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ORIGINAL RESEARCH

Phytochemical, antibacterial and synergistic potency of tissues of *Vitex grandifolia*

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ABSTRACT

This study evaluated the antibacterial and synergistic efficacy of extracts of *Vitex grandifolia* against some pathogenic microorganisms. The *V.grandifolia* were obtained within the Niger Delta University campus. Extraction was carried out using water, ethanol, methanol and hexane. Standard agar well diffusion method was employed for the sensitivity test. The experiment was arranged in a Completely Randomized Design fashion. Ethanol extracts were superior in most cases with regard to the zone of inhibition. The highest synergistic zone of inhibition for *Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus, Salmonella* and *Proteus* species were 17.00mm (root + stem), 14.00mm (root +leaf +stem), 16.00mm (stem +leaf), 16.33mm (stem +leaf) and 17.00mm (root +stem) respectively. Analysis of variance showed that there were significant differences ($P<0.05$) in efficacy of their different plant tissues against the bacteria used in the study. The presence of phytochemicals such as tannins, saponins, flavonoids, cardiac glycosides and alkaloids is an indication of the antimicrobial potential of the plant.Thus, *V. grandifolia* could be used to wade off disease conditions caused by the microorganisms under study.

KEY WORDS: *Antibacterial, Microorganisms, Phytochemical, Synergistic, Vitex grandifolia*

Introduction

The genus *Vitex* belongs to the *Verbenaceae* family. About 250 species of *Vitex* genus are found globally (Ganapaty and Vidyadhar, 2005). Of these, only about 36 have been reported (Owolabi *et al*., 2010). Of the species of Vitex i.e. *V. grandifolia* is one of the shrubs found in the tropical rain forest of the Niger Delta but predominantly found in the non-wetland region of Nigeria. Hence, *V. grandifolia* is a facultative upland plant (i.e plants with estimated probability of 66–99% and 1–33% occurring in wetland and non-wetland respectively). It can be classified as a Meso-phanerophyte (mostly tall trees with height in the range of 8 – 30 meters).

About 16 species of *Vitex* including *V. agnuscastus*, *V. negundo*, *V. rotundifolia*, *V. trifolia*, *V. gardneriana*, *V. ferrugenia*, *V. cannabifolia*, *V. doniana*, *V. polygama*, *V. leucoxylon*, *V. pinnata*, *V. scabra*, *V. mollis*, *V. altissima*, *V. glabrata*, *V. megapotamica*, *V quinata* have been assessed for their pharmacological potentials (Owolabi *et al*., 2010). Rani and Sharma (2013) reviewed the ethno pharmacology, morphology and microscopy, phytoconstituents, clinical studies, pharmacological reports and toxicological properties of the genus *Vitex*. The *vitex* genus typically has essential oils, flavonoids, iridoid, glycosides, diterpenoides and ligans (Rani and Sharma, 2013).

In the traditionally setting some species of Vitex are used for rheumatic pains, sprains, inflammation, as anti-tubercular, anticancer, diuretic, respiratory infections, in migraine, premenstrual problems, anti-fungal, and insecticidal (Ganapaty and Vidyadhar, 2005). Hemalatha *et al*. (2013) reported that the leaves of *V. negundo* are used in the treatment of cold in Tamilnadu. Tijjani *et al*. (2012) reported

the activity of *V. doniana* Stem Bark on Peripheral and Central Nervous System of rat. The leaves and stem-bark have insecticidal potentials against *Culexquinque fasciatus* (Say) and *Anopheles gambiae* Giles (Diptera: Culicidae) (Azokou *et al*., 2013). Also, Epidi and Odili (2009) reported that the leaf powder of *V. grandifolia* had biocidal effect against *Tribolium castaneum* in stored groundnut *Arachis hypogaea*.

V. grandifolia bears edible fruits which are used in the production of alcoholic drinks; the bark can also be used in the treatment of stomach ache and diarrhea, bronchial complaints, rickets, sore and fever, while the leaves can be used for the treatment of diabetes mellitus and as a diuretic in the treatment of high blood pressure; and as medication against colic, infections of the umbilical cord, toothache, rheumatism and orchitis (Owolabi *et al*., 2010), hence it has medicinal properties. According to Ere *et al.* (2014), medicinal plants have been conventionally used for the treatment of several disease conditions in humans for thousands of years in many parts of the world.

Medicinal plants which are nature's gift to humanity (Masih *et al*., 2014) form the basis of primary health care for a majority of people especially in developing countries (Iniaghe *et al*., 2012; Akinmoladun, 2007; Momoh *et al*., 2014). Statistics shows that about 80% of global population obtain health care services from traditional medicine (Fatima *et al.,* 2011; Minochecherhomji and Vyas, 2014; Gahlaut and Chhiller, 2013). The patronage to traditional medicine is due to inaccessibility of modern drugs to many people in the rural areas and economic factor (Amole and Ilori, 2010). Medicinal plants are essential in the field of pharmacognosy probably due to their clinical constituents (Devanaboyina *et al.,* 2013).

Despite the advancement in the field of microbiology (especially pharmaceutical microbiology), incidence of epidemics due to drug resistant microbes (mainly bacteria and fungi) and the occurrence of emerging and re-emerging microbes still occur (Masih *et al*., 2014). Several plants have demonstrated antimicrobial properties. Typically, medicinal plants are plants whose tissues including roots, leaves, bark possess healing properties (Minocheherhomji and Vyas, 2014; Kigigha *et al*., 2015). The above not-withstanding, there is dearth of information on the phytochemical and antimicrobial properties of *V. grandifolia* in literature hence, the need for this study.

Materials and methods

Vitex grandifolia authentication

V. grandifolia Gürke which is commonly known as Black plum, Chocolate and berry tree, (Azokou *et al*., 2013) is found in the Niger Delta region of Nigeria. The leaves of *V. grandifolia* are about 10–12 cm in length and 5–7 cm in width (Owolabi *et al*., 2010). Typical information about this plant is scarce in literature. However, based on these characteristics described by Owolabi *et al*. (2010), Lemmens (2008) and Azokou *et al*. (2013), the plant was identified.

Source and preparation of organisms

The microorganisms including *S. aureus*, *P. aeruginosa, Bacillus, Salmonella* and *Proteus* species used in this study were obtained from the stock culture in the Medical Microbiology and Parasitology Department, College of Health Sciences, Niger Delta University, Nigeria. The purity of the microbes ascertained as follows: *S. aureus* used was streaked in Mannitol salt Agar which showed yellow pigmentation. It was further grown on Nutrient Agar, the resultant growth on the Nutrient agar was subjected to biochemical test. Klingon Iron Agar was used for the confirmatory test of *P. aeruginosa, Salmonella* and *Proteus* spp. The result was compared with those of known taxa using the scheme of Cheesbrough (2004). Also, *S. aureus*, *P. aerugionas*, *Salmonella, Bacillus,* and *Proteus* species used in this study were confirmed by conducting some biochemical tests such as gram reaction, motility, catalase, coagulase, oxidase, citrate, urease and indole on the organisms using the scheme of Benson (2002) and Cheesbrough (2004).

Antimicrobial screening of the extract

Agar diffusion technique was used for the sensitivity test using the guide of Opoku and Akoto (2015), Ere *et al*. (2014) with slight modification using the guide of Kigigha *et al*. (2015). About 200 µl each of the bacteria was incubated for 24 hours at 37ºC and aseptically inoculated in Mueller Hilton agar plates which were prepared according to the manufacturer's instruction. The plates were spread using cotton swabs. Four wells of 6.0 mm diameter were made on the agar plates using sterile cock bore, each being for each solvents water, ethanol, methanol and hexane. Approximately 100µl of the concentrated extracts from the different solvents was dispensed into the wells using micropipette. The plates were masked with tape to avoid shifting. Two controls were established using known

sensitive disc. For the sensitivity disc analysis, forceps were used to pick the disc into solidified agar plates. All the plates were incubated at 37ºC for 24 hours under aerobic conditions. Thereafter, the resultant zone of inhibition was measured using a meter rule.

Phytochemical screening of the plants

Bioactive constituents including Saponins, phlobatannin, Cardiac glycosides, alkaloids, flavonoids and tannins were determined using the scheme provided by Sofowora (1993), Harborne (1973), Trease and Evans (1993), Okwu (2005) cited in Doherty *et al*. (2010).

Statistical analysis

SPSS software version 16 was used to carry out the statistical analysis. The data were expressed as Mean ± standard error. A one-way analysis of variance was carried out at α = 0.05, and Duncan Multiple Range (DMR) Test was used for the multiple comparison.

Results and Discussion

Table 1 presents the result of phytochemical screening of *V. grandifolia*. The phytochemical assessment showed that *V. grandifolia* contains saponin, alkaloids and flavonoids in all the different plant tissues. Apart from the root of *V. grandifolia,* tannins were found in other tissues of the plant. Similarly, cardiac glycoside was present in root and leaves and absent in the stem. While, phlobatannin was absent in the root but present in the stem and leaves.

Table 1: Phytochemical constituent of *V. grandifolia*

	Tannins	Saponins	Flavonoids	Cardiac Glycoside	Alkaloids	Phlobatannin
Root	-	++	+	+	++	-
Stem	+	++	+	-	++	+
Leaves	++	++	+	+	+	+

++= Highly present; + = Moderately present; - = Absence

The age and part of the plant play important role in the assessment of their bioactive constituents. The phytochemicals found in the tissues of *V. grandifolia* are comparable to those found in other plants as shown for example by the work of Doherty *et al.* (2010) and Kigigha *et al*. (2015) on *Aframomum melegueta* (Alligator pepper). The presence of phytochemicals in plants is an indication that they have functions of biological activity. For example, plants containing alkaloids have mechanisms by which they resist pests including microorganisms (Agu and Thomas, 2012). Typically, alkaloids and their synthetic derivatives are effective for the treatment of analgesic, antispasmodic and bactericidal orientated diseases (Doherty *et al.*, 2010; Opoku and Akoto, 2015; Osuntokun and Oluwafoise. 2015). Flavonoids which are hydroxylated phenolic compounds help plants to resist disease causing microbes (Opoku and Akoto, 2015). Flavonoids have several medicinal properties including antioxidant, anticarcinogens, antimicrobial and antitumor properties, while the presence of saponin in plant is an indication that such a plant could be used as expectorant, cough suppressant (Osuntokun and Oluwafoise, 2015). Scientists have variously reported the medical importance of tannins for the treatment of wounds, varicose ulcers, hemorrhoids, frostbite and burns (Doherty *et al.*, 2010; Osuntokun and Oluwafoise, 2015; Okwu and Okwu, 2004, Kigigha *et al*., 2015).

Table 2 presents the zone of inhibition of the different tissue extracts of *V. grandifolia*. The zone of inhibition of the different bacteria in the ethanolic extracts of the different plant tissues ranged from 12.00 – 15.33 mm (root), 9.00 – 13.00mm (stem), 12.67 – 13.33 mm (leaves), 10.00 – 17.00mm (root + stem), 9.33 – 14.00 (root + leaves), 12.33 – 16.33 mm (stem and leaves) and 11 .67 – 13.33 mm (stem+ root+ leaves). Typically, there were significant differences ($P<0.05$) among the bacteria tested in each of the plant tissues and their combinations. The synergistic potency of the different plants tissues showed that the highest zone of inhibition of 16.33 mm (*Salmonella* sp), 16.00mm (*Bacillus* sp), 14.00mm (*P. aeruginosa*), 14.00mm (*Proteus* sp) and 17.00mm (*S. aureus*) were obtained from stem + leaves,

stem + leaves, root + leaves, root + stem and root + stem respectively. However, there were significant variations ($P<0.05$) in the highest zone of inhibition that was achieved by each of the bacterium under study.

Table 2: Zone of inhibition by different bacteria for the various extracts of *V.grandifolia*

Plant tissues	Microorganisms	Ethanol	Methanol	Hexane	Water
Root	*Bacillus sp.*	14.33±0.33fghi	12.00±0.58bcdefgh	0.00±0.00a	0.00±0.00a
	S.aureus	14.00±0.00abcd	13.67±0.88fghij	10.67±0.33bcde	9.00±0.58b
	P.areugenosa	12.33±0.88cdef	13.00±0.58defghij	10.67±0.88bcde	11.33±0.88cdefg
	Proteus sp.	12.00±1.15bcdef	10.67±0.88bcd	13.33±0.88def	15.33±0.88i
	Salmonella sp.	15.33±0.88ghij	12.33±0.67cdefg	12.33±0.88bcde	11.00±0.58bcdefg
Stem	*Bacillus sp.*	9.00±0.58a	11.00±0.58bcde	10.00±0.58bc	10.00±0.58bcde
	S.aureus	13.00±0.58defg	13.33±0.88efghij	0.00±0.00a	10.00±0.58bcde
	P.areugenosa	12.67±0.33cdefg	15.00±0.58ij	11.33±0.88bcde	12.33±0.88fg
	Proteus sp.	12.00±0.58bcdef	11.00±0.58bcde	12.33±0.88bcde	9.33±0.88bc
	Salmonella sp.	12.00±0.58bcdef	10.67±0.88bcd	10.33±0.88bcd	11.00±0.58bcdef
Leaves	*Bacillus sp.*	13.33±0.88defgh	10.00±0.58bc	10.00±0.58bc	10.33±0.88bcdefg
	S.aureus	13.00±0.58defg	11.33±0.88bcdef	10.00±0.58bc	10.00±0.58bcde
	P.areugenosa	12.67±0.88cdefg	12.00±0.58bcdefgh	11.33±0.88bcde	11.67±0.88defg
	Proteus sp.	12.67±0.88cdefg	9.67±0.88b	11.33±0.88bcde	10.33±0.88bcdef
	Salmonella sp.	12.67±0.88cdefg	11.33±0.88bcdef	11.00±0.58bcde	9.67±0.88bcd
Root+ Stem	*Bacillus sp.*	10.00±0.58abc	12.67±0.88defghi	9.33±0.67b	0.00±0.00 a
	S.aureus	17.00±0.58j	13.00±0.00defghij	11.00±0.00bcde	11.00±0.00bcdefg
	P.areugenosa	13.33±0.33defgh	11.00±0.58bcde	13.33±2.85def	0.00±0.00 a
	Proteus sp.	14.00±0.58efghi	15.00±0.58ij	17.00±0.58g	14.33±0.33 hi
	Salmonella sp.	11.33±0.88abcde	12.00±1.15bcdefgh	10.00±0.00bc	10.33±0.33bcdef
Root+Leaves	*Bacillus sp.*	9.33±0.33ab	11.67±0.67bcdefg	0.00±0.00a	0.00±0.00a
	S.aureus	14.00±1.15efghi	14.33±0.33hij	13.67±1.76def	9.33±0.33bc
	P.areugenosa	14.00±0.58efghi	11.00±1.15bcde	10.33±0.33bcd	10.67±0.67bcdef
	Proteus sp.	12.00±1.00bcdef	0.00±0.00a	0.00±0.00a	10.67±0.67bcdef
	Salmonella sp.	13.67±0.88defghi	14.00±1.15ghij	11.67±1.20bcde	11.33±0.88cdefg
Stem+Leaves	*Bacillus sp.*	16.00±0.58hij	13.67±0.67fghij	12.33±0.88bcde	0.00±0.00a
	S.aureus	14.00±0.58efghi	13.33±0.33efghij	16.00±0.58fg	13.00±1.15gh
	P.areugenosa	13.33±2.03defgh	11.00±0.58bcde	0.00±0.00a	12.00±0.00efg
	Proteus sp.	12.33±0.33cdef	15.33±0.88j	0.00±0.00a	0.00±0.00a
	Salmonella sp.	16.33±0.88ij	13.00±0.58bcdefghij	13.00±0.58cde	13.00±0.58gh
Root+Stem+Leaves	*Bacillus sp.*	11.67±1.20bcdef	12.67±1.45defghi	11.33±0.88bcde	0.00±0.00a
	S.aureus	13.33±0.33defgh	12.00±0.58bcdefgh	13.00±1.15cde	9.33±0.33bc
	P.areugenosa	13.00±1.15defg	14.00±0.58ghij	12.67±1.45cde	10.67±0.33bcdef
	Proteus sp.	12.67±0.67cdefg	11.33±0.33bcdef	0.00±0.00a	11.00±0.58bcdefg
	Salmonella sp.	12.33±0.88cdef	12.00±0.58bcdefgh	12.00±1.15bcde	12.00±0.58efg

Each value is expressed as mean ± standard error (n = 3). Means followed by the same letters in each column is not significantly different at $P< 0.05$ according to the Duncan Statistics

The zone of inhibition of the different bacteria from the methanolic extracts of the different plant tissues ranged from 10.67 – 13.67 mm (root), 10.67 – 13.33mm (stem), 9.67 – 12.00 mm (leaves), 11.00 – 15mm (root + stem), 11.00 – 14.33mm apart from *Proteus* sp that was resistant i.e. 0.00mm (root + leaves), 11.00 – 15.33 mm (stem + leaves) and 11.33 – 14.00 mm (stem+ root+ leaves). There were significant difference (P<0.05) amongst most of the bacteria in each plant tissue and its combinations. The combinations of the different plants tissues showed zones of inhibition of 14.00 mm (*Salmonella* sp), 13.67 mm (*Bacillus* sp), 15.00mm (*P. aeruginosa*), 15.33mm (*Proteus* sp) and 14.33 mm (*S. aureus*) in respect of root+ leaves, leaves + stem, stem, leaves + stem and root + leaves respectively. There were significant variations (P<0.05) in the highest zone of inhibition that was achieved in each of the bacterium under study.

The zone of inhibition of the different bacteria from the hexane extracts of the different plant tissues ranged from 10.67 – 13.33 mm apart from *Bacillus* sp that was resistant (root), 10.33 – 12.33mm apart from *S. aureus* that was resistant (stem), 10.00 – 11.33 mm (leaves), 9.33 – 17.00mm (root + stem), 10.33 – 13.67 mm apart *Proteus* and *Bacillus* sp (root + leaves), 12.33 – 16.00 mm apart from *P. aeruginosa* and *Proteus* sp that were resistant (stem and leaves) and 11.33 – 13.00 mm apart from *Proteus* (stem+ root+ leaves). There were significant differences (P<0.05) among most of the bacteria in each of the plant tissues and its combinations. The combinations of the different plant tissues showed that the highest zones of inhibition of 13.00 mm (*Salmonella* sp), 12.33mm (*Bacillus* sp), 13.33mm (*P. aeruginosa*), 17.00 mm (*Proteus* sp) and 16.00mm (*S. aureus*) were obtained from leaves+ stem, stem + leaves, stem + root, root+ stem and stem + leaves respectively. There were significant variations (P<0.05) exist in the highest zone of inhibition that was achieved in each of the bacterium under study.

The zone of inhibition of the different bacteria from the aqueous water extracts of the different plant tissues ranged from 9.00 – 15.33 mm apart from *Bacillus* sp (root), 9.33 – 12.33mm (stem), 9.67 – 11.67 mm (leaves), 10.33 – 14.33mm apart from *P. aeruginosa* and *Bacillus sp* that was resistant (root + stem), 9.33 – 11.33 mm apart *Bacillus* sp that was resistant (root + leaves), 12.00 – 13.00 mm apart *Proteus* and *Bacillus* sp (stem and leaves) and 9.33 – 12.00 mm apart from *Bacillus* sp (stem+ root+ leaves). There were significant differences (P<0.05) amongst most of the bacteria in each of the plant tissues and its combinations. The synergistic results of the different plants tissues showed that the highest zones of inhibition of 13.00 mm (*Salmonella* sp), 10.00mm (*Bacillus* sp), 12.33mm (*P. aeruginosa*), 15.33 mm (*Proteus* sp) and 13.00mm (*S. aureus*) were obtained from leaves+stem, stem, stem, root and stem+leaves respectively. Significant variations (P<0.05) existed in the highest zone of inhibition that was achieved in each of the bacterium under study.

The significant differences (P<0.05) that existed among the different tissues or their combination against the selected microbes used in this study could be due to variation in their bioactive constituents as well as the biochemical make-up of the different microbes including their metabolism, nutrition and physiology. Also, the age of the plant and season of the study could also be contributing factors to the variation.

Generally the ethanol extracts had superior zone of inhibition. However, there were fluctuations between the highest zone of inhibition among the organisms as well as extracts (i.e. solvents). Of all the solvents used, ethanol had better results though there was fluctuation among the different solvents used. The trend agrees with the work of Masih *et al*. (2014), Ere *et al*. (2014), Akintobi *et al.* (2013). The presence of insoluble active compound found in cold water and or denaturation of the active constituents by the hot water extraction process could be the reason for the lower zone of inhibition (Opoku and Akoto, 2015).

The zone of inhibition from this study had some similarity with findings of authors who used different plant species. Kamilla *et al*. (2009) reported zones of inhibition of methanolic leaf extract (100.00mg/ml) of *Clitoria fernatea* against some pathogenic microbes including *B. cereus, B. subtilis, B. thuringiensis, S. aureus, P. aeruginosa, S. typhi* and *P. mirabilis* as 13.70 mm, 11.30 mm, 10.00 mm, 11.00 mm, 13.30 mm, 21.00 mm and 8.70 mm respectively (leaf), 12.00 mm, 12.00 mm, 14.30 mm, 12.00 mm, 10.70 mm, 18.70 mm and 19.30 mm respectively (stem), 14.00 mm, 12.70 mm, 15.70 mm, 13.00 mm, 11.30 mm, 10.30 mm and 13.70 mm respectively (flower), 12.30 mm, 12.00 mm, 14.00 mm, 12.70 mm, 12.30 mm, 11.30 mm and 15.70 mm respectively (root). Masih *et al*. (2014) studied the antimicrobial activities of four plants and reported the zones of inhibition as 29.00 mm and 30.00 mm for *S. aureus* and *P. aeruginosa* respectively (methanolic extract) and 33.00 mm and 26.00 mm for *S. aureus* and *P. aeruginosa* respectively (ethanolic extract) for *Acacia nilotica*, 15.00 mm and 5.00 mm for *S. aureus*and *P. aeruginosa* respectively (methanolic

extract) and 10.00 mm and 7.00 mm respectively for *S. aureus* and *P. aeruginosa* respectively (ethanolic extract) for *Catharanthus roseus*, 14.00 mm and 17.00 mm for *S. aureus* and *P. aeruginosa* respectively (methanolic extract) and 15.00 mm and 14.00 mm for *S. aureus* and *P. aeruginosa* respectively (ethanolic extracts) for *Sida cordifolia* and 15.00 mm and 16.00 mm for *S. aureus* and *P. aeruginosa* respectively (methanolic extracts) as 12.00 mm and 15.00 mm for *S. aureus* and *P. aeruginosa* respectively (ethanolic extract) for *Euphorbia birta*. Samidurai and Saravanakumar (2009) studied antibacterial activity of methanolic leaf extracts of *Pemphisacidula* forst and reported the zone of inhibition at different concentrations for *S. aureus* and *P. aeruginosa* as 8.00 mm and 6.00 mm respectively (5%), 4.00 mm and 8.00 mm respectively (10%).

Akrayi and Abdulrahman (2013) reported the zone of inhibition of 100% concentration distilled water extract of thyme, black dry lime and chili as 11.00 mm, 24.00 mm and 16.00 mm respectively (*P. aeruginosa* ATCC) and 0.00 mm, 22.00 mm and 11.00 mm respectively (*P. mirabilis*). Mann (2012) reported the zones of inhibition of different extracts (1000ug/disc) of *Ocimum gratissimum* against *S. typhi*, *P. aeruginosa, S. aureus, P. vulgaris and B. anthracis* as 10.20 mm, 12.00 mm, 0.00 mm and 0.00 mm respectively (ethanolic extract), 12.50 mm, 15.50 mm, 0.00 mm, 0.00 mm and 0.00 mm respectively (hexane extract), 0.00 mm, 0.00 mm, 17.00 mm, 0.00 mm and 0.00 mm (methanol extract). Akintobi *et al.* (2013) reported zones of inhibition for different extracts of *Zingiber offinale* against *S. aureus, P. mirabilis, B. subtilis, S. typhi* and *P. aeruginosa* as 9.00 mm, 11.00 mm, 0.00 mm, 13.00 mm and 0.00 mm respectively (water extracts) and 13.00 mm, 17.00 mm, 0.00 mm, 10.00 mm and 14.00 mm respectively (ethanolic extracts). However, the slight variation that exists between this study and previous study could be due to age of the plants, type and biochemical composition of the different plants, concentration of the plant extracts used as well as the amount of the extract used for the sensitivity test.

The zone of inhibitions for the known antibiotics used against *S. aureus, P. aeruginosa, Salmonella*, *Proteus* and *Bacillus* spp. are presented in Figure 1. *S. aureus* was resistant to Rifampin, Chloramphenicol and Levofloxacin, while the zone of inhibition to other antibiotics used including Ciproflox, Norfloxacin, Gentamycin, Amoxil, Streptomycin, Erthythromycin and Ampiclox ranged from 21.00 – 27.00mm. *P. aeruginosa* was resistant to Chloramphenicol and Levofloxacin, while other antibiotics sensitivity ranged from 16 – 26.00 mm. The sensitivity of *Salmonella*, *Bacillus* and *Proteus* spp to all the antibiotics used for the study ranged from 23.00 – 27.00 mm and 22.00 – 27.00 mm, 18.00 – 25.00 mm respectively. The variation in the zone of inhibition could be due to biochemical composition of both the known antibiotics as well as that of the microorganisms used for the study.

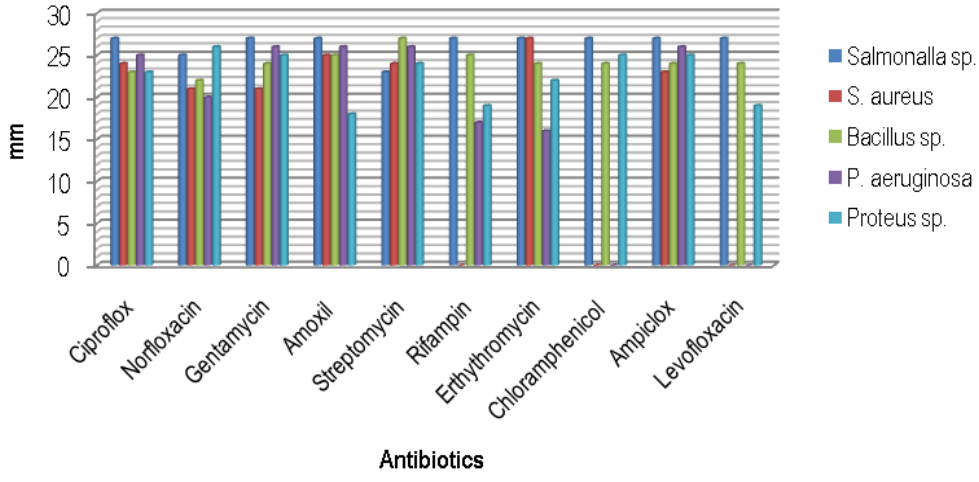

Figure 1: Antibiotics sensitivity (mm) results against the bacteria used in this study

Since the extracts of different tissues of *V. grandifolia* is effective against both gram positive and gram negative bacteria used in this study, it can inferred that they can be used as a broad spectrum antibiotics against bacteria. Some of the known antibiotics reported in this study have been previously reported to be effective against both gram negative and positive bacteria (Pondei *et al*., 2012; Kigigha *et al*., 2015; Ere *et al*., 2014; Samidurai and Saravanakumar, 2009; Amole and Ilori, 2010; Fatima *et al*., 2011; Kamilla *et al*., 2009).

Conclusion

The use of different tissues of plant for the remedy of several ailments can be dated back to several centuries. The species of plant used by traditional medicine practitioners depend on the availability of such plant in their locality especially the rural dwellers. Microorganisms are ubiquitous and are known to cause several disease conditions. This study investigated the phytochemical, antibacterial and synergistic efficacy of the different tissues of *V. gradifolia* against disease causing bacteria. The study showed that *V. grandifolia* contains bioactive constituents such as tannins, saponins, flavonoids, cardiac glycosides, alkaloids and phlobatannin. Superior zone of inhibition occurred in the synergy study. In general, the ethanol extracts was superior to methanol, hexane, and water.

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