

<https://doi.org/10.33472/AFJBS.6.9.2024.2115-2129>



African Journal of Biological Sciences



Hypolipdemic Activity of Ficus Racemosa Fruit extract in Wistar Albino Rats

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Article History

Volume 6, Issue 9, 2024

Received: 10 May 2024

Accepted : 20 May 2024

doi: 10.33472/AFJBS.6.9.2024.2115-2129

Abstract

This study investigates the hypolipidemic activity of *Ficus racemosa* extract in Wistar albino rats and its potential protective effects against diet-induced damage in the liver and heart. Rats were fed a high-fat diet to induce hyperlipidemia, followed by treatment with *Ficus racemosa* extract. Serum lipid levels and histopathological changes in the liver and heart were assessed.

Results demonstrate a significant reduction in serum lipid levels, including total cholesterol, triglycerides, and LDL cholesterol, in rats treated with *Ficus racemosa* extract. Histopathological analysis reveals amelioration of liver steatosis and cardiac hypertrophy in treated rats compared to those on a high-fat diet alone. These findings suggest that *Ficus racemosa* extract may possess hypolipidemic and cardioprotective properties, potentially through its antioxidant and anti-inflammatory effects.

In conclusion, *Ficus racemosa* extract shows promise as a natural remedy for hyperlipidemia and may offer protection against diet-induced liver and heart damage. Further research is warranted to elucidate the underlying mechanisms and to explore its therapeutic potential in humans.

Key words: Hyperlipidemia, *Ficus Racemosa*, Natural medicine, Wistar rats

Introduction

Ficus racemosa, commonly known as the cluster fig tree or gular fig, is a species of flowering plant belonging to the Moraceae family. Native to the Indian subcontinent and Southeast Asia, *Ficus racemosa* has been an integral part of traditional medicine systems such as Ayurveda,

Siddha, and Unani for centuries. This tree is characterized by its large, spreading canopy and distinctive fig fruits, which grow in clusters along the branches (1).

Throughout history, various parts of the *Ficus racemosa* plant, including the bark, leaves, fruits, and latex, have been utilized for their medicinal properties. The therapeutic potential of *Ficus racemosa* extracts lies in its rich phytochemical composition, which includes flavonoids, phenolic compounds, tannins, alkaloids, steroids, terpenoids, and polysaccharides. These bioactive compounds confer a wide range of pharmacological effects, making *Ficus racemosa* a valuable resource in traditional and modern herbal medicine. (2)

The medicinal uses of *Ficus racemosa* are diverse and encompass the treatment of various ailments. Its bark extract has been traditionally employed for its anti-inflammatory(3), antimicrobial, and wound healing properties. Additionally, *Ficus racemosa* leaves are known for their anti-diabetic (4), antioxidant, and hepatoprotective effects. The fruits of this tree are used to alleviate digestive disorders, while the latex has been used topically for skin diseases and as an anthelmintic agent.

Numerous scientific studies have corroborated the traditional uses of *Ficus racemosa* and shed light on its pharmacological activities. Research has demonstrated the antioxidant activity of its leaf and fruit extracts, as well as their potential in managing diabetes mellitus(5). Moreover, the antimicrobial properties of *Ficus racemosa* extracts have been investigated against various pathogenic bacteria and fungi, highlighting its role in combating infections.(6)

Research on hypolipidemic effects of plant extracts has gained significant attention due to the rising prevalence of hyperlipidemia and its associated cardiovascular risks. Various plant extracts have been investigated for their potential to lower lipid levels, particularly cholesterol and triglycerides, in preclinical and clinical studies (7). For example, studies have shown that extracts from plants such as garlic (*Allium sativum*) (8), fenugreek (*Trigonella foenum-graecum*)(9), and turmeric (*Curcuma longa*) (10) exhibit hypolipidemic effects through mechanisms such as inhibition of cholesterol synthesis, increased bile acid excretion, and modulation of lipid metabolism enzymes. These findings highlight the potential of plant extracts as natural alternatives for managing dyslipidemia and reducing the risk of cardiovascular diseases. The present study was undertaken to assess hypolipidemic activity of *Ficus racemosa* in wistar albino rats.

In conclusion, *Ficus racemosa* stands as a prominent botanical resource with significant medicinal value. Its long-standing use in traditional medicine systems is supported by scientific evidence, underscoring its potential as a source of novel therapeutic agents. Further exploration of its phytochemical constituents and pharmacological effects holds promise for the development of new treatments for a wide array of health conditions.

Materials and methods

Animals

All animal studies conducted in this research adhered to the guidelines set forth by the Organisation for Economic Co-operation and Development (OECD) for the testing of animals.

To ensure ethical treatment and compliance with animal welfare, the study received approval from the Institutional Animal Ethical Committee at KAMSRC, Hyderabad. The specific ethical project number for this study was KAMSRC/Pharm/IAEC/2020/1. Before the commencement of the experiments, the animals used in the study were carefully examined and allowed to acclimatize to their new environmental conditions. This acclimatization period aimed to reduce any potential stress on the animals and create a stable baseline for the study. The animal subjects in the experiment were albino rats weighing approximately 150-190 grams. Throughout the study, these rats were housed in a controlled environment with a temperature maintained at $22 \pm 3^\circ \text{C}$ and relative humidity ranging from 30% to 70%. The animals were subjected to a 12-hour light and dark cycle, simulating a natural day-night cycle.

Collection of Plant Material and extraction Procedure

In this study, all the plants used were collected specifically from the Hyderabad district. To ensure accuracy and authenticity, the identification and authentication process was conducted by Botanist, an Assistant Professor from the Department of Botany at S.V. University in Tirupati. Voucher number 0415 was assigned to the plant sample of *Ficus Racemosa* as a reference for future verification.

To begin the extraction process, the freshly collected leaves *Kalanchoe pinnata* were carefully dried in the shade. Once dried, the leaves were coarsely powdered and then passed through a sieve with a mesh size of 40, resulting in a fine powder. This powdered material was carefully stored in an airtight container for later use in the study.

For the extraction of active compounds, 100g of the dried plant material powder was macerated, or soaked, in a hydro-alcoholic solution consisting of 60% ethanol. This maceration process lasted for 7 days, allowing the solvent to draw out the desired compounds from the plant material.

After the 7-day period, the macerated mixture was filtered to separate the liquid extract from the solid residue. The solvent was then evaporated from the filtered liquid, leaving behind the concentrated extract of *Kalanchoe pinnata*. (11)

Hyperlipidemia Induction

High fat diet was procured from National institute of nutrition (NIN) for induction of hyperlipidemia

2.3 Methodology

Five groups of rats, each containing six animals, were used in this study. All the rats in these groups were fed a high-fat diet comprising specific ingredients: cholesterol (1%), cholic acid (0.5%), casein (20%), choline (0.25%), multi-vitamin mix (3.5%), and sucrose (48.4%). Alongside this high-fat diet, they were also provided with a standard pellet diet. This feeding regimen was continued for a duration of 30 days.

3.1 Experimental design:

Group I (Control)	Group II (High fat Diet)	Group III (Standard)	Group IV (Test Group I)	Group V (Test group II)
Normal saline	High fat Diet	Atorvastatin	Ficus Racemosa fruit 250mg/kgbw	Ficus racemosa 500mg/kgbw

3.2 Collection of Blood

Under mild halothane anesthesia, blood was collected through a puncture in the retro-orbital sinus. The collected samples were then centrifuged at 2000 r.p.m. for 10 minutes, and the resulting serum samples were utilized for conducting various biochemical tests.

3.3 Estimation of Biochemical Parameters.

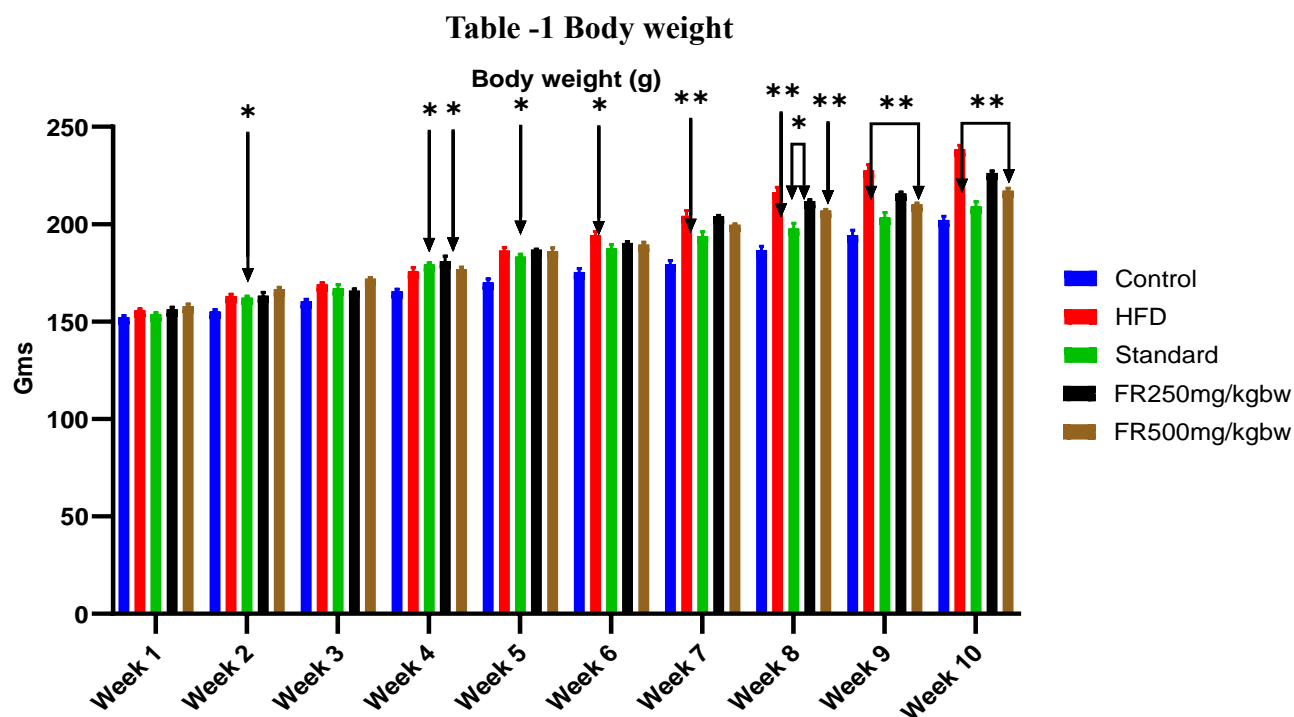
The lipid profile was assessed using standard diagnostic kits. It included the measurement of total cholesterol (TC), triglycerides (TG), and high-density lipoprotein cholesterol (HDL-C). Additionally, LDL-cholesterol and VLDL-cholesterol levels were calculated using the Friedwald formula

Liver functions were evaluated through the assessment of alkaline phosphatase (ALP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) activities.

Histopathological examination:

After the treatment period, the animals from all five groups were euthanized, and their heart, aorta, liver, and kidney were carefully removed. The collected tissues were then washed and prepared as 5 µm thick section slides. These slides were subsequently stained with haematoxylin and eosin and subjected to examination using light microscopy.

Results:



All the groups supplemented with a high-fat diet (HFD) exhibited a notable and statistically significant increase ($p < 0.05$) in serum total cholesterol, triglyceride, and LDL-C levels, while there was a significant decrease ($p < 0.05$) in HDL-C levels compared to the values before starting the high-fat diet. The serum levels of TC, TG, LDL-c, and HDL-c before administering the HFD, after HFD supplementation, and after treatment with Atorvastatin and extracts of *Kalanchoe pinnata* were presented in Table-1. Both the standard drug Atorvastatin and both doses of the hydroalcoholic extracts of *Kalanchoe pinnata* (250mg/kg, 500 mg/kg) showed a significant reduction ($p < 0.05$) in total cholesterol (Figure 1), triglyceride (Figure 2), and LDL-C (Figure 3). Additionally, they exhibited a significant increase ($p < 0.05$) in HDL-C levels (Figure 4) when compared to the control group.

Table-2 Total cholesterol

	Day 0	Day 30	Day Treatment 15	Day 30 Treatment	Day 45 Treatment
Control	95.67 ± 1.542	93.33 ± 2.376	93.67 ± 1.874	94.67 ± 2.813	94.67 ± 3.252
HFD	92.17 ± 1.558	133.83 ± 1.905	145.67 ± 3.084	157.17 ± 4.206	177.5 ± 4.918
Standard	92.33 ± 2.512	129.5 ± 2.527	120.33 ± 2.512**	116.67 ± 1.820**	107.5 ± 4.303**
FR250mg/kg bw+HFD	91.5 ± 1.310	135.83 ± 1.869	130.83 ± 2.561**	127.17 ± 6.172**	125.17 ± 1.721**
FR500mg/kg bw+HFD	95.33 ± 1.626	133.33 ± 2.512	129.17 ± 3.390**	125.83 ± 1.579**	122.17 ± 0.833**

Mean ± SEM **p<0.001

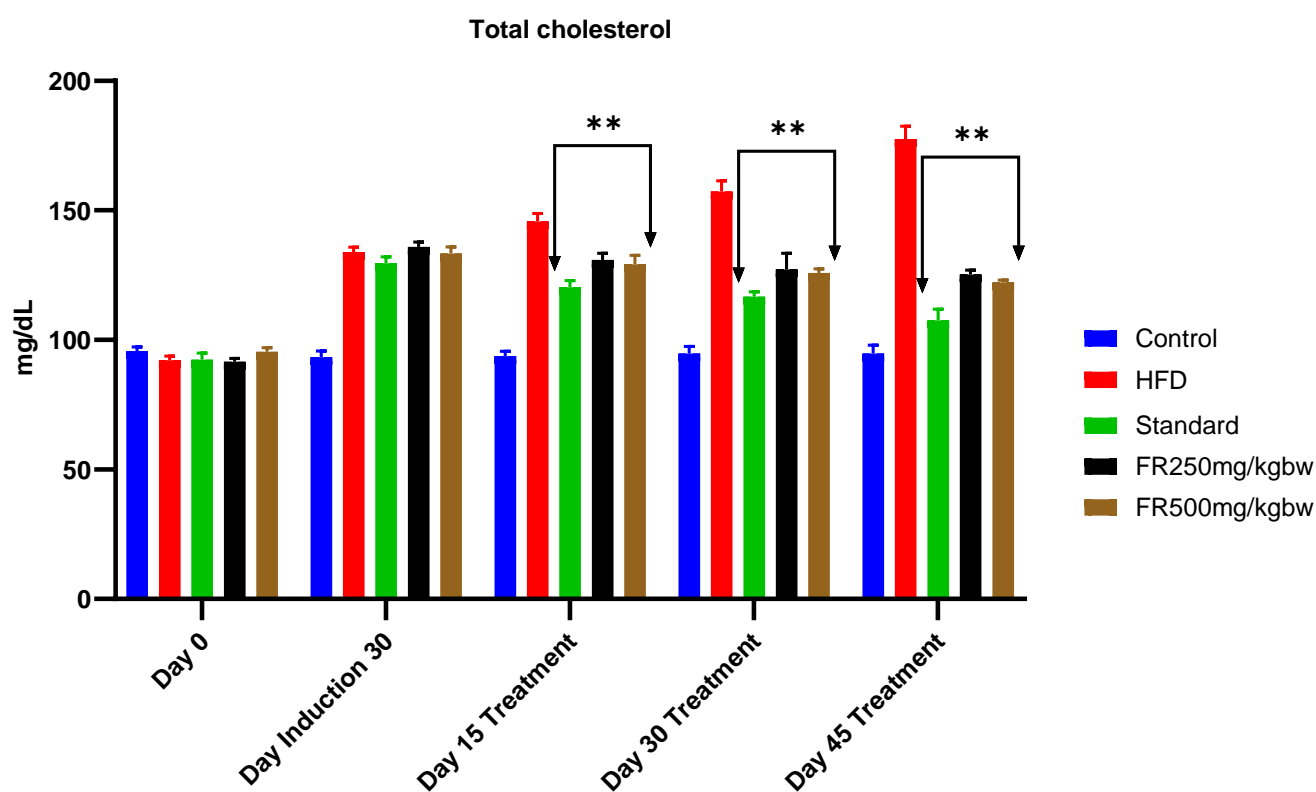


Table-3 Triglycerides

	Day 0	Day 30	Day Treatment 15	Day Treatment 30	Day Treatment 45
Control	83.17 ± 1.222	83.83 ± 1.249	84.17 ± 1.447	83.17 ± 3.301	87.83 ± 4.408
HFD	81.17 ± 1.195	102.33 ± 5.142	116.5 ± 1.821	120.67 ± 2.246	127.33 ± 2.155
Standard	79.50 ± 5.271	100.50 ± 0.847	95.83 ± 1.302	90.00 ± 1.390	82.67 ± 2.777
FR250mg/kgbw+HFD	81.00 ± 4.195	104.17 ± 1.493	102.17 ± 0.872**	100.83 ± 1.249**	96.5 ± 4.780**

FR500mg/kgbw+ HFD	86.5 ± 1.384	105.17 ± 1.797	99.33 ± 0.843**	96.83 ± 4.126**	90.5 ± 4.992**
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Mean ± SEM **p<0.01

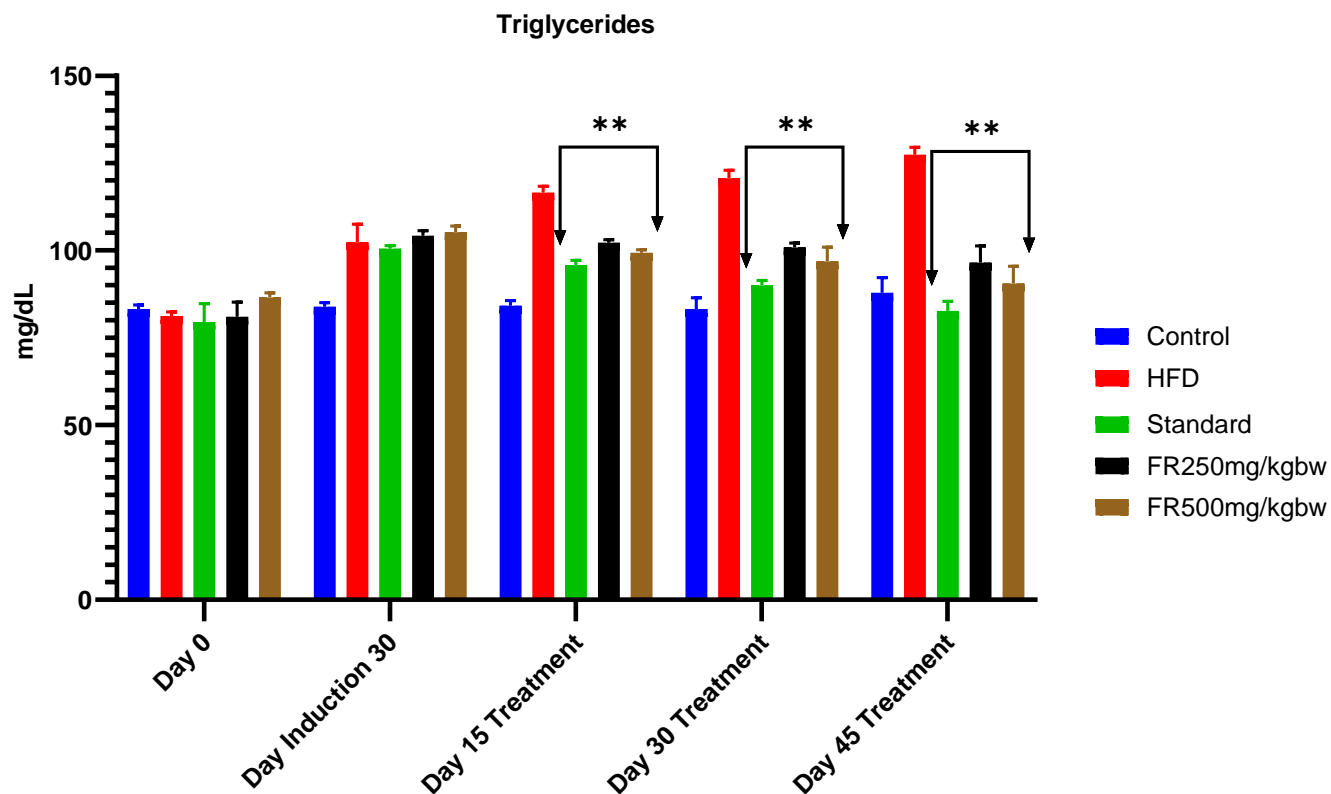


Table-4 HDL Cholesterol

	Day 0	Day 30	Day Treatment 15	Day Treatment 30	Day Treatment 45
Control	56.83 ± 1.447	56.83 ± 1.558	57.33 ± 1.022	57.67 ± 1.687	58.17 ± 1.905
HFD	57.33 ± 1.892	40.33 ± 0.558	37 ± 2.033	34.17 ± 1.302	34.17 ± 1.302
Standard	59.83 ± 2.798	41 ± 2.352	46.83 ± 0.792**	54.17 ± 1.014**	54.17 ± 1.276**
FR250mg/kgbw+ HFD	57.83 ± 1.641	40.33 ± 4.047	41.5 ± 2.705	41.17 ± 2.182**	44.5 ± 1.688**
FR500mg/kgbw+ HFD	60.5 ± 2.907	42.17 ±1.222	43.33 ± 2.216*	46.67 ± 1.256**	51.17 ± 0.601**

Mean ± SEM **p<0.01

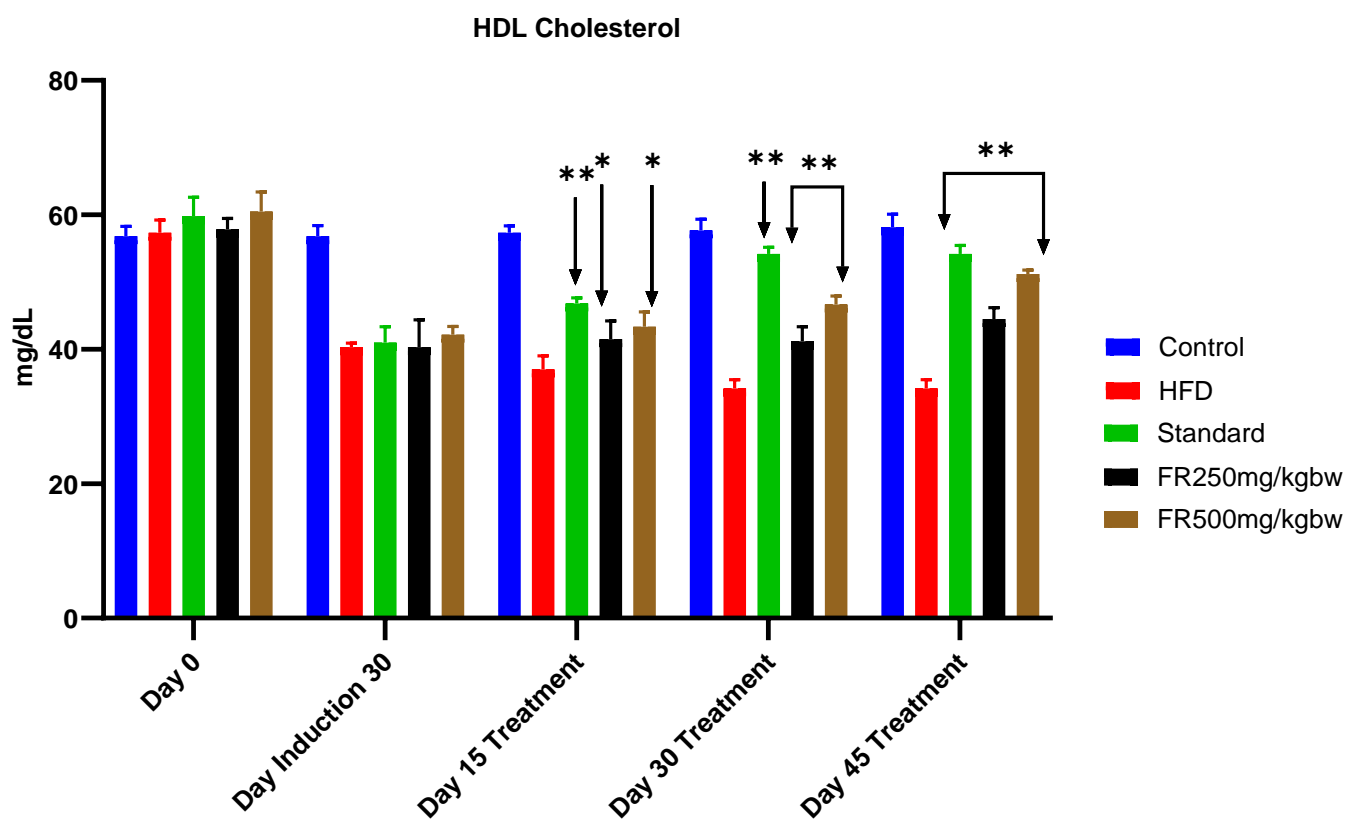
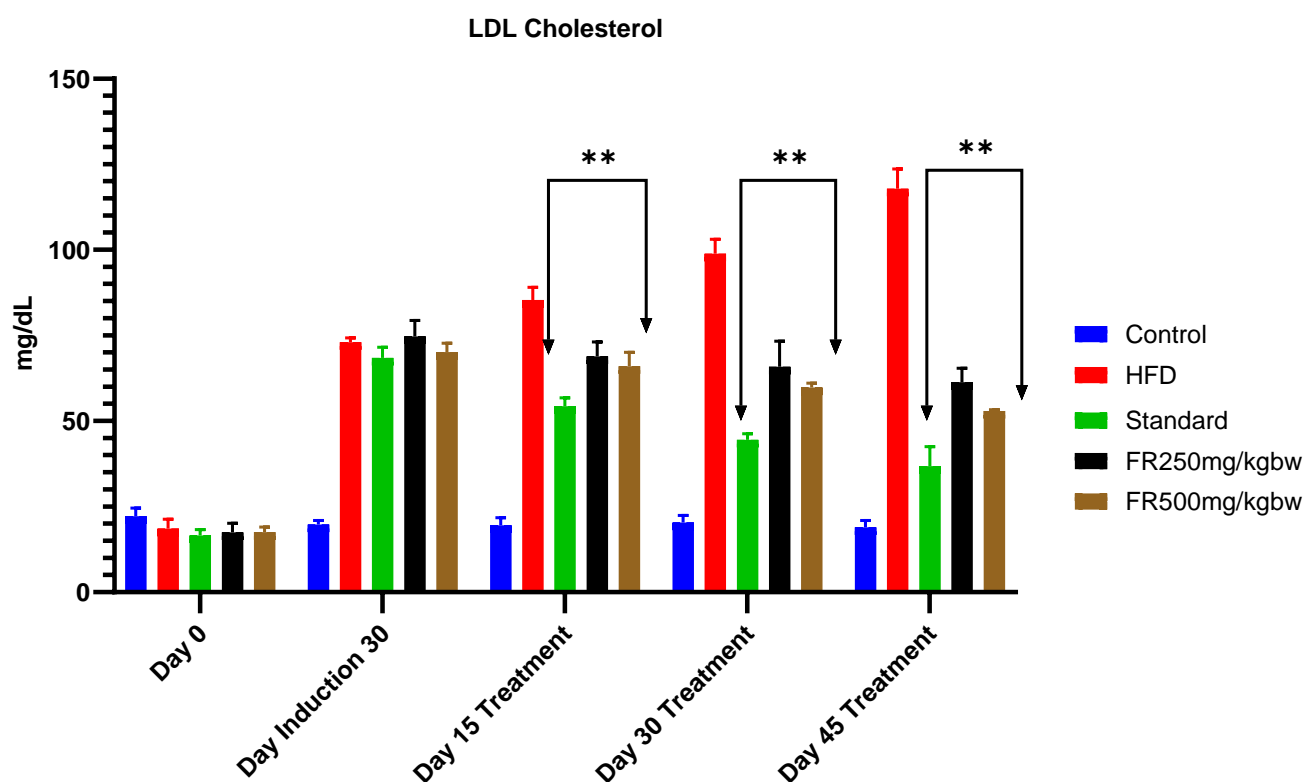


Table-5 LDL Cholesterol

	Day 0	Day 30	Day Treatment 15	Day Treatment 30	Day Treatment 45
Control	22.2 ± 2.37	19.73 ± 1.183	19.5 ± 2.277	20.37 ± 2.012	18.93 ± 1.988
HFD	18.6 ± 2.74	73.03 ± 1.244	85.37 ± 3.687	98.87 ± 4.199	117.87 ± 5.696
Standard	16.6 ± 1.711	68.4 ± 3.127	54.33 ± 2.412**	44.5 ± 1.806**	36.8 ± 5.732**
FR250mg/kgbw+ HFD	17.47 ± 2.628	74.67 ± 4.713	68.9 ± 4.152**	65.83 ± 7.443**	61.37 ± 4.01**
FR500mg/kgbw+ HFD	17.53 ± 1.516	70.13 ± 2.556	65.97 ± 4.077**	59.8 ± 1.206**	52.9 ± 0.361**

Mean ± SEM **p<0.001

**Table-6 VLDL Cholesterol**

	Day 0	Day 30	Day Treatment 15	Day Treatment 30	Day Treatment 45
Control	16.63 ± 0.244	16.77 ± 0.25	16.83 ± 0.289	16.63 ± 0.66	17.57 ± 0.882
HFD	16.23 ± 0.239	20.47 ± 1.028	23.3 ± 0.364	24.13 ± 0.449	25.47 ± 0.431
Standard	15.9 ± 1.054	20.1 ± 0.169	19.17 ± 0.26**	18 ± 0.278**	16.53 ± 0.555**
FR250mg/kgbw+ HFD	16.2 ± 0.839	20.83 ± 0.299	20.43 ± 0.174	20.17 ± 0.25	19.3 ± 0.956
FR500mg/kgbw+ HFD	17.3 ± 0.277	21.03 ± 0.359	19.87 ± 0.169	19.37 ± 0.825	18.1 ± 0.998

Mean ± SEM **p<0.001

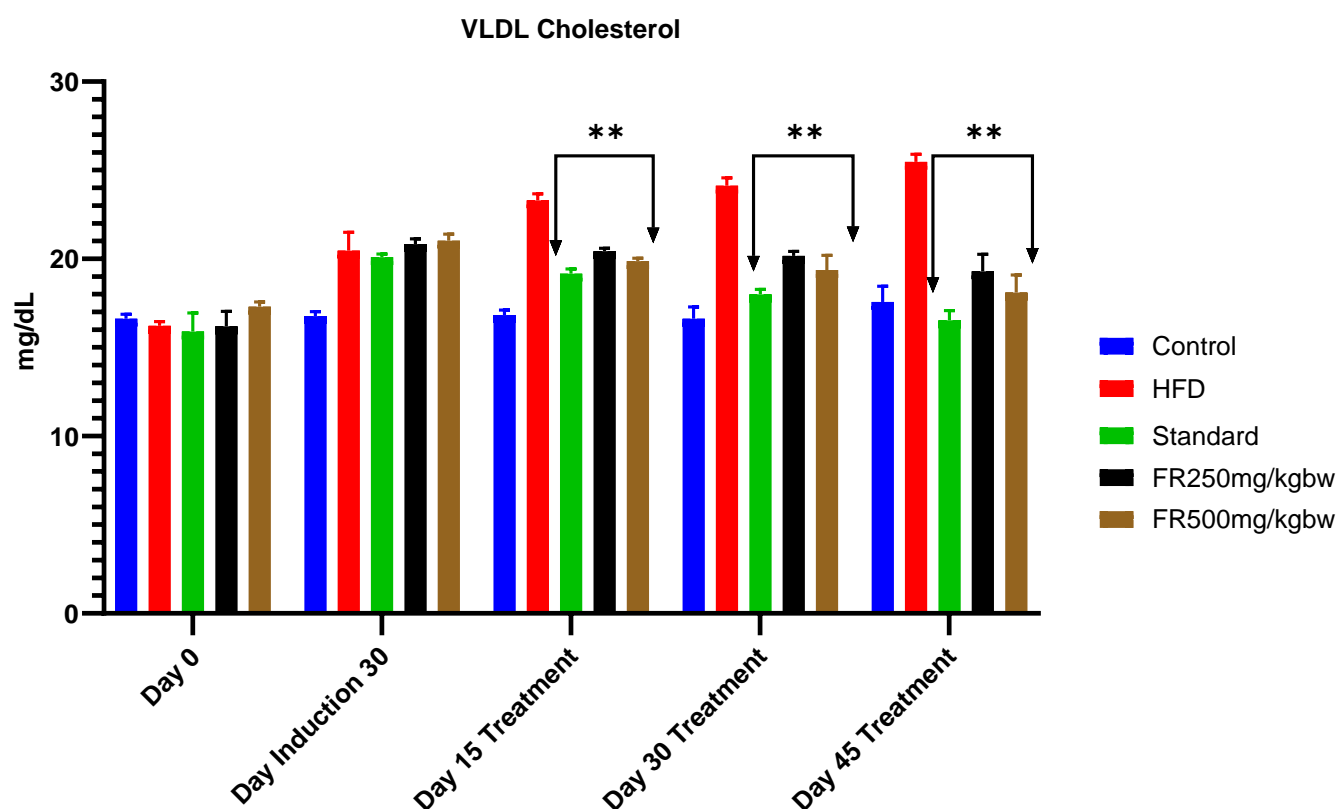
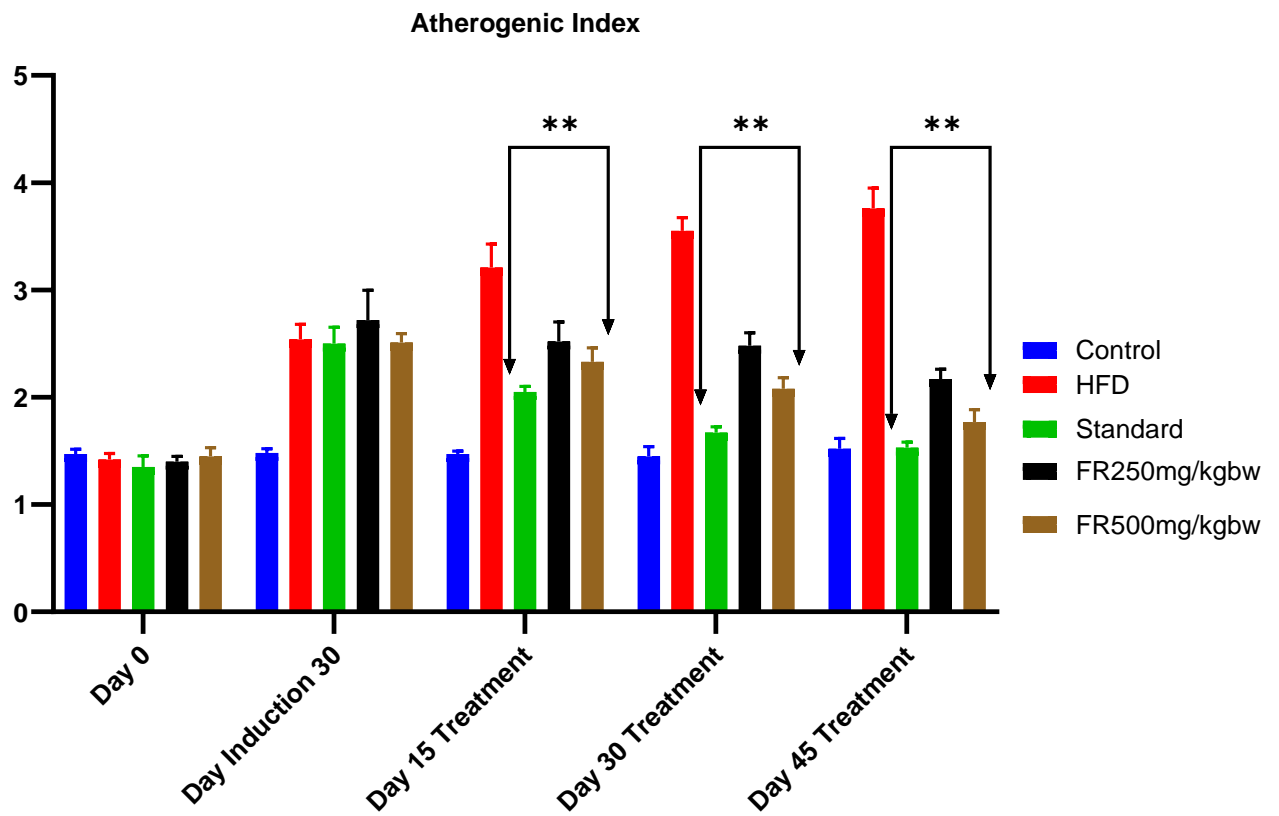


Table -7 Atherogenic Index

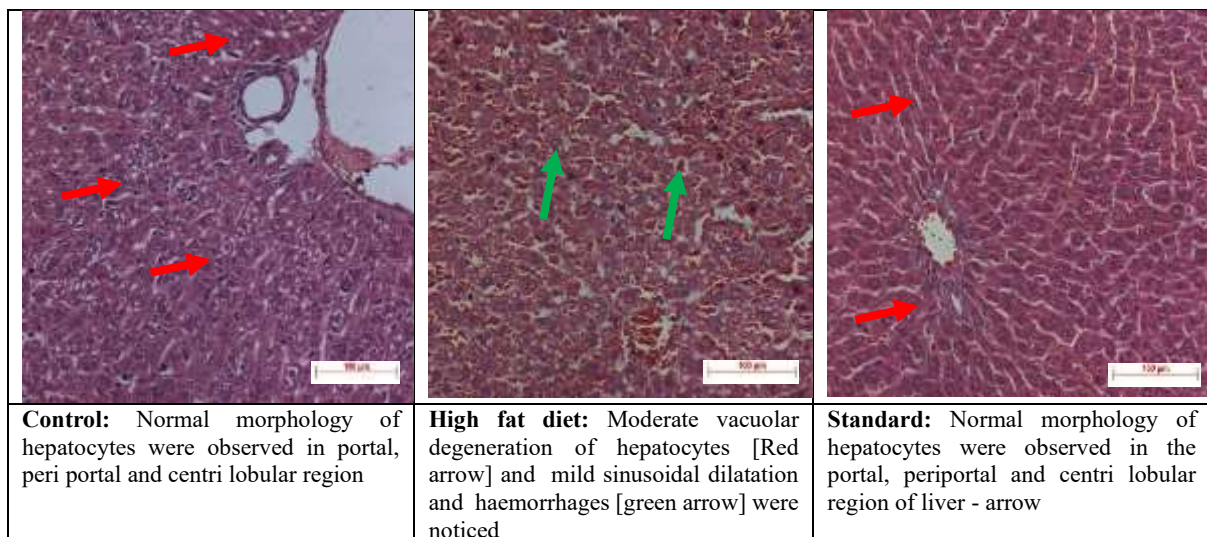
	Day 0	Day 30	Day Treatment 15	Day Treatment 30	Day Treatment 45
Control	1.47 ± 0.046	1.48 ± 0.04	1.47 ± 0.029	1.45 ± 0.089	1.52 ± 0.095
HFD	1.42 ± 0.056	2.54 ± 0.14	3.21 ± 0.217	3.55 ± 0.125	3.76 ± 0.19
Standard	1.35 ± 0.103	2.5 ± 0.152	2.05 ± 0.05**	1.67 ± 0.055**	1.53 ± 0.052**
KP 250mg/kgbw+HFD	1.4 ± 0.049	2.72 ± 0.277	2.52 ± 0.183**	2.48 ± 0.12**	2.17 ± 0.092**
KP 500mg/kgbw+HFD	1.45 ± 0.079	2.51 ± 0.084	2.33 ± 0.131**	2.08 ± 0.103**	1.77 ± 0.115**

Mean ± SEM **p<0.001



Histopathological examination

Liver

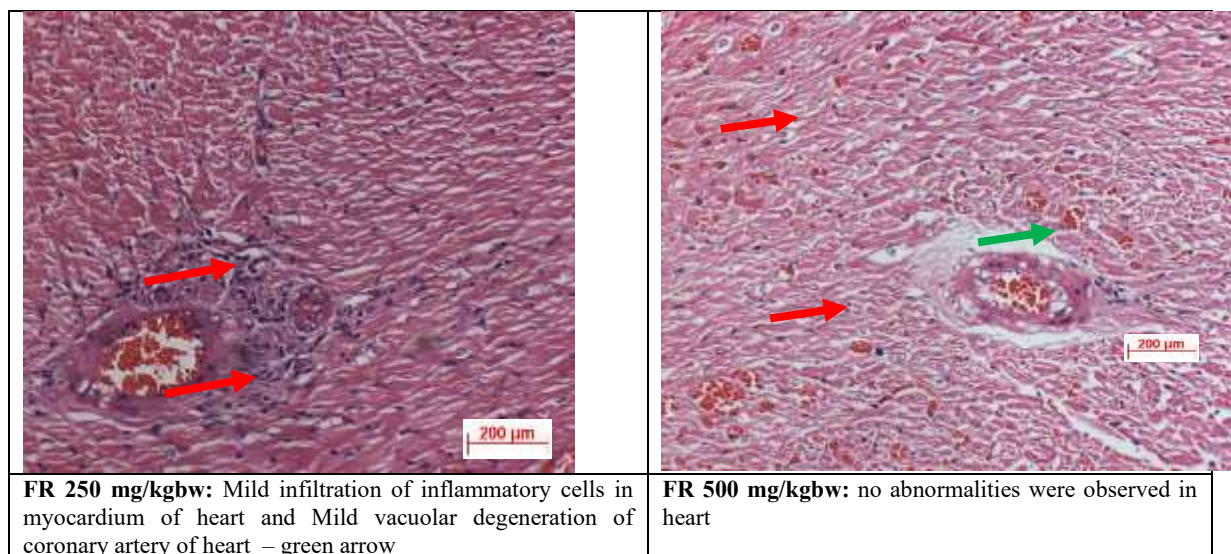


<p>FR 250 mg/kgbw: Showing Moderate vacuolar degeneration of hepatocytes and mild sinusoidal dilatation and haemorrhages [green arrow] were noticed</p>	<p>FR 500 mg/kgbw: Normal morphology of hepatocytes were observed in the portal, periportal and centrilobular region of liver - arrow</p>

Heart

<p>Control: Normal morphology of myocardium [green arrow] and coronary artery [yellow arrow] and no abnormalities were observed in heart</p>	<p>High fat diet: Multi focal vacuolar / fatty degeneration was observed in myocardium of heart – red arrow</p>	<p>Standard:Normal morphology of myocardium [Red arrow] with coronary artery of heart –</p>

		green arrow; NO abnormality were observe
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Discussion

The research article investigates the hypolipidemic activity of *Ficus racemosa* in Wistar albino rats, shedding light on its potential as a natural remedy for hyperlipidemia. The study employed a rigorous experimental design, including the use of standardized *Ficus racemosa* extract and thorough lipid profile analyses, to substantiate its findings.

The results of the study reveal a significant reduction in serum lipid levels, particularly total cholesterol, triglycerides, and LDL cholesterol, in Wistar albino rats treated with *Ficus racemosa* extract. These findings align with previous research that has demonstrated the lipid-lowering effects of various phytoconstituents present in *Ficus* species, including *Ficus racemosa*.(12)

The mechanism underlying the hypolipidemic activity of *Ficus racemosa* extract in Wistar albino rats may involve several pathways. One proposed mechanism is the inhibition of key enzymes involved in lipid metabolism, such as HMG-CoA reductase and lipoprotein lipase. By inhibiting these enzymes, *Ficus racemosa* extract may reduce the synthesis and accumulation of cholesterol and triglycerides in the body.

Moreover, *Ficus racemosa* extract may exert its hypolipidemic effects through antioxidant and anti-inflammatory mechanisms. Oxidative stress and inflammation are known to play pivotal roles in the pathogenesis of dyslipidemia, and the antioxidant and anti-inflammatory properties of *Ficus racemosa* extract may help mitigate these processes, leading to a reduction in serum lipid levels (13).

Ficus racemosa has been reported to have hepatoprotective effects, wound healing properties, and potential anti-cancer activity (14,15). These diverse pharmacological activities highlight the potential of *Ficus racemosa* as a valuable source of natural medicines.

The histopathological findings in the liver and heart tissues of Wistar rats treated with *Ficus racemosa* extract compared to those on a high-fat diet provide valuable insights into the potential protective effects of the extract against diet-induced damage.

In the liver, rats on a high-fat diet often exhibit features of steatosis, characterized by the accumulation of fat droplets within hepatocytes. This condition can progress to steatohepatitis, fibrosis, and eventually cirrhosis if left untreated. However, treatment with *Ficus racemosa* extract appears to mitigate these effects, as evidenced by a reduction in fat accumulation and overall improvement in liver architecture. This suggests that the extract may have hepatoprotective properties, possibly by reducing lipid accumulation and inflammation in the liver.

Similarly, in the heart, a high-fat diet can lead to cardiac hypertrophy, fibrosis, and dysfunction. However, rats treated with *Ficus racemosa* extract show signs of amelioration, with less pronounced cardiac hypertrophy and fibrosis. This indicates that the extract may have cardioprotective effects, potentially through its antioxidant and anti-inflammatory properties.

The histopathological findings suggest that *Ficus racemosa* extract has a beneficial impact on liver and heart health in rats fed a high-fat diet. Further studies are needed to elucidate the specific mechanisms underlying these effects and to determine the extract's potential therapeutic applications in combating diet-induced liver and heart diseases in humans.

Additionally, the hypolipidemic activity of *Ficus racemosa* extract in Wistar albino rats may be attributed to its ability to modulate gene expression related to lipid metabolism. Phytochemicals present in *Ficus* species have been shown to regulate the expression of genes involved in lipid metabolism, suggesting a potential molecular mechanism for the observed effects (16).

In conclusion, the findings of this study underscore the hypolipidemic activity of *Ficus racemosa* in Wistar albino rats, suggesting its potential as a natural remedy for hyperlipidemia. Further research is warranted to elucidate the specific bioactive compounds responsible for these effects and to explore the potential therapeutic applications of *Ficus racemosa* in the management of dyslipidemia and related cardiovascular disorders.

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