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Antimicrobial properties of the ethanolic extracts of *Zingiber officinale* (Ginger) on *Escherichia coli* and *Pseudomonas areuginosa*

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ABSTRACT

The antimicrobial properties of various extract of Zingiber officinale (ginger) against Escherichia coli and Pseudomonas aeruginosa that are common causes of gastrointestinal track infection were investigated using the Agar and tube diffusion method. The result obtained revealed that etharolic extract of ginger gave the widest zone of inhibition against one out of the two test organisms at the concentration of 20mg/ml. However, Pseudomonas aeruginosa was more sensitive to the extract. It was also observed that the solvent of extraction and its varying concentrations affected the sensitivity of the two test organisms to the plant extract, showing that ginger has antimicrobial activities on the test organisms due to its inhibitory effect thus confirming its use in folk medicine.

Keywords: Antimicrobial properties, *Zingiber officinale, Escherichia coli, Pseudomonas.*

INTRODUCTION

The use of medicinal plants to treat ailment associated with pains is well known through history [(2).] Such plants can play important role in drug discovery and this study is logical research strategies in the search for new drugs [(10).]
Ginger, the underground rhizome of the plant *Zingiber officinale* has been used as a medicinal plant in Asia, India, Jamaica and Nigeria. In China, ginger has been used to aid digestion, treat stomach upset, diarrhea and nausea, for more than 2000 years [(6 α 1).]

Ginger has a wide range of action on the human body and has been found effective in the treatment of cataract, heart disease, migraines, struck amenorrhea, athletes foot, bursitis, chronic

fatigue, cold, flu, coughs, depression, dizziness, fever, arectile difficulties, kidney stones, reynad's disease and viral infection [9].

Ginger has also been historically used to treat inflammation, which several scientific studies support through one arthritics that showed ginger to be no better that a placebo or ibuprofen. Research on rats suggests that ginger may be useful for treating diabetes; in the west, powdered derived ginger root is made into capsules and sold in pharmacies for medical use, [7 α 8].

In Venezuela, ginger is pounded into a paste and applied to the abdomen for difficult menstruation. In Costa Rica, it is used in a decoction to relieve throat inflammation and asthma, with the addition of honey; it is a valued remedy for coughs and bronchitis, and also serves as a sudorific in fever. Its natural diuretic stimulates the kidney to flush out toxins faster. In Panama, it is used to relieve rheumatism. In Guatemala and Trinidad, it is the best remedy for stomachache, malaria and indigestion; the fumes from an infusion in urine are inhaled to relieve head colds [3 α 5].

Due to prevalent resistance of micro organism to drugs and other therapeutic agent, thus research work is aimed at investigating the potentials of ginger as a medicinal plant that will provide a natural and locally produced antimicrobial agent with an effective rate compared to the standard antibiotics commercially produced against pathogenic microorganisms.

MATERIALS AND METHODS

Sampling

The test organisms, Escherichia coli and Pseudomonas auruginosa, were collected from National Institute of Pharmaceutical Research, Abuja. The pure culture was sub-cultured on nutrient agar then preserved in the refrigerator at 4^{o}C until it was required for study.

The plant material *Zingiber officinale* (Ginger) was purchased from Nasarawa market and authenticated at the College of Agriculture Lafia, Nasarawa State.

Extraction of Plant Material (Ginger)

The ginger rhizomes were washed with distilled water and allowed to dry (air-day) for two days. Extraction was done using the following procedures: Crude extraction methods, Cold water extraction and Ethanolic extraction.

i) 150g of fresh ginger was blended into zinc powder and soaked in 100ml of distilled water for 24 hours. The pulp obtained was left in a clean sterile glass container and shaken vigorously to allow proper extraction and it was filtered using sterile muslin cloth to obtain the filtrate and evaporated to dryness using water bath and stored below ambient temperature.
ii) 150g fresh ginger was soaked in 100ml of 95% ethanol for 24 hours to obtain the extract which was evaporated to dryness using water bath and stored as in (i) above
iii) 150g of fresh ginger was blended and the raw juice was extracted after standing in a clean glass container for 24 hours, it was extracted using sterile muslin cloth and the extract was evaporated to dryness using water bath stored as in (i) above.

Preparation of McFarland Standard

0.5 McFarland equivalent turbidity standard was prepared by adding 0.6ml of 1% Barium chloride dehydrate solution ($B_aCl_z2H_2O$) to 99.4ml of 1% sulphuric acid solution (H_2SO_4). A small volume of the turbid solution was transferred to capped tube of the same type that was used to prepare the test and of control inocula. This was stored in the dark at room temperature.

Preparation of Inoculum

Inoculums were prepared by direct colony suspension where a small volume of sterile was poured into a test tube to which general colonies of the test organisms taken directly from the plate, was emulsified and the suspension adjusted to match the 0.5 McFarland standard which has similar appearance of an overnight broth culture by adding distilled water [1α 4].

Antimicrobial Screening Test

The sensitivity of the test organisms to the extract of *Zingiber officinale* (ginger) was carried out using the agar and test tube diffusion method. 0.02ml of the suspension was added to an already prepared medium using a glan dropper and a sterile loop was used to spread by streaking the organisms all over the surface of the medium and allow to dry for 5 minutes. Cups of 6mm in diameter was made in the agar using sterile cork borer.

Different Dilutions of the Plant Extracts

Different dilutions of the plant extracts in the order of 20mg/ml, 5mg/ml and 1.25mg/ml were prepared respectively in five different test tubes and place in a test tube rack. About 0.3mg/litre of erythromycin was also prepared alongside which serves are a positive control and distilled water as a negative control. Each concentration was introduced into each hole on the medium, as well with disc soak with the various concentrations of *Zingiber officinale* extract on the medium and left to stand on the bench for 1 hour for proper diffusion. It was incubated at 37^{o}C for 24 hours.

RESULTS AND DISCUSSION

The results of the antimicrobial properties of the extracts on the test organisms are shown in tables below:

Table 1: Sensitivity pattern of Escherichia coli and Pseudomonas aeruginosa to raw *Zingiber officinale* extract

Concentration (mg/ml)	Zone of Inhibition Diameter (mm)		Positive control (Erythromycin)	Negative control (Distilled water)
	E. coli	*P. aeruginosa*		
20	0.0	3	24	0.0
10	0.0	5	24	0.0
5.0	0.0	7	24	0.0
2.5	0.0	0.0	24	0.0
1.25	0.0	0.0	0.0	0.0

NB: *Values are means of duplicate readings*

The result of this work indicates that the extracts of *Zingiber officinale* have antimicrobial properties when the extracts were tested on *P. aeruginosa* and E. coli. The widest zone of inhibition was obtained with *P. aeruginosa* while E. coli showed a little zone of inhibition. The difference in the zone of inhibition may be directly related to the susceptibility of each test

organism to the ginger extracts. The factors responsible for this high susceptibitity of *P. aeruginosa* to the extract may be attributed to the presence of secondary plant metabolites. Also the positive control in each of the test organism was sensitive, given a value of 24mm for E. coli and 20mm for P. aeruginosa in the raw extract of ginger, 24mm for *E. coli* and 26mm for *P. aeruginosa* in the ethanolic extract of ginger and 24mm for *E. coli* and 20mm for *P. aeruginosa* in the cold water extract of ginger. 0.5 McFarland given an equivalent approximate density of bacteria 1 x 10 – 8 cfu 28 [(1).]

Table 2: Sensitivity pattern of Escherichia coli and Pseudomonas aeruginosa to cold water extract *Zingiber officinale*

Concentration (mg/ml)	Zone of Inhibition Diameter (mm)		Positive control (Erythromycin)	Negative control (Distilled water)
	E. coli	***P. aeruginosa***		
20	12	14	26	0.0
10	10	12	24	0.0
5.0	8	9	24	0.0
2.5	5	3	24	0.0
1.25	2	2	20	0.0

NB: *Values are means of duplicate readings*

Table 3:Sensitivity pattern of Escherichia coli and Pseudomonas aeruginosa to ethanolic extract *Zingiber officinale*

Concentration (mg/ml)	Zone of Inhibition Diameter (mm)		Positive control (Erythromycin)	Negative control (Distilled water)
	E. coli	***P. aeruginosa***		
20	13	15	26	0.0
10	11	11	26	0.0
5.0	8	9	24	0.0
2.5	0.0	5	24	0.0
1.25	0.0	3	0.0	0.0

NB: *Values are means of duplicate readings.*

It became clear in this work that the solvent of extraction affected the degree of antimicrobial activity of the extracts. It was observed that the ethanolic extract of *Zingiber officinale* gave the widest zone of inhibition (15mm) and P. aeruginosa gave a zone of inhibition (13mm) using the concentration of 20mg/ml each. This credit of ethanolic extraction is due to the fact that ethanol is an organic solvent that will dissolve organic compound better (like dissolves like) hence librates the active component required for antimicrobial activity. It was observed that raw extract had activity only on P. aeruginosa and no effect was observed on E. coli. The non inhibition of growth in E. coli may be due to non liberation of the active constituents of the raw extract. The cold water extract inhibited the growth in both organism at all concentration. This is due to water ability to liberate the active constituent of the plant.

It is note worthy that the antibacterial activities of these plant is depended on the concentration of the extract.

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