LIPID LOWERING ACTIVITY OF ALCOHOLIC EXTRACT OF CYPERUS ROTUNDUS

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ABSTRACT
Synthetic hypolipidaemic agents, on prolonged usage, lead to serious side effects. Hence there is focus on Indian Traditional system of medicine for long term treatment of lipid disorders. One such plant Cyperus rotundus, commonly known as Nagarmotha, is investigated for hypolipidaemic activity on high fat diet induced hyperlipidaemic rats of wistar strain. For the study alcoholic extract of the rhizomes was used. The results demonstrated statically significant reduction in serum lipid profile.

Keywords: Lipid disorder, Cyperus rotundus, High fat diet and Serum lipid profile.

INTRODUCTION
Coronary heart disease (CHD) is the most common cause of mortality and morbidity worldwide. Over 50% of CHD is developed due to excess of blood cholesterol levels. There is closer association between CHD and hyperlipidaemia. Hyperlipidaemia is a term used to describe elevated plasma levels of lipids (triglycerides and cholesterol) and lipoproteins. Concentration of plasma total cholesterol and low density lipoprotein (LDL) cholesterol are highly correlated with the prevalence of coronary heart disease while a high plasma HDL cholesterol concentration is a powerful protective factor against coronary heart disease. The allopathic drugs available for the treatment of hyperlipidaemia are fibrate derivatives, HMG-CoA reductase inhibitors, bile acid binding agents, nicotinic acid but they show side effects like kidney and liver impairment, gall stone formation, rhabdomyolysis (destruction of skeletal muscles) and gastrointestinal disturbances.

India has got rich flora of herbs due its unique climatic and geographical condition. The Indian system of medicine have been regarded as a rich mine of ethnopharmacological knowledge. One such plant called Cyperus rotundus Family: Cyperaceae. It is mentioned in Charaka Samhita has been claimed to be effective in medoroga (lipid disorder). The Cyperus rotundus is also known as musta or nagarmotha. The rhizomes of plant have been reported to possess various medicinal properties like anthelmintic, antipyretic, antidepressant, anti-rheumatic, anti-spasmodic and anti-fungal.

Since there is no any report on hypolipaidemic activity of the drug, it was decided to screen for hypolipidaemic activity. In the present study the effect of alcoholic extract of Cyperus rotundus rhizome on the serum lipid profile of hyperlipidaemic rats of wistar strain was done. The hyperlipidaemia was induced by feeding them on high fat diet. The extract was also screen for its mode of action.
MATERIALS AND METHODS
Collection and Extraction of Plant Materials
The dried rhizomes of Cyperus rotundus were procured from local market and authenticated from Agarkar Institute Pune (Voucher no. R082). The rhizomes were coarsely powdered and subjected to extraction with absolute alcohol for eighteen hours in soxhlet extractor. The extract is further concentrated using rotavac evaporator and subsequent drying on water bath.

Preliminary Phytochemical Investigation
Preliminary phytochemical investigation revealed the presence of saponins, carbohydrates, essential oils and phenols.

Animals
Inbred adult male rats of wistar strain weighing 250–300 grams were selected for the study. The animals were housed in polypropylene cages (6 rats per cage) under good hygienic conditions natural light / dark cycle.

Acute toxicity study
For the purpose of the test, in bred wistar strain rats (250–300 g) of both sex were selected. The animals were housed in polypropylene cages (6 rats per cage) under good hygienic conditions natural light / dark cycle. The animals were given free access to standard pellet diet and water.

Acute toxicity study was carried out as per OECD guideline7.

Thus the oral acute toxicity tests revealed that the extract of Cyperus rotundus rhizomes was safe up to the administered dose 2000 mg/kg.

Hypolipidaemic Activity
Wistar rats weighing 250-300 g were selected for the study. Animals were divided into 7 groups, each group comprising of 6 rats. Rats in the group 1 received normal pellet diet and received 0.1% sodium CMC solution and served as vehicle control. The rats belonging to remaining 6 groups received high fat diet for the entire duration of the study that is for 25 days. The high fat diet was comprised of the chow enriched with high calorie and 1% cholesterol. High fat diet induced hyperlipidaemia is one of the common method to induce hyperlipidaemia. Hence hyperlipidaemia was induced by oral feeding of high fat diet. The animals were given free access to water.

After 10 days induction of hyperlipidaemia group 2 of animals was left untreated and served as high fat diet control. The rest of the groups received following treatment for 15 days.

Group 3 and group 4 treated orally with the standard drugs Simvastatin (5 mg/ kg/day) and Fenofibrate (20 mg/ kg/day) respectively. Groups 5, 6, 7 treated orally with alcoholic extract at dose level of 70 mg/ kg/day, 140 mg/ kg/day, 280 mg/ kg/day respectively. Blood samples were withdrawn from retro orbital plexus after overnight fasting. All the drugs were suspended in 0.1% Na CMC. Serum was separated from blood by centrifugation for ten minutes at three thousand rpm, subsequently analyzed for total cholesterol, triglycerides and HDL cholesterol using commercially available kits (Erba Diagnostics Germany). The serum LDL was calculated by Friedwald’s formula10.

Statistical Analysis
The data was analyzed for statistical significance by one way Analysis of Variance (ANOVA) followed by Dunnet’s t-test for comparison with the control groups. Bonferroni’s t-test was performed for pair wise comparison among the extracts and reference standards. The difference was considered to be significant at 5% level (P<0.05).

RESULTS AND DISCUSSION
Effect of alcoholic Cyperus rotundus extracts on serum lipid profile in rats
In the present study hyperlipidaemia was induced by high fat diet as it is always useful for the assessment of agents that interfere with the absorption, degradation and excretion of cholesterol.

Feeding with high fat diet caused significant (P<0.05) increase in serum TC, triglyceride and LDL levels with respect to the baseline value which shown in Table 1. Though on high fat diet feeding an increase in HDL levels were seen but they were not found to be statistically significant.

In present study treatment with the standards and different doses of extract exerted statistically significant (P<0.05) reduction in serum total cholesterol, LDL, TG, HDL levels at the end of 15 days of intervention. Table 2 describes those values.
CONCLUSION
The results found, clearly demonstrated that the bioactive compounds present in Cyperus rotundus have promising ability to attenuate the serum lipid profile in high fat diet fed hyperlipidaemic rat model. Thus the present study assess the hyperlipidaemic action of Cyperus rotundus rhizomes and also validates the claim made in the traditional system of medicine and paves way for potential evaluation of the mediators involved in hyperlipidaemia and discovering new therapeutic targets for the evaluation of lipid lowering medicinal plant.

<p>| Table 1: Effect of high fat diet on serum lipid profile at the end of 10 days induction period in rats |</p>
<table>
<thead>
<tr>
<th>Blood Parameters</th>
<th>Basal value (N=42)</th>
<th>Induction Value (N=36)</th>
<th>Increase (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum TC (mg/dl)</td>
<td>51.30±0.86</td>
<td>211.57±2.10*</td>
<td>312.41</td>
</tr>
<tr>
<td>Serum TG (mg/dl)</td>
<td>80.71±0.53</td>
<td>262.12±3.23*</td>
<td>242.19</td>
</tr>
<tr>
<td>Serum HDL (mg/dl)</td>
<td>27.93±1.37</td>
<td>59.88±0.87</td>
<td>114.93</td>
</tr>
<tr>
<td>Serum LDL (mg/dl)</td>
<td>7.23±0.77</td>
<td>99.26±2.09*</td>
<td>1272.28</td>
</tr>
</tbody>
</table>

*P<0.05 considered significant increase when compared with basal values.
(Values are mean ± SEM)
TC: Total cholesterol, TG: Triglyceride, HDL: High density Lipoprotein, LDL: Low density lipoprotein.

<p>| Table 2: Effect of alcoholic extract of Cyperus rotundus rhizomes on serum lipid profile |</p>
<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Treatment groups</th>
<th>Serum TC (mg/dl)</th>
<th>Serum LDL (mg/dl)</th>
<th>Serum TG (mg/dl)</th>
<th>Serum HDL (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Vehicle control</td>
<td>59.79 ± 3.02</td>
<td>10.68 ± 1.97</td>
<td>70.17 ± 1.24</td>
<td>35.07 ± 1.43</td>
</tr>
<tr>
<td>2</td>
<td>High fat diet control</td>
<td>296.43 ±4.27**</td>
<td>150.54 ± 11.04**</td>
<td>401.65 ± 14.16**</td>
<td>65.57 ± 6.26</td>
</tr>
<tr>
<td>3</td>
<td>Simvastatin (5 mg/kg)</td>
<td>260.85 ± 8.08*</td>
<td>132.00 ± 7.09*</td>
<td>331.16 ± 11.47*</td>
<td>62.62 ± 4.62</td>
</tr>
<tr>
<td>4</td>
<td>Fenofibrate (20 mg/kg)</td>
<td>230.23 ± 2.55*</td>
<td>99.61 ± 2.70*</td>
<td>336.43 ± 11.40*</td>
<td>63.33 ± 3.43</td>
</tr>
<tr>
<td>5</td>
<td>Alcoholic extract (70 mg/kg)</td>
<td>263.94 ± 4.27**</td>
<td>133.03 ± 11.16*</td>
<td>348.29 ± 4.56*</td>
<td>61.25 ± 1.81</td>
</tr>
<tr>
<td>6</td>
<td>Alcoholic extract (140 mg/kg)</td>
<td>249.48 ± 4.26*</td>
<td>126.56 ± 5.24*</td>
<td>301.93 ± 6.25*</td>
<td>62.53 ± 1.16</td>
</tr>
<tr>
<td>7</td>
<td>Alcoholic extract (280 mg/kg)</td>
<td>239.44 ± 4.028*</td>
<td>119.37 ± 2.60*</td>
<td>276.53 ± 9.70*</td>
<td>64.76 ± 0.62</td>
</tr>
</tbody>
</table>

*P<0.05 considered significant decrease when compared with high fat diet control
**P<0.05 considered significant increase when compared with vehicle control.
(Values are mean ± SEM) (N=6)
TC: Total cholesterol, TG: Triglyceride, HDL: High density Lipoprotein, LDL: Low density lipoprotein.

REFERENCES
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