Activities of Express Extracts of *Costus afer* Ker–Gawl. [Family COSTACEAE] Against Selected Bacterial Isolates

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ABSTRACT

Purpose: This study investigated the activity of express extracts of *Costus afer* (bush-cane) against *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aerugionsa*. **Materials and Methods:** The leave and stem sap of *Costus afer* was obtained from Wilberforce Island, Bayelsa state, Nigeria. The various plant parts (leave and stem) were separately macerated and the resultant extracts was obtained through filter-pressing using a double muslin cloth. The agar well diffusion method was employed for the sensitivity study. **Results:** The zone of inhibition exhibited by *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aeruginosa* were 10.67±1.29mm, 11.67±0.88mm, 12.67±0.33mm and 9.00±0.58mm, respectively (for synergy), 10.00±1.56mm, 11.33±1.33mm, 12.67±0.88mm and 8.66±0.66mm, respectively (for stem extract), 9.33±0.33mm, 10.00±0.58mm, 9.67±0.33mm and 7.33 ±0.33mm, respectively. Analysis of variance showed that there were no significant variations ($p>0.05$) among the various isolates for each of the plant parts and their combination except for the leaves. Between the various plant parts for each of the test organisms, there was no significant variation ($p>0.05$) except for *Bacillus subtilis.* **Conclusion:** The inhibition by the stem and leave express extracts of *Costus afer* suggests that they possess broad-spectrum antibiotics potentials.

Key Words: *Antibacterial, Costus afer, Medicinal Plants, Public health, Zone of inhibition.*

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INTRODUCTION

Costus afer which belongs to the Costaceae family is known as English ginger lily, common ginger lily, bush-cane [1] or monkey sugar cane [2]. *Costus afer* is a perennial rhizomatous herbaceous plant that can grow up to 3-4-meter high. The plant is found in several regions in Africa including Senegal, Gambia, Guinea, Sierra Leone, Ivory Coast, Ghana, Togo and Nigeria [1, 2] where they have several tribal names. The plant occurs mainly in the wild especially in regions with high rainfall. Nna et al. [2] also reported that *Costus afer* is widely distributed in western and eastern region of Nigeria.

The plant has several pharmacological properties. Burkill [1] reported that some parts of *Costus afer* can be used for the treatment of arthritis, rheumatism, venereal diseases, leprosy, dropsy, swellings, oedema, naso-pharyngeal affections, and can be used as laxatives and genital stimulants/depressants. According to Omokhua [3], Nna et al. [2], stem sap of *Costus afer* is used for the treatment of urethral discharges, Jaundice, venereal diseases and prevention of miscarriages in Akenfa, Bayelsa state, and treatment of cough, measles and malaria in Aluu, Rivers State. Omokhua [3] and Nna et al. [2] reported that the leaf extract of *Costus afer* is used for the treatment of eye inflammation, headache and malaria in Ogoni Land of Rivers State. The authors further reported that an infusion of dried aerial parts of the plant is used to treat hypertension in Ogboloma, Bayelsa State. Other uses of *Costus afer* include treatment of gonorrhoea, toothache, epilepsy, leprosy, stomach disorders, etc. [2, 3]. *Costus*

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afer is also used in combinations with some other plants for the treatment of some ailments including malaria and hunch back (using boiled leaves and stem of *Costus afer* and *Alchornea cordifolia*) among Ogba people in Rivers state, fever and malaria (using combination of boiled leave and stem of *Costus afer*, *Carica papaya*, *Citrus* species) among the Ikwerre ethnic group in Rivers State [2, 3].

Scientific studies have validated the therapeutic potentials of *Costus afer*. Some of the activities include cardioprotective potential [4], anti-oxidant [5], antimicrobial [6-8], analgesic, anti-inflammatory and CNS depressant activities [8]. Several plants have been reported to possess pharmacological potentials and have been widely used to treat several human diseases in different regions of the world. Authors have reported that approximately 70 – 80% of global population rely on traditional medicine practitioners for the treatment of different diseases [3, 9 – 17]. Most of the people that depend on tradition health care reside in rural areas in many developing nations [18].

Microorganisms are known to cause several human diseases. This is probably due to the fact that they possess diverse adaptation mechanisms and can survive in different environmental conditions. In the human body, different microbes have the tendency to invade the nasal cavity, throat, mouth, skin, stomach, small and large intestine, urethra and vagina area. For instance, *Staphylococcus aureus* can be found in the skin and nasal area in human. *Staphylococcus aureus* is among the causative agents of some blood stream infection and boils in the skin [2]. Like *Escherichia coli*, *Pseudomonas aeruginosa* is a gram negative bacteria that can be found in the large intestine in human. *Pseudomonas aeruginosa* is among the causative agents of bacteremia, pneumonia, eye infections, ear infections, etc. [2, 19]. Some pathogenic strains of *Escherichia coli* are among the causative agents of some urinary tract infections and diarrhea [2, 20]. Though some of the strains of *Escherichia coli* are harmless to both animals and humans. Some species of *Bacillus* could cause bacteremia and endocarditis in human [21]. Specifically, some species such as *Bacillus subtilis* seems to have low degree of virulence to humans, and are rarely isolated for human subjects.

Due to the challenges of drug resistance by known antibiotics, research into the discovery of alternative treatment methods have been intensified using different plant parts. Hence, this study focused on the antimicrobial activities of express extract of *Costus afer* against selected bacterial isolates.

MATERIALS AND METHODS

Samples collection and extraction

The leave and stem of *Costus afer* used for this study was obtained from Wilberforce Island, Bayelsa state, Nigeria. The plant parts (leave and stem) were macerated separately using pestle and mortar and the extract was obtained through filter-pressing using a double muslin cloth. Synergy of the leave and stem were made in 1:1 ratio [12, 13].

Sources and Preparation of organisms

The isolates (*Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Bacillus subtilis*) used for this study were obtained from Microbiology Units, Federal Medical Centre, Yenagoa, Bayelsa State, Nigeria. The purity of the isolates was determined using the guide of Cheesbrough [22]. The isolates were separately inoculated into sterile peptone water and incubated at room temperature for 24 hours prior to the sensitivity testing.

Antimicrobial screening of the extracts

The agar well diffusion method previously applied by Kigigha et al. [9 – 16, 23] was adopted for the sensitivity testing. Approximately 0.3mls of each extract was dispensed into Nutrient Agar well (of 6.00mm) containing the tests organisms. The agar plates were labelled and incubated for 24 hours and the zone of inhibition were measured using meter rule.

Statistical analysis

GraphPad prism 5 was used for the statistical analysis. The data obtained were expressed as Mean ± standard error. Single factorial analysis of variance was carried out $p<0.05$ and Tukey multiple comparison test was used to showed the source of variation between means.

RESULTS AND DISCUSSION

The zone of inhibition of express extracts of *Costus afer* is shown in Figure 1- 4. The zone of inhibition exhibited by *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aeruginosa* were 10.67±1.29mm, 11.67±0.88mm, 12.67±0.33mm and 9.00±0.58mm, respectively, being not significantly different ($p>0.05$) (for synergy) (Figure 1), 10.00±1.56mm, 11.33±1.33mm, 12.67±0.88mm and 8.66±0.66mm, respectively, being not significantly different ($p>0.05$) (for stem extract) (Figure 2), 9.33±0.33mm, 10.00±0.58mm, 9.67±0.33mm1 and 7.33 ±0.33mm, respectively, being significantly different ($p<0.05$). Tukey's multiple comparison test showed no significant variation ($p>0.05$) between *Escherichia coli* and *Staphylococcus aureus*, *Escherichia coli* and *Bacillus subtilis*, and *Staphylococcus aureus* and *Bacillus subtilis.* Furthermore, *Escherichia coli* and *Pseudomonas*

aeruginosa, *Bacillus subtilis* and *Pseudomonas aeruginosa*, and *Staphylococcus aureus* and *Pseudomonas aeruginosa* showed significant variations ($p<0.05$) (Figure 3). Based on the various parts of the plant for each of the test organisms, there was no significant variation ($p>0.05$) except for *Bacillus subtilis* (that multiple comparison test showed significant variation at $p<0.05$ between leaf and synergy extracts) (Figure 4).

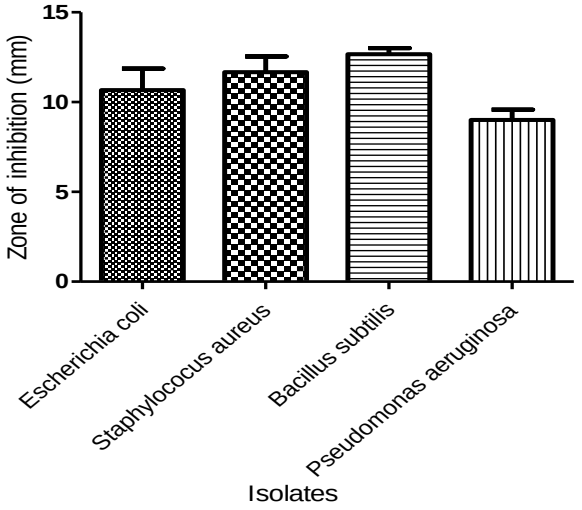


Figure 1: Zone of inhibition of synergy (leaf and stem) express extracts of *Costus afer*

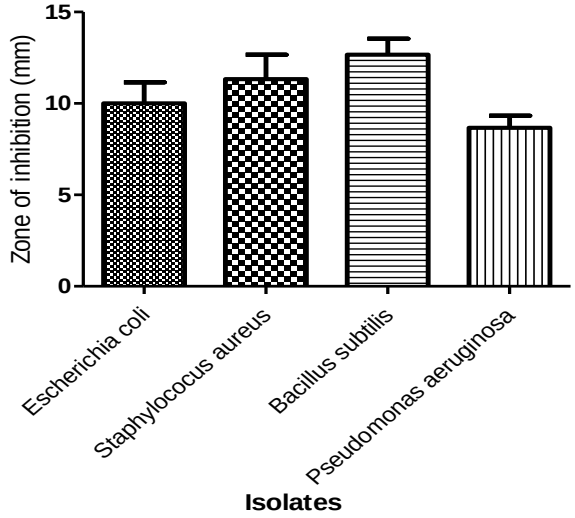


Figure 2: Zone of inhibition of stem express extracts of *Costus afer*

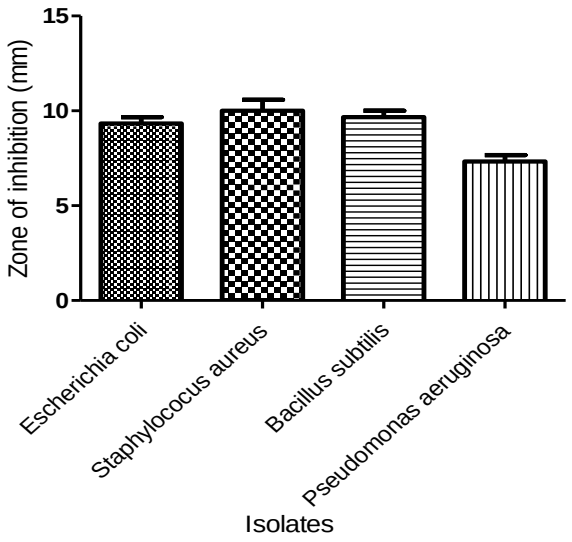
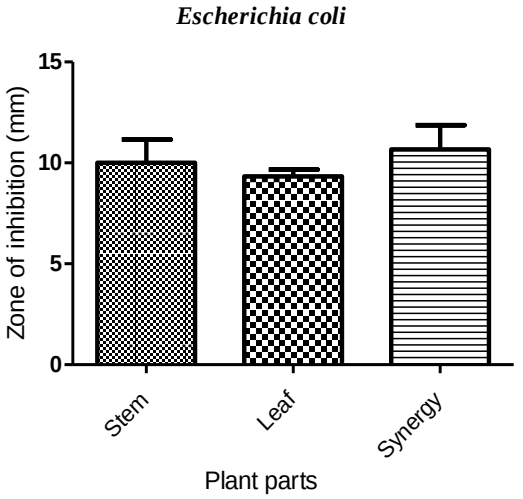
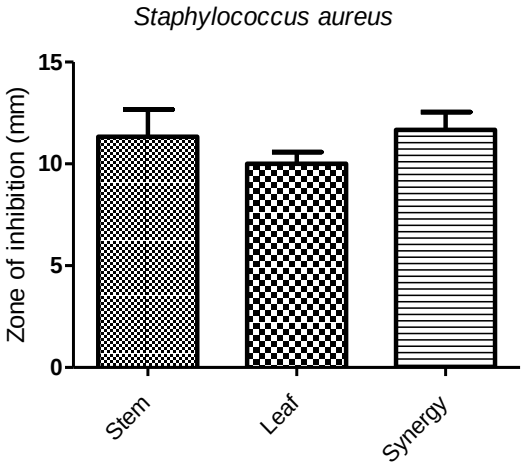


Figure 3: Zone of inhibition of leave express extracts of *Costus afer*





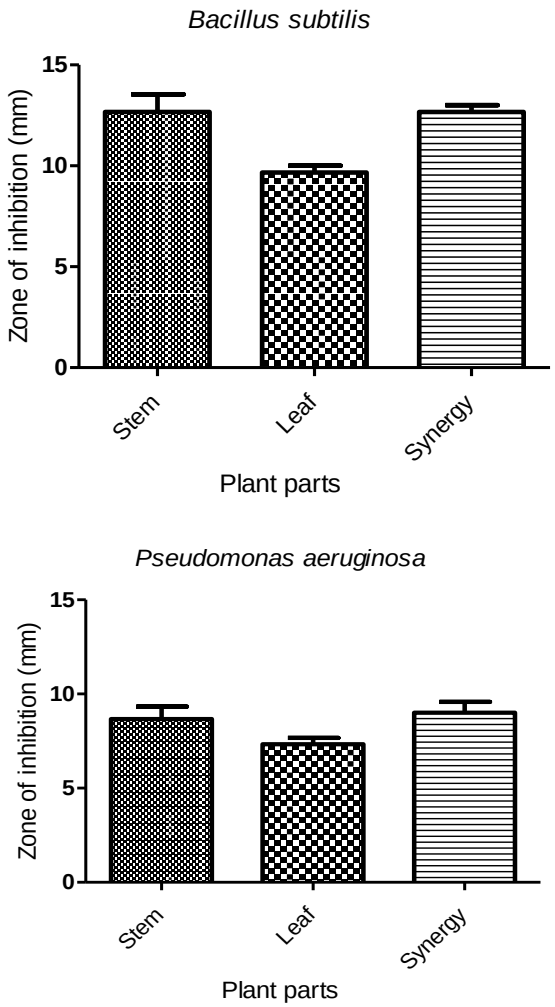


Figure 4: Zone of inhibition of the different plant parts of *Costus afer* on the test organisms

The findings of this study revealed that express extracts of *Costus afer* has antimicrobial properties. Pharmacological potentials of the plant could be due to its bioactive ingredients [12, 13]. Authors have variously reported the different phytochemical constituents of *Costus afer*. For instance, Ukpai et al. [24] reported that aqueous stem extract of *Costus afer* extract contains alkaloids, saponins, flavonoids, tannins and phenols. Anyansor et al. [25] reported that *Costus afer* extracts contain flavonoids, phenols, anthraquinones, cardiac glycosides, terpenoids, alkaloids and tannins. Akpan et al. [6] reported that ethanolic leaf extract of *Costus afer* contain tannins, saponin, alkaloids, anthraquinone, flavonoid, terpenes, phlobatanin and glycosides. Ezejiofor et al. [8] reported that ethanolic leaf extract of *Costus afer* alkaloids, saponins, flavonoids, tannins, phenols, glycosides and terpenoids. Nna et al. [2] reported that the stem of *Costus afer* contains alkaloids, flavonoids, tannins, triterpenoids, saponins, cardiac glycosides (using qualitative techniques), rutin, quinine, oxalate, naringin, catechin, anthocyanin, lunamarine, sapogenin, epicatechin, ribalinidine, sparteine, kaempferol, phytate, flavones, naringenin, anthocyanidines and tannin (using gas chromatography). Anyansor et al. [25] reported the presence of phenol, flavonoid, flavonol, proanthocyanidins, tannin, saponin and alkaloid in hexane leaf extracts of *Costus afer*. The authors further reported that Phenol, 2,4-bis(1,1-dimethylethyl)-, 2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7atrimethyl, Pentadecanoic acid, methyl ester, 1,2-Benzenedicarboxylix acid, mono (2-ethylhexyl) ester (phthalate) (detected using gas chromatography with mass spectrometry) from hexane leave extracts of *Costus afer* has antibacterial activities. In addition, the presence of alkaloids and flavonoids could be among the major contributing bioactive compounds that make the plant confer antibacterial activity.

The values obtained in this study has some similarity with previous work on *Costus afer*. Nna et al. [2] reported that stem ethyl acetate extract of *Costus afer* is sensitive to both gram positive and negative organisms with zone of inhibition of 26mm (*Bacillus cereus*), 18mm (*Staphylococcus aureus*), 14mm (*Escherichia coli*) and 12 mm (*Pseudomonas aeruginosa*). John et al. [26] reported that ethanolic leaf extract of *Costus afer* has zone of inhibition of 9mm and 8mm for *Pseudomonas aeruginosa* and *Escherichia coli* at 150mg/l concentration. Effiong and Obi [27] reported that methanolic stem extract of *Costus afer* has zone of inhibition of 15.2mm and 17.1mm for *Escherichia coli* and *Staphylococcus aureus*, respectively at 100mg/ml. Uchegbu et al. [7] reported zone of inhibition of 8mm (*Proteus mirabilis*), 10.00mm mm (*Klebsiella* pneumonia), 12.00mm (*Staphylococcus aureus*), 11.00mm (*Pseudomonas aeruginosa*) and 13.50mm (*Escherichia coli*) from stem extract of *Costus afer*. The slight variations between the result of this study and previous works could be due to the age, plant parts and its moisture content, extraction solvent and biochemistry of the isolates [28, 29]. On the overall, the findings of this study indicates that express extract of *Costus afer* has broad spectrum antibacterial potentials.

CONCLUSION

The extracts of *Costus afer* is used as an active ingredient for the treatment of several diseases by some ethnic groups in the Niger Delta region of Nigeria. In addition, it is important due to the problems of antibiotics resistance and the search for a suitable alternative. Hence, this study evaluated the antibacterial activities of stem and leave express extracts of *Costus afer*. The study found that the extracts possess antibacterial activity against *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aerugionsa*. The zone of inhibition was not significantly different ($p>0.05$) among the various

bacterium under study except for leaf extracts that showed significant variations ($p<0.05$). Based on the plant parts, there was no significant variations ($p>0.05$) for each of the organisms except for *Bacillus subtilis*. The activities of the express extracts of *Costus afer* stem and leave against gram positive and negative bacterium is indication that they have potentials for the development of broad-spectrum antibiotics.

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