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Favourable Effect of *Cleome viscosa* L. on Serum and Hepatic Lipids in Hyperlipidemic Rats

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Abstract : Hypolipidemic activity of methanolic extract of *C. viscosa* seeds was evaluated in hyperlipidemic rats. Hyperlipidemia was induced by oral administration of cholesterol (500mg/ kg b.wt./ day) suspended in coconut oil (0.5 ml/rat/day) for 60 days. Administration of *C. viscosa* seed extract (250mg/ kg b.wt.) along with cholesterol for 60 days significantly suppressed elevation of serum total cholesterol ($P<0.001$), LDL cholesterol ($P<0.001$), triglycerides ($P<0.05$) and phospholipids ($P<0.01$) concentrations when compared with cholesterol feld control rats. The HDL-cholesterol : total cholesterol ratio was significantly ($P<0.001$) increased. Hepatic total lipids ($P<0.01$), cholesterol ($P<0.01$) and triglycerides ($P<0.01$) were also lowered. Co-administration of *C. viscosa* extract raised ($P<0.01$) the level of cholesterol in feces. The results indicate favourable hypolipidemic effect of methanolic extract of *C. viscosa* seeds in hyperlipidemic rats.

Key words : *Cleome viscosa,* hypolipidemic effect, cholesterol fed rats.

Introduction :

Cleome viscosa Linn. (Syn. *C. icosanra* Linn.), a member of the family Capparidaceae, is a common weed found all over India. Various parts of *C. viscosa* plant is described in Ayurveda and other systems of medicine to cure many diseases. The seeds are antihelmintic, carminative, cardiac stimulant and are also useful in fever and diarrhoea. Externally they act as rubefacient and vesicant (Asolkar *et al.*, 1992; Nadkarni, 1998). The whole plant is useful in liver disease, chronic painful joints and mental disorders (Chatterjee and Pakrashi, 1991).

Cleome viscosa is a good source of vitamin-C and iron. It is used as a green leafy vegetable by poorer segment of the population (Theophilus and Arulanantham, 1949). The seeds are rich in unsaturated fatty acid (linoleic acid) and phytosterol and has been reported to be devoid of any toxic effect in rats on sub-chronic feeding (Rukmini, 1978). Methanolic extract of the whole plant showed the presence of steroids, triterpenoids, flavonoids and tannins (Rastogi and Mehrotra, 1991).

Data on the efficacy and biological activity of this plant for hypolipidemic activity are not available. This study was designed to examine the effect of methanolic extract of *C. viscosa* seeds on serum and liver lipids and fecal cholesterol excretion in cholesterol fed rats.

Materials and methods :

Plant material and extraction : *C. viscosa* plants were collected in the month of September - October, 2004, and authenticated at the Herbarium, Department of Botany, University of Rajasthan, Jaipur (RUBL No. 19901). The seeds of the mature plants were separated and dried in shade. The seeds were coarsely powdered and subjected to soxhlet extraction with

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methanol for 30 hrs. The extract was filtered and concentrated to dryness at low temperature. The viscous extract so obtained was used for experimental study by suspending in double distilled water.

Animal : Colony bred, adult, healthy Wistar strain male rats (175-230g) were used for this study. The rats were maintained under standard laboratory condition at 22 ± 3^0C and normal photoperiod (12 hrs dark/light cycle). Commercial pellet diet (Lipton, India Ltd.) and water were provided *ad libitum.* The study was approved by the Animal Ethical Committee of the University Department of Zoology, Jaipur and standard guidelines were followed for maintenance and use of the experimental animals.

Experimental protocol : The rats were randomly selected and divided into three groups, each of 7 rats. Group I received distilled water as vehicle (0.5 ml/rat). Group II received cholesterol (500 mg/kg b.wt/day/ orally using intragastric tube) suspended in coconut oil (0.5 ml/rat/day) for 60 days. Group III received cholesterol (as above) + *C. viscosa* seeds extract (250 mg/kg b.wt./ day, orally) suspended in distilled water (0.5 ml/rat/day) for 60 days.

Autopsy : At the end of experiment, the rats were weighed and kept for overnight fasting. These were sacrificed under mild ether anesthesia. Blood sample was collected by cardiac puncture and the serum was separated and stored at-20^0C for biochemical estimations. Liver was quickly removed, washed with cold normal saline and immediately frozen (-70^0C) for biochemical analysis.

Serum lipid profiles : Serum samples were analysed for total cholesterol (Zlatkis *et al.,* 1953), HDL-cholesterol (Burstein *et al.,* 1970), LDL-cholesterol (Friedwald *et al*., 1972) triglycerides (Gottfried and Rosenberg, 1973) and phospholipids (Zilversmit and Davis, 1950).

Liver biochemistry : Total lipids from liver samples were extracted and determined gravimetrically (Folch *et al.*, 1957). Liver total cholesterol and triglycerides were determined according to Zlatkis *et al.* (1953) and Gottfried and Rosenberg (1973) methods, respectively.

Fecal cholesterol : Fecal matter of all the rats receiving various treatments was collected during the last five days of treatment. The fecal matter was dried at 40^0C and used for analysis of total cholesterol by Zlatkis *et al.* (1953) method.

Statistical analysis : All results are presented as mean ± SEM. Statistical evaluation was done with Student 't' test. Differences were considered to be statistically significant at $P<0.05$ level.

Results :

The final body weights of all the rats of different groups showed a slight significant ($p<0.05$) increase when compared with their initial body weights (data not presented). Cholesterol feeding to rats maintained on normal diet significantly raised the levels of serum total cholesterol ($P<0.001$), LDL-cholesterol ($P<0.001$), triglycerides ($P<0.001$) and phospholipids ($P<0.001$) when compared to normal rats. The levels of serum HDL-cholesterol remained unchanged. However, the HDL-cholesterol : total cholesterol ratio was lowered significantly. Simultaneous treatment of *C. viscosa* extract along with cholesterol prevented the elevation of serum total cholesterol ($P<0.01$), LDL-cholesterol ($P<0.001$); triglycerides ($P<0.05$) and phospholipids ($P<0.01$) when compared

with cholesterol fed control rats. The HDL-cholesterol : total cholesterol ratio was significantly ($P<0.001$) increased as compared to cholesterol fed control rats. (Table 1).

Cholesterol feeding in rats caused a significant ($P<0.001$) increase in liver total lipids, cholesterol and triglycerides as compared to normal rats. Co-administration of extract with cholesterol caused significant decline in total lipid ($P<0.01$), cholesterol ($P<0.01$) and triglycerides ($P<0.01$) concentrations in liver when compared with cholesterol fed control rats. (Table 2).

Administration of cholesterol in rats caused a significant ($P<0.001$) increase in fecal excretion of cholesterol. Co-administration of *C. viscosa* extract with cholesterol, further raised ($P<0.01$) the level of cholesterol in feces (Table 2).

Discussion :

In the present study, the hypolipidemic activity of methanolic extract of *C. viscosa* seeds was evaluated in cholesterol fed rats. Cholesterol feeding in rats significantly raised the levels of serum and hepatic lipid profiles. These results are consistent with many earlier reports (Anila and Vijayalakshmi, 2002; Khanna *et al*., 2002). Excessive dietary intake of fat cause serum cholesterol to rise by down regulating LDL-receptor synthesis as a result of which the uptake of LDL-cholesterol via LDL receptor is reduced which result in an increase of blood cholesterol level (Dietschy *et al*., 1993).

Administration of *C. viscosa* seed extract (250 mg/kg b.wt) along with cholesterol, lowered serum total and LDL-cholesterol by –27.81% and –40.09%, respectively. This reduction in the levels of both total and LDL-cholesterol might be useful in reducing the risk of coronary heart disease (Lipid Research Clinics Program, 1981). The observed reduction in total cholesterol was mainly associated with lowering of LDL-cholesterol, as evidenced by non significant effect on HDL-cholesterol level. The HDL-cholesterol : total cholesterol ratio was significantly higher than that of cholesterol fed control rats which is predictive of reduced risk of coronary heart disease (Malaspina *et al*., 1981; Shin *et al*, 2004).

Administration of *C. viscosa* extract significantly reduced serum triglycerides which is also beneficial effect. The mechanism by which *C. viscosa* extract lowered serum triglycerides is not clear but it may be possible due to either a decrease in VLDL synthesis or channeling of VLDL to pathway other than to LDL or an increase in lipoprotein lipase activity. The observed reduction in hepatic total lipids, cholesterol and triglycerides in extract treated rats might be due to modulatory influence on lipogenic enzymes or by inhibition of cholesterol absorption (Devi and Sharma, 2004).

The increase in fecal cholesterol excretion in rats fed *C.viscosa* extract might be caused by a reduction in cholesterol absorption resulting in higher cholesterol catabolism in the liver, consequently lowering of serum cholesterol concentration (Kim and Shin, 1998).

Nicola et al., (1991) also observed lowering of total and LDL- cholesterol and an increase in HDL cholesterol : LDL-cholesterol ratio in rats treated with *Cleome droserifolia,* which is very closely related to present plant.

Table 1 : Effect of *C. viscosa* seeds extract on serum lipid profiles.

Treatments	Total cholestrol (mg/dl)	HDL-cholesterol (mg/dl)	LDL-cholesterol (mg/dl)	Triglycerides (mg/dl)	Phospholipid (mg/dl)	HDL-c/Tc ratio
Normal	107.89 ± 3.07	39.21 ± 1.58	53.86 ± 1.57	76.31 ± 5.11	121.40 ± 6.24	0.35 ± 0.009
Cholesterol fed Control (% change)	198.11 ± 7.32[a] (+83.62%)	42.20 ± 1.62 (+ 7.62%)	130.07 ± 5.12[a] (+ 148.92%)	109.21 ± 4.85[a] (+ 43.12%)	184.32 ± 8.44[a] (+ 51.83%)	0.21 ± 0.003[a] (- 40.0%)
Cholesterol + *C. viscosa* extract (% change)	143.01 ± 4.64[**] (-27.81%)	43.81 ± 1.96 (+3.81%)	80.31 ± 2.27[***] (-40.09%)	94.40 ± 3.38[*] (-13.56%)	148.50 ± 6.28[**] (-19.43%)	0.30 ± 0.005[***] (+ 42.85%)

Values are mean+SEM of 5 out of 7 animals. HDL-cHDL-cholesterol, TcTotal cholesterol

Levels of Significance

[a] P < 0.001 when compared to normal rats

[*] P < 0.05 ; [**] P<0.01 ;

[***] P < 0.001 when compared with the cholesterol fed control rats.

Table 2. Effect of *C. viscosa* seeds extract on liver total lipids, total cholesterol, triglycerides and fecal cholesterol levels in rats.

Treatments	Liver (mg/g)			Fecal cholesterol (mg/g)
	Total lipids	Total cholesterol	Triglycerides	
Normal	46.67 ± 3.93	9.78 ± 1.07	8.63 ± 0.79	8.60 ± 1.41
Cholesterol fed Control	82.01 ± 4.42 [a]	20.31 ± 1.72 [a]	18.04 ± 1.92 [a]	26.84 ± 3.09 [a]
Cholesterol + *C. viscosa* extract	62.82 ± 2.12[**]	14.80 ± 0.90[**]	11.55 ± 0.55 [**]	42.80 ±+ 2.71[**]

Values are mean+SEM of 5 out of 7 animals

Levels of Significance

[a] P < 0.001 compared to normal rats

[**] P < 0.01 compared with the cholesterol fed control rats.

Phytochemical studies of this plant have shown the presence of ascorbic acid, plant sterols, polyunsaturated fatty acid, saponins and flavonoids (Rukmini, 1978; Rastogi and Mehrotra, 1991). The observed hypolipidemic effect might be due to individual or synergistic action of these components (De Silva *et al.*, 2001;De Jong *et al*., 2003; Ristic and Ristic, 2003). The exact mechanism of hypolipidemic effect of *C. viscosa* is yet to be understood.

From these results it is concluded that methanolic extract of *C. viscosa* seeds effectively decreases serum and hepatic lipid levels in cholesterol fed rats. Further, studies are required to gain more insight in to the mechanism of hypolipidemic action.

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