

Alterations in Serum Urea, Creatinine and Electrolytes Concentrations in Wister Rats Following Repeated Administration of Methanol Extracts *Ofanza garckeana* Pulp

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Abstract

The effect of air-dry and sun-dry methanol extracts of *Azanza garckeana* pulp on serum urea, creatinine and electrolytes concentrations of Wister rats were evaluated. A total of thirty-five (35) rats were grouped into seven (7) of five rats each. Group A-C were given 150, 300 and 600 mg/kg bw air-dried methanol extract of *A. garckeana* pulp respectively, groups D-F were given 150, 300 and 600 mg/kg bw sun-dried methanol extract of *A. garckeana* pulp respectively while group G serve as the normal control. All treatments were administered orally twice a day (morning and evening) for a period of 2 weeks. Both extracts of *A. garckeana* pulp caused a dose dependent significant ($p < 0.05$) increase in serum sodium, potassium and, decreases urea and uric acid levels when compared with the control rats. However, the extracts did not cause any significant ($p > 0.05$) alterations to the levels of chloride and bicarbonate concentration when compared with the control group. The creatinine levels in rats treated with air-dry extract decreases ($p < 0.05$) while those treated with the sun-dry extract compared well ($p > 0.05$) with the control group. In conclusion, the extracts alter some functional integrity of kidney with more pronounced effect at higher dose, thus caution should be exercise when using *A. garckeana* for oral remedy.

Keywords: *Azanza garckeana*, urea, creatinine, electrolyte.

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INTRODUCTION

Plants are recognized as the most common form of alternative medicine [1]. In Africa the use of medicinal plants to alleviate specific ailments is in practice from ancient time's onwards [2]. Traditional medicine has maintained greater popularity all over developing world and the use for treatment of several diseases is rapidly on the increase [3-6]. The World Health Organization (WHO) has listed 21,000 plants, which are used for medicinal purposes around the world [7]. Despite the profound therapeutic advantages possessed by some of the medicinal plants, some constituents of medicinal plants have been found to be potentially toxic to multiple organs

The kidney is highly susceptible to toxicants because of its high volume of blood flow through it and

it filters large amounts of toxins which can concentrate in the kidney tubules [8]. The safety of herbal medicine use has recently been questioned due to reports of illness and fatalities particularly nephrotoxicity [9]. Although there are many traditional herbal medicines available, only a few have been verified by clinical trials, their efficacy and safety are still questioned by consumers.

Azanza garckeana also known as Goron Tura, the fruits are spherical and woody in nature of about 2.5-4cm in diameter. The plant is native to Tula village in Kaltungo Local Government Area of Gombe State and have been reportedly used for treatments of several ailments [10]. Pharmacological studies carried out with different extracts of *Azanza garckeana* have reported many effects which include anti-epileptic, sedative, and hypnotic actions, analgesic, antipyretic, anti-

inflammatory effects, antispasmodic, emmenagogue, antihelminthic, antiarrhythmic and antibacterial actions [11-13].

Azanza garckeana is widely consumed in Northern Nigeria owing to its fast action in boosting energy for sexual satisfaction. The plant is consumed without being mindful of the dose and potential toxic effect. No adequate scientific evidence exists on safety or toxicity of *Azanza garckeana* on organs particularly, the kidney. Therefore, the evaluation of the safety of *Azanza garckeana* intended to be used in humans and animals is of greatest importance. The present study therefore aimed at evaluating the effect of air-dry and sun-dry methanol extract of *Azanza garckeana* pulp on biomarkers of kidney integrity in wister rats

MATERIALS AND METHODS

Collection of Plant Materials

Azanza garckeana was collected in June 2019, from Tula, Village of Gombe State in Northern Nigeria, and it was identified by a botanist in the Department of Plant Biology, School of Life Science, Federal University of Technology Minna, Nigeria

Experimental Animals

A total of thirty five (35) adult Swiss albino albino rats (*Rattus norvegicus*) weighing 150.65 ± 5.89 g were obtained from Animal Holding Unit, School of Life Science, Federal University of Technology Minna, Nigeria. These animals were kept in the animal house, at a temperature of 20°C and a natural photoperiod cycle. The animals were housed in plastic cages (5 rats per cage) and had free access to standard commercial diet and tap water. The animal handling and experimentation were in concordance with the guidelines for laboratory animal use and care as contained in the European Convention on Animal Care Guidelines and Protocol.

Preparation and extractions of plant extracts

The whole nuts of *Azanza garckeana* were rinsed under clean running water and the shaft was removed. The pulp was separated into 2 portions, first portion was air-dried for two weeks while the second portion was sun-dried for 2 weeks. The dried materials were pulverized into coarse powder using a grinder mill. A 200 g of the plant material was extracted with 200 mL of methanol using Soxhlet apparatus, and the resulting extract was concentrated using rotary

evaporator. The resulting extract was placed in air-tight container and refrigerated until when required.

Experimental design for Sub-chronic Study

The thirty-five (35) albino rats were grouped into seven (7) of five rats each. All treatments were administered orally twice a day (morning and evening) for a period of 2 weeks. Group A-C were given 150, 300 and 600 mg/kg bw air-dried methanol extract of *A. garckeana* pulp respectively, groups D-F were given 150, 300 and 600 mg/kg bw sun-dried methanol extract of *A. garckeana* pulp respectively while group G serve as the normal control

Blood sample collection

On the fifteenth day and following an overnight fast, rats were anaesthetized under diethylether vapor and euthanized. The blood samples were collected by carotid puncture into sample bottles and left for fifteen minutes to clot, and then centrifuged at 3000 rpm for 15 minutes in order to obtain the serum [14]. The sera were stored in the refrigerator at -20°C for subsequent analysis.

Analysis of Biochemical Parameters

Assay kits used for creatinine and urea were products of Randox Laboratories, Co-Antrim, UK. All other kits used were product of Agape Diagnostic AGAPPE DIAGNOSTICS SWITZERLAND GmbH. The concentrations of serum urea and creatinine were determined using standard procedures [15-16], while the concentrations of sodium, potassium, and chloride ions were determined as described by Tietz [17].

Statistical analysis

All values were expressed as the Mean \pm SEM of three replicate analyses. The analysis was performed using SPSS statistical package for WINDOWS (version 16.0; SPSS Inc, Chicago). Results were subjected to ANOVA followed by Duncan Multiple range test and $p < 0.05$ were considered to be statistically significant

RESULTS

Serum Creatinine Concentration

The air-dry methanol extract of *A. garckeana* pulp caused a significant ($p < 0.05$) and dose-dependent decrease in serum creatinine concentrations when compared with the control rats. However, the creatinine concentrations of rats administered sun-dry extract were similar ($p > 0.05$) with the control group (Table-1).

Table-1: Effects of repeated administration of air-dried and sun-dried methanol extract of *A. garckeana* pulp on serum creatinine concentration in wister rats

	Sun-dry	Air-dry
150 mg/kg bw <i>A. garckeana</i>	5.89 \pm 0.24 ^a	5.04 \pm 0.04 ^c
300 mg/kg bw <i>A. garckeana</i>	5.89 \pm 0.15 ^a	4.73 \pm 0.06 ^b
600 mg/kg bw <i>A. garckeana</i>	4.84 \pm 0.06 ^a	3.70 \pm 0.03 ^a
Control	5.89 \pm 0.05 ^a	5.89 \pm 0.05 ^c

Data are Mean \pm SEM of triplicate determinations. Values along the same column with different superscripts alphabet are significantly different ($p < 0.05$).

Serum Urea and Uric acid Concentration

Both the air-dry and sun-dry methanol extract of *A. garckeana* pulp caused a dose dependent significant ($p < 0.05$) decrease in serum uric acid concentrations when compared with the control rats. However, urea concentrations increase significantly ($p < 0.05$) when compared with the control group.

Table-2: Effects of repeated administration of air-dry and sun-dry methanol extract of *A. garckeana* pulp on serum urea and uric acid concentration in wister rats

	Uric Acid (mg/dL)	Sun Dry	Air Dry	Urea(mg/dL)	Sun Dry	Air Dry
150 mg/kg bw <i>A. garckeana</i>	6.65 \pm 0.36 ^b	6.87 \pm 0.78 ^b		51.00 \pm 3.45 ^b	55.02 \pm 4.45 ^b	
300 mg/kg bw <i>A. garckeana</i>	5.40 \pm 0.67 ^{ab}	4.04 \pm 0.67 ^a		49.89 \pm 1.45 ^b	52.48 \pm 3.45 ^b	
600 mg/kg bw <i>A. garckeana</i>	4.49 \pm 0.62 ^a	4.03 \pm 0.43 ^a		50.37 \pm 4.35 ^b	50.31 \pm 4.56 ^b	
Control	7.80 \pm 0.75 ^c	7.80 \pm 0.75 ^c		45.38 \pm 2.34 ^a	45.38 \pm 2.34 ^a	

Data are Mean \pm SEM of triplicate determination. Values along the same column with different superscripts alphabet are significantly different ($p < 0.05$).

Serum Electrolyte

Both the air-dry and sun-dry methanol extract of *A. garckeana* pulp caused a dose dependent

significant ($p < 0.05$) increase in serum sodium, potassium concentrations when compared with the control rats. However, the extracts of *A. garckeana* pulp did not cause any significant ($p > 0.05$) alterations to the levels of chloride concentration when compared with the control group (Table-3).

Table-3: Effects of repeated administration of air dry and sundry methanol extract of *A. garckeana* pulp on serum sodium, potassium and chloride concentrations in wister rats

	Sodium (mg/dL)		Potassium (mg/dL)		Chloride (mg/dL)	
	Sun-dry	Air dry	Sun-dry	Air dry	Sun-dry	Air dry
150 mg/kg bw <i>A. garckeana</i>	181.36 \pm 6.56 ^b	170.42 \pm 4.56 ^b	5.26 \pm 0.35 ^a	6.40 \pm 0.43 ^{ab}	72.76 \pm 3.46 ^a	70.24 \pm 3.45 ^a
300 mg/kg bw <i>A. garckeana</i>	171.20 \pm 6.54 ^b	189.38 \pm 4.56 ^{bc}	7.89 \pm 0.42 ^b	8.10 \pm 0.17 ^b	78.31 \pm 5.43 ^a	75.38 \pm 5.43 ^a
600 mg/kg bw <i>A. garckeana</i>	180.21 \pm 5.44 ^b	190.20 \pm 3.72 ^c	9.10 \pm 0.56 ^c	12.30 \pm 0.35 ^c	70.82 \pm 5.35 ^a	70.08 \pm 4.34 ^a
Control	131.43 \pm 5.43 ^a	131.43 \pm 5.43 ^a	4.38 \pm 0.02 ^a	4.38 \pm 0.02 ^a	74.38 \pm 5.43 ^a	74.38 \pm 5.43 ^a

Data are Mean \pm SEM of triplicate determination. Values along the same column with different superscripts alphabet are significantly different ($p < 0.05$).

Bicarbonate

The air-dry and sun-dry methanol extract of *A. garckeana* pulp did not cause any significant ($p > 0.05$) alterations to the concentrations of bicarbonate when compared with the control group

Table-4: Effects of repeated administration of air dry and sundry methanol extract of *A. garckeana* pulp on serum bicarbonate concentration in wister rats

	Sun-dry	Air-dry
150 mg/kg bw <i>A. garckeana</i>	23.44 \pm 0.56 ^a	21.08 \pm 1.89 ^a
300 mg/kg bw <i>A. garckeana</i>	25.42 \pm 0.87 ^a	26.35 \pm 2.43 ^a
600 mg/kg bw <i>A. garckeana</i>	29.04 \pm 0.56 ^a	20.32 \pm 2.34 ^a
Control	25.35 \pm 2.23 ^a	25.35 \pm 2.23 ^a

Data are Mean \pm SEM of triplicate determination. Values along the same column with different superscripts alphabet are significantly different ($p < 0.05$).

DISCUSSION

The kidney is highly susceptible to toxicants because of a high volume of blood flow through it and it filters large amounts of toxins which can concentrate in the kidney tubules [18]. Nephrotoxicity can result in systemic toxicity causing decreased ability to excrete

body wastes, inability to maintain body fluids, electrolytes balance and decreased synthesis of essential hormones [19]. Therefore, the measurement of the levels of electrolytes, creatinine and urea plays important roles in determining the synthetic and excretory roles of the kidney [20, 21].

Creatinine is the catabolic products of creatinine phosphate which is used by the skeletal muscle. It is a metabolite of muscle – creatinine, whose amount in serum is proportional to the body's muscle mass. The amount of creatinine is usually constant, it is

easily excreted by the kidneys, and thus elevated levels indicate diminished renal function [22].

The significant ($p < 0.05$) and dose-dependent decrease in serum creatinine following the administration of the extracts indicates a compromised renal functional capacity. The extract might have either interfered with creatinine metabolism leading to decreased synthesis or might have compromised tubular excretion [23].

Urea is an end product of protein metabolism and its product reflects diet protein intake and protein catabolic rate [8]. It is a waste product that is left over from the breakdown of protein. Urea circulates in the blood until it is filtered out by the kidneys and excreted in the urine [2]. If the kidneys are not functioning properly, there will be excess urea levels in the bloodstream. Impaired balance of nitrogen coupled with lowered protein synthesis leads to increased concentration of urea and uric acid in serum [24] which indicates progressive renal damage. The significant alteration in serum urea and uric acid following the administration of the extracts may be due to decreased protein catabolism or renal dysfunction [25]. This therefore suggests that the continuous administration of the air-dry and sun-dry methanol extract of *A. garckeana* pulp could induce renal impairment.

The non-significant effects in the level of chloride and bicarbonate concentrations following administrations of extracts at all doses tested suggests that normal functioning of kidney tubules as regard to these electrolytes was preserved [24]. However, serum potassium and sodium concentrations in albino rats following repeated administration of air dry and sundry methanol extract of *A. garckeana* pulp significantly ($p < 0.05$) increase when compared with the control rats. These findings suggest that some aspects of tubular functioning as it relates to potassium and sodium have been compromised [26].

CONCLUSION

The repeated administration of both air dry and sun-dry methanol extract of *A. garckeana* pulp caused alterations to the normal levels of urea, uric acid, creatine and some electrolyte with more pronounced effect at higher dose, thus caution should be exercised when using *A. garckeana* for oral remedy

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