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Phytochemical and Hypolipidemic activity of ethanolic leaves extract of Ficus racemosa Linn

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ABSTRACT

Ficus racemosa (Moraceae family) is used in traditional system of medicine for the treatment of several disorders including diabetes mellitus. The present study was carried out to evaluate hypolipidemic activity of ethanolic leaves extract of Ficus racemosa Linn against high cholesterol diet induced hyperlipidemia in rats for 30 days. Rats were fed with ethanolic extract of Ficus racemosa (250mg/kg and 250mg/kg p.o) and atorvastatin (30 mg/kg, p.o) along with hyperlipidemic diet for 30 days. Ficus racemosa and atorvastatin were found to lower the serum cholesterol, triacylglyceride, VLDL, LDL, and atherogenic index, but were found to increase the HDL as compared to the corresponding high fed cholesterol diet group. The hypolipidemic activity of Ficus racemosa can be ascribed to its inhibitory effect on the liver HMG CoA reductase activity. Thus, the study demonstrates that Ficus racemosa possesses a hypolipidemic effect.

Keywords: *Ficus racemosa, HMG Co-A reductase, Hypolipidemic Effect, Lipid Profile.*

INTRODUCTION

Lipids and lipoproteins abnormalities are preceding risk factor for cardiovascular diseases and prevalence of this in general population has increased considerably in last few decades. Hyperlipidemia contributes significantly in the prevalence and severity of atherosclerosis and coronary heart diseases [1]. Cardiovascular diseases are the primary cause of mortality and morbidity worldwide [2]. Numerous factors, such as diet rich in saturated fats and cholesterol, age, family history, hypertension and life style play an important role in the development of high cholesterol and LDL levels, which are primarily responsible for the onset of atherosclerosis and coronary heart diseases[3]. Lowering of lipids levels, by a drug or diet management could reduce the risk of cardiovascular diseases. Current awareness of medicinal plants in the management of cardiovascular diseases has encouraged the researchers for exploring novel lipid lowering pharmaceuticals [4].

Ficus racemosa is a medium tall tree with quite rich green foliage that provides good shade. It is popularly known as "Country fig" in English and "Atti" in Tamil. The leaves, bark and fruits of *F. racemosa* are employed in native medicine to treat several diseases [5]. Experimental studies have demonstrated its anti-inflammatory, hepatoprotective and hypoglycemic effects [6-8]. However, there were no reports on antihyperlipidemic effect of *F. racemosa* bark in alloxan induced diabetic rats. In view of the above, it seems necessary to investigate the hypolipidemic activities of ethanolic extract of *F. racemosa* bark in alloxan-induced diabetic rats.

MATERIALS AND METHODS

Preparation of plant material and ethanolic extract:

The leaves were dried under shade at room temperature for seven days and powdered by the means of grinder and were sieved through sieve no.40 to get the coarse powder (750gm) and was extracted with ethanol by Soxhlet apparatus and obtained extract was concentrated and stored in vacuum desiccator. The obtained yield was calculated. Then the ethanolic extract of *Ficus racemosa* was subjected to qualitative and phytochemical analysis.

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Preliminary Phytochemical Screening:

The ethanolic leaves extracts of *Ficus racemosa* were subjected to preliminary phytochemical screening for their presence or absence of active phytochemical constituents by the following methods(Kokate 2007 and Khandelwal 2004).

Experimental Animals:

Wistar albino rats (150-200 g) of both sexes were obtained from the animal house. Before and during the experiment, rats were fed with standard diet (Gold Moher, Lipton India Ltd). After randomization into various groups and before initiation of experiment, the rats were acclimatized for a period of 7 days under standard environmental conditions of temperature, relative humidity, and dark/light cycle. Animals described as fasting were deprived of food and water for 16 h ad libitum. All animal experiments were carried out in accordance with the guidelines of CPCSEA and study was approved by the IAEC (Institutional animal ethical committee).

Acute Toxicity studies:

The acute oral toxicity of ethanolic extract of leaves of the *Ficus racemosa* was carried out as per OECD 423 – guidelines

Evaluation of Anti-Hyperlipidemic activity:

Animals - Wistar rats of either sex weighing 200-250 g

Grouping and treatment---- Animals are to be divided into **5** groups containing **6** in each group

Group I **-** Administered with normal saline along with normal diet
Group II **-** Administered with normal saline along with cholesterol diet (2% cholesterol, 1% sodium cholate and 2% arachis oil) for 30 days.
Group III **-** Administered with ethanolic extract of *Ficus racemosa* (250mg/kg, p.o) along with high cholesterol diet for 30 days.
Group IV **-** Administered with a different effective dose of ethanolic extract of *Ficus racemosa*(500mg/kg, p.o) along with high cholesterol diet for 30 days.
Group V **-** Administered with simvastatin (20 mg/kg, p.o) along with high cholesterol diet for 30 days.

On the 31st day, blood samples were collected from the retro orbital sinus and serum samples were analyzed for serum total cholesterol (TC), triglyceride (TG),High density lipo-protein(HDL-C), Very low density lipoprotein (VLDL), Low density lipo-protein (LDL) and blood parameters like Alanine amino transaminase (ALT / SGPT), Aspartate amino transaminase (AST / SGOT) and Alanine phosphotase (ALP) was analyzed using diagnostic kits.

Statistical analysis:

Results are expressed as mean±SEM (standard error mean) and subjected to one-way analysis of variance (ANOVA) followed by Dunnett's test and values with $p<0.05$ were considered to be statistically different.

RESULTS

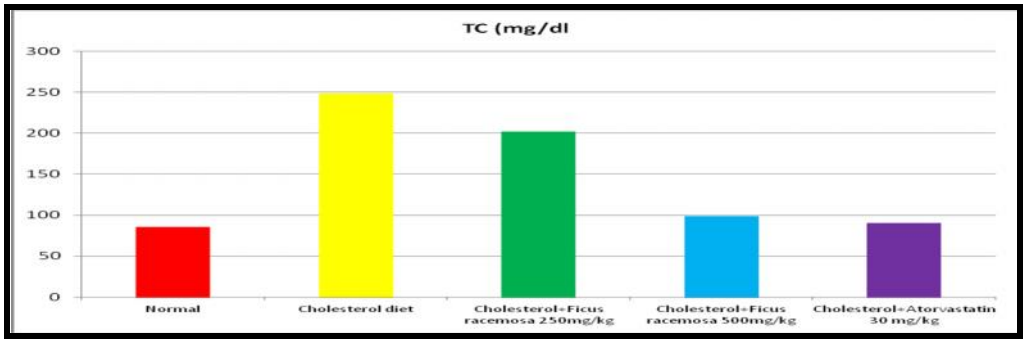
Preliminary phytochemical studies of ethanolic leaves extract *of Ficus racemosa* revealed the presence of flavanoids, triterpenoids, steroids and carbohydrates.

The acute toxoicity studies were conducted according to OECD 423 guidelines. The ethanolic leaves extract *of Ficus racemosa* found to be non toxic up to 2000 mg/kg.

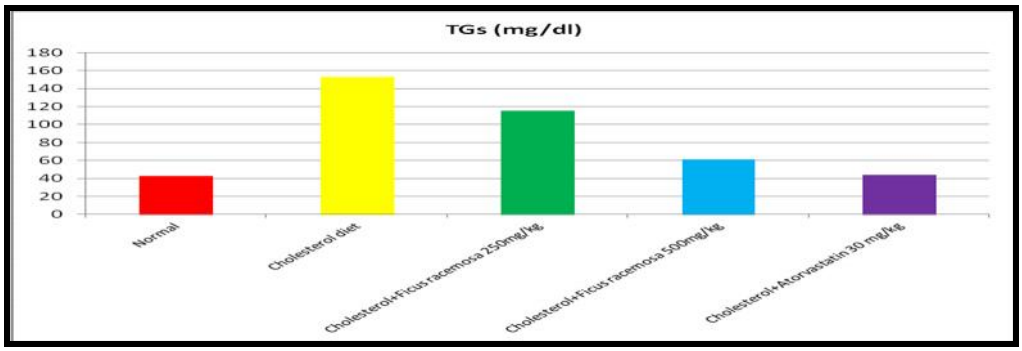
The effect of ethanolic leaves extract of *Ficus racemosa* on TC , TGs , HDL-C , VLDL and LDL in rats are summarized in Table 1. There was a significant increase in TC , TGs , VLDL and LDL in Cholesterol diet group II rats, when compared to the normal control group. The HDL-C levels were significantly decreased to 18.73 mg/dl in Cholesterol diet rats from the level of 44.28mg/dl in normal group. On the other hand the group with received both leaves extract 250mg/kg and 500mg/kg + Cholesterol diet (Group III and Group IV) and Cholesterol+Atorvastatin (Group V) showed significantly decreased the elevated TC , TGs , VLDL and LDL when given orally and reversed the altered HDL-C to almost normal level (Table 1 and Graph I-V).

Table No. 1: The effect of *Ficus racemosa* on TC , TGs , HDL-C , LDL and VLDL.

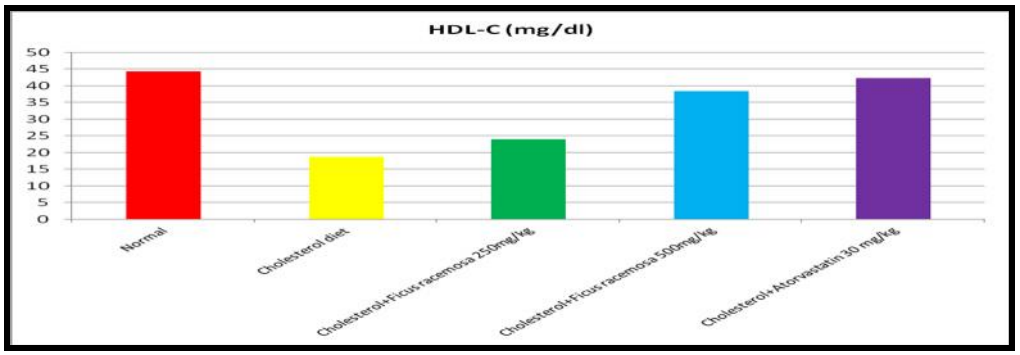
Groups	TC (mg/dl	TGs (mg/dl)	HDL-C (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
Normal	85.27 ±3.48	42.43± 2.36	44.28±1.56	33.19±2.42	11.43±0.33
Cholesterol diet	248.37±4.28	152.72±6.72	18.73±2.93	182.92±1.3	28.91±2.86
Cholesterol+*Ficus racemosa* 250mg/kg	201.62±8.23	115.48±2.73	23.93±2.20	161.26±2.9	24.72±1.21
Cholesterol+*Ficus racemosa* 500mg/kg	98.62±3.84	60.83±2.29	38.41±3.02	41.65±1.82	15.23±1.34
Cholesterol+Atorvastatin 30 mg/kg	90.72±1.92	44.20±1.82	42.22±1.92	38.42±3.42	12.20±0.28



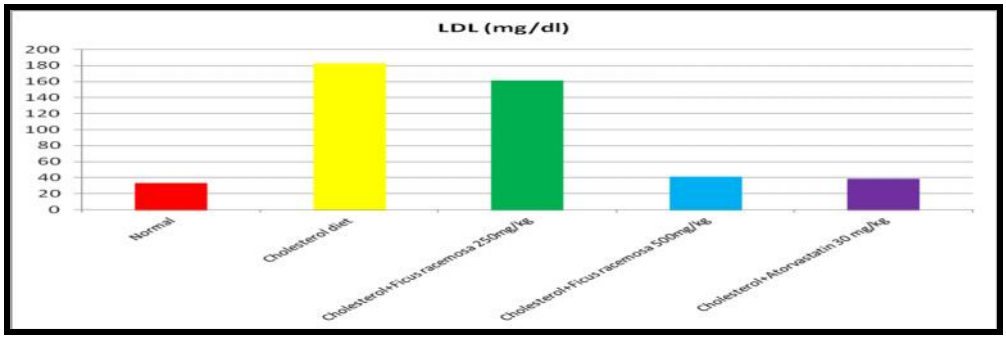
Graph. I: The effect of *Ficus racemosa* on TC



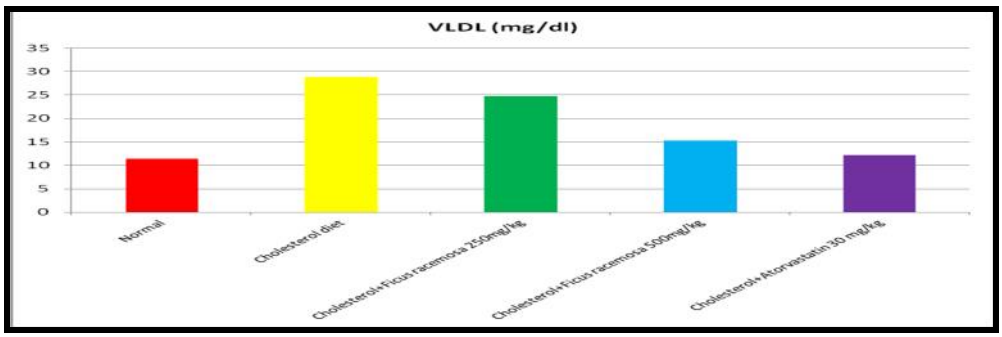
Graph. II: The effect of *Ficus racemosa* on TGs



Graph. III: The effect of *Ficus racemosa* on HDL-C



Graph. IV: The effect of *Ficus racemosa* on LDL



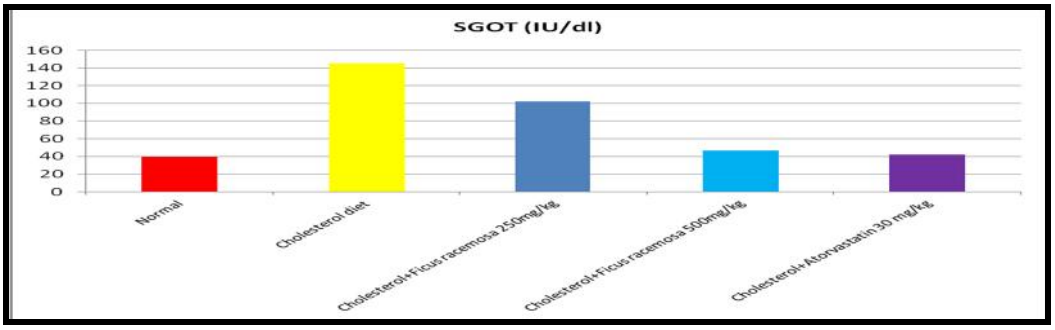
Graph. V: The effect of *Ficus racemosa* on VLDL

Blood parameter:

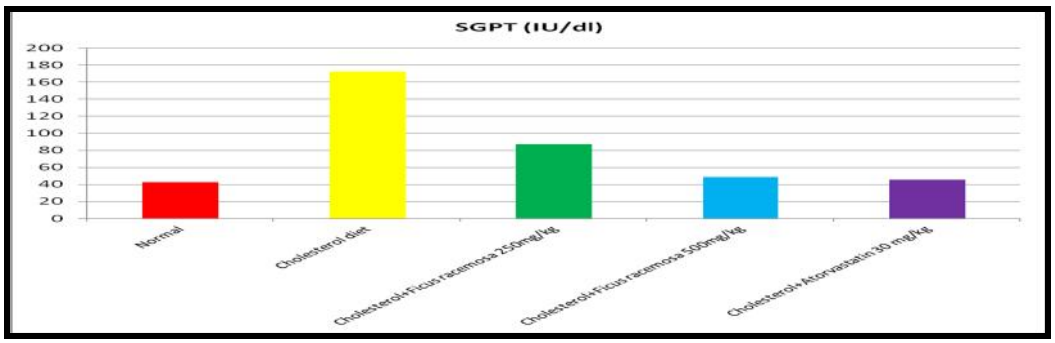
In relation to liver function test, alanine amino transferase (ALT), aspartate amino transferase (AST), and ALP were assessed and the results are given in Table 3. It is observed that ALT, AST and ALP levels were found elevated in high fat diet group (Group II) animals. The activities of ALT, AST and ALP in ethanolic leaves extracts of *Ficus racemosa* treated group are significantly lowered than those of the high fat diet group animals. (Table 2 and Graph:VI-VIII).

Table No. 2: The effect of *Ficus racemosa* on SGOT, SGPT and ALP.

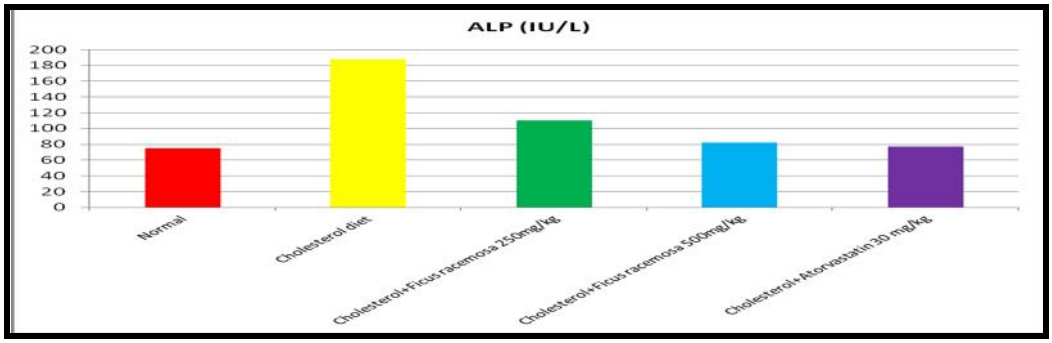
Treatment groups	SGOT(IU/dl)	SGPT (IU/dl)	ALP (IU/L)
Normal	39.36±1.82	42.37±0.26	75.24 ± 2.46
Cholesterol diet	145.2±2.62	172.32±4.46	188.29±4.19
***Ficus racemosa* 250mg/kg, + Cholesterol diet**	102.4±4.37	87.25±1.60	110.42±5.15
***Ficus racemosa* 500mg/kg+ Cholesterol diet**	46.72±3.51	48.56±2.91	82.30±2.37
Atorvastatin30mg/kg,+ Cholesterol diet	42.35±2.61	45.61±1.02	77.20±3.10



Graph. VI: The effect of *Ficus racemosa* on SGOT



Graph. VII: The effect of *Ficus racemosa* on SGPT



Graph. VIII: The effect of *Ficus racemosa* on ALP

DISCUSSION

Recently, a number of clinical studies suggest that the increased risk of coronary heart disease is associated with a high serum concentration of TC, LDL-C and triglyceride. The abnormally high concentration of serum lipids is mainly due to the increase in the mobilization of free fatty acids from the peripheral depots. Previous works on *Ficus racemosa* were carried out by many researchers and significance of *Ficus racemosa* in various disease treatment was illustrated through various animal models. The present work were carrying out on antioxidant and anti hyperlipidemic activity of *Ficus racemosa,* a direct approach of treating hypercholesterolemia in animal model is shown with various parameters. The previous work done on hyperlipidemia only show that the relation between blood glucose and lipid profile modifications. But from this research project this has been illustrated that *Ficus racemosa* could directly be effective in treating hyperlipidemia.

In recent years, considerable interest has been directed towards the investigation of plasma lipids and lipoproteins pattern in diabetes mellitus due to the fact that abnormal lipid level leads to the development of coronary artery disease in diabetic patients [9]. Reduced insulin secretion and defect in insulin function results in enhanced metabolism of lipids from adipose tissue to the plasma. Impairment in insulin sensitivity due to high concentration of lipids in the cells is responsible for the elevated cardiovascular risk in diabetes mellitus [10, 11].

As insulin has a profound role in the regulation of key enzymes involved in the lipid and lipoprotein metabolism, its deficiency causes major changes in the activity of these enzymes and thereby affecting overall lipid metabolism and lipid profile of various tissues [12]. Insulin has also profound influence on the synthesis and expression of apolipoproteins in hepatic and extra hepatic tissues [13]. The higher concentration of plasma total cholesterol observed in diabetic rats is probably due to mobilization of free fatty acids from the peripheral fat depots [14]. HDL removes cholesterol from non-hepatic tissues to liver through the process known as reverse cholesterol transport. Several studies have documented reduction in plasma HDL cholesterol in diabetic rats and diabetic patients due to defect in reverse cholesterol transport [15].

In the present preliminary phytochemical studies of ethanolic leaves extract *of Ficus racemosa* revealed the presence of steroids, tannins, flavonoids, saponins, alkaloids and flavanoids,. The acute toxoicity studies *of Ficus racemosa* leaves found to be non toxic up to 2000 mg/kg.

The possible mechanism involved in the atherogenesis in rat may be due to enhance cholesterol biosynthesis by increasing activity of HMGCoA reductase. In addition, this could be associated with a down regulation in LDL receptors by the cholesterol and saturated fatty acids in the diet [16]. The biochemical estimations shown that the extracts MEMD & FFMD increased the protective HDL-C level and decreased the atherogenic LDL and VLDL levels. The possible mechanism of test drug may involve increase of HDL-C, which can lead to the mobilization of cholesterol from peripheral cells to the liver [17].

Flavonoids activate multi enzyme systems, such as citocrome P450 and b5 and this action affects the whole metabolism, as these systems are involved in the metabolism of xenobiotics, including drugs, insecticides, and pollutants, that have great importance on pharmacology and toxicology [18]. Due to this effect, flavonoids act on body lipid constituents like steroids and bile acids, and influence lipid metabolism. They increase bile acid excretion because cytocrome P-450 enzymes bind some compounds to the bile acids and therefore reduce cholesterol level in the body [19]. The physiological effect of flavonoids include possible antioxidant activity, therefore suggestion their role in prevention of coronary heart disease including atherosclerosis [20]. Flavonoids may also work by making liver cells more efficient to remove LDL-C from blood by increasing the LDL-C receptor densities in liver and by binding to apolipoprotein B [21].

In the present study, the in vitro antioxidant activity of *Ficus racemosa* by DPPH methods showed significant results when compared to Vitamin C. The results shown suggest that the study carried out on *Ficus racemosa* (500mg/kg) is found to more effective than *Ficus racemosa* (250mg/kg) against hyperlipidemia in reducing the levels of low density and very low density lipoproteins and increase in the HDL levels in the present model of research.

CONCLUSION

Administration of *Ficus racemosa* (250/500mg/kg) for 30 days in high cholesterol diet successfully prevented the elevation of VLDL, TG, TC and LDL-C levels. While administration of *Ficus racemosa* leaves extracts (MC) (250mg/kg and 500mg/kg) for 30 days successfully prevented decrease of serum HDL-C in high cholesterol diet model rats, as compared with the standard atorvastatin (30 mg/kg, p.o).Ethanolic leaves extract of *Ficus racemosa* also showed significant antioxidant effect done by DPPH in vitro method. Hence from the above result it is conclusion that the hyporlipidemic activity is might be due the presence of flavonoids in leaves extract of *Ficus racemosa*.

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