Anti-Hyperlipidemic Activity of Ethanolic Extract of Salvia Hypoleuca in Rats

Jasem Estakhr[1]*, Nasim Javdan[1], Shahla Najafi[2]

[1] Science and Research Branch, Islamic Azad University, Fars, Iran.

[2] Department of Biology, Faculty of Science, University of Zabol, Zabol, Iran.

***Corresponding Address:** Jasem Estakhr, j.estakhr@yahoo.com. Tel: +989179283966.

Summary

Excess of lipids (cholesterol and triglycerides) in the blood is called hyperlipidemia. The main purpose of treatment in patients with hyperlipidemia is to reduce the risk of developing ischemic heart disease or the occurrence of further cardiovascular or cerebrovascular disease. In this study the main aim is to reduce the levels of triglycerides by using ethanolic extract of Salvia hypoleuca. Results showed that the food intake and body weight was increased in high cholesterol fed diet rats as compared to normal control. Data indicated that food intake did not change in rats treated with ethanolic extract of Salvia hypoleuca but there was a significant decrease in weight gain as compared to high cholesterol fed diet rats. Results showed that there was a significant reduction in ($p<0.001$) Total Cholesterol (TC), LDL, and Triglyceride ($p<0.001$), but significant increase ($p<0.01$) in HDL-C as compared to high cholesterol diet fed rats. In conclusion, Salvia hypoleuca has a strong anti-hyperlipidemic activity in rats and potential for manufacturing drugs.

Keywords: Salvia hypoleuca, Anti-hyperlipidemic activity, Rat.

Introduction

Hyperlipidemia includes several conditions, but it usually means that the high cholesterol and high triglyceride levels. One of the main causes of atherosclerosis and atherosclerosis-associated conditions, such as Coronary Heart Disease (CHD), ischemic cerebrovascular disease and peripheral vascular disease is hyperlipidemia. Reduction in serum lipids (cholesterol and triglycerides) levels reduces the risk for CHD. Reduction the risk of developing ischemic heart disease or the occurrence of further cardiovascular or cerebrovascular disease is the major aim of treatment in patients with hyperlipidemia. High lipid levels can accelerate a process called atherosclerosis, or hardening of the arteries. Lipoproteins are the “packages” in which cholesterol and triglycerides (lipids) are transport throughout the body by combining with proteins. Lipoproteins contain cholesterol, phospholipids and triglycerides at the core and an outer layer of protein called “apolipoproteins/apoproteins” (1). The lipid constituents are water insoluble and apoproteins are water soluble (2).Depending on upon their lipid carrying capacity Lipoproteins are classified into (3).Depending on the complexity of the disease, hyperlipidemia is classified into two types. Primary Hyperlipidemia and Secondary / Acquired Hyperlipidemia (4).

Today, medicinal plants are increasingly being used in most parts of the world for gaining of better health. Hypolipidemic agents like medicinal plants have been used in the management of Hyperlipidemia. The genus Salvia, one of the most important genuses of Lamiaceae family, is widely used in flavouring and folk medicine all around the world. Fifty-eight species of this genus are documented in the Flora of Iran; 17 of them are endemic (5-7). The plants of the genus Salvia, which consist about 900 species are generally known for their multiple pharmacological effects

such as analgesic and anti-inflammatory (8) hepatoprotective (9), hypoglycemic activities (10), and antiischemia (11, 12). Survey of the literatures revealed that the anti- hyperlipidemic activity of Salvia hypoleuca has not been clinically evaluated so far. In view of this, the present study was aimed at evaluating the anti- hyperlipidemic activity of the ethanolic extract of Salvia hypoleuca in rats.

Materials and Methods

Preparation of the extract of Salvia hypoleuca

Salvia hypoleuca was collected from Guilan province (Iran), and authenticated at Medicinal Plants & Drugs Research Institute, Shahid-Beheshti University, Tehran, Iran. Its leaves and fruits were dried, under shade and powdered. Air dried Powdered plant was divided in to two equal parts (1 kg each); one part was macerated with ethanol (90 % v/v) in glass percolator and allowed to stand at room temperature for about 24 hours. Then the extract obtained was filtered, concentrated by rotary vacuum pump to get the solid mass. The percentage yield was 19.6 %, second part of powdered was cold macerated with distilled water then the same procedure was repeated as mentioned above. The percentage yield was 10.5%.

Animals' treatment

Albino mail wistar rats weighing 150-200 g of either sex, 4 months of age were used for this study. The experimental animals were housed in polypropylene cages and maintained under standard conditions (12 h light and dark cycles, at 25±3º C and 35-60% humidity). Standard pelletized feed and tap water were provided ad libitum. Male wistar rats were induced for hyperlipidemic by the oral administration of cholesterol (400 mg/kg) along with cholic acid (50 mg/kg) in coconut oil for 20 days, once daily. The rats with elevated cholesterol level were divided into 5 groups of 6 animals each and given drug/ vehicle treatment for 7 days. Group – I: Normal control - received vehicle (Acacia 4%) 5 ml/kg.p.o., Group – II: Hyperlipidemic Control – received vehicle (Acacia 4%) 5 ml/kg. p.o., Group – III: Standard - received atorvastatin10mg/kg p.o., Group- IV: received ethanolic extract of Salvia hypoleuca 200 mg/kg; p.o., Group- V: received ethanolic extract of Salvia hypoleuca 400 mg/kg; p.o. During these days all groups also received cholesterol in same dose as earlier. After the experimentation period, blood sample was collected by retro orbital sinus puncture under mild ether anaesthetic condition and allowed to clot for 30 minutes at room temperature. Blood samples were centrifuged at 3000 rpm for 20 minutes. Serum was separated and stored at - 20°C for analyzing cholesterol, triglycerides, LDL, VLDL and HDL levels using standard methods.

Statistical analysis

For statistical evaluation, data were expressed as mean ±SEM (n=6) and One-way ANOVA and student t-test were performed.

Results and Discussion

Balance between synthesis and degradation of biological tissues is maintained by Lipid metabolism. Coronary heart diseases (CHD) are the clinical manifestation of atherosclerosis. Development of hyperlipidemia disease is a complicated process involving accumulation of lipid containing particles in the walls of coronary arteries & other major arteries within the body. A high-fat diet causes cholesterol levels to increase in susceptible people, which leads to obesity. The weight gain in high cholesterol diet (HCD) group of rats was significantly higher than control rats reflecting the influence of high cholesterol diet. High fat diet fed rats showed

significant increase ($p<0.001$) in Serum cholesterol, triglyceride, LDL-C, but significant decrease ($p<0.001$) in HDL-C level as compared to 0 day and normal control rats. Treatment with Salvia hypoleuca extract (200mg/kg) exhibited the significant reduction ($p<0.05$) in triglyceride, LDL-C. Treatment with Salvia hypoleuca extract (400mg/kg) showed significant reduction ($p<0.001$) Total Cholesterol (TC), LDL, and Triglyceride ($p<0.001$), but significant increase ($p<0.01$) in HDL-C as compared to high cholesterol diet fed rats. In this study there was also a significant weight gain in cholesterol control as compared to normal control group. Treatment with Salvia hypoleuca significantly reduced the weight gain. Lowering high cholesterol levels significantly reduced the risk of heart attacks, strokes, and death. Normally hepatocyte initiate synthesis of triglycerides and cholesterol during states of increased free fatty acid flux to the liver but due to anti-hyperlipidemic drug, there may be inability of hepatocytes to increase cholesterol synthesis and decrease hepatocyte cholesterol concentration by increases the catabolic conversion of cholesterol to bile acids in liver. High cholesterol diet increased serum cholesterol and LDL-C level significantly. A rise in LDL may cause deposition of cholesterol in arteries and aorta and hence it is a direct risk factor for coronary heart disease (13).In the present study, there was an elevation in serum and tissue cholesterol, LDL-C, in response to high cholesterol diet as compare to normal control group. Treatment with ethanolic extract of Salvia hypoleuca significantly reduced serum and tissue cholesterol, LDL-C and triglyceride. Recent studies show that triglycerides are independently related with coronary artery disease (14). Treatment with ethanolic extract of Salvia hypoleuca shows significant decreased in triglyceride. HDL is synthesized mainly in intestine and liver. HDL is considered to be a beneficial lipoprotein as it has an inhibitory effect in the pathogenesis of atherosclerosis. Low level of HDL is associated with high risk of coronary artery disease. In the present investigation, HDL-C level in both serum and tissue were significantly increased by ethanolic extract of Salvia hypoleuca.

Table 1: Effect of ethanolic extract of Salvia hypoleuca on body weight gain of hyperlipidemic rats.

Groups	0^{th} Day	8^{th} Day	16^{th} Day	24^{th} Day	28^{th} Day
Normal	181±11.41	181±13.42	181±13.43	181±11.89	181±11.13
Control	180±12.58#	192±13.06#	236 ± 13.38#	275±23.23#	297±11.58#
S. hypoleuca (200 mg/kg)	183±13.03**	177±17.11**	174±23.12**	165±18.16**	164±9.36**
S. hypoleuca (400 mg/kg)	189±15.13**	184±32.04**	171±13.05**	169±12.05**	165±15.22**
Atorvastatin	183±14.20**	171±12.03**	148±12.36**	124±11.02**	124±11.02**

Values are as mean ± SEM, #P < 0.05, **P < 0.01, Vs Control. (n=6)

Table 2: Effect of ethanolic extract of Salvia hypoleuca on lipids level in hyperlipidemic rats.

Groups	TC (mg/dl)	HDL (mg/dl)	TGL (mg/dl)	LDL (mg/dl)
Normal	85±4.745	37±2.324	71±2.085	31±2.865
Control	177±3.326###	16±2.322###	151±3.241###	126±3.432###
S. hypoleuca (200 mg/kg)	164±4.643	19±3.121	133±5.324*	122±3.842*
S. hypoleuca (400 mg/kg)	151±4.321**	24±1.112*	111±1.854***	102±2.532**
Atorvastatin	105±5.432**	33±1.333***	91±4.453***	89±2.632***

Values are as mean ± SEM, ###P < 0.05Vs Normal; *P<0.05,**P<0.01, ***P<0.001 Vs Control.

References

1- Goodman and Gilman, Ed. The pharmacological basis of therapeutics, 10th Edn, McGraw hill medical publishing division, New York, 2001; 242-265.
2- Nancy J Nordensos and Teresa G odle., Ed. Lipoprotein test, Health A to Z Encyclopedia index L Source. Gale encyclopedia of medicine, by gale group, 2002; 211-219.
3- Raja chattopadhyaaya, Devender Pathak and Dharam Paul Jindal., Antihyperlipidemic agents – a review. Indian drugs, 1996; 33 (3): 85-97.
4-Sharpless KB, Snyder TE, Spencer TT, Maheswari KK, Guhn G, and Clayton RB., Terpenoid Biosynthesis: the Stereochemistry of Squalene Cyclisation. Journal of American Chemical Society, 1969; 91 (13): 3394.
5- Zargari A. Medical plants, Tehran: Tehran University. 2003; 3595–6.
6- Rustayan A, Masoudi S, Monfared A, Komilizadeh H. Volstile constituents of three Salvia species grown wild in Iran. Flavor Fragrance J. 1999; 14:267–78.
7- Brickell C. Encyclopedia of garden plants. London: Dorling Kindersley. 1996; 926.
8- Hernandez-perez M, Rabanal RM, de la Torre MC, Rodriguez B. Analgesic, anti inflammatory, anti pyretic and haematological effect of aethiopinone, an o-naphthoquinone diterpeniod from Salvia anthiopis roots and two hemisynthetic derivatives. Planta Med. 1995;61:505–9.
9- Cuppett SL, Hall CA. Antioxidant activity of the Labiatae. Adv Food Nutr Res. 1998;42:245–71.
10- Wasser S, Ho JM, Ang HK, Tan CE. Salvia miltiorrhiza reduce experimentally-induced hepatic fibrosis in rats. J Hepatol. 1998;29:760–71.
11- Jimenez J, Risco S, Ruiz T, Zarzuelo A. Hypoglycemic activity of Salvia lavandulifolia. Planta Med. 1996;4:260–2.
12- Akbar S, Tariq M, Nisa M. A study on CNS depressant activity of Salvia haematodes wall. Int J Crude Drug Res. 1984;22:41–4.
13- Christie M. Ballantyne, Anders G. Olsson, Thomas J. Cook, Michele F. Mercuri, Terje R. Pedersen, John Kjekshus, Influence of Low High-Density Lipoprotein Cholesterol and Elevated Triglyceride on Coronary Heart Disease Events and Response to Simvastatin Therapy in 4S. Circulation; The journal of American Heart Association, 2001; 104, 3046-3051.
14- Boden WE, Pearson TA., Raising low levels of high-density lipoprotein cholesterol is an important target of therapy. American Journal Cardiology, 2000; 85 (5): 645-50.