



Antioxidant and Antimicrobial Activities of Floral Methanolic Extract of *Costus lucanisianus*

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

The chemical and biological profiling of the methanolic extract of the floral part of *Costus lucanisianus* have been carried out. The floral part was collected from Ogbia Local Government Area of Bayelsa State. The floral part was air dried, grinded and extracted at room temperature in methanol. The methanol extract mixture was then concentrated under pressure. The concentrated extract, MCL was subjected to quantitative phytochemical screening, antioxidant and antimicrobial profiling. The result of the quantitative analysis showed the presence of phenols (14.853 ± 0.350), tannins (6.638 ± 0.051), alkaloids (3.72 ± 0.02), flavonoids (59.444 ± 0.000) and saponins (2.93 ± 0.06). The antioxidant activity increases as the concentration of the methanolic extract increases. Biologically, the floral part has shown activities against the following strains of microbes: *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Candida albican*, *Klebsiella pneumonia*, and *Fusarium spp.* with the highest activity observed against *Staphylococcus aureus*. The least antimicrobial activity was observed for *Escherichia coli*. The present study has established the phytoconstitution, *in vitro* antioxidant and antimicrobial activities.

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1. INTRODUCTION

The biodiversity of phytochemicals in the plant kingdom has been of immersed blessing to mankind and his living counterparts. The spread and wide range of phytochemical applications have heightened research and development towards reproducing what nature has endowed the ecosystem with. One of such nature depots of therapeutic significant secondary metabolites is the plant kingdom. Notably, the plant kingdom has been basis for phytochemicals with vast activities such as antioxidant, antimicrobial, larvicidal, analgesic, antistress, antiprotozoal and cytotoxicity [1,2,3,4]. The need to constantly look for complementary medicines has necessitated the explorations of the various parts of plants for their pharmacological relevance.

Costus lucanisianus is a tropical herbal perennial plant commonly known as 'spiral ginger' or locally referred to as 'monkey sugarcane' in the Southern part of Nigeria [5,6]. Ethnomedicinally, the leaves have been employed in the treatment of prolapsed rectum, diarrhea, pyrexia, pain and dysmenorrhea [7]. Pharmacologically, the antihyperglycemic, hepatoprotective, renoprotective [6], uterine-relaxant [8,9] and oxytocic [10] potentials of *Costus lucanisianus* have been established. However, the pharmacological properties of the floral part have not been investigated. There is need to probe its phytoconstitution and establish the *in vitro* bioactivities (antioxidant and antimicrobial) as platform for drug targets in complementary medicines.

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

Chemicals and reagents employed in the course of this work were of analar grade and products of Sigma-Aldrich. Chemicals and reagents used include: methanol, 2, 2-diphenyl-1-picrylhydrazine (DPPH), distilled water, dimethylsulfoxide (DMSO), Folin-Ciocalteu, Na₂CO₃, AlCl₃, gallic acid and rutin.

2.2 Collection of Plant Material

The floral part of the plant material was collected from Ogbia Local Government Area of Bayelsa State, Nigeria. The complete plant material was identified by a botanist with the Department of

Biology, Federal University Otuoke, Bayelsa State, Nigeria.



Plate 1. *Costus lucanisianus*

2.3 Extraction of Plant Material

40 g of air dried floral part of the plant material was placed in a 1000 ml standard flask and then extracted with methanol at room temperature for 72 hrs, shaken at intervals and decanted. The mixture was concentrated under pressure and constant temperature of 40°C with the aid of a rotatory evaporator (R-250 Pro) to yield the methanolic extract.

2.4 Quantitative Phytochemical Screening of the Methanolic Extract

The quantitative phytochemical screening on the methanolic extract was carried out using standard methods as described by researchers [11,12].

2.5 Determination of Total Phenolic Content

Diluted solution of methanolic extract (0.5 ml) was dissolved in 100 µl of Folin-Ciocalteu reagent and 6 ml of distilled water. It was vortexed for 1 min, before adding 2 ml of 15% sodium carbonate (Na₂CO₃) and the mixture vortexed once again for 30 sec. The solution was then made up to 10 ml with distilled water. The absorbance was read at 750 nm via UV spectrometer (Perkin-Elmer) after one and half hours [13,14]. The total phenolic was expressed as milligrams of gallic acid equivalent.

2.6 Total Flavonoid Content

5 ml of 2% AlCl₃ in methanol was mixed with 0.4 mg/ml of the methanolic extract and the absorption read at 415 nm with the aid of a UV spectrometer (Perkin-Elmer) after 10 min. The

blank for the assay is a mixture of equal volumes of both the extract and methanol. The total flavonoid content was determined using a standard curve with rutin as standard [14,15].

2.7 Antioxidant Analysis of the Methanolic Extract

The antioxidant analysis of methanolic extract was done using the 2,2-Diphenyl-1-picrylhydrazine, DPPH free radical technique with the wavelength of absorbance read at 517 nm using ascorbic acid as an organic standard as reported in earlier studies [16,17]. The inhibitory percentage, IP was calculated using equation 1.

$$IP = \frac{P_b - P_s}{P_b} \times 100 \quad (1)$$

Where: P_b is the absorbance of blank,
 P_s is the absorbance of the methanolic extract

2.8 Antimicrobial Analysis of the Methanolic Extract

The antimicrobial analysis was carried out using the Agar diffusion method [18,19,20] Microbial strains of *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Candida albican*, *Klebsiela pneumonia*, and *Fusarium spp.* were cultured and probed for the antimicrobial efficacy of the methanolic extract.

2.9 Agar Diffusion Method

Methanolic extract for the susceptibility test was prepared by dissolving 2 g methanolic extract in 10 ml of sterile 1% peptone water. Serial dilution was carried out to obtain 100, 50, 25 and 6.25 mg/ml respectively. Antimicrobial activities of the various concentrations were tested by inoculating an aliquot in a well bored on Muller Hilton culture agar sealed with test bacteria. The

plates were incubated for 72 hours at 37°C. The relative susceptibility of the microorganisms in the various extract was indicated by clear zones and growth of inhibition around the well. The zone of inhibition was recorded in millimeter. Ciprofloxacin and Ketoconazole were also inoculated as a positive control.

3. RESULTS AND DISCUSSION

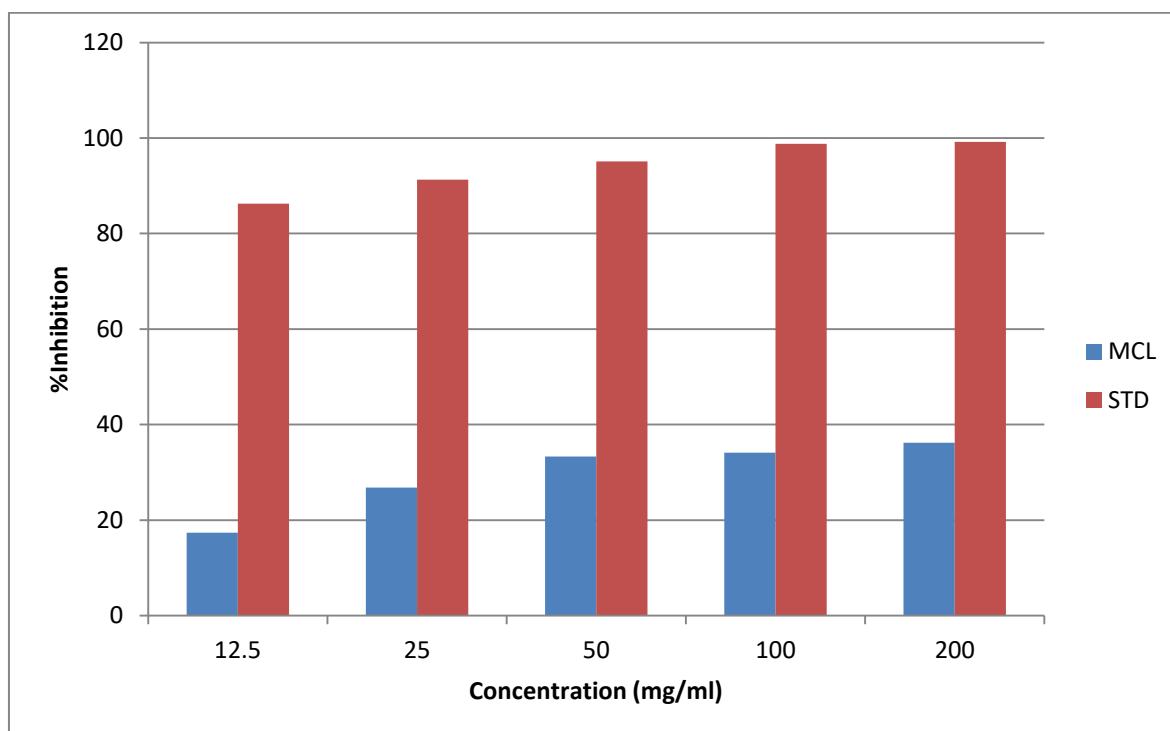
3.1 Quantitative Phytochemical Screening of Methanolic Extract

The presence of phenolics (14.853±0.350), tannins (6.638±0.051), alkaloids (3.72±0.02), flavonoids (59.444±0.000) and saponins (2.93±0.06) have been identified and reported in Table 1. The presence of these phytochemicals has been reported in literature to be present in the stem of *Costus lucanisianus* [21]. The floral part of *Costus lucanisianus* is rich in phytochemicals that can mediate in complementary medicines. The phenolics are phytochemicals with established therapeutical bioactivities such as the antioxidant, anticancer, antimutagenic and anti-inflammatory activities [22]. Tannins are known phytochemicals with wide range of activities such as antimicrobial, fasten blood clotting, lower blood pressure and regulate serum lipid level [23]. Alkaloids are known amines of natural products and have been implicated in the physiological functions such as anesthetics, cardioprotective and anti-inflammatory [24]. Flavonoids are polyhydroxylated aromatic compounds with proven antioxidant, anticancer, antiviral, anti-inflammatory, neuro- and cardio-protective activities [25]. Saponins are known for their characteristic foamy nature and have been explored in clinical science as inhibitor of tooth caries, platelet aggregation, acute lead poisoning and in the treatment of hypercalciuria [26]. The phytoconstitution identified in the floral part of *Costus lucanisianus* is a justifiable fact for bioactivities outcome.

Table 1. Quantitative phytochemical screening of Methanolic extract

Phytochemical	Concentration
Total Phenolics (mg/g GAE)	14.853±0.350
Tannins (m/g GAE)	6.638±0.051
Alkaloids (mg/g)	3.72±0.02
Flavonoids (mg/g RE)	59.444±0.000
Saponins	2.93±0.06

*GAE- Gallic Acid Equivalent, RE- Rutin Equivalent



*MCL- methanolic extract, STD- standard

Fig. 1. Antioxidant analysis of Methanolic extract

Table 2. Antimicrobial analysis of Methanolic extract

Organisms	100	50	25	12.5	6.25	10ng/ml Cipro	1% Ket
S.a	22	18	16	14	12	28	NA
B.s	20	14	12	10	10	24	NA
E.c	14	12	10	0	0	20	NA
P.a	16	14	12	12	10	25	NA
S.t	18	12	10	8	6	18	NA
K.p	16	10	10	8	6	22	NA
C.a	16	14	10	10	8	0	NA
Fus	12	10	8	0	0	NA	20

*S.a- *Staphylococcus aureus*, B.s- *Bacillus subtilis*, E.c- *Escherichia coli*, P.a- *Pseudomonas aeruginosa*, S.t- *Salmonella typhi*, C.a- *Candida albican*, K.p-*Klebsiela pneumonia*, Fus-*Fusarium spp.*, Cipro-Ciprofloxacin, Ket-Ketoconazole

3.2 Antioxidant and Antimicrobial Analysis of the Methanolic Extract

The profiling carried out on the antioxidant activity of the methanolic crude extract (Fig. 1.) shows that *Costus lucanisianus* floral has antioxidant potential as it's obtainable in the family [21]. The antioxidant capacity is however, lowered when compared to those of ascorbic acid. This antioxidant potential general increases as concentration decreases. The presence of phytochemicals such as the flavonoids, phenols

and tannins are supporting evidence for antioxidant activity of the floral part of *Costus lucanisianus*. Inferentially, the floral part of *Costus lucanisianus* can be employed in the scavengers of free radicals that are associated with some endogenous metabolic activities within living systems. Free radicals are species with associated unpaired electrons with biochemical implications in the oxidative modifications of macromolecules. Macromolecules such as the lipids, nucleic acids (DNA, RNA) and the proteins once interfered with could lead to clinical cases

such as aging, diabetes, cancers and neuro-degenerative conditions in mankind [27].

Bacteria such as *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Candida albican* and *Klebsiela pneumonia* are single cell organism that are associated with day-to-day engagement. However, some are beneficial and others are pathogenic. Bacteria like *Staphylococcus aureus*, *Salmonella typhi* and *Escherichia coli* are known be causative agents of syphilis, typhoid, urinary tract infection and diarrhea [28]. The antimicrobial activity of methanolic crude floral extract of *Costus lucanisianus* is reported in Table 2. The highest antimicrobial activity was witnessed against *Staphylococcus aureus*. The least antimicrobial activity was observed for *Escherichia coli*. The antimicrobial activity of methanolic crude extract generally decreases as the concentration decreases. The antimicrobial activity of methanolic crude extract against tested strains of bacteria was generally lowered than that of the standard, ciprofloxacin. Susceptibility was observed at the highest concentration of 100 mg/ml for *Staphylococcus aureus*, *Bacillus subtilis* and *Salmonella typhi*. Those of *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albican* and *Klebsiela pneumonia* were of intermediates between susceptibility and resistance. *Fusarium spp* is a fungus with notable pathological relevance. It causes a wide range of infections such as keratitis, onychomycosis and both the invasive and disseminated ones [29], sinusitis [30] and mycotoxicosis [31,32]. Table 2, reveals the antimicrobial activity of *Fusarium spp* which witnessed resistance at concentrations lower than 12.5 mg/ml and intermediate activity at the higher concentrations.

4. CONCLUSION

The methanolic extract of the floral part of *Costus lucanisianus* has been profiled for its phytoconstitution, antioxidant and antimicrobial activities. Phytochemically, the floral part of *Costus lucanisianus* contains phenols, tannins, alkaloids, flavonoids and saponins. The presence of these phytoconstituents is responsible for the antioxidant and antimicrobial activities of the floral part. Thus, there are evidence for the therapeutical applications of the floral part of *Costus lucanisianus* in complementary medicines. The toxicological and further pharmacological studies should be carried so as

to ascertain toxicity and pharmaceutical relevance of the floral part of *Costus lucanisianus*.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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