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PHARMACOLOGICAL EVALUATION, ANTIOXIDANT AND HEPATOPROTECTIVE ACTIVITY OF *SESBANIA CANNABINA* LEAVES EXTRACT IN EXPERIMENTAL RATS

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ABSTRACT

The present study evaluates the hepatoprotective potential of the methanolic extract of *Sesbania cannabina* against paracetamol-induced hepatotoxicity. Preliminary phytochemical screening confirmed the presence of alkaloids, glycosides, carbohydrates, flavonoids, saponins, phenolics and tannins. Acute oral toxicity assessment revealed no mortality up to 2000 mg/kg, allowing selection of 100 and 200 mg/kg doses for pharmacological studies. The extract demonstrated significant antioxidant activity, with 60.13% DPPH radical inhibition and an IC_{50} value of 49.37 µg/mL, compared to ascorbic acid (85.41%; IC_{50}: 20.77 µg/mL). Hepatoprotective evaluation was carried out using paracetamol-induced hepatotoxicity, wherein serum biomarkers including SGOT, SGPT, ALP and total bilirubin were significantly elevated following toxic insult. Treatment with *Sesbania cannabina* extract produced a dose-dependent restoration of these biochemical markers toward normal values. The 200 mg/kg dose exhibited more prominent hepatoprotective activity than 100 mg/kg. Histopathological findings supported the biochemical results, showing reduced tissue injury in treated groups. Overall, the methanolic extract of *Sesbania cannabina* demonstrated significant hepatoprotective and antioxidant effects, supporting its traditional use in liver-related ailments.

KEYWORDS: *Sesbania cannabina*; paracetamol-induced hepatotoxicity; hepatoprotective activity; antioxidant activity; DPPH assay; liver biomarkers; phytochemicals; traditional medicine.

1. INTRODUCTION

The liver is a vital metabolic organ responsible for detoxification, protein synthesis, and biochemical regulation essential for maintaining homeostasis (Trefts et al., 2017). Exposure to hepatotoxic agents, alcohol, environmental chemicals, or therapeutic drugs can lead to hepatic cellular injury, thereby compromising physiological function. Among various hepatotoxins, paracetamol (acetaminophen) remains one of the most widely used analgesic–antipyretic agents capable of inducing dose-dependent liver damage (Lancaster & Lawrence, 2021). Paracetamol overdose triggers oxidative stress, glutathione depletion, and formation of a reactive metabolite, N-acetyl-p-benzoquinone imine (NAPQI), ultimately causing hepatocyte necrosis (Jaeschke et al., 2012).

Due to the limitations and adverse effects associated with current hepatoprotective drugs, there is continuous scientific interest in identifying new, safe, and effective hepatoprotective agents from natural sources (Madrigal-Santillán et al., 2014). Medicinal plants have long been recognized in traditional systems of medicine for their ability to mitigate hepatic injury due to their rich phytochemical composition, including flavonoids, alkaloids, glycosides, and phenolic compounds exhibiting antioxidant and anti-inflammatory properties (Muthuraman et al., 2020).

Sesbania cannabina (Fabaceae), traditionally used for the treatment of inflammatory conditions, fever, and hepatic disorders, contains flavonoids, tannins, alkaloids, saponins, and phenolic compounds known for their free-radical scavenging and cytoprotective properties (Kaur & Kalia, 2021). Preliminary phytochemical studies reveal that these phytoconstituents may contribute to hepatoprotective effects by enhancing antioxidant defense and protecting cell membrane integrity (Saha et al., 2022). Moreover, antioxidant assays such as DPPH have confirmed the radical-scavenging potential of *Sesbania cannabina*, suggesting that its biological activity may be associated with phytochemical synergy.

Given the ethnomedicinal significance and reported phytoconstituents of *Sesbania cannabina*, the present study investigates the hepatoprotective potential of its methanolic extract against paracetamol-induced hepatotoxicity in animal models. Serum hepatic biomarkers, antioxidant activity, and histopathological assessment were conducted to evaluate the protective efficacy of the plant extract.

2. MATERIALS AND METHODS

Plant Material and Authentication

Fresh fruits of *Sesbania cannabina* (300 g) were collected, washed, and shade-dried for three days, followed by oven-drying at 45 °C for three additional days. The dried material was stored in airtight glass containers at room temperature. The plant was authenticated by a qualified plant taxonomist, and a voucher specimen no. **203/Saif./Sci./Clg/Bpl** was retained for reference.

Extraction of Plant Material

Dried fruits were powdered and extracted by continuous hot percolation using a Soxhlet apparatus. The material was first extracted with petroleum ether (non-polar solvent) at 60 °C, followed by methanol. Each extraction was continued until the solvent in the siphon tube was colorless. Extracts were concentrated under reduced pressure at 40 °C using a rotary evaporator, weighed, and percent yield was calculated. Extracts were stored in airtight containers under refrigeration until use. **(Baidya *et al*. 2002).**

Phytochemical Screening

Preliminary phytochemical screening of the methanolic extract of *Sesbania cannabina* was performed using standard qualitative procedures to determine the presence of major phytoconstituents, including carbohydrates, alkaloids, saponins, steroids, phenolics, tannins, flavonoids, glycosides, and fixed oils **(Kokate et al., 2000).**

DPPH

The antioxidative capacity of *Sesbania cannabina* using the DPPH test, the extract was found for free radical detoxification 1 mg/ml methanol solution of extracts/standard was prepared.

Different concentration of *Sesbania cannabina* extracts/standard (20 – 100µg/ml) was prepared from 1mg/mL stock solution and 2mL of 0.1mM DPPH solution was added. The resultant mixture was vortexed, allowed to sit at standard temperature in a somewhat dark environment for 30 minutes, and then its wavelength was measured at 517 nanometer using a UV spectrophotometer (Shimadzu 1700). For control, Take 3 millilitre of 0.1mM DPPH mixture, then cultivated for 30 min at satndard temperature in dark condition. Absorbance of the regulating was taken against methanol (as blank) at 517 nm **(Athavale *et al.*, 2012).**

Percentage antioxidant capacity of sample/standard was calculated by using formula:

% Inhibition = [(Ab of control- Ab of sample)/ Ab of control x 100]

FT-IR

To establish the existence of the functional divisions within the methanolic extract, FT-IR spectroscopy was carried out with a Perkin Spectrum BX spectrophotometer. The sample was dried and ground with KBr pellets and analyzed on Thermo Nicolet model 6700 spectrum instrument. A mixture of 2% finely dried sample was used to make a disk containing 100 mg of KBr, which was subsequently analyzed using an IR spectrometer. There were infrared spectra reported in the vicinity of 400 - 4,000 cm-1 **(Luciene *et al.*, 2008).**

Study of Acute Toxicity

The acute toxicology course approach set out in guideline is a methodical process using three animals of the same sex for each step. An assessment of the test substance's acute toxicity may need, on average, two to four phases, contingent on the animals' mortality and/or moribund state. One of the specified doses of the material is given orally to a group of experimental animals. Three animals of the same sex are used in each phase of the methodical testing process for the chemical. The next course of action, which entails treating three more animals at the same dose and three more animals at the next higher or lower dose level, will depend on whether compound-related mortality of the animals dosed at one step is present or absent. For every step, three animals are utilized. The dose level to be used as the starting dose is selected from one of four fixed levels, body weight in mg/kg, 5, 50, 300, and 2000 **(Guideline Document., 1996).**

In-vivo Experimental work

IAEC Approval: All animal experiments were approved by the Committee on Institutional Animal Ethics (IAEC). CCSEA Registration No.**1647/PO/Re/S/l 1/CCSEA**

Housing Condition: Albino wistar rats, male aged 2-3 months and weighing 200-250 were taken for the study. The animals were maintained in standard animal house conditions with ad libitum food and water access under a 12 hour light dark cycle prior to treatment.

Paracetamol-induced hepatotoxicity

When accidentally overdosed (which can happen in elderly people and alcoholics), the commonly used analgesic-antipyretic medication paracetamol (acetaminophen) causes severe liver damage. The oxidation product of paracetamol, N-acetyl-p-benzoquinoneimine, covalently binds to protein sulphydryl groups, resulting in lipid peroxidation and necrosis in cells. This hepatotoxic effect raises serum marker enzyme levels, including SGOT, SGPT, ALP and TB, or total bilirubin **(Parmar *et al.*, 2010).**

Experimental design

Either sex of Wistar albino rats were split up into five groups of 6 animals each and are given orally the following treatment for seven days.

Group 1: Functioned as a typical control group and were given 1% sodium. carboxy methyl cellulose (Na.CMC) 1 mL/kg bw, p.o.

Group 2: Taken 1 g/kg bw p.o. of paracetamol (paracetamol control).

Group 3: Received both 50 mg/kg bw, p.o. silymarin and paracetamol dose.

Group 4: Represented test treatment in which rats received treatment with the 1st dose of *Sesbania cannabina* extracts at dosages of 100 mg/kgbw along with paracetamol.

Group 5: Represented test treatment in which rats received treatment with the 2nd dose *Sesbania cannabina* extracts at dosages of 200 mg/kgbw along with paracetamol.

Biochemical studies

The chemical parameters were determined following a 24-hour fasting after administration of last dose of treatment. Blood was acquired from all animal by puncturing retro-orbital plexus. The sample of blood is allowed to for 45 minute at room temperature. Serum was separated by centrifugation at 4^0 C for 15 minute and utilized for the estimation of various bio-chemical parameters namely SGOT, SGPT, ALP, and serum bilirubin.

ALP (Alkaline phosphatase)

The pH level 10.3 alkaline phosphatase (ALP) catalyses the breakdown of colourless p- Nitrophenyl phosphate (pNPP) to yellow coloured p-Nitrophenol and phosphate. Change in At 405 nm, fluorescence as a consequence of the creation of yellow color is determined energetically and is proportional to the ALP action within the sample **(Brien *et al.*, 2002).**

Prepare "Working reagent" by reconstituting one Reagent 2 vial (pNPP substrate) with 5.5 mL Reagent 1 (AMP buffer) were added. To 20 µL of animal serum sample added 1000 µL of Reagent A and mixed well. The mixture is kept incubate at the assay temperature of 370C for 1minute. The absorbance was read after 30 seconds. Repeat reading following each 30 seconds i.e. upto 120 seconds at 405 nanometer wavelength. The change in mean absorbance per minute was calculated.

AST (Aspartate Transaminase) or SGOT (Glutamic-oxalacetic transaminase)

Aspartate Transaminase or AST catalyses the transamination of L-Aspartate and α-ketoglutarate to produce oxaloacetate and L-glutamate. In subsequent reaction, MDH, or malate dehydrogenase reduces Oxaloacetate to Malate with simultaneous oxidation of Nicotinamide Adenine Dinucleotide (reduced) (NADH) to Nicotinamide Adenine Dinucleotide (NAD). The rate of oxidation of NADH is measured kinetically by monitoring the decrease in 340 nm absorbance and is directly proportional to AST activity in sample. Lactate Dehydrogenase (LD) is added to enzyme system to prevent endogenous pyruvate interference, which is normally present in the serum.

Prepare "Working reagent" by reconstituting reagent 1 in 4 mL + Reagent 2 in 1 mL were added. Up to 100 µLof animal serum sample added 1000 µL about Reagent 1 and mixed well. Incubation of the mixture occurs at the assay temperature of 37^oC for 1minute. Then added Reagent 2 in a milliliter and mixed well. The reading for absorbance after 60 seconds. Repeat perusing following each 30 second i.e. up to 120 seconds at 340 nanometer wavelength. The change in mean absorbance per minute was calculated**.**

AST activity (IU/L) =Δ A/minute × Kinetic Factor

Where

Δ A/minute = Change in absorbance per minute

Kinetic factor (K) = 1768.

ALT (Alanine Transaminase) or SGPT, or glutamic-pyruvic transaminase

Alanine Transaminase or ALT catalyses the transamination of L-Alanine and α-Ketoglutarate to produce L-glutamate and pyruvate. In subsequent reaction, Lactate Dehydrogenase (LD) reduces Pyruvate to Lactate conversion with simultaneous oxidation of Nicotinamide Adenine Dinucleotide [reduced] (NADH) into Nicotinamide Adenine Dinucleotide (NAD). The rate of oxidation of NADH is measured kinetically by monitoring the decrease in at 340 nm in absorbance. During the first incubation phase, LD quickly and totally eliminates endogenous sample pyruvate to prevent interference with the test**.** Working Reagent 1 of 4 mL + Reagent 2 of 1mL**.** Up to 100 µL of animal serum sample added 1000 µL of working ALT Reagent 1 and mixed well. The blend is kept incubating at 37^0C for 1minute. The absorbance was read after 60 seconds. Repeat perusing following each 30 seconds i.e. up to 120 seconds at 340 nanometerwavelength. The average change in absorbance per minute was calculated**.**

ALT activity (IU/L) = ΔA/minute × Kinetic factor

Where

Δ A/minute = Change in absorbance per minute

Kinetic factor (K) = 1768.

Total Bilirubin

Total bilirubin in serum or plasma is determined using jendrassik and Grof method by coupling with the addition of sodium acetate, sodium benzoate, and caffeine, with diazotized sulfanilic acid. Alkaline Fehling solution II forms a blue azobilirubin, which is measured photometrically **(Dybkaer *et al.*, 1970).** Mix Reagent 1 and Reagent 2 in the ratio of 4 + 1 (e. g. 400 µl of sulfanilic acid solution and 100 µl of sodium nitrite solution) to make a diazo solution. The mixing ratio should be observed exactly. Always use freshly prepared diazo solution. The absorbance was read at 546 nm wavelength.

3. RESULTS AND DISCUSSIONS

Result of Percentage Yield

In phytochemical extraction the yield percentage is very crucial to ascertain the standard efficiency of extraction for a specific plant, various sections of the same plant or different solvents used. The yield extracts received from the *Sesbania cannabina* shown in Table: 1

Table 1: The percentage yield of *Sesbania cannabina* crude extracts.

S.No	Plant name	Solvent	Theoretical weight	Yield(gm)	% yield
1	*Sesbania cannabina*	Pet ether	285	1.48	0.51%
2		Methanol	299	6.07	2.03%

Results of Preliminary Phytochemical study

Table 2: Phytochemical testing of *Sesbania cannabina.*

S. No.	Experiment	Presence or absence of phytochemical test	
		Pet. Ether extract	Methanolic extract
1	Alkaloids	-	+
2	Glycoside	+	+
3	Carbohydrates	-	+
4	Proteins and Amino Acids	-	-
5	Flavonoids	-	-
6	Tannin and Phenolic Compounds	-	+
7	Saponin	+	+
8	Triterpenoids and Steroids	-	-

Result of *In vitro* Antioxidants assay

In the current study, *in vitro* anti-oxidant activity including *Sesbania cannabina* extracts was evaluated by activity of DPPH radical scavenging.

DPPH 1, 1- diphenyl-2-picryl hydrazyl Assay

Table 3: Std's ability to scavenge DPPH radicals with ascorbic acid.

Concentration (µg/ml)	Absorbance	% Inhibition
20	0.479	51.810
40	0.433	56.438
60	0.342	65.593
80	0.283	71.529
100	0.145	85.412
Control	0.994	
IC50	**20.77**	

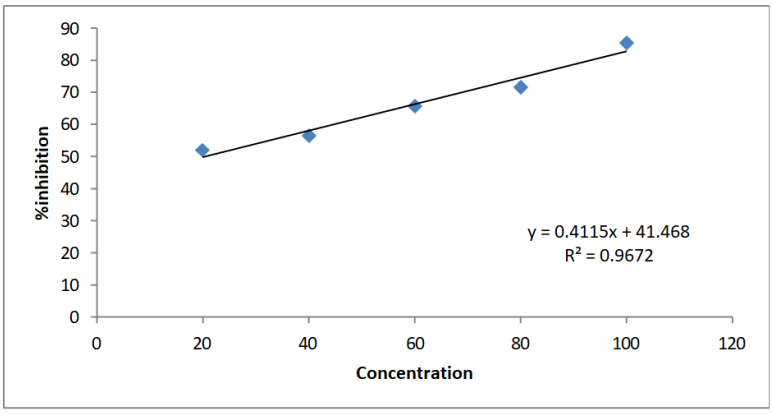


Figure 1: Std. Ascorbic acid's ability to scavenge DPPH radicals.

Table 4: The ability of *Sesbania cannabina's* methanol extract to scavenge DPPH radicals.

Concentration (µg/ml)	Absorbance	% Inhibition
20	0.519	43.770
40	0.463	49.837
60	0.451	51.137
80	0.412	55.362
100	0.368	60.130
Control	0.923	
IC50	**49.37**	

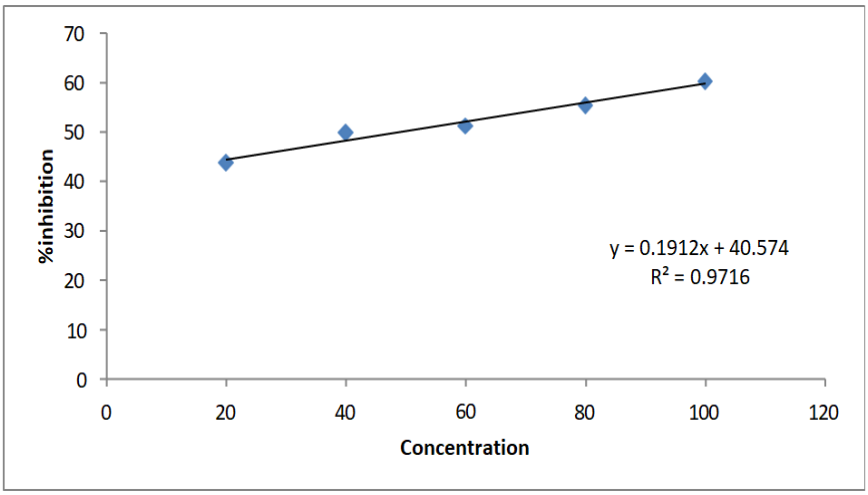


Figure 2: Represents the Percentage Inhibition *Vs* Concentration of Sesbania cannabinoid extract.

Results of Functional group identified by FTIR Study

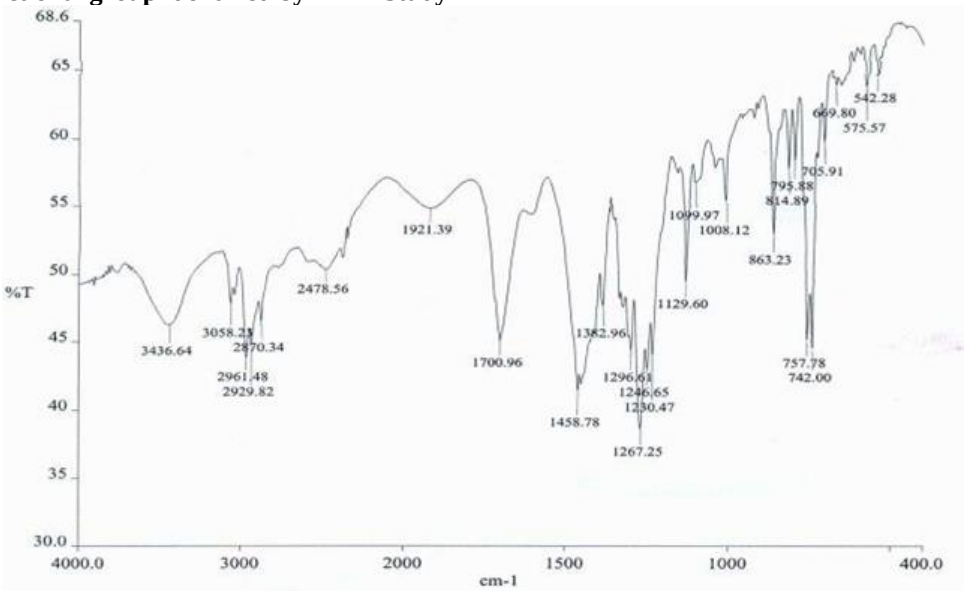
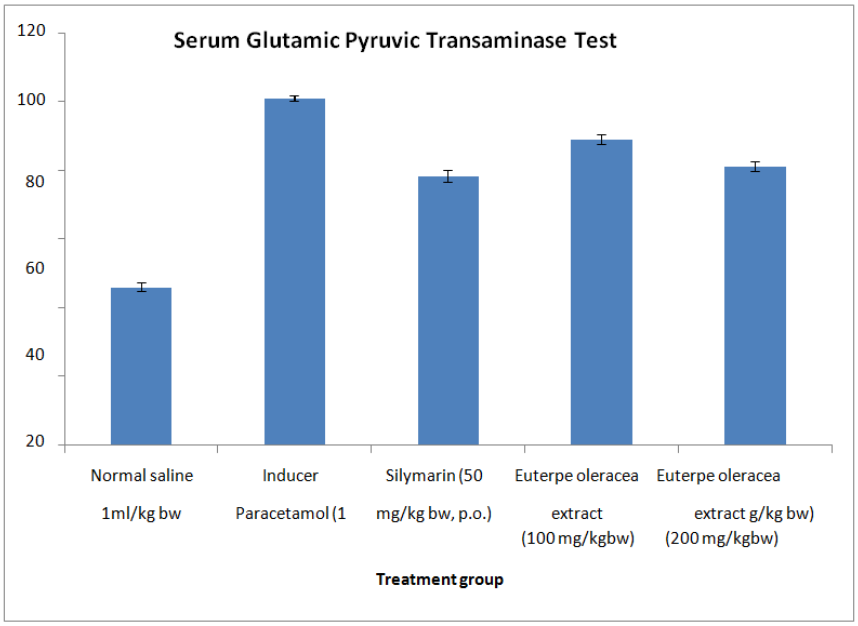


Figure 3: FTIR of *Sesbania cannabina.*

Results of SGPT, or serum glutamic pyruvic transaminase test

Table 5: Results of the serum glutamic pyruvic transaminase test.

S.No	Treatment Group	Results (Mean±SD)
1.	Normal saline 1ml/kg bw	45.88±1.26
2.	Inducer Paracetamol(1 g/kg bw)	101±0.91
3.	Silymarin (50 mg/kg bw, p.o.)	78.39±1.72
4.	*Sesbania cannabina* extract(100 mg/kgbw)	88.99±1.33
5.	*Sesbania cannabina* extract(200 mg/kgbw)	80.99±1.41

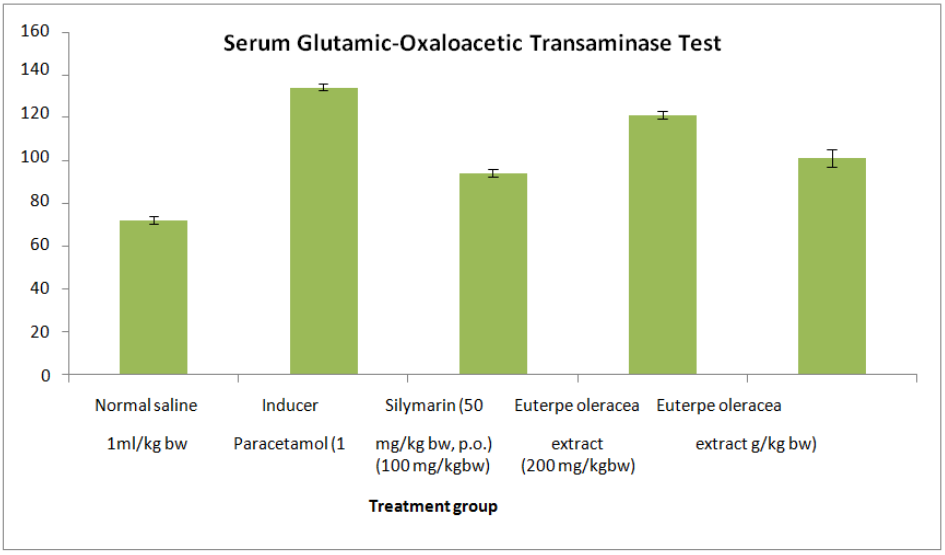


Graph 1: The findings of the serum glutamic pyruvic transaminase test are displayed as a bar graph.

Results of test for serum glutamic-oxaloacetic transaminase (SGOT)

Table 6: Results of the serum glutamic-oxaloacetic transaminase test.

S.No	Treatment Group	Results (Mean±SD)
1.	Normal saline 1ml/kg bw	71.99±1.74
2.	Inducer Paracetamol(1 g/kg bw)	134±1.61
3.	Silymarin (50 mg/kg bw, p.o.)	93.98±1.74
4.	*Sesbania cannabina* extract(100 mg/kgbw)	120.99±1.83
5.	*Sesbania cannabina* extract(200 mg/kgbw)	100.91±4.01

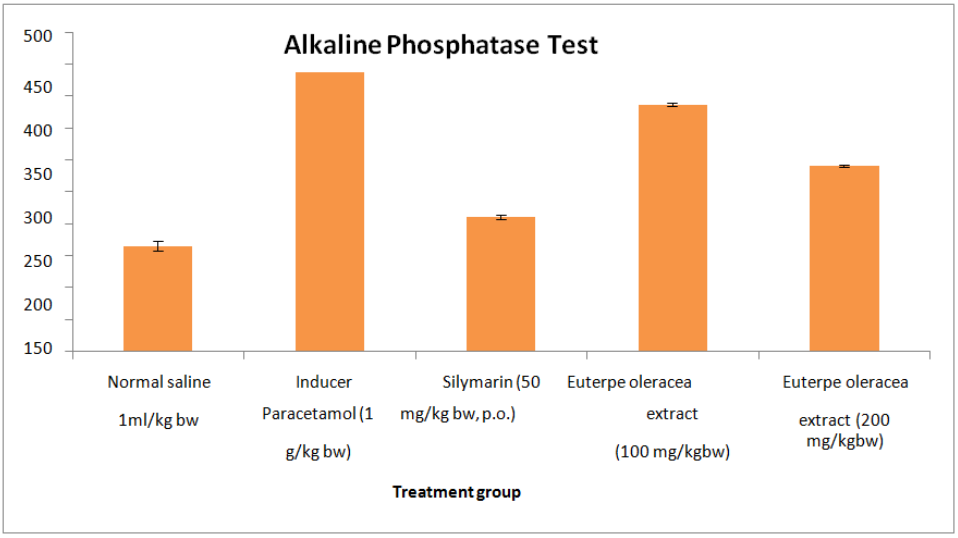


Graph 2: **Bar graph represents the Serum Glutamic Oxaloacetic Transaminase test results.**

Results of Alkaline Phosphatase *(ALP)* Test

Table 7: **Alkaline Phosphatase *(ALP)* Test results.**

S.No	Treatment Group	Results (Mean±SD)
1.	Normal saline 1ml/kg bw	164±7.48
2.	Inducer Paracetamol(1 g/kg bw)	437±5.04
3.	Silymarin (50 mg/kg bw, p.o.)	209.95±2.80
4.	*Sesbania cannabina* extract(100 mg/kgbw)	386.10±2.25
5.	*Sesbania cannabina* extract(200 mg/kgbw)	290.34±2.09

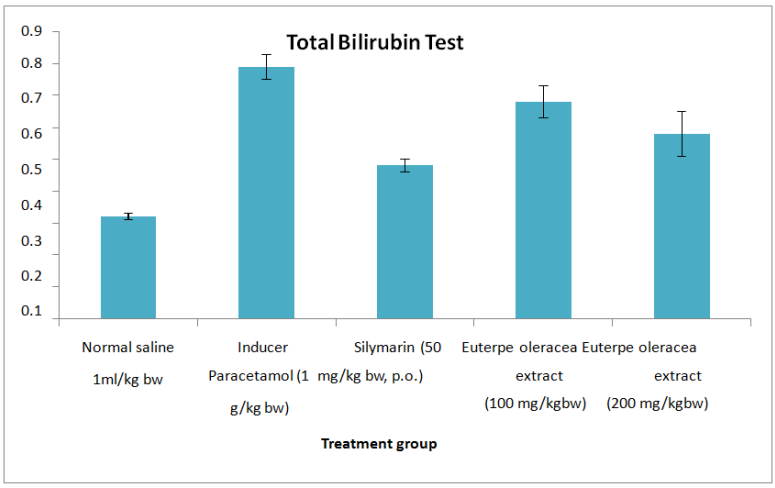


Graph 3: **Bar graph represents the Alkaline Phosphatase *(ALP)* test results.**

Results of Total Bilirubin test

Table 8: **Total Bilirubin Test results.**

S.No	Treatment Group	Results (Mean±SD)
1.	Normal saline 1ml/kg bw	0.32±0.01
2.	Inducer Paracetamol(1 g/ksgbw)	0.79±0.04
3.	Silymarin (50 mg/kg bw, p.o.)	0.48±0.02
4.	*Sesbania cannabina* extract(100 mg/kgbw)	0.68±0.05
5.	*Sesbania cannabina* extract(200 mg/kgbw)	0.58±0.07



Graph 4: Bar graph represents the results of the total bilirubin test.

Total bilirubin concentration = A × 10.3 mg/d

4. CONCLUSION

The methanolic extract of *Sesbania cannabina* exhibited significant hepatoprotective activity against paracetamol-induced liver damage. Treatment effectively restored serum enzyme levels—including SGOT, SGPT, ALP, and total bilirubin—toward normal values, suggesting preservation of hepatic cellular integrity. The hepatoprotective effect was dose-dependent, with the 200 mg/kg dose producing stronger responses than 100 mg/kg. Antioxidant activity demonstrated through DPPH radical scavenging further supports its protective mechanism. Histopathological observations corroborated the biochemical findings, indicating reduced hepatic injury following extract administration. The hepatoprotective effects may be attributed to the presence of various bioactive phytoconstituents such as flavonoids, phenolics, saponins, and tannins. These results validate the traditional use of *Sesbania cannabina* in liver disorders and suggest its potential as a natural therapeutic agent for hepatoprotection. Further studies are warranted to isolate active constituents and elucidate underlying molecular mechanisms.

5. REFERENCES

1. Jaeschke, H., Xie, Y., & McGill, M. R. Acetaminophen-induced liver injury: From animal models to humans. *Journal of Clinical and Translational Hepatology,* 2012; *1*(3): 1–11.
2. Kaur, R., & Kalia, A. N. Phytochemical and pharmacological potential of *Sesbania cannabina*: A review. *Journal of Ethnopharmacology,* 2021; *273:* 113992.
3. Lancaster, E. M., & Lawrence, S. P. Acetaminophen toxicity: Epidemiology, clinical features, and management. *New England Journal of Medicine,* 2021; *384*(24): 2401–2410.
4. Madrigal-Santillán, E., Madrigal-Bujaidar, E., Álvarez-González, I., Sumaya-Martínez, M. T., & Morales-González, J. A. Review of natural products with hepatoprotective effects. *World Journal of Hepatology,* 2014; *6*(10): 614–621.
5. Muthuraman, A., Ramesh, M., & Sen, S. Medicinal plants with hepatoprotective activities: A review. *Pharmacognosy Reviews,* 2020; *14*(28): 56–70.
6. Saha, P., Banerjee, S., & Ghosh, S. Phytochemical analysis and antioxidant capacity of *Sesbania cannabina* extracts. *Asian Journal of Pharmaceutical and Clinical Research,* 2022; *15*(6): 89–95.
7. Trefts, E., Gannon, M., & Wasserman, D. H. The liver. *Comprehensive Physiology,* 2017; *7*(1): 577–610.
8. Baidya, B., Gupta, S. K., & Mukherjee, T. An extraction-based verification methodology for MEMS. *Journal of Microelectromechanical Systems*, 2002; *11*(1): 2-11.
9. Kokate CK, Purohit AP and Gokhale SB (2000). Textbook of Pharmacognosy, NiraliPrakashan.
10. Athavale, A., Jirankalgikar, N., Nariya, P., & Des, S. Evaluation of *In-vitro* antioxidant activity of panchagavya: a traditional ayurvedic preparation. *Int J Pharm Sci Res.*, 2012; *3:* 2543-9.
11. Parmar, S. R., Vashrambhai, P. H., & Kalia, K. Hepatoprotective activity of some plants extract against paracetamol induced hepatotoxicity in rats. J Herbal Med Toxicol, 2010; 4(2): 101-106.
12. O'brien, P. J., Slaughter, M. R., Polley, S. R., & Kramer, K. Advantages of glutamate dehydrogenase as a blood biomarker of acute hepatic injury in rats. *Laboratory animals*, 2002; *36*(3): 313-321.
13. Dybkaer, R., & Hertz, H. A reference for determination of bilirubin concentration in serum using bilirubin in cyanide-formamide for enrichment of serum. *Scandinavian Journal of Clinical and Laboratory Investigation*, 1970; *25*(2): 151-160.
14. Guideline Document on Acute oral Toxicity Testing, Series on Testing and Assessment No. 423. Paris: OECD Environment, Health and Safety Publications; 1996. Available from: http://www.oecd.org/ehs