

EVALUATION OF ANTI-HYPERLIPIDEMIC ACTIVITY OF LEAVES EXTRACT OF AEGLE GLUTINOSA PLANT

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ABSTRACT

The present study was designed to perform preliminary phytochemical screening, acute oral toxicity and to evaluate antihyperglycemic activity of hydroalcoholic extract of *Aegle Glutinosa* Plant. Hypercholesterolaemia is the state been illustrate by an increased fatty substances called lipids, rise in plasma TCs and TGs levels it is as well called hyper lipoproteinemia. *Aegle Glutinosa* whole plant was extracted using ethanol and water mixture as solvent by cold maceration process. The extract was subjected to preliminary phytochemical screening. Acute oral toxicity studies were performed to determine dose test. Preliminary phytochemical screening revealed the presence of alkaloids, carbohydrates, glycosides, saponins, tannins, flavonoids, Triterpenes. The evaluation of antihyperlipidemic activity was done using High Fat Diet induced hyperlipidemia models in Wistar albino rats. for 15 Days, and then they are checked for the blood parameter

levels like TC, TGs, LDL, VLDL, and HDL. Hydroalcoholic extracts at low dose (250 mg/kg) and high dose (500 mg/kg) significantly at high doses it reduced the levels of TC, TGL, LDL, VLDL & increased the levels of HDL and reduced the body weights.

KEYWORD: *Aegle Glutinosa* Plant, Hypercholesterolaemia, HDL, LDL, VLDL, SGOT, SGPT.

1. INTRODUCTION

High levels of circulating lipids are considered to be hyperlipidemia, which raises the risk of atherosclerosis and other serious illnesses. Specific types of hyperlipidemia comprise hyperlipoproteinemia, elevated levels of low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL), elevated levels of cholesterol (hypercholesterolemia), and elevated levels of triglycerides (hypertriglyceridemia). The majority of the time, hyperlipidemia is asymptomatic and is found by routine screening. Sometimes there are xanthomas and xanthelasmas. These are subcutaneous fatty deposits that are often present in people with hereditary disorders such as familial hypercholesterolemia. Hyperlipidemia is frequently caused by either inefficient or delayed liver production of VLDL, which is then reshaped into LDL. Defective hepatic and nonhepatic LDL receptors are a factor in familial hypercholesterolemia.

Saturated fat overconsumption raises the liver's production of triglycerides and VLDL via a molecular process involving protein activators. Animal products including meat, whole milk, fat, and tropical oils like coconut, palm kernel, and palm are sources of saturated fats. Cardiovascular risk is predicted in both men and women by high concentrations of total and low-density lipoprotein (HDL) cholesterol and low levels of HDL cholesterol. Higher risk has only been associated with elevated triglyceride levels in women. For every 1% increase in total cholesterol, there is an average 2% increase in the risk of cardiovascular disease.

The disorder known as hyperlipoproteinemia, sometimes known as hyperlipidaemia, is characterized by abnormally high blood levels of any or all lipids and/or lipoproteins. This type of dyslipidemia, which also involves lower lipid levels, is primarily well-known. The density of lipids, or fat-soluble molecules, is determined by the size of the protein capsule that contains them, or lipoprotein.

2. METHODOLOGY

2.1 Collection of plant material

Present work carried out on plant species *Aegle glutinosa*. Whole plant of *Aegle glutinosa* was collected from Vindhya herbal nursery in the months of September, 2023.

2.2 Extraction procedure

Following procedure was adopted for the preparation of hydroalcoholic extracts from the shade dried and powdered herbs. 40 gm dried powdered *Aegle glutinosa* has been extracted

with hydroalcoholic solvent (20:80) using maceration process for 48 hrs, filtered and dried using vacuum evaporator at 40°C. The percentage yield of each extract was calculated by using following formula

$$\text{Percentage yield} = \frac{\text{Weight of Extract}}{\text{Weight of powder drug Taken}} \times 100$$

2.3 Phytochemical Screening

The *Aegle glutinosa* extract acquire was subjected to the precursory phytochemical analysis following standard methods by Khandelwal and Kokate. The extract was screened to identify the presence of various active principles of alkaloids, glycosides, phenols, flavonoids, Terpenoids, Saponins, Steroids.

2.4 Estimation of total Phenolic, flavonoid and alkaloid Content

2.4.1 Total Phenolic content estimation

The total phenolic content of the extract was determined by the modified Folin-Ciocalteu method. 10 mg Gallic acid was dissolved in 10 ml methanol, various aliquots of 5- 25µg/ml was prepared in methanol. 10 mg of dried extracted dissolve in 10 ml methanol and filter. Two ml (1mg/ml) of this extract was for the estimation of phenols. 2 ml of extract or standard was mixed with 1 ml of Folin-Ciocalteu reagent (previously diluted with distilled water 1:10 v/v) and 1 ml (7.5g/l) of sodium carbonate. The mixture was vortexed for 15s and allowed to stand for 15min for colour development. The absorbance was measured at 765 nm using a spectrophotometer.

2.4.2 Total flavonoids content estimation

Determination of total flavonoids content was based on aluminium chloride method. 10 mg quercetin was dissolved in 10 ml methanol, and various aliquots of 5- 25µg/ml were prepared in methanol. 10 mg of extract dissolved in 10 ml methanol and filter. Three (1mg/ml) of this extract was for the estimation of flavonoid. 1 ml of 2% AlCl₃ methanolic solution was added to 3 ml of extract or standard and allowed to stand for 15 min at room temperature; absorbance was measured at 420 nm.

2.5 *In vivo* anti-Hyperlipidemic activity of *Aegle glutinosa* extract

Wistar rats (180–250 g) were group housed (n= 6) under a standard 12 h light/dark cycle and controlled conditions of temperature and humidity (25±2 °C, 55–65%). Rats received

standard rodent chow and water *ad libitum*. Rats were acclimatized to laboratory conditions for 7 days before carrying out the experiments. All the experiments were carried in a noise-free room between 08.00 to 15.00 h. Separate group (n=6) of rats was used for each set of experiments.

Acute oral toxicity was conducted according to the method of Organisation for Economic Co-operation and Development (OECD) (OECD, 2001). Animals were kept fasting providing only water, hydroalcoholic extract of leaves of *Aegle glutinosa* (50,100,150,200,300 mg/kg/day) was administered orally for 4 days of six groups of rats (n=6) and the animals were kept under observation for mortality as well as any behavioral changes for evaluation of a possible hepatoprotective effect.

Experimental designs

Group –I: Normal (vehicle alone)

Group –II: Hyperlipidemic rats treated with vehicle alone

Group -III: Hyperlipidemic rats treated with hydroalcoholic extract of *Aegle glutinosa* (250mg/kg, p.o.)

Group –IV: Hyperlipidemic rats treated with hydroalcoholic extract of *Aegle glutinosa* (500mg/kg, p.o.)

Group –V: Hyperlipidemic rats treated with Orlistat (60 mg/kg/day p.o.).

3. RESULTS AND DISCUSSION

3.1 Result of Percentage Yield

Table 3.1: % Yield of leaves of *Aegle glutinosa*.

S.No.	Part	% Yield (W/W)
1.	Whole plant	3.92

3.2 Phytochemical screening of extract

The outcomes of the results are discussed separately in the table 3.2.

Table 3.2: Phytochemical screening of hydroalcoholic extract of *Aegle glutinosa*.

S.No.	Constituents	Hydroalcoholic extract
1.	Alkaloids Dragendroff's test	+ve

	Hager's test	+ve
2.	Flavonoids Lead acetate Alkaline test	+ve +ve
3.	Phenolics FeCl ₃	+ve
4.	Steroids Libermann Burchard's test	+ve
5.	Glycoside Legals test	+ve
6.	Carbohydrates Fehling's test	+ve
7.	Saponins Foam test	+ve
8.	Triterpenes Salkowski's Test	+ve

3.3 Results of Estimation of Total Phenolic Contents

Table 3.3: Total Phenolic and Total flavonoid content of *Aegle glutinosa* leaves extracts.

S. No.	Extracts	Total Phenol (GAE) (mg/g)	Total flavonoid (QE) (mg/g)
1.	Hydroalcoholic extract	208.3	51.7

3.4 Results of antihyperlipidemic effect of extract of *Aegle glutinosa*

Antihyperlipidemic effect of the hydroalcoholic extract *Aegle glutinosa* on the high fat diet induced rats. The mean body weight as shown in Table 3.4. The activity levels of serum total cholesterol (TC), triglycerides (TG) and Serum high density lipoprotein (HDL) were observed in normal and experimental animals. In group II animals, the activity levels of serum total cholesterol (TC) and triglycerides (TG) were significantly elevated when compared to that of normal groups. On the other hand the serum level of Serum high density lipoproteins (HDL) were significantly depleted in the HFD fed rat. In group III, IV and V animals, the activity levels of serum total cholesterol (TC) and triglycerides (TG) were significantly decreased when compared to that of normal groups. Also HDL level was significantly increased in the same groups.

Table 3.4: Mean Body Weight Change.

Group	Drug	Dose	Body weight (g)	
			Onset of study	End of study

I	Normal	Normal saline	175.10±7.50	195.00±7.50
II	Control	HFD	185.05±8.50	238.10±8.50
V	Extract of <i>Aegle glutinosa</i>	250 mg/kg p.o.	190.00±7.00	198.00±7.00
VI	Extract of <i>Aegle glutinosa</i>	500 mg/kg p.o.	190.05±8.00	191.00±8.00
IV	Orlistat	60 mg/kg p.o.	190.00±8.00	183.50±8.00

Table 3.5: Effect of the hydroalcoholic extract of *Aegle glutinosa* on serum lipid profile levels (mg/dL) in HFD induced rat.

Treatment	Dose	Total cholesterol (mg/dL)	Triglycerides (mg/dL)	High density lipoproteins (mg/dL)
Normal	Normal saline	75.00 ± 5.00	80.00 ± 5.50	33.00 ± 4.50
Control	HFD	133.10 ± 5.00	140.3 ± 5.22	22.00 ± 4.60
<i>Aegle glutinosa</i>	250 mg/kg p.o.	91.20 ± 5.70**	92.82 ± 5.10**	28.40 ± 4.50**
<i>Aegle glutinosa</i>	500 mg/kg p.o.	87.10 ± 5.10***	86.71 ± 5.50***	30.50 ± 4.60***
Orlistat	60 mg/kg p.o.	83.40 ± 5.60***	82.59 ± 5.70***	32.10 ± 4.50***

Aegle glutinosa a well known traditional medicinal plants possesses diverse biological activities and pharmacological function including reducing blood glucose and serum lipids. It has long been used to treat diabetes mellitus and related hyperlipidemia. Hypercholesterolemia, a high cholesterol diet and oxidative stress increase serum levels resulting in increased risk for development of atherosclerosis. Cholesterol is synthesized in all animal tissue. It is important to relate to its role in the stabilization of membrane structures because of its rigid planar structure. It also as a precursor for the synthesis of steroid hormones. In the present study, feeding rats with diets rich in cholesterol resulted in increased TC and TG levels. From this study, we found that daily oral administration hydroalcoholic extract of whole plan of *Aegle glutinosa* shows significantly reduced total cholesterol levels in plasma after 15 days of administration. This result agrees with literature where depleted level of HFD fed hyperlipidemia. HDL is directly anti-androgenic and it is believed to remove cholesterol from the developing lesions. The intense interest in this area results in part from the generally low toxicity of antioxidants and the hope that treatment with antioxidants might be additive with cholesterol lowering regimes. In the present study serum TG levels were significantly elevated in HFD rat. The excess of fat diet increased the TG level which is one of the causes of hardening of arteries. In conclusion, it could be said that the hydroalcoholic extract of whole plant of *Aegle glutinosa* exhibited a significant hypolipidemic activity. Administration of HFD produced a highly significant increase in weight mesenteric fat pads. A reduction in the raised weight in the fat pads as observed in the groups of animals treated

with hydroalcoholic extract of whole plant *Aegle glutinosa* may be attributed to increased thermogenesis and decreased lipogenesis.

CONCLUSION

Folk medicine claims that *Aegle glutinosa*, a member of the Rutaceae family, has therapeutic qualities. *Aegle glutinosa* is a treatment for hepatitis virus. In the current study, biochemical assays were used to investigate the phytochemicals found in the entire *Aegle glutinosa* plant. It was discovered that phenolics, flavonoids, glycosides, carbohydrates, saponins, and triterpenes were among the many phytochemicals that were present in large concentration. In conclusion, the observation suggests that a decrease in cholesterol production and an increase in cholesterol excretion may be responsible for the hydroalcoholic extract of *Aegle glutinosa*'s protective impact on high fat-induced hyperlipidemia.

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