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Phytochemical screening and antibacterial properties of *Garcinia kola*

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Abstract

Extract of bark, seed and root of *Garcinia kola* were used to determine the antibacterial properties of the extract as well as determine which extract is most effective. Methanol and ethanol were used for the extraction. Minimum inhibitory concentration (MIC; mg/ml) of the extract as well as zones of inhibition of the extract were determined. Results show that the extracts from bark of *Garcinia kola* contain higher % Saponins (3.2±0.04), while the root contain higher % of phenol (0.44±0.11). Bark extract of Garcinia kola was more effective in inhibiting the growth of *Pseudomonas auraginosa*, *Esherichia coli* and *Staphylococcus aureus* when compared with the extract from the root and seed. Results also show that inhibition of the test extract in (mm) was higher in extract from the bark, followed by the seed and the least zone of inhibition was found in the root extracts. The variation in the antibacterial activities is presumed to be due to different active compounds present in the extract.

Keywords: *Garcinia kola*, Minimal Inhibitory Concentration (MIC), Zones of Inhibition, Phytochemical Composition, Isolates, Therapeutic.

Introduction

The medicinal values of plants lie in the chemical substances presents in the parts of the plant such as seed, leaves bark and root. These substances produce definite physiological action in the human body.

Despite modern development in the treatment of diseases, herbal remedies have been continuous and universal. Modern medicines have always depended on herbal extracts from plants as fundamental source of therapeutic ingredients. Some naturally occurring substances in plants play significant role in plant disease resistance and thus most bacteria are sensitive to extract from these plants. Plants are pools of potential antimicrobial compounds for pharmaceutical need. The array of active compounds derived from them have impressive pharmaceutical properties such as analgesics, aesthetic, antibiotics, anti–parasitic, anti-inflammatory, oral contraceptive, hormones, ulcer therapeutic laxative. Seeds, herbs, vegetables, bark, roots accumulate in their cells a great variety of Phytochemical compounds such as alkaloids, tannins, saponins, phenolic compounds.[1]

Garcinia kola is highly valued because of its medicinal use as the stem, root and bark serve as raw material for pharmaceutical properties.[1, 2] *Garcinia kola* is popular in south eastern Nigeria as it is extensively used in herbal medicine.[3] This study aimed at determining the antibacterial properties of extracts of G. kola in south eastern

nigeria is therefore propriety.

Materials and Methods

Sample Preparation

The sample used in the research work was obtained from Umuokiri Village in Umunumo Ehime Mbano Local Government Area, Imo State, while the laboratory used for the practical was the central laboratory service unit of Imo State Teaching Hospital Orlu, Imo State.

The seed, bark and root of *Garcinia kola* were used for the analysis. The seeds of *Garcinia kola* were removed from the seed coat and placed into a neatly washed and dried tray. The seeds were cut into bits for fast drying. The seeds were sun dried and crushed to coarse powder using a neatly washed local mortar and pestle.

The bark of the *Garcinia kola* was thoroughly washed to remove the sand, after which it was cut into bits to aid fast drying. The same procedure was also carried out for the root. The powdered form of the seed, root and bark of *Garcinia kola* were placed in different containers and were properly labelled.

Clinical pure isolates of *Staphlococcus aureus*, *Echerichia coli*, and *Pseudomonas auroginosa* were subculture to obtain pure isolates and identified on the basis of the colonial reactions and biochemical characteristics.

Extraction

Absolute ethanol 90% and methanol were used as solvent, for extraction of active compounds in the plant materials. 15g of processed plant material (*Garcinia kola*) was soaked in 100 ml of 90% absolute ethanol in 250 ml flask. The same was done with methanol. This was shaken vigorously and allowed to stand for 48 hours to effect proper extraction of active ingredient. The suspension was filtered with Whitman's filter paper to obtain the supernatant while the debris was discarded. The extract after distillation was analysed according to Harbone.[4]

Standard filter paper was used to prepare 2 mm paper disc with the aid of perforating machine. These discs were thereafter sterilized by autoclaving at 121^0C for 15minutes and later interpregnated with the plant extract under aseptic conditions.

The antibacterial activity of the different extracts was determined against test organisms using agar diffusion method. 0.2 ml of a 24 hours subculture of each test organisms was uniformly spread over the surface of a sterile nutrient agar and allowed to dry. The bacteria isolated were first sub-cultured in a nutrient agar. Serial dilution of the plant extracts was made. Number 4 filter papers were perforated with the aid of paper perforators. These filter papers now in the form of disc perforated out were impregnated into the serial dilutions of the plant extracts with various concentrations of 1.0, 0.8, 0.6, 0.4, 0.2 ug/dl.

The plant extracts were tested at various concentrations. With the aid of wire loop, the isolates of the bacteria were aseptically streaked in a nutrient agar media. The prepared different concentrations of the plant extracts with the aid of a forceps were placed on the surface of the streaked nutrient agar media. The agar plates were randomly placed in the oven incubated at 37^oC for 24 hours.

The impregnated nutrient agar media were placed in an incubator at 37^oC for 24 hours. The experiments were done in triplicates. The zones of inhibition were measured in mm diameter and recorded.

Results

The Phytochemical compositions of *Garcinia kola* extracts of the bark, seed and root are shown in table 1. Result shows that the bark, seed and root sample of *Garcinia kola* contains Phytochemical of interest, such as alkaloids, tannins, flavonoids and Saponins in different concentration. The bark extracts containing higher quantity of Saponins, flavonoids, tannin, alkaloid and phenol. The table also shows that the active compound tannins, Saponins and alkaloid were more abundant in the extract of methanol than the ethanol.

Table 1: Phytochemical Composition

Sample	Solvent	Tannin	Flavonoids	Saponins	Alkaloid	HCl	Phenol
Seed	Ethanol	+	+	++	+	+++	+
	Methanol	+++	+	++	+	+++	++
Bark	Ethanol	++	++	++	++	+	+
	Methanol	+++	++	+++	+++	+	+
Root	Ethanol	++	+	+	+	++	+
	Methanol	+++	+	++	+	++	++

+ = presence of photochemical
++ = strong presence of photochemical
+++ = abundant presence of photochemical

Table 2 shows the % active compound in the extract of ethanol and methanol. The result show that the active compounds extracted was higher in the extract of bark than those of the seed and root. The bark produced highest active compound in Saponins (3.2%) than those of the seed and root (1.76 and 1.54 respectively).

Table 2: Quantitative Photochemical Components in %

Sample	Solvent	Saponins	Flavonoids	Tannin	Alkaloids	Phenol
Bark	Ethanol	2.70±0.03	0.47±0.01	0.72±0.01	1.03±0.72	0.13±0.007
	Methanol	3.20±0.04	0.77±0.03	1.02±0.03	1.54±0.81	0.35±0.010
Seed	Ethanol	1.32±0.02	0.22±0.02	0.44±0.01	0.03±0.02	0.14±0.007
	Methanol	1.76±0.03	0.34±0.04	0.98±0.02	0.07±0.02	0.35±0.009
Root	Ethanol	1.013±0.03	0.17±0.01	0.03±0.02	0.025±0.01	0.16±0.07
	Methanol	1.54±0.4	0.78±0.02	0.06±0.03	0.077±0.02	0.44±0.11

Table 3 shows the minimal inhibitory concentration and the zones of inhibition of the isolates. Results show that *Pseudomonas auroginosa* was more sensitive to the bark extract than the extract of seed and root.

Table 3: Minimal Inhibitory Concentration (MIC) and Zones of Inhibition of the Extracts of *G. kola*

		Seeds		Bark		Root	
Bacteria	**Solvent**	**Minimal Inhibitory Concentration**	**Zones of Inhibition**	**Minimal Inhibitory Concentration**	**Zones of Inhibition**	**Minimal Inhibitory Concentration**	**Zones of Inhibition**
Staphylococcus aureus	Ethanol	0.02	19.5	0.03	20.5	0.01	16.0
	Methanol	0.06	21.5	0.07	21.50	0.07	17.5
Esherichia coli	Ethanol	0.05	15.0	0.08	23.5	0.03	18.0
	Methanol	0.08	17.5	0.11	25.0	0.08	20.50
Pseudomonas auraginosa	Ethanol	0.09	29.0	0.50	40.0	0.05	34.0
	Methanol	0.11	30.25	0.90	42.50	0.09	35.0

Discussion

Results show that methanol and ethanol used as solvents were effective in extracting the active compounds with methanol being more effective in extracting more active compounds as shown in table 2 particularly Saponins (3.20±0.04) and alkaloid (1.54±0.81). This could be because of high volatile property of methanol. Table 3 shows the zones of inhibition of the extract on the test organisms. Extracts from bark generally had higher zones of inhibition on all the test isolates, although it was more effective on *Pseudomonas auraginosa* (42.5 mm) than other isolates.

The antibacterial activity of the extracts can be attributed to the synergistic action of some bio reactive substances such as the alkaloids, tannins, Saponins, flavonoids among others in the extracts.[1] Many higher plants are known to posses antibacterial agents and indeed extracts of plants from different parts of the world have been known to produce antimicrobial properties as observed in this work.[1]

Results of Phytochemical analysis revealed the predominance of tannins and scanty presence of alkaloids, Saponins, flavonoids in both ethanol and methanolic extracts of *Garcinia kola*. Also (table 3) shows the zones of inhibition of the test organism by extracts of the bark, seed and the root of *Garcinia kola*. It was observed that the inhibition of the tests extract in millimetre was higher in the bark, followed by the seed and the least zone of inhibition was found in the root extracts. This show that *G. kola* can be extensively used in treatment of bacteria disease, particularly with the bark extract.[2]

It was also shown that the methanolic and ethanolic extracts of *Garcinia kola* inhibited the growth of gram positive and the gram negative organism. Although extract produced with methanol exhibited more inhibitory effect when compared with ethanol, this could be because of high active compounds present in the extract. The result in this work shows that there is variation in the degree of antibacterial activities of the extracts. The variation in the antibacterial activities is presumed to be due to difference in the quantity of compounds present in those plant extracts.

Generally, methanol extracted more active ingredients than ethanol. This may be due to higher volatility of methanol as the highest extraction was observed in the methanolic extraction of Saponins (3.2%).

The results obtained on bacteria test isolates indicate that the lethal effect was greater on *P. auroginosa* (45.6 mm). Attempt has been made to show that the crude nature of this extracts as well as the age long problem of dosage is obstacle in the general acceptance of this plant extract. The presence of impurity, which may negatively modify the effect of this extracts may ultimately lead to loss of therapeutic value. Although results have revealed the potentials of this medicinal plant as having antibacterial effect, a lot need to be done before they can be accepted by a greater majority of the populace.

Conclusion

The observed inhibitory effect of extract of *G. kola* explained their utilization in traditional medicine.[1, 2] High phenolic compounds in the bark extract and seeds are an indication of the plant effectiveness in being used as anti – bacterial agent, as it is used in disinfections. The high Saponins content in G. kola explains its high antibacterial effects. The zones of inhibition therefore indicate that G. kola has potent antiseptic or antibacterial properties.[1] Findings from this work support the use of extracts from *G. kola* in treating wound as it prevents the wound from being septic.

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