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Antibacterial activity and phytochemical screening of Chrysophyllum albidum leaves

Duyilemi O.P* and Lawal I.O.

Forestry Research Institute of Nigeria, P.M.B 5054, Ibadan, Oyo State, Nigeria.

*Author to whom correspondence should be addressed, email: bumexd@yahoo.com

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Abstract

The aim of this study was to evaluate the antibacterial activity of water and methanolic extracts from *Chrysophyllum albidum* leaves. The crude extracts were screened against *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhimurium* and *Shigella* spp. at different concentrations (125 µg/mL, 250 µg/mL and 500 µg/mL) using the agar well diffusion technique. The methanolic extracts had stronger inhibitory effects on test organisms than the water extracts. The antimicrobial activity observed with the water extract ranged between 10 (±0.19) and 27 (±0.25) mm with no detectable activity at 125 µg/mL and 250 µg/mL on *Escherichia coli*. Stronger antimicrobial activity was observed with the methanolic crude extracts at all concentrations with all test organisms. The antimicrobial activity ranged between 25 (±0) and 35 (±0.24) mm. Preliminary phytochemical screening showed that these extracts contained anthraquinone, tannin and cardiac glycoside and with no traces of reducing sugars, saponin and alkaloids,. The result of this study provides the scientific basis for developing a novel broad spectrum of antimicrobial herbal formulation.

Keywords : *Chrysophyllum albidum*, antibacterial activity, phytochemical analysis

Introduction

African star apple (*Chrysophyllum albidum* G. Don) is a tropical edible fruit tree. It belongs to the family of Sapotaceae which has up to 800 species and make up almost half of the order (Ehiagbonare et al., 2008). It is primarily a forest tree species and its natural occurrences have

been reported in diverse ecozones in Nigeria, Uganda, Niger Republic, Cameroon and Cote d'Ivoire (Bada, 1997). The plant often grows to a height of 36 though it may be smaller (Keay, 1989). The African star apple fruit is a large berry containing 4 to 5 flattered seeds or sometimes fewer due to seed abortion. The plant has in recent times become a crop of commercial value in Nigeria (Oboh et al., 2009). The fleshy pulp of the fruits is eaten especially as snack and its fruit has been found to have higher contents of ascorbic acid than oranges and guava (Amusa et al., 2003). It is also reported as an excellent source of vitamins, irons, flavours to diets (Adisa, 2000). The seeds are also used for local games or discarded (Bada, 1997). *C. albidum* fruit is common in both urban and rural centres especially during the months of December to April. The fruits are not usually harvested from the trees, but left to drop naturally to the forest floor where they are picked up (Amusa et al., 2003).

The roots and leaves of *C. albidum* have been widely used for medicinal purposes (Adewusi, 1997). In addition, its seeds are a source of oil, which is used for diverse purposes.(Ugbogu and Akukwe, 2008) The fruits also contain 90% anacardic acid, which is used industrially in protecting wood and as source of resin, while several other components of the tree including the roots and leaves are used as a remedy for yellow fever and malaria. The leaves are used as emollients and for the treatment of skin eruptions, diarrhea and stomacheache, which are as a result of infections and inflammatory reactions (Adisa, 2000).

The people of south western Nigeria have been using *C. albidum* leaves for the management of infections / ailments since prehistoric times, although scientific evidence for its antimicrobial effect is still lacking. Studies have been carried out on seeds of *C. albidum*; for example seed storage and its food value, physical properties of the seed, use of the shell of seeds for the removal of metal ions and antimicrobial effect of oil from its seeds against some local clinical bacteria isolates (Amusa et al., 2003; Oyelade et al.,2005; Ugbogu and Akukwe, 2008; Oboh et al., 2009). Studies have also examined its antinociceptive, anti-inflammatory and antioxidant activities of eleagnine: an alkaloid isolated from the seed cotyledons (Idowu et al., 2006).

Relatively little information is available on the antimicrobial and phytochemical screening of *C. albidum* leaves. This study investigates the antimicrobial and phytochemical screening of *Chrysopyllum albidum* leaves using water and methanolic extracts against local clinical bacteria isolates.

.Materials and Methods

Fresh leaves of *Chrysopyllum albidum* were collected in Ibadan, Oyo State, Nigeria. The botanical identification of the plant was done at the Department of Taxonomy of Forestry Research Institute of Nigeria (FRIN).

Extraction

The leaves were cleaned, oven dried at 70 °C and ground into powder. The dried powdered plant materials were extracted separately with methanol and sterile water using soxhlet apparatus for 48 hrs. The extract was stored in the refrigerator until required for testing.

Phytochemical analysis

Chemical tests were carried out on the extracts using standard procedures to identify the constituents as described by Sofowora (1993) and Trease and Evans (2002) .

Microbial strains and growth

The species of microorganisms; *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhi* and *Shigella* spp. were used in this study. They were maintained on agar slant at 4 °C in the Laboratory of Microbial Physiology and Biochemistry of the Department of Botany and Microbiology, University of Ibadan. These strains were sub - cultured on a fresh appropriate agar plate 24 hrs prior to any antimicrobial test. The identity of the tested microbial species was confirmed before use by culturing on the specific media followed by biochemical characterization.

Antimicrobial assay activity

The *in vivo* antimicrobial activity was performed by agar well diffusion method technique. The antimicrobial diffusion test was carried out as described by Okeke et al., 2009 using a cell suspension of about 3.0 x 10^8 cfu/mL obtained from a McFarland turbidity standard no 0.5. Antimicrobial activity was evaluated by measuring the diameter of inhibition zone around the extract. The assay was repeated four times the results recorded as mean ± SD of the experiment(s).

Results and Discussion

Preliminary phytochemical screening of *Chrysophyllum albidum* leaves for secondary metabolites showed the presence of tannin, anthraquinone and cardiac glycosides in the plant samples. The samples were devoid of saponin, alkaloids and reducing sugars (Table 1). Further the phytochemical screening was also shown posses medicinal activity as well as exhibiting physiological activity (Sofowora, 1993) and shows common components in medicinal plants. Biological actions are primarily due to these components in a very complicated concert of synergistic or antagonistic activities. Mixtures of such chemicals show a broad spectrum of biological effects and pharmacological properties (Felix, 1982; Kubmarawa et al., 2008). The antimicrobial properties of the plant observed in this study could be attributed to the presence of tannin, anthraquinone and cardiac glycosides. Table 2 shows the antimicrobial activity of water extract of *Chrysophyllum albidum*. The results showed that the test organisms were susceptible to the water extracts based on their zones of inhibition which ranged from 10 (±0.19) to 27 (±0.25) mm. *Escherichia coli* showed the highest susceptibility of 27 (±0.25). All the organisms were sensitive at all concentrations except *Escherichia coli* which was not sensitive at 125 µg/mL and 250 µg/mL. The least activity 10 (±0.19) mm was detected at 125 µg/mL concentration with. The methanolic extract of *Chrysopyllum albidum* leaves (Table 3) exhibited some antimicrobial activity against all the microorganisms tested as assessed by zones of inhibition that ranged from (25±0 to 35±0.40mm). *Escherichia coli* had the highest susceptibility at 250 µg/mL concentrations with zone of inhibition of 35±0.40 mm. There was increase in the zone of inhibition for *Escherichia coli* from 30 (±0.15) mm at 125 µg/mL to 35 (±0.40) mm and 35±0.21 at 250 µg/mg and 500 µg/mL respectively. *Salmonella typhi* and *Shigella* spp had a zone of Inhibition of 35 mm at all concentrations except at 500 µg/mg where *Shigella* spp. which had zone of inhibition of 30 (±0.30) mm.

Table 1: Preliminary phytochemical analysis of the aqueous extract of *Chrysophyllum albidum* leaves.

Saponin	-
Sapofenin	++
Reducing sugar	-
Tannin	++
Alkaloids	-
Anthraquinone	++
Cardiac glycoside	+

Key: (+++) Highly concentrated (++) concentrated (+) Trace (-) Absent.

Table 2: Antimicrobial activity of water extract of *Chrysopyllum albidum* leaves.

Organisms/concentration	**125 µg/mL**	**250 µg/mL**	**500 µg/mL**
Staphylococcus aureus	10±0.19	12±0.25	27±0.25
Escherichia coli	-	-	25±0.15
Salmonella typhi	15±0.21	20±0.19	20±0.70
Shigella spp	12±0.30	15±0.47	20±0.10

Values are mean of three replicates ± standard error

Table 3: Antimicrobial activity of methanolic extract of *Chrysopyllum albidum* leaves.

Organisms/concentration	**125 µg/mL**	**250 µg/mL**	**500 µg/mL**
Staphylococcus aureus	30±0.15	35±0.12	25±0.00
Escherichia coli	30±0.15	35±0.40	35±0.21
Salmonella typhi	35±0.24	35±0.10	35±0.00
Shigella spp	35±0.10	35±0.00	30±0.30

Values are mean of three replicates± standard error

These results suggest that, water and methanolic extracts from the leaves of *Chrysophylum albidum* may contain active agent(s). This provides the basis for their folkloric use as cure for some human ailments like skin infection, diarrhea and stomachache which are as a result of infections and inflammatory reactions (Adisa, 2000). It is suggested that more research be conducted to further isolate, identify, characterize and elucidate the bioactive compounds from *C. albidum*

Conclusions

In conclusion, the result of this study justifies the traditional uses of the leaves of *C. albidum* for therapeutic purposes. The findings could also be of commercial interest to both pharmaceutical companies and research institute in the production of new drugs

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