

Short Communication

Preliminary studies on the antimicrobial properties of *Buchholzia coriacea* (wonderful kola)

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In this work, the antimicrobial properties of fresh *Buchholzia Coriacea* (wonderful kola) and its extracts was investigated. The proximate composition of the fresh kola was also determined. *B. coriacea* was dried, milled and extracted using two different solvents of varying polarity: hexane and methanol. The effect of the fresh kola, hexane and methanol extracts was tested on some food borne pathogens. This was evaluated by measuring the zone of inhibition on nutrient agar for bacteria and malt extract agar for fungi. The food borne pathogens used in this study are *Esherichia coli*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Trichoderma viride* and *Aspergillus niger*. The proximate composition showed that the fresh kola consists of 53.13% moisture, 9.8% ash, 3.46% crude fibre, 2.20% fat, 13.22% crude protein and 28.19% carbohydrate. The fresh kola showed inhibitory zones with the test bacteria: *E. coli* (62 mm), *E. faecalis* (40 mm) and *S. aureus* (50 mm). The growth of the two test fungi *T. viride* and *A. niger* was completely inhibited. The hexane extract showed inhibitory zones ranging from 20 to 40 mm with the test bacteria: *E.coli* (21 mm), *E. faecalis* (20 mm) and *S. aureus* (40 mm). It however showed no inhibitory effect on *T. viride* and *A. niger*. The methanolic extract of *B. coriacea* also showed inhibitory zones ranging from 20 to 30 mm with some of the test pathogens: *E.coli* (30 mm), *E. faecalis* (25 mm) and *S. aureus* (20 mm), *T. viride* (15 mm). It however showed no inhibitory effect on *A. niger*.

Key words: Antimicrobial, *Buchholzia coriacea*, pathogens, hexane extracts, methanol extracts.

INTRODUCTION

Food preservation to ensure food safety continues to be a challenge for the food industry and regulatory agencies. Among the strategies used to achieve food preservation by inhibiting growth of undesirable microorganisms is the use of chemical agents exhibiting antimicrobial activity. These chemicals may be either synthetic compound intentionally added to foods or naturally occurring biologically-derived substances. However, consumer perception that use of industrially synthesized food antimicrobials may be associated with potential toxicological problems has generated interest in the food industry for the utilization of naturally occurring compounds. To meet this demand is a difficult challenge for the food industry; thus recent trends in food preservation are oriented in that direction. Most food preservation techniques like dehydration, freezing and thermal treatment can success-

fully inhibit or destroy pathogens and spoilage microorganisms but are not so good as to keep overall food quality. For this reason, the food industry is exploring the use of natural antimicrobial substances to replace "chemical" preservatives in order to achieve a higher degree of retention of overall food quality. There is also a growing interest among food technologists in Nigeria to conduct research on the nutritional, medicinal and industrial uses of less studied and largely indigenous plants. Unlike exotic fruits and seeds, literatures on the purely indigenous species are scarce. An example of such an indigenous plant is *Buchholzia coriacea* popularly known as wonderful kola.

B. coriacea was named after R. W Buchholz who collected plants in Cameroon in the late 19th century (Keay et al., 1989). It belongs to the Capparaceae family. The seed of *B. coriacea* has medicinal values. These seeds gave the plants its common name of "wonderful kola" because of its usage in traditional medicine. The seeds are covered in a purple aril which is chewed in Ivory Coast and has a sharp pungent taste. Burkill (1985)

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carried out a research on the medical uses of this plant in different parts of Africa. It has been used for years to meet a variety of illnesses; since it has been used continually over many generations it is likely that wonderful kola actually has an effect against illnesses. As a result of its supposed broad-spectrum activity, there is need to conduct studies on potential utilization of wonderful kola in foods.

The objectives of this study are to determine the proximate composition of *B. coriacea* and evaluate the effect of the fresh kola and its extracts on some food borne pathogens.

MATERIALS AND METHODS

B. coriacea (wonderful kola)

Fresh *B. coriacea* (wonderful kola) was obtained from Bodija market in Ibadan and was identified by Dr. (Mrs) A. Akinyele at the Department of Forestry Management University of Ibadan, Ibadan, Nigeria. The fresh wonderful kolanuts were cleaned by the double disinfection method. They were washed thoroughly with distilled water to remove adhering particles after which they were soaked in 80% ethanol for 30 min. They were rinsed with distilled water and then washed with aqueous sodium hypochlorite (NaClO₄) to reduce surface contamination. This was followed by rinsing with distilled water. The kolanuts were diced to facilitate drying in an air-draught oven at 60°C for 72 h. The dried kolanuts were pulverized using a hammer mill. The powder was stored in polyethylene bags to prevent moisture absorption and contamination.

Hexane and methane extracts from *B. coriacea* powder were obtained using soxhlet oil extraction method (A.O.A.C., 1990). The percentage yield of the extracts was calculated as: Total yield (%) = (weight of extracts/original weight of sample) x 100.

Microorganisms

The microorganisms used were obtained from Department of Microbiology. The bacteria include *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, *Shigella dysenteriae* and *Klebsiella aerogenes*. These were maintained on nutrient agar slants and stored at 4°C. The fungi used were *Aspergillus niger* and *Trichoderma viride*. These were maintained on malt extract agar

Screening for antimicrobial activities

The process involves the use of test organisms to screen for the inhibitory properties of the extracts by measuring the diameters of slants and stored at 4°C. The organism was sub-cultured once every three months inhibition zones. Control experiment was set up the same way but without the addition of the fresh kola nor any of the extracts.

Determination of antibacterial activity of the extracts: Nutrient agar was poured into Petri dishes, allowed to set and bored with a Durham tube. Bacterial culture was used to inoculate each of the agar plates after which about 0.01 ml of the extract was added. Incubation was done at 37°C for 24 h after which the plates were inspected for zones of inhibition.

Determination of antifungal activity of the extracts: Nutrient agar was poured into Petri dishes, allowed to set and bored with a Durham tube. Fungal culture was used to inoculate each of the agar plates after which about 0.01 ml of the extract was added. Incubation was done at 28°C for 120 h after which the plates were

Table 1. Proximate composition of *Buchhlozia coriacea*.

Parameter	Composition (%)
Moisture content	53.13 ± 0.5
Crude protein	13.22 ± 0.4
Crude fat	2.20 ± 0.1
Crude fibre	3.46 ± 0.2
Carbohydrate	71.32 ± 0.4
Ash	9.8 ± 0.2

inspected for zones of inhibition.

Fresh wonderful kolanut: The above procedure was repeated using fresh wonderful kolanut of 10 mm thickness.

Proximate analysis of *B. coriacea*

Moisture content, crude protein, crude fat, ash, and carbohydrate were determined using (AOAC) 1990 method.

RESULTS AND DISCUSSION

Table 1 showed the proximate composition of fresh *B. coriacea*. Values obtained for ash, fat crude fibre, protein and moisture content of *B. coriacea* showed that its incorporation in foods as an additive can improve the nutritional composition of such foods.

The hexane extract appeared as a brownish viscous liquid with a yield of 1.3% while the methane extract appeared as a greenish yellow viscous liquid with a yield of 23.3%. Methanol, although not a recommended food solvent because of its toxicity, gave a higher yield than hexane due to its higher polarity. It has been observed that the more polar the solvent the higher the yield of extraction (Chang et al., 1977).

The hexane extracts of *B. coriacea* showed inhibitory zones ranging from 20–40 mm with three of the test bacteria (*E. coli*, *E. faecalis* and *S. aureus*). It had no inhibitory effect on *Klebsiella aerogenes*, *Shigella dysenteriae* and the two test fungi *T. viride* and *A. niger*. The methanol extract of *B. coriacea* showed inhibitory zones of 20-30 mm with three of the test bacteria (*E. coli*, *E. faecalis* and *S. aureus*). It showed no inhibitory effect on *K. aerogenes* and *S. dysenteriae*. It had an inhibitory effect of 15 mm on *T. viride* but had no inhibitory effect on *A. niger*. Zaika (1988) noted that extracting solvents could bring about variation in spice extractive components, which may influence their antimicrobial activities. The relatively smaller zone of inhibition obtained for the test fungi may be due to the multicellular and filamentous nature of the organisms (Plempel et al., 1987). *T. viride* resisted the hexane extract of *B. coriacea* but could not resist the methanol extract. Stem bark fractions of *B. coriacea* have been found to inhibit *S. aureus*, *E. coli*, *S. typhii*, *P. aeruginosa*, *Candida albicans* and *A. flavus* (Ajayeoba et al., 2003). The fresh kolanut exhibited greater inhibitory effect on the test organisms than the hexane

Table 2. Antimicrobial activities of fresh *Buchholzia coriacea* and its extracts.

Test organism	Mean diameters of inhibition zones (mm) \pm standard deviation ^a			
	Hexane extract	Methanol extract	Fresh wonderful kola	Control
Bacteria				
<i>E. coli</i>	21.0 \pm 0.5	30.0 \pm 1.0	50.0 \pm 0.5	+
<i>E. faecalis</i>	20.0 \pm 0.5	24.0 \pm 0.5	62.0 \pm 0.3	+
<i>S. aureus</i>	40.0 \pm 0.2	20.0 \pm 0.5	40.0 \pm 0.2	+
Fungi				
<i>T. viride</i>	+	+	-	+
<i>A. niger</i>	+	+	-	+

^aN = 3; - = complete inhibition; + = growth of microorganism.

and methanol extracts (Table 2). It showed inhibitory zones ranging from 40-62 mm with the three test bacteria (*E. coli*, *E. faecalis* and *S. aureus*) it was exposed to and it completely inhibited the growth of *T. viride* and *A. niger*. Hitokoto et al. (1980) similarly observed that powdered allspice completely inhibited the growth of three species of mycotoxigenic fungi; *A. flavus*, *Aspergillus versicolor* and *Aspergillus ochraceus* when incorporated into culture media for mycotoxin production. Growth of fungi has also been found to be inhibited by extracts of *Allium* species such as garlic and onion extracts (Moore and Atkins, 1977). Azzouz and Bullerman (1982) also reported that a 2% level of oregano in potato dextrose agar completely inhibited the growth of seven mycotoxigenic moulds.

The relatively poor inhibitory effect of the extracts of *B. coriacea* compared with the fresh wonderful kola could be attributed to the heat applied during drying (Savitri et al., 1986). The unit operations during the production of powder from the kola might have influenced their activity as some of the active ingredients may be volatile in nature (Desrosier, 1977).

Conclusion

The fresh kola was found to be more active on the test food borne pathogens than the hexane and methanol extracts. The lower inhibitory properties of the extracts confirms that over exposure to air, sunlight, too much artificial heat and rapid drying can cause a loss of essential oils. This study indicates clearly that *B. coriacea* possesses an invaluable but yet to be tapped potentials which, if exploited, will benefit the food industry. Column chromatography assay of the essential oils in *B. coriacea* can further be done in order to isolate and characterize their active components.

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