

Effects of Ethanolic Root Extract of *Spondias mombin* (Hog plum) on Wistar Rats

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ABSTRACT

Spondias mombin has been used for a variety of therapeutic purposes. The leaves of *S. mombin* are used to treat a variety of microbiological illnesses, including those caused by viruses, fungus, bacteria and yeast. Its leaves exhibit sedative and anti-inflammatory properties. The aim of this study is to determine the side effects of *Spondias mombin* leaf extracts on the liver of adult wistar rats. The animals were numbered, weighed and randomly selected into 5 groups; Group A served as the control group, group B received 100 mg/kg body weight of *Spondias mombin* extract, group C received 200 mg/kg body weight of *Spondias mombin* extract, group D received 300 mg/kg body weight of *Spondias mombin* extract. Group E received 400 mg/kg body weight of *Spondias mombin* extract. The experiment lasted for 28 days after which the animals were sacrificed and their liver tissues were excised, grossed, washed in normal saline, The results showed that the experimental animals fed with varying doses (100 mg/kg, 200 mg/kg, 300 mg/kg bw and 400 mg/kg bw) of *Spondias mombin* showed no significant structural changes in the architecture of the liver tissues. This result suggests that although the use of the leaf extracts of *Spondias mombin* is relatively safe, , high dosages could lead to marked structural damages of the liver.

Keywords: *Spondias mombin*, leaves adult Wistar rats. Ethanol, FT-IR, FRAP and Randox kits

INTRODUCTION

Traditional medicine is defined as indigenous medicine that is used to maintain health, prevent, and treat physical and mental illnesses based on theories, beliefs, and experience [1,2]. Traditional medicine has been used for thousands of years with great contributions made by practitioners to human health, particularly primary healthcare providers at the community level and has maintained its popularity worldwide [2]. The use of medicinal plants as alternative therapies has been an age-long practice in traditional medicine [3].

Natural products have shown potential as alternative therapies for liver diseases due to their antioxidant and hepatoprotective properties. One such plant is (*Spondias mombin*), which is widely used as a traditional medicine for the treatment of various ailments such as diarrhoea,

fever, and inflammation [4]. Previous studies have shown that *S. mombin* possesses several bioactive compounds with potent antioxidant, anti-inflammatory, and antimicrobial properties. *Spondias mombin*, commonly known as yellow mombin, is a tropical tree with various medicinal properties, including antioxidant and anti-inflammatory effects [5-7]. *Spondias mombin* has been traditionally used in folk medicine to treat liver disorders and has demonstrated potential hepatoprotective effects. Moreover, studies have indicated that *Spondias mombin* possesses antioxidant properties attributed to its phenolic and flavonoid content.

The aim of this study is to determine the effect of ethanolic root extract of *Spondias mombin* on liver function and the *in vivo* antioxidant activity in albino rats.

MATERIALS AND METHODS

Sample Collection

Fresh roots of *S. mombin* were collected from Nurul Yaqeen Primary School, Mubi, Mubi South L.G.A. of Adamawa State. The plant was identified in Forestry Department, Federal Polytechnic, Mubi. The healthy thick roots were sorted out and washed with distilled water to eliminate dust and other foreign particles and air dried. The washed root was dried for about 30 days till they were completely dried., The dried root was ground to powder using mortar and pestle and kept in an airtight container.

Preparation of Plant Extracts

Powdered sample weighing 1000 g was soaked in 1000 ml of ethanol in a conical flask. The flask which contained the root sample was shaken, covered with a cork and allowed to stand for 72 hours at room temperature. Then, the mixture was filtered and the extract was collected and concentrated to dryness by evaporation in an evaporating dish.

Phytochemical Screening

The phytochemical screening of the plants constituents (alkaloid, tannins, phlobatannins, phenol, flavonoids, glycoside, saponins, volatile oil, hydrolysable tannin and protein) were assessed by using qualitative methods [8] and quantitative methods [9].

***In Vitro* Antioxidant Activity**

Thiobarbituric Acid (TBA) method used for evaluating the extent of lipid peroxidation, and Ferrous Reducing Antioxidant Capacity (FRAP) property were determined by the method of Sepideh et al [10]. The extract sample was tested for its antioxidant activity by 2,2-diphenyl-1-picrylhydrazyl (DPPH).

Acute Toxicity

The acute toxicity (LD₅₀) of *Spondias mombin* aqueous extract was estimated orally in rats (n = 13) following Lorke's method [11].

Experimental Design

Twenty five albino rats each weighing between 150 and 200 g were purchased from Mubi, Adamawa State. They were housed in a metal cage and allowed access to a standard laboratory pellet diet and water throughout the experimental period. The albino rats were divided into five (5) groups of five (5) rats each.

The rats were randomly sampled into five of five rats each and were treated as follows:

Group A: Normal control, were treated with normal food and water

Group B: Were administered with 100 mg/kg ethanolic root extract of *Spondias mombin*

Group C: Were administered with 200 mg/kg ethanolic root extract of *Spondias mombin*

Group D: Were administered with 300 mg/kg ethanolic root extract of *Spondias mombin*

Group E: Were administered with 400 mg/kg ethanolic root extract of *Spondias mombin*

After 28 days of administering ethanolic root extract of *Spondias mombin*, the rats were sacrificed by cardiac puncture and blood sample was collected and allowed to clot and centrifuge at 4000 rpm for 10 minutes.

Determination of Liver and Kidney Function Test

The method of Bergmeyer [12] was used to determine Aspartate Aminotransferase (AST), Alanine Aminotransferase (ALT), Bilirubin, Total protein, Albumin. Urea and creatinine were determine respectively.

RESULTS AND DISCUSSION

Table 1. Qualitative phytochemical screening of aqueous and ethanolic root extract of *S. mombin*

Parameters	Ethanolic root extract
Tanins	+
Saponins	-
Terponoids	-
Flavonoids	-
Alkaloids	-
Glycosides	++
Steroids	-
Phenols	+

Keys: Positive = (+), Undetected = (-), Excess = (++)

Table 2. Quantitative Phytochemical Screening

PARAMETERS	Root extract
Phenols	0.116 ± 0.01
Tannins	0.024 ± 0.01
Glycosides	22.68 ± 0.02

Results are mean ± SEM of three replicates

Table 3. Effect of ethanol root extract of *S. mombin* on TBA and FRAP Antioxidant activities

	20 µg/mL	40 µg/mL	60 µg/mL	80 µg/mL	100 µg/mL
TBA	18.33 ± 0.01	35.41 ± 0.02	59.58 ± 0.01	69.58 ± 0.03	74.17 ± 0.03
FRAP	3.17 ± 0.02	28.80 ± 0.01	55.66 ± 0.03	62.53 ± 0.05	80.51 ± 0.02

The values are mean ± SEM of three replicates

Table 4: Effects of ethanolic root extract of *Spondias mombin* on liver enzyme markers

	AST (µL)	ALT (µL)	ALP (µL)
NORMAL	15.11± 6.41	15.00± 1.00	660.97± 2.63
100 mg/kg BW	47.73± 25.76	22.67± 8.29	703.06± 17.04
200 mg/kg BW	28.97± 3.32	28.40± 3.52	667.4± 17.82
300 mg/kg BW	31.43± 18.51	28.67± 4.41	687.2± 168.1
400 mg/kg BW	45.40± 5.05	25.53± 5.22	751.3± 110.6

The values are mean ± SEM of three replicates at 0.05 significant levels; ALP is Alkaline Phosphatase.

Table 5: Effects of ethanolic root extract of *Spondias mombin* on product of the liver

	Total (mmol/L)	Bilirubin Direct (mmol/L)	Bilirubin	Albumin (g/L)	Protein (g/L)
NORMAL	11.73± 1.54 ^c	11.37± 1.32 ^b		25.43± 7.43	175.6± 2.36 ^b
100 mg/kg bw	16.03± 0.32 ^b	35.80± 1.78 ^{ab}		27.50± 0.61	196.8± 1.14 ^{ab}
200 mg/kg bw	17.77± 1.507 ^b	22.80± 1.135 ^b		32.50± 1.15	235.1± 4.36 ^a
300 mg/kg bw	13.73± 1.52 ^{bc}	61.63± 20.79 ^a		33.40± 1.70	188.1 ± 25.78 ^b
400 mg/kg bw	22.07± 0.87 ^a	44.30± 0.954] ^{ab}		33.30 ± 0.90	237.6± 3.57 ^a

The values are Mean ± SEM of three replicates at 0.05 significant levels

Superscript a, b, ab, & c, indicate significance difference at p< 0.05 down the column (across the groups)

Table 6: Effects of ethanolic root extract of *Spondias mombin* on product of the liver

	Urea (mmol/L)	Creatinine (mmol/L)	Potassium (mmol/L)	Sodium (mmol/L)	Chloride (mmol/L)
NORMAL	4.80 ± 0.22	48.97 ± 1.45 ^a	9.40 ± 0.21	134.20 ± 3.12 ^b	130.67 ± 2.11 ^b
100 mg/kg bw	4.10 ± 0.23	49.77 ± 2.42 ^a	8.40 ± 0.12	127.93 ± 2.23 ^b	127.30 ± 1.01 ^b
200 mg/kg bw	4.83 ± 0.41	58.37 ± 1.21 ^a	7.70 ± 0.32	123.40 ± 3.12 ^b	105.97 ± 1.31 ^{ab}
300 mg/kg bw	4.46 ± 0.32	63.92 ± 1.23 ^{ab}	7.27 ± 0.42	115.33 ± 1.22 ^{ab}	105.27 ± 2.11 ^{ab}
400 mg/kg bw	4.43 ± 0.16	83.50 ± 0.56 ^b	6.50 ± 0.24	94.43 ± 1.11 ^a	97.43 ± 1.04 ^a

The values are Mean ± SEM of three replicates at 0.05 significant levels; Superscript a, b, ab, & c, indicate significance difference at $p < 0.05$ down the column (across the groups)

Plants contain secondary metabolites that can be detected through phytochemical screening, a process used to identify these compounds in plants. Secondary metabolites are organic compounds that are not directly involved in the growth, development, reproduction, or other primary functions of an organism. While the absence of these compounds may not cause immediate plant death, it can result in long-term impairment of other functions, such as defensive mechanisms, aesthetic appearance, or in some cases, overall impact [13].

A qualitative preliminary phytochemical analysis was conducted using in the root extract of *S. mombin*. Results indicating the presence of primary and secondary metabolites are shown in Table 1. The phytochemical screening revealed that the presence of some chemical constituents, including phenolic compounds, tannins and glycosides, which have demonstrated anti-diabetic properties in the root extract. The chemical composition of the *S. mombin* extract aligns with the findings of Bishir et al [14] who reported the presence of secondary metabolites such as tannins, phenols, and glycosides in *S. mombin*.

The in-vitro antioxidant activity of the ethanolic extract of the root from *S. mombin* was evaluated using FRAP and TBA. The results are shown in Table 3, and the Total Antioxidant Activity assay result is shown in Figure 1. Oxidative stress has been implicated in several diseases, including diabetes, rheumatoid arthritis, cardiovascular diseases, atherosclerosis, neurodegenerative diseases (Parkinson's, Alzheimer's, and Huntington's), cancer, and aging [15]. Natural antioxidants such as phenolic acids from plants may offer resistance against oxidative stress by scavenging free radicals, inhibiting lipid peroxidation, and through other mechanisms [16]. Antioxidants play an important role in the human body system by reducing the concentration of reactive oxygen species (ROS), as reported in previous studies that antioxidants, especially phenolic compounds, FRAP, and TBA, are suggested to be major and essential

compounds that protect human cells from oxidative stress, damage, and cell proliferation [17-18]. A previous study on *S. mombin* revealed that the root has potent antioxidant activity [19-20]. Antioxidant capability and free radical scavenging properties are essential in using plant materials for curative purposes.

Tissue enzymes can indicate cellular damage long before conventional histological techniques can determine structural damage of tissues. The serum levels of ALP, ALT, and AST are helpful in identifying inflammation, necrosis, and determining liver function [21]. In chronic liver diseases like cirrhosis, AST tends to be released more than ALT [22]. The administration of ethanolic root extract of *Spondias mombin* at doses of 100, 200, 300, and 400 mg/kg bw did not show a significant increase in liver enzyme markers, as presented in Table 4. However, there was a significant increase in bilirubin and protein levels in the group treated with the extract, as indicated in Table 5. This increase may suggest impairment in liver function.

The measurement of electrolytes, creatinine, and urea aims to assess kidney functionality. There was a significant increase in creatinine levels when the extract was administered, compared to normal, and this increase showed a dose-dependent relationship, as shown in Table 6. The elevated level of creatinine in serum is primarily indicative of glomerular filtration, and it may also point to a problem with proximal tubular secretion [23]. Moreover, there was a significant reduction in sodium, potassium, and chloride levels in the groups administered with the extract. It is important to note that sodium plays a critical role in maintaining the normal distribution of water and osmotic pressure in various fluid compartments [24]. Ishaku et al [25] reported that alteration in the concentration of these body electrolytes is indicative of poor renal functions or renal impairment.

CONCLUSION

Spondias mombin, also known as Hog plum, is a plant with significant medicinal importance, utilized for various purposes. Phytochemical screening revealed that the root ethanol extract is rich in chemical constituents. The presence of these secondary metabolites validates the plant's use as an herbal drug. Studies on the in-vitro antioxidant activity of this extract revealed potent antioxidant properties. However, the effects of the plant's root on liver function and body weight indicate that its daily intake may have adverse effects on vital organs and body weight, depending on the dosage.

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