

To Evaluate the Antihyperlipidemic Activity of *Flagellaria Indica* Leaves in High-Fat Diet Induced Hyperlipidemia in Wistar Rats

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Abstract

Background: Hyperlipidemia is a major metabolic disorder associated with increased serum lipids, oxidative stress, hepatic dysfunction, and an increased risk of cardiovascular complications. The present study was designed to evaluate the antihyperlipidemic, antioxidant, and hepatoprotective effects of *Flagellaria indica* leaf extract in high-fat diet-induced hyperlipidemic Wistar rats.

Methods: Leaves of *Flagellaria indica* were collected, authenticated, shade dried, powdered, and extracted with ethanol. Preliminary phytochemical screening was performed to identify the major phytoconstituents. Hyperlipidemia was induced in Wistar rats using a high-fat diet. Animals were divided into normal control, hyperlipidemic control, standard atorvastatin-treated, and *F. indica* extract-treated groups. The extract was administered orally at doses of 100, 200, and 400mg/kg body weight, while atorvastatin 10 mg/kg was used as the standard drug. Body weight, serum lipid profile, liver function parameters, antioxidant markers, and hepatic histopathology were evaluated.

Results: Phytochemical screening revealed the presence of several important secondary metabolites, including flavonoids, phenolics, tannins, saponins, alkaloids, glycosides, steroids, and terpenoids. High-fat diet administration produced marked dyslipidemia, characterized by increased total cholesterol, triglycerides, LDL and VLDL with decreased HDL levels. Treatment with *F. indica* extract produced a dose-related improvement in the lipid profile. The extract also improved altered liver function parameters, including AST, ALT, ALP, total bilirubin, total protein, and albumin. Antioxidant evaluation demonstrated improvement in SOD, CAT, and GSH levels compared with the hyperlipidemic control group. Histopathological examination further showed progressive restoration of hepatic architecture and reduction of fatty and degenerative changes, particularly at the higher extract dose.

Conclusion: The present findings indicate that ethanolic leaf extract of *Flagellaria indica* possesses promising antihyperlipidemic, antioxidant, and hepatoprotective activities against high-fat diet-induced metabolic alterations in Wistar rats. The observed effects may be associated with the combined action of its bioactive phytoconstituents. Further studies are warranted to isolate the active constituents and elucidate their precise molecular mechanisms.

Keywords: *Flagellaria indica*, anti-hyperlipidemic activity, high-fat diet, hyperlipidemia, Wistar rats, lipid profile.

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Introduction

Hyperlipidemia: Hyperlipidemia is a metabolic disorder characterized by an abnormal elevation of lipids and lipoproteins in the blood, including total cholesterol (TC), triglycerides (TG), low-density lipoprotein (LDL), and very low-density lipoprotein (VLDL), along with a reduction in high-density lipoprotein (HDL). It is one of the major risk factors for the development of cardiovascular diseases such as atherosclerosis, coronary artery disease, and stroke.

The imbalance between lipid synthesis and degradation leads to lipid accumulation in the bloodstream, which deposits in arterial walls and contributes to plaque formation.

Pathophysiology of Hyperlipidemia: Hyperlipidemia is a complex metabolic disorder resulting from disturbances in lipid synthesis, transport, and clearance, leading to elevated levels of circulating lipids such as cholesterol and

triglycerides. However, disruption of this balance results in excessive lipid accumulation in the bloodstream, ultimately contributing to pathological conditions such as atherosclerosis and

cardiovascular diseases.

Plant Profile: *Flagellaria indica*



Figure 1: Plant Profile of *Flagellaria indica*

Pharmacological Activities

Antioxidant Activity: The plant exhibits strong antioxidant activity due to the presence of flavonoids and phenolic compounds, which neutralize free radicals and reduce oxidative stress.

Anti-inflammatory Activity: Extracts of *Flagellaria indica* have shown anti-inflammatory effects by inhibiting inflammatory mediators.

Antimicrobial Activity: The plant demonstrates antimicrobial activity against various bacterial and fungal strains.

Wound Healing Activity: Traditionally used in wound healing due to its ability to promote tissue regeneration.

Materials and Methods

Collection and Authentication: Fresh leaves of *Flagellaria indica* was collected from a Local area of the jaipur during the appropriate growing season in month of april. The collected plant material were thoroughly washed with water to remove soil particles, dust, and other extraneous matter. The plant was identified and authenticated by a qualified botanist.

Preparation of Extract: The dried powdered leaves of *Flagellaria indica* were subjected to Soxhlet extraction using ethanol as solvent. A known quantity of the powdered plant material was packed in a thimble and placed in the Soxhlet apparatus.

Preliminary Phytochemical Screening: The ethanolic extract of *Flagellaria indica* leaves were

subjected to qualitative phytochemical screening to detect the presence of major secondary metabolites and Standard qualitative chemical tests was carried out according to established pharmacognostic procedures.

Experimental Animals: Healthy adult Wistar albino rats of either sex, weighing between 150–200gm, was used in the present investigation. The entire experimental protocol was approved by the Institutional Animal Ethics Committee (IAEC).

All animal handling and experimental procedures were carried out in accordance with CCSEA guidelines for the care and use of laboratory animals. The animals was housed in clean polypropylene cages under standard laboratory conditions.

Acute Oral Toxicity Study: An acute oral toxicity study of the ethanolic extract of *Flagellaria indica* leaves was carried out according to OECD guideline 423.

Induction of Hyperlipidemia: Hyperlipidemia was induced in Wistar rats by feeding them with a high-fat diet (HFD) for a period of 28 days. The diet consisted of cholesterol (1%), cholic acid (0.5%), and coconut oil or animal fat (10%) mixed with standard pellet diet. The prepared diet was administered daily, while the normal control group received only standard pellet diet.

The animals was randomly divided into six groups, each comprising six animals.

- **Group I – Normal Control:** Received normal pellet diet and vehicle

- **Group II** – Disease Control: Received high-fat diet only
- **Group III** – Standard Drug: Received high-fat diet + Atorvastatin (10 mg/kg, p.o.)
- **Group IV** – Test Low Dose: Received high-fat diet + Ethanolic extract (100mg/kg, p.o.)
- **Group V** – Test Medium Dose: Received high-fat diet + Ethanolic extract (200mg/kg, p.o.)
- **Group VI** – Test High Dose: Received high-fat diet + Ethanolic extract (400mg/kg, p.o.)

The treatment were administered once daily for 28 consecutive days. This grouping allowed evaluation of disease progression, standard drug effect, and dose-dependent activity of the plant extract.

Biochemical Parameters

Observation of Body Weight: Body weight of all animals was recorded on day 0 and at regular intervals throughout the experimental period. Final body weight were noted on the last day of treatment.⁶³

Collection of Blood Samples: At the end of the treatment period, animals were fasted overnight. Blood samples was collected under light anesthesia from the retro-orbital plexus using a capillary tube.

Estimation of Serum Lipid Profile: Serum lipid profile were estimated using standard diagnostic kits and spectrophotometric methods.

Estimation of Liver Function Parameters: Liver function was assessed by estimating AST, ALT, ALP, total bilirubin, total protein, and albumin.

Estimation of Antioxidant Markers: Antioxidant status was assessed in tissue homogenate by estimating SOD, CAT, and GSH.

Histopathological Examination: After completion of the experiment, rats was sacrificed and liver tissues were isolated immediately. The tissues was washed with normal saline to remove blood and fixed in 10% formalin. Fixed tissues were dehydrated in graded alcohol, cleared with xylene, embedded in paraffin wax, and sectioned at 5 μ m thickness using a microtome. The sections were stained with hematoxylin and eosin and examined under light microscope.

Statistical Analysis: All experimental results was expressed as Mean $\pm$ SEM for six animals in each group. Statistical analysis were performed using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test. A value of $p < 0.05$ were considered statistically significant.

Results

Preliminary Phytochemical Screening: The preliminary phytochemical screening of the ethanolic extract of *Flagellaria indica* leaves was carried out to identify the presence of major secondary metabolites.

These phytoconstituents are known to possess significant pharmacological activities, particularly antioxidant and lipid-lowering properties, which support the antihyperlipidemic potential of the plant extract.

Table 1: Preliminary Phytochemical Screening of *Flagellaria indica* Leaf Extract

S. No.	Phytoconstituent	Result
1	Alkaloids	+
2	Flavonoids	+
3	Tannins	+
4	Phenolic Compounds	+
5	Saponins	+
6	Glycosides	+
7	Carbohydrates	+
8	Proteins	–
9	Amino Acids	–
10	Steroids	+
11	Terpenoids	+

(+): Present, (–): Absent

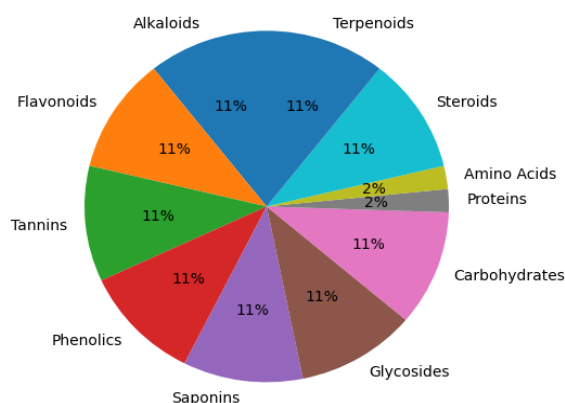


Figure 2: phytochemical constituents in Flagellaria indica leaf extract

Effect on Body Weight: In the present study, body weight of all animals was recorded at regular intervals during the experimental period. The disease control group showed a significant increase in body weight compared to the normal control

group, indicating successful induction of hyperlipidemia. Treatment with Flagellaria indica leaf extract at different doses resulted in a dose-dependent reduction in body weight gain.

Table 2: Effect of Flagellaria indica Leaf Extract on Body Weight (g)

Group	Treatment	Initial Body Weight (g)	Final Body Weight (g)	Change in Body Weight (g)
Group I	Normal Control	180 ± 5	195 ± 6	+15
Group II	Disease Control (HFD)	182 ± 6	230 ± 8	+48
Group III	Standard (Atorvastatin 10 mg/kg)	181 ± 5	200 ± 6	+19
Group IV	Extract (100 mg/kg)	180 ± 4	215 ± 7	+35
Group V	Extract (200 mg/kg)	182 ± 5	205 ± 6	+23
Group VI	Extract (400 mg/kg)	181 ± 6	198 ± 5	+17

Values are expressed as Mean ± SEM (n = 6)

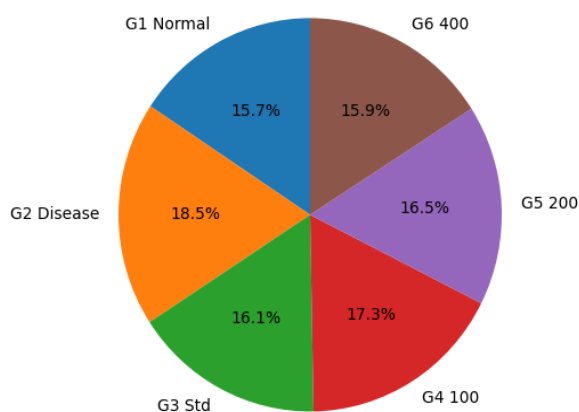


Figure 3: Flagellaria indica leaf extract on body weight

Treatment with Flagellaria indica leaf extract showed a dose-dependent reduction in body weight gain.

The higher dose (400 mg/kg) exhibited maximum reduction, which was comparable to the standard drug atorvastatin.

This suggests that the extract may reduce lipid accumulation and improve metabolic function. The observed effect may be attributed to the presence of phytoconstituents such as flavonoids, saponins, and

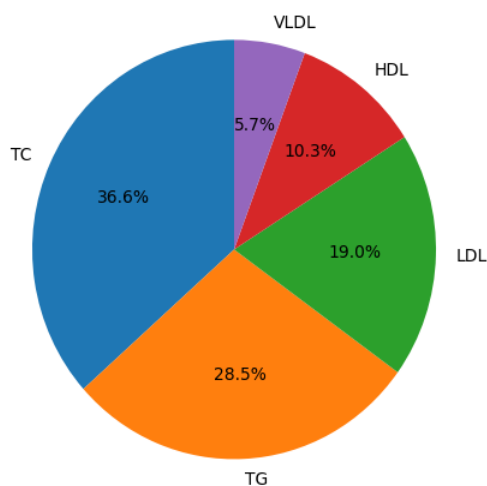
phenolic compounds, which are known to regulate lipid metabolism and reduce fat deposition.

Effect on Lipid Profile: The effect of Flagellaria indica leaf extract on serum lipid profile was evaluated in high-fat diet induced hyperlipidemic Wistar rats. The lipid parameters studied included Total Cholesterol (TC), Triglycerides (TG), Low-Density Lipoprotein (LDL), High-Density Lipoprotein (HDL), and Very Low-Density Lipoprotein (VLDL).

Table 3: Effect of *Flagellaria indica* Leaf Extract on Lipid Profile

Group	Treatment	TC (mg/dL)	TG (mg/dL)	LDL (mg/dL)	HDL (mg/dL)	VLDL (mg/dL)
Group I	Normal Control	110 ± 5	90 ± 4	50 ± 3	45 ± 2	18 ± 1
Group II	Disease Control (HFD)	220 ± 8	180 ± 7	140 ± 6	25 ± 2	36 ± 2
Group III	Standard (Atorvastatin 10 mg/kg)	130 ± 6	100 ± 5	65 ± 4	40 ± 3	20 ± 1
Group IV	Extract (100 mg/kg)	180 ± 7	140 ± 6	110 ± 5	30 ± 2	28 ± 2
Group V	Extract (200 mg/kg)	150 ± 6	120 ± 5	85 ± 4	35 ± 3	24 ± 1
Group VI	Extract (400 mg/kg)	135 ± 5	105 ± 4	70 ± 3	38 ± 2	21 ± 1

Values are expressed as Mean ± SEM (n = 6)

**Figure 4: Lipid profile parameters in the *Flagellaria indica***

Treatment with *Flagellaria indica* leaf extract resulted in a significant and dose-dependent improvement in lipid profile.

The extract at 400mg/kg dose showed maximum lipid-lowering activity, which was comparable to the standard drug atorvastatin.

The observed antihyperlipidemic activity may be attributed to the presence of bioactive phytoconstituents such as flavonoids, phenolics,

and saponins, which are known to possess antioxidant and lipid-lowering properties.

Effect on Liver Function Tests: The effect of *Flagellaria indica* leaf extract on liver function parameters was evaluated in high-fat diet induced hyperlipidemic Wistar rats. The liver function markers studied included serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), total bilirubin, total protein, and albumin.

Table 4: Effect of *Flagellaria indica* Leaf Extract on Liver Function Tests

Group	Treatment	AST (U/L)	ALT (U/L)	ALP (U/L)	Total Bilirubin (mg/dL)	Total Protein (g/dL)	Albumin (g/dL)
Group I	Normal Control	42 ± 2	35 ± 2	110 ± 5	0.6 ± 0.1	7.2 ± 0.2	4.1 ± 0.1
Group II	Disease Control (HFD)	82 ± 4	76 ± 3	190 ± 8	1.5 ± 0.1	5.2 ± 0.2	2.8 ± 0.1
Group III	Standard (Atorvastatin 10 mg/kg)	50 ± 3	43 ± 2	125 ± 6	0.8 ± 0.1	6.8 ± 0.2	3.9 ± 0.1
Group IV	Extract (100 mg/kg)	68 ± 3	61 ± 3	165 ± 7	1.2 ± 0.1	5.8 ± 0.2	3.2 ± 0.1
Group V	Extract (200 mg/kg)	58 ± 3	50 ± 2	145 ± 6	1.0 ± 0.1	6.3 ± 0.2	3.5 ± 0.1
Group VI	Extract (400 mg/kg)	52 ± 2	45 ± 2	130 ± 5	0.9 ± 0.1	6.7 ± 0.2	3.8 ± 0.1

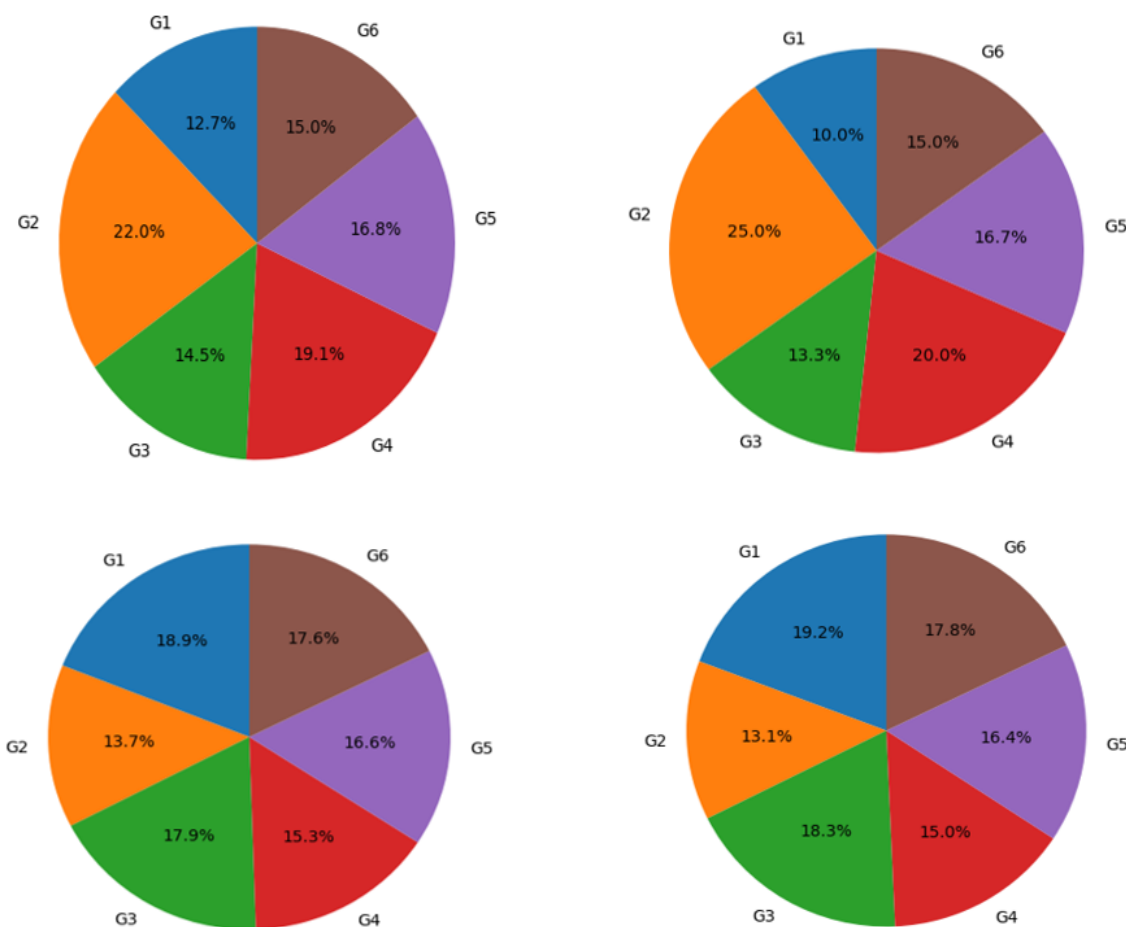


Figure 5: Effect of *Flagellaria indica* leaf extract on Liver Function Tests

Treatment with *Flagellaria indica* leaf extract significantly improved liver function in a dose-dependent manner.

Among the tested doses, the 400 mg/kg extract-treated group showed maximum protection, with values approaching those of the standard drug atorvastatin.

The reduction in AST, ALT, and ALP levels suggests stabilization of hepatocellular membrane integrity and reduced leakage of enzymes into the bloodstream. The decrease in total bilirubin level

indicates improvement in hepatic excretory function.

Effect on Antioxidant Markers: In the present study, the disease control group showed a significant reduction in SOD, CAT, and GSH levels compared to the normal control group, indicating enhanced oxidative stress due to high-fat diet administration. Treatment with *Flagellaria indica* leaf extract produced a dose-dependent increase in antioxidant enzyme levels, suggesting restoration of antioxidant defense mechanisms.

Table 5: Effect of *Flagellaria indica* Leaf Extract on Antioxidant Markers

Group	Treatment	SOD (U/mg protein)	CAT ($\mu\text{mol H}_2\text{O}_2$ decomposed/min/mg protein)	GSH ($\mu\text{g/mg protein}$)
Group I	Normal Control	8.5 ± 0.3	62 ± 2	7.8 ± 0.2
Group II	Disease Control (HFD)	4.2 ± 0.2	32 ± 1	3.6 ± 0.1
Group III	Standard (Atorvastatin 10 mg/kg)	7.6 ± 0.2	56 ± 2	7.0 ± 0.2
Group IV	Extract (100 mg/kg)	5.6 ± 0.2	40 ± 1	4.8 ± 0.1
Group V	Extract (200 mg/kg)	6.7 ± 0.2	48 ± 2	5.9 ± 0.2
Group VI	Extract (400 mg/kg)	7.3 ± 0.3	54 ± 2	6.8 ± 0.2

Values are expressed as Mean $\pm$ SEM (n = 6)

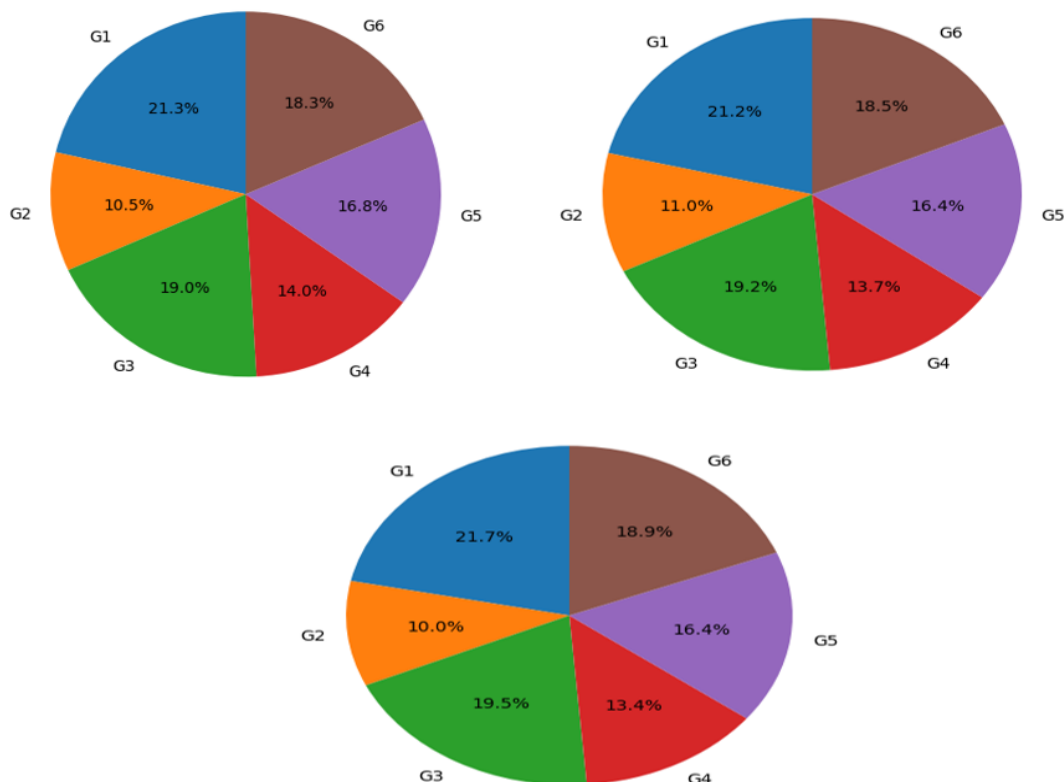


Figure 6: Effect of Flagellaria indica leaf extract on Antioxidant Markers

Treatment with *Flagellaria indica* leaf extract significantly increased antioxidant enzyme levels in a dose-dependent manner. Among the tested groups, the 400 mg/kg dose produced maximum improvement and showed values close to the standard drug atorvastatin.

The increase in SOD levels indicates enhanced dismutation of superoxide radicals, while the rise in CAT levels reflects improved breakdown of hydrogen peroxide. Similarly, restoration of GSH levels suggests improved intracellular antioxidant capacity and detoxification mechanisms.

Histopathological Study: Histopathological examination of liver tissue was carried out to assess the structural changes produced by high-fat diet induced hyperlipidemia and the protective effect of *Flagellaria indica* leaf extract. The microscopic observations revealed marked pathological alterations in the disease control group, whereas treatment with the extract showed dose-dependent improvement in hepatic architecture.

Group I – Normal Control: Microscopic examination of liver tissue of the normal control group showed normal hepatic architecture with well-preserved hepatocytes arranged in cords around the central vein. The sinusoidal spaces appeared normal, and no evidence of fatty degeneration, necrosis, or inflammatory infiltration was observed. This group confirmed the normal structural integrity of liver tissue.

Group II – Disease Control (HFD): The liver section of the disease control group showed severe pathological alterations characterized by marked fatty degeneration, ballooning of hepatocytes, sinusoidal congestion, inflammatory cell infiltration, and disorganization of normal hepatic architecture.

These findings indicate significant liver damage produced by high-fat diet induced hyperlipidemia.

Group III – Standard Drug Treated Group: The liver tissue of rats treated with atorvastatin (10 mg/kg) showed near-normal hepatic architecture with only mild fatty changes. Hepatocytes appeared mostly normal, central vein structure was preserved, and inflammatory infiltration was minimal. This indicates significant hepatoprotective activity of the standard drug.

Group IV – Extract Treated Group (100 mg/kg): Microscopic examination of liver tissue of rats treated with *Flagellaria indica* extract at 100 mg/kg showed moderate improvement compared to the disease control group. Mild fatty degeneration and slight hepatocyte swelling were still present, but congestion and inflammatory changes were reduced.

Group V – Extract Treated Group (200 mg/kg): The liver section of rats treated with 200 mg/kg extract showed significant restoration of hepatic architecture. Hepatocytes were better arranged,

fatty changes were minimal, and sinusoidal spaces appeared more normal. Inflammatory infiltration was markedly reduced, indicating better protection than the lower dose.

Group VI – Extract Treated Group (400 mg/kg): Microscopic examination of liver tissue of

the 400 mg/kg extract-treated group revealed almost normal hepatic architecture. Hepatocytes appeared intact with very mild fatty change, normal central vein, and minimal inflammatory infiltration. This group showed maximum hepatoprotective effect, comparable to the standard drug treated group.

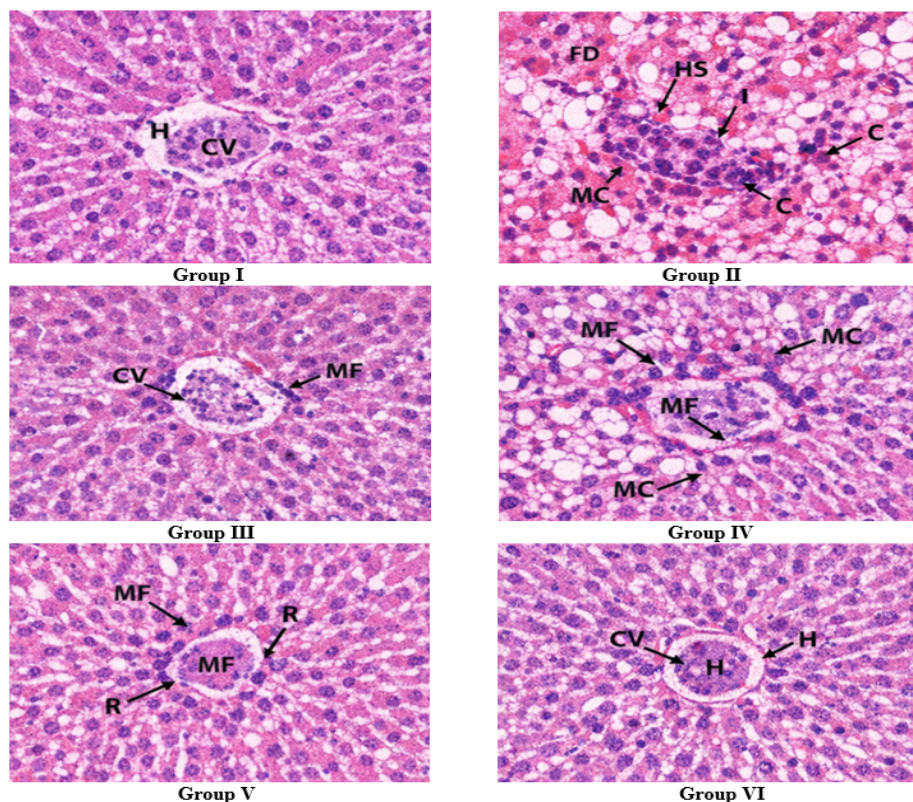


Figure 7: Photomicrograph of liver section

The histopathological findings clearly supported the biochemical results. The disease control group showed severe hepatic damage due to high-fat diet administration, while treatment with *Flagellaria indica* leaf extract produced dose-dependent protection against liver injury. The higher dose of the extract showed almost complete restoration of normal liver structure, indicating strong hepatoprotective and antihyperlipidemic activity.

Discussion

The ethanolic leaf extract of *Flagellaria indica* showed dose-dependent antihyperlipidemic activity, improving lipid profile and liver function parameters in high-fat diet-induced hyperlipidemic rats. It also enhanced SOD, CAT and GSH levels, indicating improved antioxidant defense.

Histopathological findings supported the protective effect, with greater restoration of hepatic architecture at the higher dose.

Conclusion

The findings showed a clear dose-dependent effect of *Flagellaria indica* leaf extract, with increasing doses producing greater improvement in lipid and liver parameters. The 400mg/kg dose showed the most pronounced antihyperlipidemic and antioxidant effects among the treated groups. Improved SOD, CAT, GSH levels and hepatic architecture further supported its protective activity. These results suggest that *F. indica* has promising dose-related antihyperlipidemic and hepatoprotective potential.

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