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(RESEARCH ARTICLE)



Preliminary phytochemical screening, antibacterial and antioxidant activities of *Azanza garckeana* (Fruits)

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Abstract

Azanza garckeana is a source of food, fodder for animals, fuel (fire wood), timber, medicine (roots are taken orally for pain relief, to treat cough and chest pains) and for shelter. The n-hexane, ethyl acetate and ethanol extracts were obtained by serial extraction using shaker apparatus. The results showed that; the phytochemical screening indicated the presence of alkaloids, cardiac glycosides, reducing compounds, flavonoids, saponin, Steroids and Triterpens, Tannins and phenols. The results of antibacterial recorded that there were significant antibacterial activity against all types of bacteria that used in this study. The result of the DPPH radical scavenging activity of the fruits extracts of *Azanza garckeana* showed that the n-hexane extract has high value 86 µg/ml, ethyl acetate extract value was 61 µg/ml and ethanol extract value was 42 µg/ml.

Keywords: *Azanza garckeana*; Malvaceae; Fruits; Phytochemical; Antibacterial; Antioxidant.

1. Introduction

Azanza garckeana is the species of the Malvaceae family, the generic name "Azanza" is derived from the word "Azania", which is meaning black and surviving in Zanzibar [1]. *Azanza garckeana* has been identified as one of the few plant species that should be integrated in the domestication process in farming systems in sub-Saharan Africa to support nutritional, medicinal and income security of local communities [2, 3, 4, 5]. Plants are potential sources of antimicrobial compounds and several researchers have investigated the antimicrobial activities of medicinal plants utilized in traditional or alternative healthcare systems [6,7]. The fruits of *A. garckeana* have potential in the development of new food and beverage products [8]. The species used as herbal medicine for diseases and ailments such as chest pains, cough, infertility, liver problems, menstruation problems and sexually transmitted infections.

This study aimed to determine the phytochemical screening, antibacterial and antioxidant activities of the fruits extract of *Azanza garckeana*.

2. Materials and methods

2.1. Collection of Plant sample

The plant sample (Fruits) of *Azanza garckeana* was collected from, South Kordofan State- Sudan. The dry fruits were obtained in the laboratory at room temperature and crashed then the extraction was carried out by using shaker extraction method with the following solvents: n-hexane, ethanol and ethyl acetate [9].

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2.2. Preparation of Plant Extracts

A weight of 150 gram of the coarsely powdered–dried sample was successively extracted with n-hexane by using orbital mixer (shaker) for 48 hours , Then the n-hexane extract was evaporated by transferring it from conical flask to plate ; then the residual powdered fruits was air dried and extracted again with ethyl acetate for 48 hours, the extract was evaporated by transferring it in to conical flask to plate ,Finally the air dried residual powdered fruits extract again with 70% ethanol for 48 hours and extract evaporated .Any solvent has different amount of extract from each other. Each evaporated extract was transferred from plates to bottles and stored in refrigerator.

2.3. Phytochemical screening

Qualitative chemical analysis for chemical constituents of *Azanza garckeana* fruits were determined using the method of [10].With some modifications.

2.4. Antibacterial activity

The extract of *Azanza garckeana* fruits at concentrations (100mg/ml, 50mg/ml, 25mg/ml, 12.5mg/ml), were subjected to antimicrobial tests by using cup plate agar diffusion method and inhibition zone were measured in (mm) against four bacterial strains. The range of inhibition was found 12-20mm.

2.4.1. Preparation of media

28 g of powdered nutrient agar was weighted, dispersed in 1 liter of distilled water and allowed to soak for 10 minutes, "swirl to mix then sterilized by autoclaving for 15 minute at 121c, cooled to 47 °C", mixed well then poured into Petri dishes.

2.4.2. Testing of bacteria organisms

One ml of the standardized bacterial stock suspension 10⁸-10⁹ C.F.U/ml was thoroughly mixed with 100ml of molten sterile nutrient agar which was maintained at 45 °C. 20ml aliquots of the inoculated nutrient agar were distributed into sterile Petri dishes. The agar was left to set and each of these plates ,4 cups (10mm in diameter) were cut using a sterile cork borer (No.5) and agar disk were removed .Alternate cups were filled with 0.1ml sample of each of the extract dilution in ethanol using automatic micro-liter pipette, and allowed to diffuse at room temperature for two hours. The plates were incubated in the upright position at 37 °C for 18 hours. Three replicates were carried out for each extract against each of the test organism. After incubation the diameters of the resultant growth inhibition zones were measured, averaged and mean values were tabulated.

2.5. Antioxidant Activity

2.5.1. DPPH (radical scavenging assay)

DPPH radical scavenging was determined according to the methods of [11].With some modification. In 96-wells plate the test samples were allowed to react with 2,2-Di (4-tert- octyl-phenyl)-1-picryl-hydrazyl stable free radical (DPPH) for half an hour at 37 °C. The concentration of DPPH was kept at (300 µM) the test sample were dissolved in DMSO while DPPH was prepared in ethanol after incubation , decrease in absorbance was measured at 517nm using multiplate reader spectrophotometer. Percentage radical scavenging activity by samples was determined in comparison with a DMSO treated control group. All tests and analysis were run in triplicate.

3. Results and discussion

Azanza garckeana have potential in the development of new food and beverage products [8]. The species used as herbal medicine for diseases and ailments such as chest pains, cough, infertility, liver problems, menstruation problems and sexually transmitted infections. Alkaloids are significant for the protecting and survival of plant because they ensure their survival against microorganisms [12]. Flavonoids constitute a wide range of substances that play important role in protecting biological systems against the harmful effects of oxidative processes on macromolecules, such as carbohydrates, proteins, lipids and DNA [13,14,15].

Phytochemical Screening of *Azanza garckeana* fruits consist of the following metabolites as indicated in Table (1). Saponins were tested positive in *Azanza garckeana* fruit extracts are responsible for numerous pharmacological properties in addition to antimicrobial activity, it inhibit mould, and protect plants from insect attack [16]. The results of phytochemical groups of *Azanza garckeana* fruit agree with [3, 17, 18, 19, 20].

Table 1 The Chemical Constituents of *Azanza garckeana* (Fruits).

Phytochemical	Results
Alkaloids	+
Cardiac glycosides	+
Reducing compounds	+
Phenols	+
Saponin	+
Tannins	+
Flavonoids	+
Steroids and Triterpens	+

The solvents extracts of *Azanza garckeana* showed significant antibacterial activity against all types of bacteria that used in this study, however it showed higher inhibition zone against *E coli* (20-18) mm in n-hexane extraction. The lower in habitation zone was considered against *Bacillus subtilis* and *pseudomonas aeruginosa* (12 mm) at n-hexane extraction. Generally, the fruits extracts showed good inhibition zone for all tested microbes, these results confirm the presence of antibacterial compounds in *Azanza garckeana* and it may be used to fulfill which is need for alternative antibiotic.

Table 2 Antibacterial activity of *Azanza garckeana* (Fruits).

Plant extraction	Standard bacterial strains					
	Concentration mg/ml	in	<i>E.c</i>	<i>P.s</i>	<i>S.a</i>	<i>B.s</i>
70% Ethanol	100		17	16	18	16
	50		16	16	18	15
	25		16	14	17	14
	12.5		14	13	16	13
Ethyl acetate	100		19	17	15	17
	50		18	17	14	17
	25		17	16	13	16
	12.5		15	15	12	15
n-hexane	100		20	19	15	15
	50		20	17	14	15
	25		19	16	14	14
	12.5		18	16	12	12

Key: B.s, *Bacillus subtilis*; S.a -*Staphylococcus aureus*; E.c - *Escherichia coli*; P.a - *Pseudomonas aeruginosa* .Concentration of extracts (100, 50, 25, 12.5mg/ml). Zone of inhibition in (mm), - no inhibition, <9mm inactive, 9-12 mm partially active, 13-18mm active, >18mm very active.

As indicated in table (3), the most potent activity on the DPPH scavenging activity was observed by n-hexane extract (86±0.04) as compared with standard Propyl gallate.

Table 3 The results of Antioxidant activity

NO	Extracts	% RSA \pm SD (DPPH)
1	Ethyl acetate	61 \pm 0.02
2	Ethanol	42 \pm 0.09
3	n-hexane	86 \pm 0.04
Standard	Propyl Gallate	91 \pm 0.01

4. Conclusion

Azanza garckeana is rich in a diverse variety of Phytochemicals, The various biological activities of this plant is due to the presence of a varied degree of active constituents, these constituents were supported the traditional uses of the extracts of this plant.

Compliance with ethical standards

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Disclosure of conflict of interest

Authors have no any conflict of interest.

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