Antihyperlipidemic Activity Of *Emblica Officinalis* Leaves On Obese Male Albino Wistar Rats

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**ABSTRACT**

The present study aimed to evaluate antihyperlipidemic action and reduce the time duration of experiment; the aqueous extract of *Emblica officinalis* (EO) leaves against high fat diet along with propylthiouracil (PTU) induced hyperlipidemia in rats. Wistar albino male rats weighing between 150-200 gm were used, rats were divided into 5 groups (n= 6). Group I served as Normal control received normal diet with water for 7 days. Group II served as standard control received atorvastatin 7.2mg/kg body weight. Group III served as toxic control received high fat diet (250g/kg) along with propylthiouracil (PTU) for 7 days and Group IV and V received aqueous extract of *Emblica officinalis* 200mg/kg, and 500mg/kg respectively for 7 days. At the end on day 7th animals were fasted for whole night and on 8th day blood sample was collected by heart puncturing method for the estimation of lipid profile such as low density lipoproteins (LDL), very low density lipoproteins (VLDL), triglycerides (TRIG), total cholesterol (TCH) and high density lipoproteins (HDL). Liver was isolated for histopathological estimation. The result indicates that aqueous extract of *Emblica officinalis* at the dose of 500mg/kg is effective and shows significant decrease in the level of lipid profile (TCH, LDL, TRIG and VLDL) and increase of HDL level.
INTRODUCTION:
In current state of affairs lipemia may be a commonest developing issue in world. Lipidemia is 1st for the foremost risk issue for causing cardiovascular diseases. There numerous plants that have shown to have the chemical constituents that lower the lipid concentration to the normal level. Lipidemia is that the major issue for developing it Conjointly results in coronary artery disease (CAD), cerebro-vascular stroke, and peripheral vascular illness [1]. Lipidemia illness has affected humanity since an extended time. In 2002, coronary heart epidemiologic report has completed the positive association between the blood lipids, hyperlipidemia and its complications, primarily CHD [2]. The lipidemia is outlined because the condition within which the quantity of steroid alcohol or triglyceride-carrying lipoproteins in plasma exceeds on top of the conventional limit of the plasma. These lipoproteins stores within the area between cells of arteries connected from arterial blood vessel that restricts the blood flow to the guts. The mechanism is termed to be arterial sclerosis. Higher storage of lipoproteins oft blocks the blood flow to the guts, and so myocardial infarction (MI) happens, that is just well-known to be viscous attack (Goodman Gilman; 1970) [3].

Hyperlipidemias are often of primary or secondary sorts. The first kind are often treated by the hypolipidemic medicine rather then, the secondary kind elicited from polygenic disease, malady gland disease glandular disorder (adenosis) or excretory organ lipoid death might treated by treating the inflicting disease instead of hyperlipaemia itself [4]. Normally hyperlipaemia don't have any obvious symptoms and that they are typically been discovered throughout the routine health check or till it reaches on top of the conventional vary that results in occur stroke or internal organ attack. Patients having raised blood sterol level or patients having the disorder from their ancestors will develop xanthomas that are keep variety of sterol might collect at a lower place the skin, oft below the eyes. At identical time, patients with raised vary of triglycerides might develop several pimple-like scares at completely different sites in their body [5].

Low-density lipoprotein (LDL) is that the major at heterogenic lipoprotein, the reduction of compound protein would be expected to cut back arterial sclerosis and reduces vessel adverse effects [6]. *Emblica officinalis* (EO) have associate degree holy notation in ayurvedic system– associate degree Indian culture of medication. It’s curative role in cancer, diabetes, liver treatment, heart trouble, ulcer, anemia and various other diseases [7]. Many pharmacological studies have demonstrated the ability of *Emblica officinalis* fruit shows antioxidant, anti-carcinogenic, anti-tumour, anti-genotoxic, anti-inflammatory activities [8].

MATERIALS AND METHODS
The experimental protocols was been approved by the Institutional Ethics Committee and Animal Ethics Committee of Shambhunath Institute of Pharmacy, Prayagraj, Uttar Pradesh, India.

Plant collection
Fresh leaves of *Emblica officinalis* were collected in winters, during the of months November to December 2018 from the local area of Jhalwa, Prayagraj, authenticated and specimen kept at Botanical Survey of India, Prayagraj (Specimen No. 103973). The leaves were washed in running tap water in laboratory and kept 30 days for shade dry.

Animals
30 healthy male albino rats of wistar strain weighing 150-200g of male sex. Animals been housed in the polypropylene cages, and kept in room by providing proper condition (12h day and 12h light cycle; temperature at 23-25°C, 50% RH), standard customary pellet diets and water was been given to the rats. The Institutional Animal Ethical Committee (SIP-IAEC) approved this study (Registration No. SIP-IAEC/003/09/18).

Preparation of high fat diet (HFD)
Concentrated starch, flour, vanaspati ghee®, butter, cholesterol, casein, salt, 0.01% w/v PTU (propylthiouracil). Diet ratio is 12% carbohydrates, 65% fat, 20% protein, 3% minerals. A dose of 250g/kg body weight was been fed to the rats by orally daily within adding up to a normal diet for 1 week.

Preparation of extract
The dried leaves were been crushed into fine particles and then extracted with ethanol, ethyl acetate, distilled water, chloroform and petroleum ether. The prepared extracts were tested for various chemical constituents like flavonoids, phenols, vitamin C, etc. The highest percentage yield was obtained in aqueous extract.

**STUDY DESIGN**

For the study, the animals were weighed, recorded, numbered, and randomly divided into 5 groups (n=6) for a period of 1 week according to CPCSEA (Committee for the purpose of control and supervision of experiments on animals) for laboratory animal facilities [9, 10].

**Grouping and Treatment Schedules**

Group 1: Normal saline with along normal diet

Group 2: High fat diet (250g/kg/day) + PTU (10mg/kg/day) along with 0.01% solution of PTU throughout the day + atorvastatin (7.2mg/kg/day).

Group 3: High fat diet (250g/kg/day) + PTU (10mg/kg/day) along with 0.01% w/v solution of PTU throughout the day

Group 4: High fat diet (250g/kg/day) + PTU (10mg/kg/day) along with 0.01% w/v solution of PTU throughout the day + EO leave powder (200mg/kg/day)

Group 5: High fat diet (250g/kg/day) + PTU (10mg/kg/day) along with 0.01% w/v solution of PTU throughout the day + EO leave powder (500mg/kg/day)

The animals were kept under supervision for daily ingestion of high fat diet provided to them. The drugs were administered to the animals for 7 days by an oral cannula tube. At the end of 7th day, nightlong abstinence was done of all teams of animals. On 8th day, 3ml of blood was collected from the guts with the help of a syringe and the liver of the animals was been extracted. The blood was been collected and biochemical investigation was done for lipid profile, serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT)[11].

**Statistical analysis**

The statistical analysis between groups was done by using one way ANOVA, followed by Dunnet’s multiple comparison test. \( p < 0.001 \) taken as significance value (Table 1).

**Histopathological Study**

The 10% formalin solution was accustomed to resolute the various tissue samples of liver was dehydrated in ascending grades of ethyl radical alcohol, cleared in dissolver and embedded in paraffin. Sections of 5 \( \mu m \) thickness were cut by rotator microtome [12, 13]. Minimum 25 tissue sections for each organ were assessed. The sections were processed and passed through alcohol series, stained with hematoxylin and eosin, cleaned in xylene and cover-slipped in dibutylphthalate polystyrene xylene (DPX). Histological examination was done under magnification (10X) using microscope and further obtained from 10 random microscopic fields per animal at X45 and X100 objective [14].

**RESULT AND DISCUSSION**

The result obtained are been cumulated in the Table 1. The values were expressed in specific units of those parameters as shown in the table. The results of estimation were reported in the form of Mean ± SEM (standard error of mean) of animals (n=6) at a time from each group. It was seen that, there was a significant increase in all the lipid parameters \( (p < 0.001) \), excluding HDL, following administration of high fat diet with PTU.

It was also seen that concurrent administration of the EO leaves powder extract (aqueous) at a dose of 500mg/kg body weight along with HFD in the study animals; showed a significant decrease in all the lipid parameters \( (p < 0.001) \), excluding HDL, following administration of high fat diet with PTU.

The standard drug atorvastatin at a dose of 7.2mg/kg was administered along with a high fat diet which has showed a significant decrease \( (p < 0.001) \) in all the lipid parameters besides there was a significant rise in serum HDL. The hypolipidemic activity of the test drug was found to be slightly less effective than that of the standard drug, in comparison to the control group (Fig. 1).
Table 1: Effects of aqueous extract of EO leaves on blood lipid at the 8\textsuperscript{th} day (One way ANOVA followed by Dunnet’s multiple comparison test, data mentioned as Mean ± SEM)

<table>
<thead>
<tr>
<th>Group</th>
<th>SGOT (U/L)</th>
<th>SGPT (U/L)</th>
<th>TCH (mg/dl)</th>
<th>HDL (mg/dl)</th>
<th>LDL (mg/dl)</th>
<th>VLDL (mg/dl)</th>
<th>Trig (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>66.38±5.2</td>
<td>43.8±0.66</td>
<td>182.61±6.86</td>
<td>48.75±0.68</td>
<td>115.43±6.5</td>
<td>18.3±1.63</td>
<td>92±8.22</td>
</tr>
<tr>
<td>II</td>
<td>47.3±0.56</td>
<td>45.1±0.41</td>
<td>193.17±5.82</td>
<td>54.13±4.42</td>
<td>116.4±5.65</td>
<td>22.4±1.27</td>
<td>115.28±4.7</td>
</tr>
<tr>
<td>III</td>
<td>88.38±7.3</td>
<td>66.96±5.4</td>
<td>265.7±4.42</td>
<td>29.35±0.68</td>
<td>197.3±4.88</td>
<td>38.85±0.76</td>
<td>194.28±3.8</td>
</tr>
<tr>
<td>IV</td>
<td>84.76±7.3</td>
<td>64.48±5.5</td>
<td>257.42±4.41</td>
<td>35.61±1.1</td>
<td>186.17±4.5</td>
<td>36.23±0.19</td>
<td>181.15±0.9</td>
</tr>
<tr>
<td>V</td>
<td>62.85±5.7</td>
<td>52.97±2.16</td>
<td>197.17±5.25</td>
<td>44.9±1.32</td>
<td>121.64±6.35</td>
<td>30.63±1.36</td>
<td>153.16±6.2</td>
</tr>
<tr>
<td>F†</td>
<td>8.467</td>
<td>8.928</td>
<td>52.234</td>
<td>21.573</td>
<td>52.608</td>
<td>56.99</td>
<td>61.987</td>
</tr>
<tr>
<td>Df††</td>
<td>25, 4</td>
<td>25, 4</td>
<td>25, 4</td>
<td>25, 4</td>
<td>25, 4</td>
<td>25, 4</td>
<td>25, 4</td>
</tr>
<tr>
<td>P†††</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

† : Statistic Value; †† : Degree Of Freedom; †††: Significance value of compared groups p<0.01

(Fig. 1: Graph showing mean serum lipid parameters in 5 groups at the end of 8\textsuperscript{th} day)
Liver function assessment activity levels of serum (SGOT, EC 2.6.1.1 and SGPT, EC 2.6.1.2) were assayed in control and other group experimental rats (plasma) using kits. Liver function assessment activity levels of plasma SGOT and SGPT were significantly elevated in toxic control group (SGOT-88.38, SGPT-66.96) compared to the normal group (SGOT-66.38, SGPT-43.8) and significantly decrease in Group-V (SGOT-62.85,SGPT-52.97) compared to the control group (hyperlipidemia). Group-V recorded decrement in SGOT levels whereas; there was significant change in SGPT levels as compared to toxic control (Fig. 2).

**HISTOPATHOLOGY**

Histopathological studies explicit that hyperlipidemia is a result of an oxidative abuse due to free radicals, formed by the interaction of high fat diet. They further stated that, an augmentation in the concentration of serum cholesterol and triglycerides of hyperlipidemic rats may be result of lipid peroxidation elicited by high fat diet [15]. In the present study, atorvastatin was used as a standard drug [16].

EO powder, administered in hyperlipidemia rats can elicit a profound influence on the lipid metabolism. An enhancement in the concentration of total serum cholesterol, serum triglycerides, serum LDL, atherogenic index of hyperlipidemia rats was observed, which was probably due to lipid peroxidation caused by high fat diet. Lipid peroxidation is a free radical mediated procedure which has been occupied in a variety of diseased conditions [17].
Dyslipidemia is one of the autonomous major risk factors for cardiovascular disease (CVD) throughout the world. It is mainly caused by the sedentary lifestyle, genetic, behavioural and nutritional modifications. As the synthetic hypolipidemic drugs presently available are costly and possess various side effects upon long term treatment, there exists a continuous demand for the development of promising plant based alternative hypolipidemic agents which are easily available and claimed to be cheaper and comparatively safer.

Aqueous extract displayed the presence of flavonoids, tannins and saponins, while in petroleum ether extract, only tannins and phenolic compounds could be detected. Latest epidemiological studies have concealed that the intake of flavonoids is reciprocally connected to the risk of coronary heart disease. EO powder, rich in flavonoids and polyphenols may also be causative towards its hypolipidemic effect, because of its ability to combat oxidative stress by quenching free radicals generated in the body because of high fat diet. It may also act by triggering the emission of anti-oxidant enzymes: superoxide dismutase, catalase and glutathione peroxidase in an enhanced level, which in turn blocked the oxidative damage due to hyperlipidemia.

In a latest study reveals that in type II diabetes mellitus patients, the leaves of EO exhibit lipid lowering properties. The study also demonstrated improved endothelial function, reduction in biomarkers of oxidative stress and systemic inflammation in those patients [18].

The phyto-constituents of EO have been shown to possess hypolipidemic activity. Hypolipidemic effects have been occurred due to flavonoids, alkaloids, saponins and tannins. Flavonoids are been reported to have the potential to increase the HDL-C concentration and decreases LDL and VLDL levels in hypercholesteromic rats. Flavonoids from lotus were shown to reduce hyperglycemia and hyperlipidemia in alloxan-induced diabetic mice. Total flavonoids of *Perilla frutescens* leaves have showed lipid lowering properties in hyperlipidemic rats [19, 20].

**CONCLUSION**

The present study showed that aqueous extract of *Emblica officinalis* exhibits antihyperlipidemic action against high fat diet along with PTU. Antihyperlipidemic activity was carried out on male wistar albino rats. The level of dose given was 200mg/kg and 500mg/kg body weight out of which the dose level 500mg/kg has shown the significant antihyperlipidemic action than 200mg/kg body weight.

Histopathological examination of the liver section of rats treated with extract dose has shown the antihyperlipidemic actions evident by the absence of necrosis and vacuoles. Thus, it can be concluded that extract exhibits dose dependent antihyperlipidemic action. From the above preliminary study, it is concluded that aqueous extract of *Emblica officinalis* can be one of the herbal remedy for antihyperlipidemic purpose. Further studies are though recommended to characterize the active chemical constituent responsible for the antihyperlipidemic action and to elucidate the mechanism.

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**REFERENCES**


