



Nutritional and Medicinal Potential of *Ziziphus mauritiana* Lam. Leaf and Fruit Extracts

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ABSTRACT

Ziziphus mauritiana (LAM.) is a small, semi deciduous, drought resistant, usually spiny shrub or tree up to 12 m tall. The leaves are generally underutilized but the fruits for various purposes. Screening of leaves and fruits was conducted in 2018 to determine its nutritional and medicinal potential. The study evaluated phytochemical, proximate, mineral and antibacterial activity of the plant extracts. Qualitative phytochemical screening indicated high saponins in leaves and fruits, moderate alkaloids in fruits, trace flavonoids and tannins in leaves and fruits with trace alkaloids in fruits. Quantitative results (mg/100g dry weight) indicated saponins content of 92.65 ± 6.20 and flavonoids (25.80 ± 1.73) are higher in leaves whereas alkaloids content is more in fruits. No significant difference ($P > 0.05$) in tannins' content of leaves (0.50 ± 0.03) and fruits (0.30 ± 0.02) but significantly higher alkaloids in fruits (3.40 ± 0.23) than leaves (2.20 ± 0.15). Proximate analysis revealed higher contents of Moisture (5.8 ± 0.01), Ash (8.2 ± 0.00), Crude Fibre (11.7 ± 0.001) and Carbohydrates (34.05 ± 0.00) in leaves, while fruits had significantly ($P < 0.05$) higher Crude protein (27.18 ± 0.00) and Lipids (6.65 ± 0.05). Mineral content's analysis showed significantly ($P < 0.05$) higher sodium (154.79 ± 10.50), potassium (389.08 ± 10.69), manganese (2.0 ± 0.14) and iron (1.21 ± 0.15) in leaves, whereas fruits had significantly ($P < 0.05$) higher phosphorus (585.90 ± 41.29), magnesium (7.0 ± 0.12) and zinc (15.52 ± 0.05). Antimicrobial screening indicated increase in zone of inhibition at higher concentrations of leaf and fruit extracts on the test organisms with greater effect in leaf than fruit extract. The observed nutritional and mineral constituents in both leaf and fruit of the plant indicate its potential for utilization in food and medicine.

Keywords: *Ziziphus mauritiana*, Leaf & Fruit extracts, Nutritional & Medicinal potentials, Sudan Savanna, North western Nigeria

INTRODUCTION

Ziziphus mauritiana, commonly known as Chinese apple or Indian plum, is mostly a shrub, sometimes a tree up to 20 m height. It is one of the species that is grown in dry areas and generally used for feeding of cattle, sheep, camel, goats and is believed to have some resistance against different types of pathogens (Gupta *et al.*, 2012). The plant is readily available to the majority of people in rural and urban areas. It is widely used for medicinal purposes especially its leaf and fruit. The leaves are useful in the treatment of liver diseases, asthma and fever, diarrhoea and as poultice on wounds or mixed with oil and rubbed on rheumatic areas. They are also helpful in liver troubles and fever (Zhao, 2012). The roots help in hair growth, reduce fever associated with measles and chicken pox. The bark makes perfect eye drop for curing inflammation while the seeds make medicines for liver, heart and digestive organs (Mojtaba *et al.*, 2016).

The fruit is eaten raw, pickled or used in beverages. It is quite nutritious and rich in vitamin C (Gupta *et al.*, 2012). The fruits which taste like a mixture of dates and apples may also be dried and kept for later use (Nyanga *et al.*, 2013). The leaves are readily eaten by camels, cattle and goats, and are considered nutritious. The flowers are rated as a minor source of nectar for honey bees, the honey obtained is light and of fair flavour (Orwa *et al.*, 2009). Despite its numerous health and nutritional benefits, eating of *Z. mauritiana* fruits has been regarded in some quotas as a practice limited mostly to poor rural communities and only a fewer number of people in the urban communities (Nyanga *et al.*, 2013).

Quite a number of research works have been carried out on nutritional composition and pharmacological properties of *Z. mauritiana* fruits (Egwim *et al.*, 2011). Little attention has been paid to leaf, root and bark extracts which constitute an important source of food and medicine (Abdallah *et al.*, 2016). The present study sought to investigate *Z. mauritiana* for its medicinal and nutritional composition of leaf extract in relation to the fruit. It is envisaged that additional utilization of the leaf component of the plant would be encouraged for its higher leaf proportion relative to fruit. This is required in charting the course for popularising the use of the plant among people in parts of the world where intake of fruits and vegetables is still low due to harsh economic realities, especially among women and children.

MATERIALS AND METHODS

Collection and Preparation of Plant Material

The matured fruits and leaf were collected from *Z. mauritiana* plant obtained at different locations in Sokoto, North western Nigeria. The samples were crushed mildly using pestle and mortar and then shade dried to constant weight. The samples were later crushed into powder, further dried for two days and then packaged in a clean well ventilated cupboard for subsequent analyses. Sample for antimicrobial studies was pulverised to small pieces using pestle and mortar and 100 g each of the leaf and fruit material was extracted with 95 % methanol. The mixture was kept at room temperature for 72 hours and filtered with Whatman filter paper No. 1. The filtrate was evaporated at 45^o C using rotary evaporator. The methanol residue of the leaf and fruit was each dissolved in 50 % aqueous methanol

Proximate Analysis

Ash content, crude fibre, crude lipids and moisture contents were determined using Soxhlet extraction method as described by Gupta *et al.* (2012). The crude protein was determined by Kjeldahl Distillation Technique (Abdallah *et al.*, 2016). Carbohydrates were determined by difference of 100 (that is, the sum of moisture, ash protein and lipids contents). The determinations were carried out in triplicates and the results were expressed as dried weight bases.

Determination of Mineral Ions

The method explained in Aguinaldo *et al.* (2005) was adopted. Mineral ions, sodium, magnesium, phosphorus, potassium, zinc, manganese and iron were determined by Atomic Absorption Spectrometry and sample solutions analysis was done using Buck Scientific Atomic Spectrophotometer VGP (variable giant pulse) system Model 210. The optical system was set up with a hollow cathode lamp for the elements with appropriate slit wavelength selected for the elements. The solution with known concentration of the analyte was aspirated and the absorbance reading noted.

Phytochemical Analysis

The phytochemical screening of aqueous extracts of the leaves and fruits were carried out according to the method of Trease and Evans (1989) and Sofowora (1993) to determine presence and proportions of alkaloids, flavonoids, tannins and saponins.

Antibacterial Studies

The methods of Ogbonna *et al.* (2003), Yusuf *et al.* (2005) and Aliero and Wara (2009) was used in the antibacterial assay. The bacterial cultures of Gram

positive bacteria (*Staphylococcus aureus* and *Streptococcus pyogenes*) and Gram negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*) obtained from Microbiology Department, Usmanu Danfodiyo University, Sokoto were pre-cultured in nutrient broth (NB) at 37^o C. The methanol leaf and fruit extracts were tested using agar well and disc diffusion methods. Concentrations of 50, 100 and 150 mg/ml were prepared by constituting with methanol. Streptomycin sulphate (100 mg/ml) was used as positive control and methanol solvent (100 mg/ml) as negative control. The diameter measurement (mm) of the inhibition zones was done using a transparent meter rule.

DATA ANALYSIS

Data obtained from the screening of leaf and fruit extracts and their antibacterial action were subjected to analysis of variance ANOVA) using SPSS Software version 21 (Terna *et al.*, 2016). Results were presented as mean \pm standard deviation of triplicate determinations. Statistical comparisons between the mean values were done by Mann-Whitney 2 pair sample T-test (Mukhtar and Aliyu, 2008). Values obtained at P < 0.05 were regarded as significant (Sokal and Rohlf, 1995).

RESULTS

Qualitative Phytochemical Proportions

The result in Table 1 shows the phytochemical proportions of *Z. mauritiana* leaf and fruit. The result indicates the presence of trace amounts of flavonoids and tannins in the leaves and fruits. Moderate amount of alkaloids was found in the leaves with trace amount in fruits, whereas high proportion of saponins was found in both leaves and fruits.

Table 1: Qualitative phytochemical proportions in leaf and fruit of *Z. mauritiana*

Constituents	Leaf	Fruit
Saponins	+++	+++
Tannins	+	+
Flavonoids	+	+
Alkaloids	++	+

KEY: +++ = Present in high amount ++ = Moderate amount, and + = Trace amount

Table 2: Quantitative phytochemical composition of leaf and fruit extracts of *Z. Mauritania*

Extract	Constituents (mg/100g dry weight) of plant extract			
	Saponins	Tannins	Flavonoids	Alkaloids
Leaf	92.65 \pm 6.30 ^a	0.50 \pm 0.03 ^a	25.80 \pm 1.73 ^a	2.20 \pm 0.15 ^b
Fruit	70.89 \pm 4.70 ^b	0.30 \pm 0.02 ^a	11.20 \pm 0.75 ^b	3.40 \pm 0.23 ^a

Results are presented as mean \pm standard deviation of three replicates. Means with the same letters in each column are not significantly different (P< 0.05)

Quantitative Phytochemical Constituents

The result of quantitative phytochemical screening of leaves and fruits extract of *Z. mauritiana* are presented in Table 2. Higher proportions of saponins (92.65%), flavonoids (25.8%) and tannins (0.5%) were found in the

leaf than in fruit with 70.89%, 11.20% and 0.30% saponins, flavonoids and tannins respectively. Fruit had more alkaloids (3.4%) than the leaf (2.20%).

Proximate/Nutritional Composition

The nutritional composition (mg/100g dry weight) in leaf and fruit of *Z. mauritiana* are presented in table 3. Higher proportions of carbohydrate (30.05), crude fibre (11.70),

ash (8.20) and moisture content (5.80) were obtained in leaf. The fruit extract had significantly higher ($P < 0.05$) amounts of crude protein (33.10) and lipids (25.80).

Table 3: Proximate composition (mg/100g dry weight) of leaf and fruit extracts of *Z. mauritiana*

Extract	Constituents (mg/100g dry weight) of plant extract					
	Moisture	Ash	Crude Protein	Crude Fibre	Lipids	Carbohydrates
Leaf	5.80±0.01 ^a	8.20±0.00 ^a	27.18±0.00 ^b	11.70±0.001 ^a	6.65±0.00 ^b	34.05±0.00 ^a
Fruit	5.21±0.30 ^a	2.79±0.27 ^b	33.10±0.57 ^a	10.04±0.88 ^a	25.80±0.11 ^a	22.26±0.63 ^b

Results are presented as mean± standard deviation of three replicates. Means with the same letters in each column are not significantly different ($P < 0.05$)

Mineral Content

Result of mineral content in leaf and fruit extracts of *Z. mauritiana* is shown in Table 4. Higher contents (mg/100 g dry weight) of sodium (154.79), potassium (389.08),

manganese (2.0) and iron (1.21) were obtained in the leaf extract. Fruit extract on the other hand indicated higher contents (mg/100 g dry weight) of phosphorus (585.90), magnesium (7.0) and zinc (15.52).

Table 4: Mineral content (mg/100g dry weight) in fruit and Leaf extracts of *Z. mauritiana*

Extract	Constituents (mg/100g dry weight) of plant extract						
	Sodium	Magnesium	Potassium	Zinc	Manganese	Iron	Phosphorus
Leaf	154.79±10.50	6.23±0.12	389.08±10.69	6.00±0.05	2.0±0.14	1.21±0.15	465.43±41.29
Fruit	140.8±10.50	7.0±0.12	316.08±10.69	15.52±0.05	15.52±0.05	0.8±0.15	585.90±41.29

Results are presented as mean± standard deviation of three replicates. Means with the same letters in each column are not significantly different ($P < 0.05$)

Antibacterial Studies

The result of agar well diffusion method in table 5 indicates that methanol leaf extract (50 mg/ml), like the positive control (25.00 – 35.00 mm) had significantly high inhibition ($P < 0.05$) on *S. aureus* (30.33 mm), *P. aeruginosa* (20.67) and *E. coli* (30.33 mm) but low inhibition on *S. pyrogenes* (19.33 mm). Fruit extract (50 mg/ml) had low inhibition on both the test organisms (7.67 – 22.33 mm). At 100 mg/ml concentration, methanol leaf extract had significant inhibition ($P < 0.05$) on both the test organisms (25.33 – 35.33 mm) in comparison with the positive control (25.33 – 35.00 mm) but methanol fruit extract had significantly low ($P < 0.05$) inhibition on both the test organisms (7.33 – 27.33 mm). At 150 mg/ml concentration, methanol leaf extract also had high inhibition ($P < 0.05$) in comparison with the positive control (25.33 – 35.00 mm) but fruit extract had high inhibition on three out of the four test organisms: *S. aureus* (35.67 mm), *P. aeruginosa* (20.33 mm) and the highest, *E. coli* (55.00 mm) but had significantly low ($P < 0.05$) inhibition on *S. pyrogenes* (16.33 mm) compared with the positive control (30.00 mm).

The result of disk diffusion method (table 6) indicated high inhibition of the methanol leaf extract (15.33 mm) and fruit extract (9.33 mm) at 50 mg/ml concentration on *E. coli* compared with the positive control (7.76 mm). At 150 mg/ml concentration, the methanol leaf extract with inhibition of 21.00 mm and fruit extract with inhibition of 25.00 mm on *E. coli* are as effective as the positive control with inhibition of 20.33 mm. At 100 mg/ml concentration, the leaf extract (8.10 – 21.30 mm) and fruit extract (6.67 – 9.33 mm) had significantly low inhibition on all the test organisms compared with the positive control. The fruit to leaf extracts at 50 and 150 mg/ml concentrations respectively indicated low inhibition on *S. aureus* (6.67 – 16.00 & 16.33 – 21.00 mm); *P. aeruginosa* (6.67 & 15.67 – 16.00 mm) and *S. pyrogenes* (7.67 – 14.00 & 8.67 – 18.00). These values are significantly low ($P < 0.05$) compared to the positive control with 7.76 – 30.33 mm at 50 mg/ml and 20.33 – 45.33 mm at 150 mg/ml concentrations.

DISCUSSION

The study indicated presence in the plant's leaves and fruits of constituents which are known to exhibit medicinal as well as physiological activities (Orwa *et al.*, 2009). The leaf and fruit extracts of *Z. mauritiana* have indicated the presence of phytochemicals in varying amounts. These plant constituents detected include saponins, tannins, flavonoids and alkaloids. This finding agrees with that of Mojtaba *et al.* (2016), which reported that *Z. mauritiana* possesses antioxidant-activity related polyphenolic compounds widely distributed in different parts of the plant. Saponins have been

The high saponin and flavonoid contents in the plant's leaves, justifies the reason for its utilization for feeding of cattle, sheep, camel, goats and according to Morton (1987), it is believed to have some resistance against different types of diseases. High alkaloids content in fruits compared to the leaves shows the usefulness of its consumption by man. Alkaloids are reported to have dramatic physiological activities, which account for their wide use in the development of medicines (Aguinaldo *et al.*, 2005). Phytochemicals have either antioxidants (e. g. flavonoids) or hormones (e. g. phytoestrogens) like actions. With these two mechanisms of action the various health effects of phytochemicals in humans could be accounted for (Akpabio and Ikpe, 2013). Antioxidants play a major role in the stability of biological systems (Egwim *et al.*, 2011). The beneficial effect of fruits, vegetables, tea and red wine have been attributed to the presence of flavonoids, alkaloids and other polyphenolic compounds (Kebu and Fassil, 2006). The leaf extract of *Z. mauritiana* contains flavonoids, saponins, alkaloids and tannins as major chemical constituents (Hemant *et al.*, 2014); have immunostimulant and cardiovascular properties (Gupta *et al.*, 2018); demonstrates important biological activities and can therefore be explored for potential uses as antioxidant, antitumor and anticancer agent for pharmaceutical industry (Ashraf *et al.*, 2010).

The result of proximate analysis of the leaf of *Z. mauritiana* revealed higher contents of carbohydrates, crude fibre, crude ash and moisture which are both considered important for the nutritional wellbeing of both animals and humans. Zhao (2012) reported that the dietary fibre in *Z. mauritiana* can lower serum cholesterol level, risk of coronary heart disease, hypertension, constipation, diabetes, cancer of the breast and colon. The higher contents of crude protein and lipids in the fruits of *Z. mauritiana* are of paramount nutritional importance. As earlier reported by Zhao (2012), energy value of 100 g of the fruit containing 33.10 g of proteins, 25.80 g of lipids and 22.26 g of carbohydrates is 470.28 cal (1967.66 KJ). This exceeds the energy value of *Z. mauritiana* pericarp with only 1,547.14 KJ/100g. According to Anderson *et al.* (2003) Children, ages 1 to 10 years (19.33 kg) requires an average of 2,267 KJ of protein per day, meaning, they need to take about 68.47 g of *Z. mauritiana* fruits, if an allowance of 25% digestibility and limiting sulphur and amino acids requirements were to be met.

Result of mineral constituent analysis has indicated higher amounts of phosphorus, zinc and magnesium in *Z. mauritiana* fruits. The plant's leaves however showed greater amounts of sodium, potassium, iron and zinc. Similar result was earlier reported by Abdallah *et al.* (2016).

reported to possess the ability to strengthen the constriction of heart muscles. Flavonoids detected have been known to provide protection against oxidative stress induced diseases and are major factors responsible for the anti-oxidative activity of the plant. The presence of alkaloids also suggests the importance of the plant in the pharmaceutical industry, as alkaloids have been reported to have a stimulating effect, acting as topical anaesthetics in Ophthalmology, powerful pain reliever and antipyretic action among other uses (Orwa *et al.*, 2009).

According to Bahtia and Mishra (2009), adult male human beings weighing 66.60 kg requires about 15 mg of zinc per day and this can be obtained from 0.426 kg of *Z. mauritiana* fruits. For pregnant and lactating women, about 1,200 mg of phosphorus is required per day: This implies that, they need to take about 0.2 kg of *Z. mauritiana* fruit per day to maintain the recommended dietary allowances for phosphorus.

Antibacterial studies revealed that the methanol leaf extract generally produced higher zones of inhibition than fruit extract on test bacteria across concentration gradient and the inhibition increased with increased concentration (50, 100, 150 mg/ml). The result corroborates the work of Ogbonna *et al.* (2003), whose work on *Ximenia americana* showed increased bacterial inhibition at increased concentrations of the leaf extract (125, 250, 500 mg/ml). Like *Z. mauritiana*, the activity of *Euphorbia hirta* and *Ximenia americana* have been attributed to the presence of tannins and saponins in their extracts (Rojas *et al.*, 1992; Ogbonna *et al.*, 2003). High susceptibility of *S. aureus* to ethanol leaf extract has been reported (Yusuf *et al.*, 2005). The increased sensitivity observed for the Gram negative bacteria *E. coli* up to 55.00 mm inhibition against the positive control (35.00 mm) at 150 mg/ml concentration has been attributed to the thin lipid layer that is more polar than the thick-cell walled Gram positive bacteria which have tough non-polar peptidoglycan layer (Nyam *et al.*, 2009).

CONCLUSION

The presence in the plant's leaves and fruits of phytochemical compounds coupled with the nutritional and mineral constituents has been confirmed by previous researches. The higher nutritional contents such as crude proteins and lipids as well as, mineral constituents particularly phosphorus, zinc and magnesium in the fruits give an indication of its importance in the provision of energy and protection of body against diseases. The inhibitory effects of the plant's extracts especially the leaves on bacterial cultures have confirmed the status ascribed to it by traditional healers and local populace who make use of the plant in treatment of various infections.

RECOMMENDATIONS

Based on findings of this study, the consumption of *Z. mauritiana* fruits particularly by children aged 1 – 10 and nursing mothers is encouraged in order to meet the limiting sulphur and amino acid requirements and for the pregnant and lactating women to meet their phosphorus requirements. The utilization of *Z. mauritiana* leaves through incorporation as food supplement, in addition to its utilization as animal feed and herbal medicine ingredient, would enhance the wellbeing and nutritional needs of humans. Based on the results of antibacterial screening of the plant's extracts, pharmaceutical products can be produced from its constituents and patented after thorough screening and certification.

Table 5: Antimicrobial activity of methanol leaf and fruit extracts of *Z. mauritiana* on test organisms against control by Agar well diffusion method

Extract con. Test organism	50 mg/ml				100 mg/ml				150 mg/ml			
	A	B	C	D	A	B	C	D	A	B	C	D
	Zone of inhibition (mm)				Zone of inhibition (mm)				Zone of inhibition (mm)			
Leaf extract	30.33±0.2 9 ^a	20.67±0.2 a	30.33±0.1 5 ^a	19.33±0.0 08 ^b	35.33±0.0 28 ^a	25.67±1.0 04 ^a	33.00±0.0 34 ^a	25.33±0.0 23 ^a	39.00±0.0 25 ^a	31.00±0.0 23 ^a	36.00±0.0 25 ^b	29.33±0.0 36 ^a
Fruit extract	15.33±0.2 6 ^b	7.67± 0.29 ^b	22.33±0.7 6 ^b	7.67±0.0 5 ^b	27.33±0.0 58 ^c	7.33± 0.18 ^b	26.33±0.0 25 ^b	7.67±0.4 4 ^b	35.67±0.0 58 ^a	20.33±0.0 26 ^b	55.00±0.0 50 ^a	16.33±0.0 16 ^b
Streptomycin Sulphate (+ve control)	30.33±0.5 8 ^a	25.00±0.1 6 ^a	35.00±0.3 3 ^a	30.00±0.0 44 ^a	30.33±0.0 58 ^b	25.33±0.0 23 ^a	35.00±0.0 33 ^a	30.00±0.0 43 ^a	30.33±0.0 58 ^b	25.33±0.0 25 ^b	35.00±0.0 54 ^b	30.00±0.0 47 ^a
Methanol (-ve control)	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI

Results are mean ± standard error of three replicates. Means with the same superscripts in each column are not significantly different (P<0.05); NI = no inhibition; A = *S. aureus*; B = *P. aeruginosa*; C = *E. coli*; D = *S. pyrogenes*

Table 5: Antimicrobial activity of methanol fruit and Leaf extracts of *Z. mauritiana* on test organisms against control by Disc diffusion method

Extract conc. Test organism	50 mg/ml				100 mg/ml				150 mg/ml			
	A	B	C	D	A	B	C	D	A	B	C	D
	Zone of inhibition (mm)				Zone of inhibition (mm)				Zone of inhibition (mm)			
Leaf extract	16.00±0.0 29 ^b	6.67± 0.2 ^b	15.33±0.0 15 ^a	14.00±0.0 08 ^b	21.80±0.0 28 ^b	8.33±0.2 7 ^b	8.10±0.34 b	16.00±0.1 2 ^b	21.00±0.0 50 ^b	16.00±0.0 23 ^b	21.00±0.0 25b	18.00±0.0 31 ^b
Fruit extract	6.67±0.2 6 ^c	6.67± 0.29 ^b	9.33±0.5 8 ^b	7.67±0.0 5 ^c	6.67±0.5 8 ^c	6.67±0.4 7 ^c	9.33±0.56 c	8.67±0.44 c	16.33±0.0 58 ^c	15.67±0.0 58 ^b	25.00±0.0 87a	8.67±0.3 2 ^c
Streptomycin Sulphate (+ve control)	30.33±0.0 58 ^a	45.33±0.0 16 ^a	7.76±0.3 3 ^b	25.00±0.0 23 ^a	30.33±0.0 10 ^a	30.33±0.0 28 ^a	20.33±0.3 4 ^a	25.00±0.2 7 ^a	30.33±0.0 58 ^a	45.33±0.0 25 ^a	20.33±0.0 54b	25.00±0.0 46 ^a
Methanol (-ve control)	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI

Results are as mean ± standard error of three replicates. Means with the same superscripts in each column are not significantly different (P<0.05); NI = no inhibition; A = *S. aureus*; B = *P. aeruginosa*; C = *E. coli*; D = *S. pyrogenes*

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