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Hepatorenal Consequences of *Spondias mombin* Root Extract: A Histo-Biochemical Study in Animal Model

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ABSTRACT

Spondias mombin, a tropical medicinal plant widely used in ethnomedicine, has been reported to possess different therapeutic properties, yet its safety profile on vital organs remains insufficiently defined. This study investigated the histomorphological and biochemical effects of *S. mombin* root extract on the liver and kidneys of Wistar albino rats. Twenty-four adult male rats (150-200 g) were randomly divided into six groups (n = 4): a control group and five treatment groups administered 200, 400, 600, 800, or 1000 mg/kg body weight of *S. mombin* root extract orally for 28 days. Serum biochemical parameters, including electrolytes, urea, creatinine, and liver enzymes (AST, ALT, ALP), were analyzed using standard methods, while liver and kidney tissues were processed for histological evaluation with hematoxylin and eosin staining. Significant variations in body weight were observed ($p < 0.05$), with the 800 mg/kg group showing the highest gain and the 200 mg/kg group the least. Organ weights, however, were not significantly affected ($p > 0.05$). Biochemical assays indicated stable levels of urea, bilirubin, and liver enzymes across groups, although creatinine was significantly reduced at 1000 mg/kg ($p = 0.006$). Histological analysis showed preserved hepatic and renal architecture at doses up to 600 mg/kg. At higher doses (800-1000 mg/kg), ballooning degeneration and microvesicular steatosis were observed in hepatocytes, whereas renal tissues remained morphologically intact. The findings suggest that *S. mombin* root extract is relatively safe at low-to-moderate doses but may induce dose-dependent hepatotoxicity at higher concentrations. Further long-term studies are needed to establish safe therapeutic limits and identify the bioactive compounds responsible.

Keywords: hepatotoxicity, histomorphology, medicinal plants, nephrotoxicity, *Spondias mombin*.

INTRODUCTION

Medicinal plants have long been central to global healthcare, providing both preventive and therapeutic benefits. The World Health Organization estimates that approximately 80% of the world's population relies on medicinal plants for primary healthcare needs, highlighting their importance in sustaining human health and improving quality of life [1]. Among these, *Spondias mombin* Linn. (Family: Anacardiaceae), commonly known as hog plum, is widely distributed across tropical regions including Nigeria, Ivory Coast, Brazil, and Mexico [2]. In Nigerian ethnomedicine, the plant is valued for its broad therapeutic applications. Different parts—such as leaves, bark, fruit,

and roots—are traditionally used for the management of diarrhea, dysentery, gonorrhea, postpartum hemorrhage, tuberculosis, and inflammatory disorders [3, 4]. Phytochemical analyses of *S. mombin* have revealed the presence of bioactive constituents, including alkaloids, glycosides, flavonoids, saponins, tannins, steroids, and terpenoids, which have been linked to antibacterial, antioxidant, antidiabetic, haematinic, and oxytocic properties [5, 6]. These pharmacological characteristics suggest promising therapeutic potential. However, despite its widespread use, there is insufficient scientific evidence regarding its safety profile, particularly its structural and functional effects on important organs such as the liver and kidneys. The liver and kidneys are key metabolic and excretory organs responsible for detoxification, biotransformation of xenobiotics, maintenance of electrolyte balance, and regulation of blood pressure. Damage to these organs from unregulated herbal remedies may result in profound physiological dysfunction [7]. Evaluating histological and biochemical alterations in these organs therefore provides a critical basis for determining the toxicological or protective effects of medicinal plants.

This study therefore aimed to investigate the histomorphological and biochemical effects of graded doses of *S. mombin* root extract on the liver and kidneys of Wistar albino rats. Findings are expected to provide scientific validation for its ethnomedicinal applications, define its potential dose-related risks, and contribute evidence for the safe integration of *S. mombin* into therapeutic use.

MATERIALS & METHODS

Animals and Ethical Approval

Twenty-four adult male Wistar rats (150-200 g) were obtained from the University of Benin animal facility. Animals were housed under standard laboratory conditions (12-hour light/dark cycle, 25 ± 2 °C, free access to food and water) and acclimatized for one week before experimentation. Ethical approval for this study was sought from the Ethics and Research Committee of the Ministry of Agriculture, Edo State, Nigeria with ethical number (MAFSAEC: 025-08/13/0042).

Experimental Design

Twenty-four adult male Albino Wistar rats (150-200 g) were randomly assigned into six groups (n = 4 per group). Experimental groups received graded oral doses of *Spondias mombin* root extract at 200, 400, 600, 800, and 1000 mg/kg body weight daily for 28 consecutive days, while the control group received equal volumes of distilled water. Oral administration of plant extract at these dosage ranges and duration [8] has been previously validated as sufficient for assessing dose-dependent biochemical and histological changes in rat liver and kidney tissues [8-11].

Organ Collection and Processing

After the four (4) weeks treatment period, the animals were anesthesized with chloroform and sacrificed by fenestration 24 hours after the last day of administration. Livers and kidneys were excised, trimmed of adherent fat, blotted, and weighed. Organs were fixed in 10% neutral buffered formalin for 24 hours, dehydrated in ascending grades of ethanol, cleared in xylene, and embedded in paraffin wax.

Histological Staining

Paraffin blocks were sectioned at 5 µm using a rotary microtome (Leica RM2235, Germany). Sections were stained with hematoxylin and eosin (H&E) following standard procedures [11]. Sections were stained with hematoxylin for 10 minutes, followed by counterstaining with 1% eosin. Stained sections were dehydrated, cleared in xylene, and mounted in DPX. Microscopic examination was performed using an Olympus CH20 light microscope (Olympus, Japan) at ×40 and ×100 magnifications. Representative photomicrographs were obtained using a microscope-linked digital camera.

Scoring System

Histological quality was assessed semi-quantitatively using parameters of nuclear clarity, cytoplasmic contrast, background staining, and overall histoarchitecture. Scoring was independently performed by two blinded histopathologists.

STATISTICAL ANALYSIS

Data were analyzed using IBM SPSS Statistics software version 20.0 (IBM Corp., Armonk, NY, USA). Results were expressed as mean ± standard deviation (SD). Comparisons among groups were performed using one-way analysis of variance (ANOVA). Where significant differences were detected, Duncan's multiple-range post hoc test was applied to identify pairwise group differences. A p-value of less than 0.05 was considered statistically significant.

RESULT

General Observations

Liver

Sections of the liver from control rats revealed a well-preserved hepatic architecture, with clearly visible lobular organization. Hepatocytes appeared polygonal, arranged in cords radiating from the central vein, and separated by narrow sinusoidal spaces. The cytoplasm of the hepatocytes was uniformly eosinophilic, and nuclei were centrally located, normochromic, and round, with indistinct but regular nucleoli, consistent with healthy hepatocellular morphology. The sinusoidal lining cells appeared intact, and no evidence of vacuolar degeneration, fatty change, necrosis, or inflammatory cell infiltration was observed. Portal tracts were normal in appearance, with preserved bile ducts and blood vessels, and no signs of fibrosis or congestion (Plate 1.1A). Given an interpretation like the above-Section of the liver from the control rats showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli (Plate 1.1A).

Sections of the liver from rats administered *S. mombin* root extract at a dose of 200 mg/kg revealed hepatocytes (arrow) with well-preserved eosinophilic cytoplasm surrounding centrally placed normochromic nuclei, some with indistinct nucleoli. The hepatic cords appeared intact and radiated normally from the central vein, while sinusoidal spaces remained open and patent. No evidence of vacuolation, degeneration, inflammatory infiltrates, or fibrosis was observed (Plate 1.1B).

Sections of the liver from rats administered *S. mombin* root extract at a dose of 400 mg/kg revealed hepatocytes (arrow) with preserved eosinophilic cytoplasm surrounding centrally placed normochromic nuclei, some exhibiting indistinct nucleoli. The hepatic cords were orderly arranged and radiated normally from the central vein, while sinusoidal spaces appeared patent without evidence of compression. No cytoplasmic vacuolation, ballooning degeneration, inflammatory infiltrates, or fibrosis were observed (Plate 1.1C).

Sections of the liver from rats administered *S. mombin* root extract at a dose of 600 mg/kg revealed hepatocytes (arrow) with preserved eosinophilic cytoplasm surrounding centrally placed normochromic nuclei, some displaying indistinct nucleoli. The hepatic cords remained intact and radiated normally from the central vein, while sinusoidal spaces were widely patent. No evidence of cytoplasmic vacuolation, ballooning degeneration, inflammatory cell infiltration, or fibrosis was observed (Plate 1.1D).

Sections of the liver from rats administered *S. mombin* root extract at a dose of 800 mg/kg revealed hepatocytes with abundant eosinophilic cytoplasm containing multiple fine microvacuoles, consistent with ballooning degeneration. The vacuolated cytoplasm surrounded centrally placed normochromic nuclei, although occasional nuclear displacement was observed due to cytoplasmic expansion. Hepatic cords showed focal disruption, with hepatocytes losing their uniform arrangement compared

to controls. Sinusoidal spaces remained patent with mild compression. No appreciable inflammatory cell infiltrates or fibrotic changes were observed at this dose (Plate 1.1E).

Sections of the liver from rats administered *S. mombin* root extract at a dose of 1000 mg/kg revealed hepatocytes (arrow) with eosinophilic cytoplasm containing multiple microvacuoles, indicative of ballooning degeneration. The vacuolated cytoplasm surrounded centrally placed normochromic nuclei, with occasional displacement due to cytoplasmic expansion. Hepatic cords appeared focally disrupted compared to controls, while sinusoidal spaces remained patent with mild compression. No significant inflammatory infiltrates or fibrotic changes were evident at this dose (Plate 1.1F).

Kidney

Sections of the kidney from control rats revealed normal glomeruli (thick arrow) with intact mesangial cells, well-preserved capillary loops, and a normal epithelial lining. The renal tubules (thin arrow) appeared oval-shaped and were lined by cuboidal epithelium, with some tubules containing pale eosinophilic material within their lumina. The renal interstitium showed no evidence of edema, inflammation, or fibrosis (Plate 1.2 A).

Sections of the kidney from rats administered *S. mombin* root extract at a dose of 200 mg/kg revealed normal glomeruli (thick arrow) with intact mesangium, well-preserved blood vessels, and a normal epithelial lining. The renal tubules (thin arrow) were oval and lined by cuboidal epithelium, with some tubules containing pale eosinophilic material in their lumina. No evidence of tubular degeneration, necrosis, inflammation, or interstitial changes was observed (Plate 1.2B).

Sections of the kidney from rats administered *S. mombin* root extract at a dose of 400 mg/kg revealed normal glomeruli (thick arrow) with intact mesangium, preserved blood vessels, and normal epithelial lining. The renal tubules (thin arrow) appeared oval and were lined by cuboidal epithelium, with some tubules containing pale eosinophilic material within their lumina. No evidence of tubular degeneration, necrosis, inflammation, or interstitial pathology was observed (Plate 1.2C).

Sections of the kidney from rats administered *S. mombin* root extract at a dose of 600 mg/kg revealed normal glomeruli (thick arrow) with intact mesangium, well-preserved blood vessels, and a normal epithelial lining. The renal tubules (thin arrow) were oval and lined by cuboidal epithelium, with some tubules containing pale eosinophilic material in their lumina. No signs of tubular degeneration, necrosis, inflammatory infiltrates, or interstitial alterations were observed (Plate 1.2D).

Sections of the kidney from rats administered *S. mombin* root extract at a dose of 800 mg/kg revealed normal glomeruli (thick arrow) with intact mesangium, preserved blood vessels, and normal epithelial lining. The renal tubules (thin arrow) appeared oval and were lined by cuboidal epithelium, with some tubules containing pale eosinophilic material in their lumina. No evidence of tubular injury, necrosis, inflammation, or interstitial changes was observed (Plate 1.2E).

Sections of the kidney from rats administered *S. mombin* root extract at a dose of 1000 mg/kg revealed normal glomeruli (thick arrow) with intact mesangium, preserved blood vessels, and a normal epithelial lining. The renal tubules (thin arrow) were oval and lined by cuboidal epithelium, with some tubules containing pale eosinophilic material in their lumina. No histological evidence of tubular degeneration, necrosis, inflammatory infiltrates, or interstitial alterations was observed (Plate 1.2F).

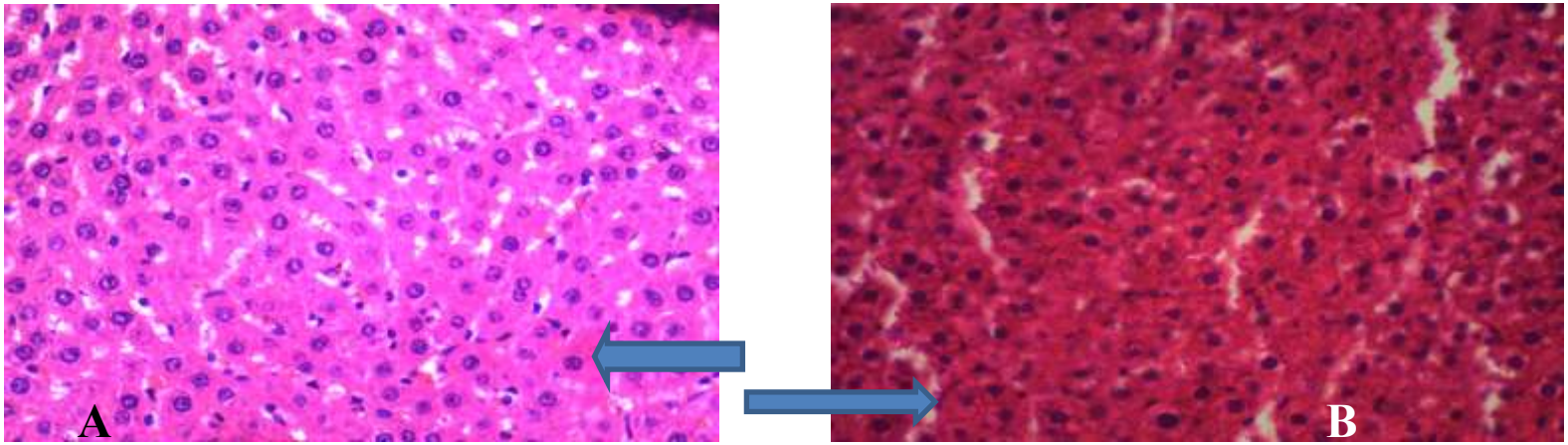

Plate 1.1A: Section of the liver from the control rats showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli. Features In Keeping with Normal Hepatocytes H and E Mag x400.
Plate 1.1B: Section of the liver of rats administered 200mg/kg body weight of *S. mombin* root extract showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli. Features In Keeping with Normal Hepatocytes H and E Mag x400.

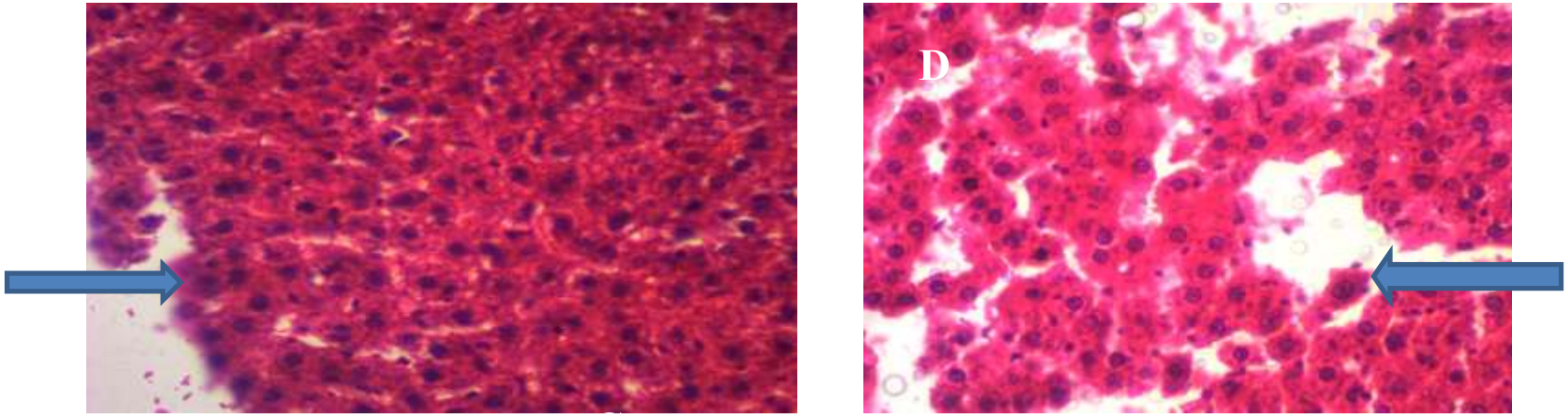

Plate 1.1C: Section of the liver of rats administered 400mg/kg body weight of *Spondias mombin* root extract showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli. Features In Keeping with Normal Hepatocytes H and E Mag x400.
Plate 1.1D: Section of the liver of rats administered 600mg/kg body weight of *Spondias mombin* root extract showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli. Features In Keeping with Normal Hepatocytes H and E Mag x400.

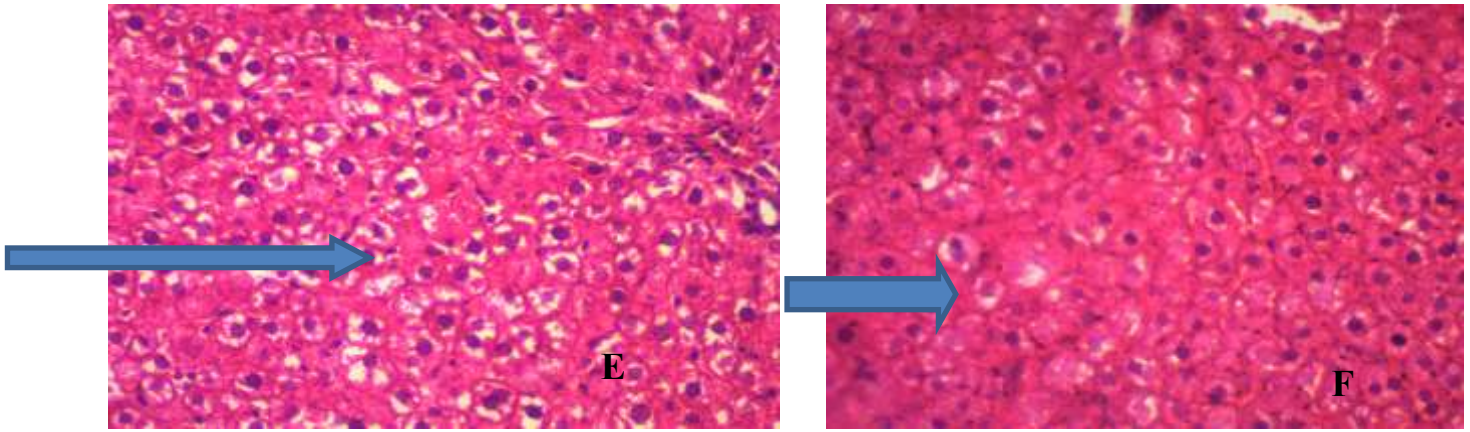

Plate 1.1E: Section of the liver of rats administered 800mg/kg body weight of *S. mombin* root extract showed hepatocytes (arrow) with eosinophilic cytoplasm containing microvacuoles (ballooning degeneration), the cytoplasm surrounds a centrally placed nucleus. Features In Keeping with Steatosis H and E Mag x400.

Plate 1.1F: Section of the liver of rats administered 1000mg/kg body weight of S. mombin root extract showed hepatocytes (arrow) with eosinophilic cytoplasm containing microvacuoles (ballooning degeneration), the cytoplasm surrounds a centrally placed nucleus. Features In Keeping with Steatosis H and E Mag x400.

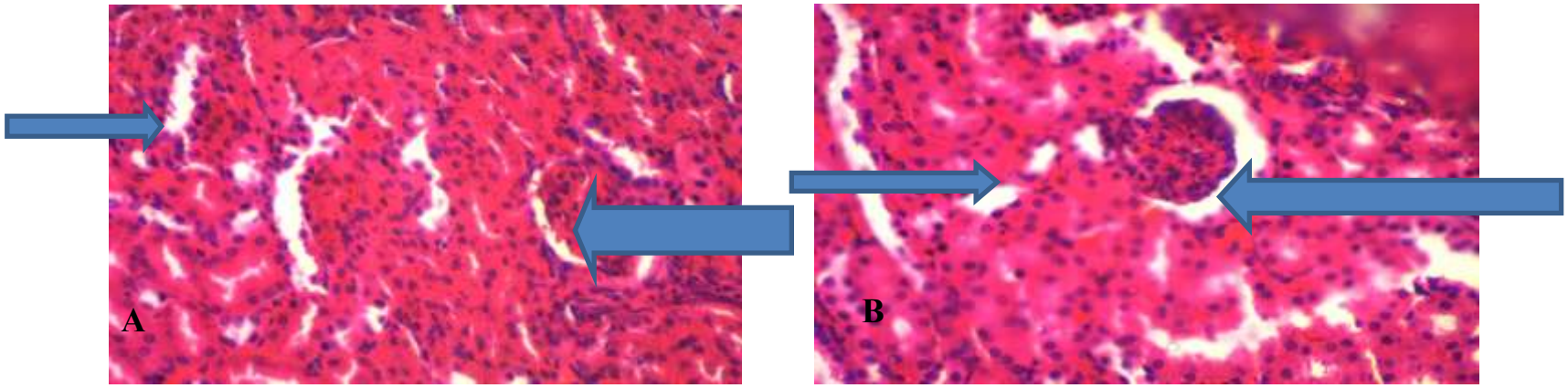

Plate 1.2A: Section of the kidney from the control rats showed normal glomeruli (thick arrow) containing normal mesangium, blood vessels and epithelium. The tubules (thin arrow) are oval shaped and lined by cuboidal epithelium with some tubules containing pale eosinophilic material. Features are in keeping with Normal Kidney H and E Mag x400.

Plate 1.2B: Section of the kidney from rats administered *S. mombin* root extract at a dose of 200 mg/kg revealed normal glomeruli (thick arrow) containing normal mesangium, blood vessels and epithelium. The tubules (thin arrow) are oval shaped and lined by cuboidal epithelium with some tubules containing pale eosinophilic material. Features are in keeping with Normal Kidney

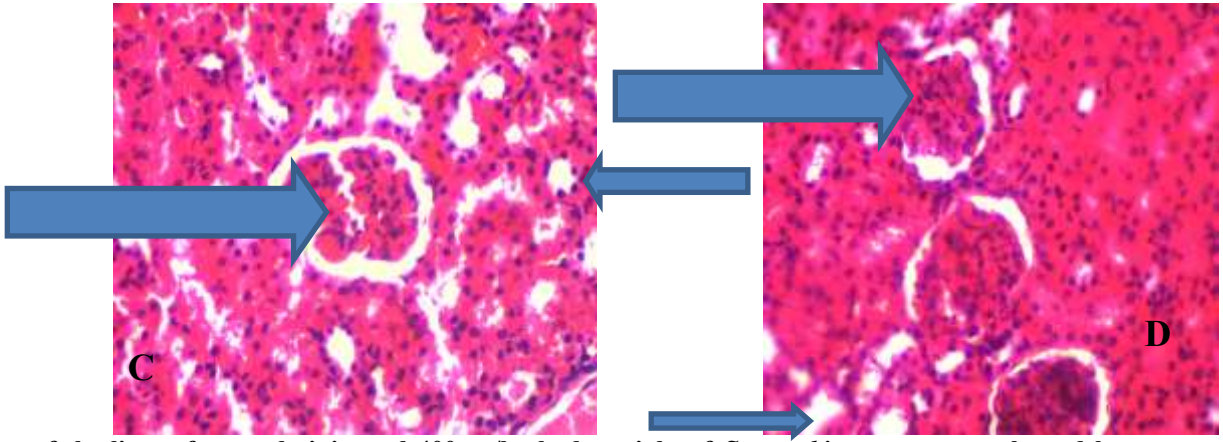

Plate 1.2C: Section of the liver of rats administered 400mg/kg body weight of *S. mombin* root extract showed hepatocytes (arrow) with eosinophilic cytoplasm surrounding a centrally placed normochromic nuclei with indistinct nucleoli. Features In Keeping with Normal Hepatocytes H and E Mag x400.

Plate 1.2D: Section of the kidney from rats administered *S. mombin* root extract at a dose of 600 mg/kg revealed normal glomeruli (thick arrow) containing normal mesangium, blood vessels and epithelium. The tubules (thin arrow) are oval shaped and lined by cuboidal epithelium with some tubules containing pale eosinophilic material. Features are in keeping with Normal Kidney H and E Mag x400.

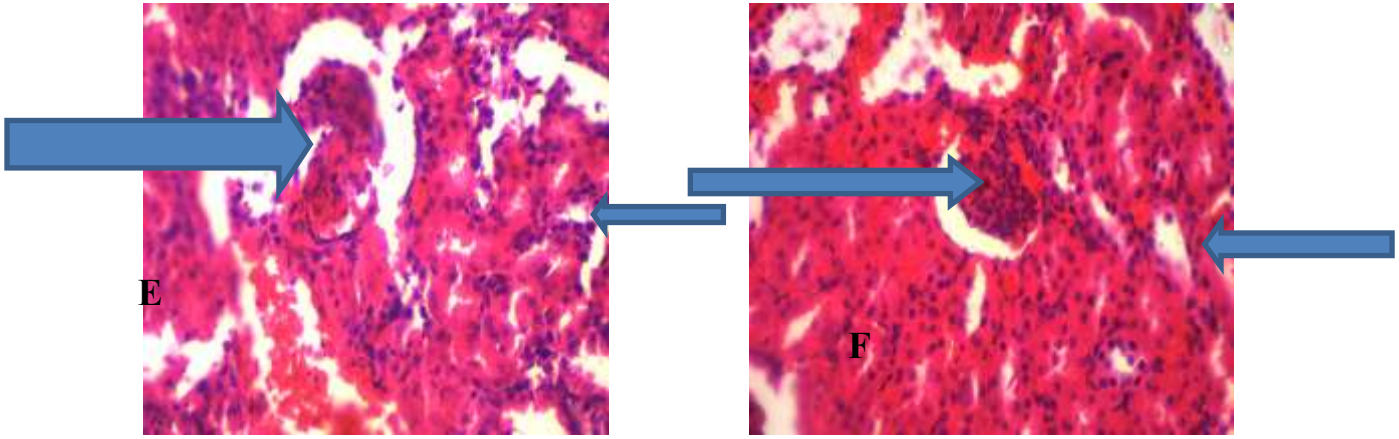

Plate 1.2E: Section of the kidney from rats administered *S. mombin* root extract at a dose of 800 mg/kg revealed normal glomeruli (thick arrow) containing normal mesangium, blood vessels and epithelium. The tubules (thin arrow) are oval shaped and lined by cuboidal epithelium with some tubules containing pale eosinophilic material. Features are in keeping with Normal Kidney H and E Mag x400.

Plate 1.2F: Section of the kidney from rats administered *S. mombin* root extract at a dose of 1000 mg/kg revealed normal glomeruli (thick arrow) containing normal mesangium, blood vessels and epithelium. The tubules (thin arrow) are oval shaped and lined by cuboidal epithelium with some tubules containing pale eosinophilic material. Features are in keeping with Normal Kidney stained with H and E (x400 magnification).

Table 1.1: Initial and Final Body Weights, Liver and Kidney Weights of Adult Wistar Rats Exposed to Lead and Treated with *Spondias mombin* Root Extract

Group	Initial Weight (g)	Final Weight (g)	Liver Weight (g)	Kidney Weight (g)
A	130.50 ± 4.50^{b}	162.00 ± 1.00^{b}	7.45 ± 1.65	1.20 ± 0.00
B	116.33 ± 3.28^{a}	134.33 ± 4.37^{a}	7.23 ± 0.46	0.70 ± 0.06
C	130.25 ± 7.88^{b}	151.50 ± 6.98^{ab}	8.38 ± 0.55	1.18 ± 0.11
D	129.00 ± 6.31^{b}	163.00 ± 4.32^{b}	9.03 ± 0.70	1.05 ± 0.09
E	156.00 ± 3.89^{c}	196.25 ± 6.43^{c}	7.40 ± 0.53	0.88 ± 0.19
F	138.00 ± 3.63^{b}	161.00 ± 6.87^{b}	7.28 ± 0.30	0.83 ± 0.09
p-value	0.004	<0.001	0.253 (ns)	0.080 (ns)

Values are presented as mean ± SEM (n = 2–4 per group). Superscript letters indicate statistical groupings from Tukey HSD post hoc test; groups sharing the same letter are not significantly different at p<0.05.

Table 1.2: Biochemical Parameters of Adult Wistar Rats Exposed to Lead and Treated with *Spondias mombin* Root Extract

Parameter	A	B	C	D	E	F	p-value
Urea (mg/dL)	26.00 ± 4.00	24.50 ± 0.50	33.50 ± 0.50	28.50 ± 1.50	32.50 ± 0.50	30.50 ± 3.50	0.153
Creatinine (µmol/L)	130.50 ± 0.50^{b}	136.50 ± 0.50^{b}	138.50 ± 0.50^{b}	136.50 ± 2.50^{b}	136.00 ± 1.00^{b}	127.50 ± 1.50^{a}	0.006*
Total Bilirubin (mg/dL)	3.65 ± 0.25	4.00 ± 0.30	4.10 ± 0.20	4.40 ± 0.10	4.30 ± 0.30	3.90 ± 0.00	0.309
Conjugated Bilirubin (mg/dL)	0.30 ± 0.10	0.45 ± 0.05	0.55 ± 0.05	0.55 ± 0.05	0.50 ± 0.10	0.50 ± 0.00	0.227
AST (U/L)	0.075 ± 0.025	0.075 ± 0.025	0.100 ± 0.000	0.100 ± 0.000	0.026 ± 0.024	0.100 ± 0.000	0.126
ALT (U/L)	0.050 ± 0.000	0.0025 ± 0.000	0.026 ± 0.024	0.0025 ± 0.000	0.0025 ± 0.000	0.026 ± 0.024	0.212
ALP (U/L)	9.00 ± 2.00	8.50 ± 0.50	8.00 ± 1.00	8.00 ± 0.00	8.50 ± 0.50	7.00 ± 0.00	0.762

Values are expressed as Mean ± SEM (n = 2 per group). Superscripts (^{a},^{b}) denote statistically significant differences between groups according to Tukey's post hoc test at p < 0.05, with groups sharing the same superscript letter not significantly different, while * indicates a significant ANOVA result at p < 0.05.

DISCUSSION

This study evaluated the histomorphological and biochemical effects of *S. mombin* root extract on the liver and kidneys of Wistar albino rats. The findings demonstrated that the extract induced dose-dependent alterations, particularly in hepatic tissues, while renal morphology and function were largely preserved.

Significant body-weight changes were observed, with the highest gain in the 800 mg/kg group and the lowest in the 200 mg/kg group. This suggests a possible metabolic effect of the extract at certain doses, which aligns with previous studies attributing weight modulation to phytochemicals such as saponins and flavonoids that influence nutrient absorption and metabolism [1,2]. Organ weights did not differ significantly across groups, consistent with reports that relative liver and kidney weights are less sensitive markers of subacute toxicity compared to biochemical and histological changes [3].

Biochemical analysis revealed stable serum electrolytes, urea, bilirubin, and liver enzymes across most treatment groups. However, a significant reduction in serum creatinine at the 1000 mg/kg dose indicates a possible nephroprotective influence of *S. mombin* or enhanced renal clearance mechanisms, corroborating previous reports of its antioxidant and haematinic activities [4]. Nevertheless, histological evaluation showed preserved renal architecture across all groups, confirming that the kidney may be less vulnerable to short-term exposure.

In contrast, the liver exhibited dose-dependent alterations. While hepatic architecture remained intact up to 600 mg/kg, higher doses (800-1000 mg/kg) induced ballooning degeneration and

microvesicular steatosis. These findings suggest hepatocellular lipid accumulation and injury at higher exposures, in agreement with earlier studies that documented hepatotoxicity associated with excessive intake of polyphenol-rich plant extracts [5,6]. The dual hepatoprotective and hepatotoxic effects observed may be attributed to the concentration-dependent actions of phytoconstituents such as flavonoids and tannins, which exhibit both antioxidant and pro-oxidant properties under different metabolic conditions [7].

The overall findings indicate that *S. mombin* root extract is relatively safe at low to moderate doses but carries hepatotoxic risks at higher concentrations. These results extend existing literature [8-11] by providing histological evidence of dose-dependent hepatic alterations, while also confirming renal tolerance within the same exposure window. Importantly, this study emphasizes the need for caution in the unregulated use of *S. mombin* and other herbal preparations, especially at high doses and prolonged durations.

Further studies should include chronic toxicity assessments, mechanistic investigations of the implicated phytoconstituents, and evaluation in other organ systems. Such data will be crucial for establishing safe therapeutic ranges and harnessing the medicinal benefits of *S. mombin* while minimizing adverse outcomes.

CONCLUSION

This study demonstrates that *Spondias mombin* root extract is relatively safe at low-to-moderate doses, but prolonged exposure to higher doses may induce dose-dependent hepatocellular injury while sparing renal architecture. A key strength of this work lies in its combined biochemical and histological evaluation, which provides complementary insights into organ function and structure. However, the relatively small sample size and limited duration represent important constraints that may not capture chronic or subtle toxicological changes. Overall, the findings provide valuable preliminary evidence for the safe use of *S. mombin*, while highlighting the need for extended studies to establish long-term safety and define therapeutic limits.

Declaration by Authors

Ethical Approval: Approved

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Conflict of Interest: The authors declare no conflict of interest.

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