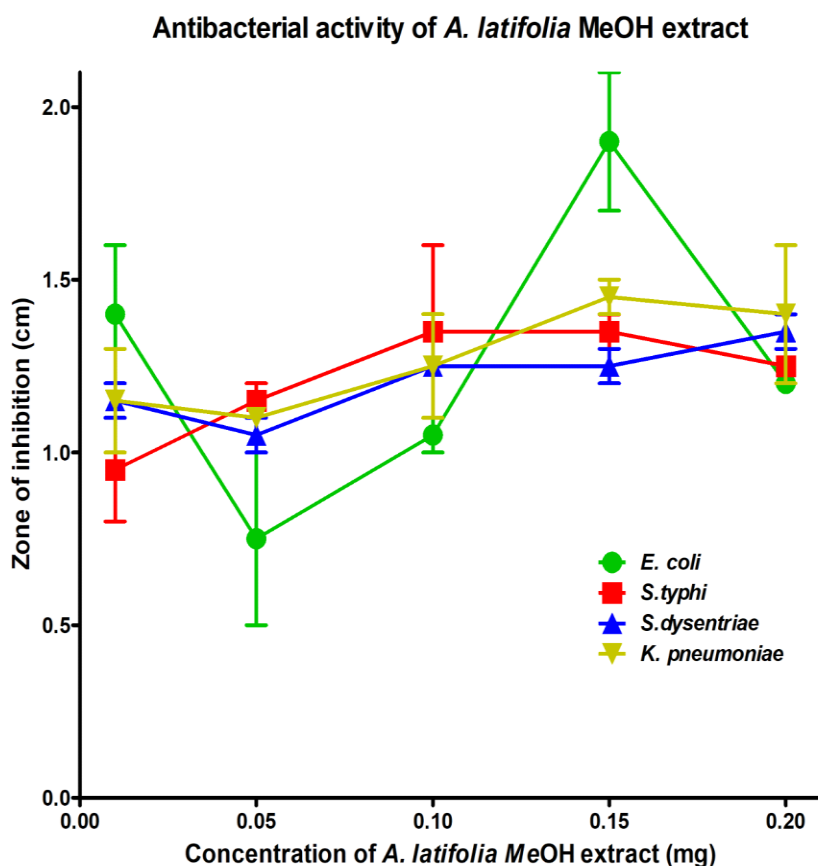


Figure 11 (shown above), we can understand that the methanolic extract of *A. latifolia*

DPPH radical scavenging activity of *Anogeissus latifolia* and *Opilia amentaceae* in methanolic extract

DPPH radical scavenging activity of *Anogeissus latifolia* and *Opilia amentaceae* in methanolic extract was analysed. In *Anogeissus latifolia* maximum percentage of inhibition (70.800) was observed at 200 µg/ml concentration and minimum percentage (36.000) of inhibition was recorded in 350 µg/ml concentration.

In *Opilia amentaceae* maximum percentage of inhibition (66.426) was observed at 500 µg/ml concentration and minimum percentage (19.521) of inhibition was recorded in 50 µg/ml concentrations (Table 6).

Table 6: DPPH radical scavenging activity of *Anogeissus latifolia* and *Opilia amentaceae* in methanolic extract.

Number of concentration (µg/ml)	Ascorbic acid	*Anogeissus latifolia*	*Opilia amentaceae*
50	90.00	54.800	19.521
100	90.93	67.200	34.262
150	91.56	48.000	60.159
200	92.18	70.800	63.346
250	92.81	44.000	52.191
300	93.75	44.000	63.312
350	93.12	36.000	62.112
400	90.31	52.000	63.721
450	93.75	50.800	64.412
500	96.25	56.000	66.426

IV. Discussion

In this study Phytochemical analysis of active extract was demonstrated by the presence of common phytoconstituents like tannins, glycosides, saponins, flavonoids and alkaloids. These compounds are believed to be responsible for the observed antibacterial properties. Some studies have also attributed to their observed antimicrobial affect of plant extracts in the presence of these secondary plant metabolites (Nweze et al., 2004). The presence of tannins suggests the ability of this plant to play a major role as anti diarrhoeic and anti haemorrhagic agents (Price et al., 1987). Presence of saponins has revealed the enormous and significant anti hypercholesterol, hypotensive and cardiac depressant properties (Asquith and Butler 1986).

The presence of cardiac glycosides in medicinal plants were under usage for more than two centuries and used as stimulants for cardiac failures (Sood et al., 2005). This could justify and well established with the local practitioners and traditional healers to cure and treatment of hypertension with viable plant functionalities. The presence of these photochemical bases in *Opilia amentaceae* (Roxb.) and *Anogeissius latifolia* accounts for its usefulness as medicinal plant. The most important of these bioactive compounds of plants are alkaloids, flavonoids, tannins and phenolic compounds (Edeoga *et al.,* 2005). Secondary metabolites, which are loaded with diverse kinds of chemical compounds, may aid in either bacteriostasis or bactericidal killing of the pathogenic bacteria used as test species in our study. Govindarajan *et al*., 2006 reported the ethno botanical importance of *Anogeissus latifolia*, the bark of *Anogeissus latifolia* (Roxb. ex DC.) Wall.exGuill. & Perr. (Combretaceae) has reported to be used in the treatment of various disorders including stomach and skin diseases.

Patil and Gaikwad., 2010, studied the importance of *Anogeissus latifoli. as* one of the important medicinal plants used in Ayurveda for cardiac disorders. Thhis plant is also useful in Urinary tract infections, skin diseases, liver complaints, fever, epileptic fits etc. The plant is rich in pharmacologically active phenolic phytoconstituent-ellagic acid. It contains the healing potential, microbicidal activities, antiulcer potential, hypolipidemic activities and hepatoprotective potential.

In the present investigation *Opilia amentaceae* (Roxb.) and *Anogeissius latifolia are* having the viable inhibition properties against the growth of several human pathogens, due to the presence of Anthraquinones, Tannins, Saponins, Flavonoids, Catachins and Sugar.

V. Conclusion

Herbal medicines are becoming essential and had vastly growing tremendous potential in international pharmacopeia. Knowledge of the medicinal properties is growing as a result of research and testing, which will make them an increasingly safe alternative or a preferred option to allopathic medicine. Today, there is a renewed interest in traditional medicine and an increasing demand for more drugs from plant resources. This revival of interest in plant-derived drugs is mainly due to the current widespread belief, “green medicine” is safe and more dependable than expensive synthetic drugs that had wide adverse side effects. There is a growing interest in correlating phytochemical constituents of a plant with its pharmacological activity. Scientists have even started correlating the botanical properties of plants with their pharmacological activity. In future, more coordinated multidimensional research is aimed at correlating botanical and phytochemical properties to

elucidate more pharmacological activities. Thus, determining the biological activities of plants used in traditional medicine is helpful to the rural communities and informal settlements.

References

[1]. Asquith TN, Butler LG (1986). Interaction of condensed Tannins with selected proteins. *Phytochemistry*, 25 (7) 1591-1593.

[2]. Dejong, J. (1991). Traditional medicine in Sub- Saharan Africa: Its importance and potential Policy options, Policy, Research and external Affairs, World Bank Publication.

[3]. Edeoga H.O., Okwu D. E. and Mbaebie B.O. (2005). Phytochemical constituents of some Nigerian medicinal plants. *African Journal of Biotechnology*. 4 **(7)**: 685-688.

[4]. Govindarajan R, Vijayakumar M, Singh Ch.V. Rao .V. ShirwaikarA.Rawat, Pushpangadan (2006).Antiulcer and antimicrobial activity of *Anogeissuslatifolia.J. Ethanopharmacology,*106 (1): 57-61.

[5]. Kokwaro JO (2009). Medicinal plants of East Africa, 3rdEd.University of Nairobi press.

[6]. Nweze EI, Okafor JI, Njoku O (2004). Antimicrobial activities of Methanolic Extracts of Tremaguineensis (Schumm and Thorn) and MorindalucidaBenth used in Nigerian Herbal Medicinal Practice. *J. Biol. Res. Biotech*, 2(**1**): 39-46.

[7]. Palombo EA (2006). Phytochemicals from traditional medicinal plants used in the treatment of diarrhoea: Modes of action and effects on intestinal function. *Phytother Res*. 20(**9**):717-724.

[8]. Patil, Udaysing&Gaikwad, Dattatraya. (2010). Phytochemical profile and Antibacterial activity of stem bark of *Anogeissuslatifolia. Pharmacognosy Journal*. 2. 70–73.

[9]. Price KR, Johnson TI, Fenwick GR (1987). The Chemistry and Biological Significance of Saponins in Food and feeding stuffs. *Crit Rev Food SciNutr*, 26:22-48.

[10]. Sood S, Narang D, Dinda AK, Maulik SK (2005). Chronic oral administration of Ocimum sanctum Linn. augments cardiac endogenous antioxidants and prevents isoproterenol-inducedmyocardial necrosis in rats. *J Pharm Pharmacol*, 57(**1**):127-33.

IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS) is UGC approved Journal with Sl. No. 5012, Journal no. 49063.

Punitha M. "Antimicrobial, Antioxidant Properties And Phytochemical Analysis Of Opilia Amentaceae." IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS) 13.5 (2018): 72-81.