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ALLIUM SATIVUM ESSENTIAL OIL (ASEO);

EFFECT OF SUPPLEMENTATION ON SERUM TRIGLYCERIDES, TOTAL CHOLESTEROL, HDLC, LDLC AND BLOOD CELL COUNTS IN ALBINO RATS

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ABSTRACT... Objectives: To determine the effects of Allium sativum essential oil (ASEO) phytotherapy on serum triglycerides, total cholesterol, HDLc, LDLc and blood cell counts in albino rat model. **Study design:** Experimental study. **Setting and Duration:** Animal House, Sindh Agriculture University and Isra University Hyderabad from May 2014 to January 2015. **Materials and Methods:** 60 albino rats were divided into four groups. Controls were given Placebo. Experimental rat groups were given ASEO 100 mg/kg, 200 mg/kg and 300 mg/kg orally for 30 days. Cardiac puncture was performed for blood sampling. Research variables were analyzed on Statistix 10.0 (USA). **Results:** Blood lipids showed significant reduction in various blood lipid fractions. Serum LDLc exhibited with a concomitant rise in serum HDLc (p = 0.0001) in high ASEO treated rats. Red blood cells, white blood cells and platelet showed significant improvement ASEO fed rats (p=0.001). **Conclusion:** Allium sativum essential oil (ASEO) phytotherapy showed a rise in HDLc and a reduction in LDLc, triglycerides and total cholesterol with improvement in red blood cell counts.

Key words: Allium sativum essential oil Blood lipids LDLc HDLc Blood cell counts Rats.

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INTRODUCTION

Allium sativum is a traditionally used herb for cooking purpose. Publicly, it is known as the Adrak (Garlic). Currently, Allium sativum essential oil (ASEO) is available for lipid lowering in coronary artery disease patients. ASEO is purified oil of Allium sativum extract (ASE) which is recommended for both the prevention and therapeutic purpose.1-5 Anti-microbial activity of ASE has been reported.² Therapeutic potential of ASE has been reported against the cancer, diabetes mellitus and cardiovascular disease. ASE has shown promising results as an anti- oxidant, anti- peroxidant and as antibiotic.^{3,4} Previous studies reported the Allium sativum reduced total blood cholesterol and inhibited the liver cholesterol synthesis.4,5 Systemic reviews have shown Allium sativum possess hypolipidemic hypocholesterolemia activity.5-7 Allium and sativum protects against coronary artery disease (CAD) through amelioration of LDLc and HDLc cholesterol. An increase in HDLc protects against the CAD by preventing atherosclerosis.^{8,9} ASEO improves the blood lipoproteins with a reduction in bad cholesterol (LDLc) thus inhibiting the atherogenesis. Recent research shows the ASEO protects against the in human being suffering from CAD.¹⁰⁻¹² Research evidence from developed countries is available while research from developing countries is lacking.^{11,13} It is reported to decrease systemic hypertension¹³⁻¹⁴ and alimentary disorders.¹⁵⁻¹⁶ Another previous study¹⁷ reported ASE oil reversed the liver steatosis. The active ingredient of ASE is the "allicin". Biochemically, the allicin is a "diallyl-disulfideoxide" and a potent vasodilator.¹⁸ Effects on the alleviation of diastolic blood pressure (DBP) in systemic hypertension¹⁹ and platelet aggregation inhibition of human has been reported in an invitro study.20

Previous studies reported immune boosting effects on the NK (natural killer) and T cell, an increase in interleukin-2 $(IL-2)^{21}$ and immune enhancing

effects both in-vitro and in-vivo studies.²² The previous studies have reported effect of Allium sativum extract, but not the Allium sativum essential oils (ASEO), which were being used for the first time in the present study. The ASEO phytotherapy was used to determine effects on the serum triglycerides, total cholesterol, HDLc, LDLc and blood cell counts in albino rat model. The CAD and Diabetes mellitus are increasing: hence easily available remedy should be search for the metabolic problems which should be easily available, cost effective and inexpensive. The present study reports for the first time on the effects of Allium sativum essential oil on serum triglycerides, total cholesterol, HDLc, LDLc and blood cell counts in albino rat model.

MATERIALS AND METHODS

Prior permission was taken from the ethical review committee of the institutes. Animals were housed at the Sindh Agriculture University, Tando Jam. Study covered period from May 2014 to January 2015. Rats of 150-200 grams of male gender were the inclusion criteria. Female rats. sick rats. and rats showing sluggish physical activity not feeding well were excluded during study period. NIH guidelines were followed for housing and handling of experimental rats. Animals were exposed to 12/12 hours dark - light cycles at 25°C and 55-60% humidity. Animal houses were well ventilated, with free access to chow diet and fresh water.

60 albino rats were divided into four groups. Rats were distributed equally into 4 groups i.e. 15 rats in each group. Controls (group I) were given placebo (0.9% isotonic saline). Experimental rat groups were given ASEO 100 mg/kg (group II), 200 mg/kg (group III) and 300 mg/kg (group IV) body weight (b.w) orally for 30 days. Commercially available Allium sativum essential oil was purchased from Pharmacy (Garlish oil soft gel) (High Q International, Pakistan). Calculated ASEO dose was given to each rat in all groups. ASEO was administered for 30 days duration.

At the end of 30 day experiment period, a 12 hours fasting was necessary for collection of blood samples. Cardiac puncture was performed

for blood sampling (Butterfly cannula B.D USA). Blood samples were taken in heparinized tubes. Centrifugation of blood was carried out at 350 rpm for 15 minutes. Sera were separated out. Blood was stored at 4°C for later use.

Blood lipids - triglycerides, total cholesterol and HDLc were detected by an enzyme-linked (Pharmacia-Biotech. immuno-sorbent assav Cambridge, UK) on Roche Chemistry analyzer. LDLc was detected by Friedewald's formula. Blood complete counts were estimated on Roche Hematoanalyzer. Data was preserved in a pre structured designed proforma. Research variables were analyzed on statistical software Statistix 10.0 (USA). Intra- and inter group comparisons were analyzed by one-way ANOVA. Post Hoc Bonferonni test was employed for the statistical significance between groups. All data analysis was performed at confidence interval of 95% CI ($P \le 0.05$).

RESULT

Blood lipids showed significant reduction in various blood lipid fractions. Serum LDLc exhibited with a concomitant rise in serum HDLc (p =0.0001) in high ASEO treated rats. Red blood cells, white blood cells and platelet showed significant improvement ASEO fed rats (p=0.001). Hemoglobin and hematocrit were improved by high dose ASEO supplementation. Blood glucose, serum creatinine and blood urea nitrogen were also studied as shown in Table-I. Figure-1 and 2 show the blood lipid and blood count distribution.

160 140 125.613.4 120 100,019.5 100 TAG (mg/dl) mg/dl. 80 TC (mg/d) 60 LDLc (mg/dl) 41.5 6.1 (6) 40HDie (mg/di) 20 ð ASE0 [300 Controls ASEO (100 ASE0 (200 mg/dl)

mg/dl)

Figure-1. Blood lipid levels in Rat groups

mg/dl}

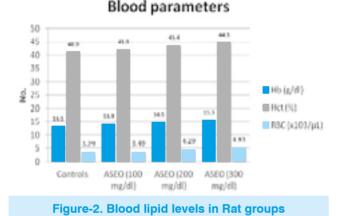
Blood lipids in Rat groups

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	Group I	Group II	Group III	Group IV
B. Glucose (mg/dl)	111.9±23.6	131.8±12.7	119.7±31.5	100.9±45.7
Creatinine (mg/dl)	0.92 ± 0.14	0.55 ± 0.23	0.8±0.41	0.81 ± 0.14
Blood Urea (mg/dl)	23.6±8.1	22.19±4.5	27.2±3.6	19.5±5.1
TAG (mg/dl)	117.2± 23.5	149.4±31.5	115.6±34.7	105.8±21.5
TC (mg/dl)	139±45.2	141.4±35.4	113.4 ± 23.5	109.5±19.7
LDLc (mg/dl)	42.5 ± 4.5	41.1±4.3	35.6±5.2	30.81 ± 4.9
HDLc (mg/dl)	37.4 ± 6.1	39.9±5.3	42.9±4.23	44.79±3.5
Hb (g/dl)	13.1±2.3	13.9±3.1	14.5±3.5	15.3±2.5
Hct (%)	40.9±10.34	41.9±9.32	43.4±8.7	44.5±5.1
RBC (x10 ³ /µL)	3.29 ± 1.34	3.49 ± 0.98	4.29±0.75	4.93 ± 0.57
WBC (/µL)	9011±110.3	8350±90.5	8571 ± 80.12	9021 ± 56.9
Platelets (x10 ³ /µL)	4.25±97.3	4.23±89.5	4.19±91.1	5.18±87.5

 Table-I. Blood lipids and Blood cell counts distribution in animal groups

 TAG- triglycerides, TC- total cholesterol, LDLc- Low density lipoprotein cholesterol, HDLc- High density lipoprotein cholesterol, Hb- Hemoglobin, Hct-Hematocrit, RBC- Red blood cells, WBC- White blood cells



DISCUSSION

The present study reports for the first time on the effects of ASEO on the blood lipids and blood cell counts in-vivo in Wistar albino rat model. To the best of web search, it is the first study which reports the hypolipidemic and hematopoiesis stimulating effects of ASEO. It is for the first time, the effects of ASEO on the blood cell counts were evaluated in-vivo. Hemoglobin (Hb), hematocrit (Hct), red blood cell (RBC), white blood cells (WBC) and platelets showed a significant increase in ASEO treated rat groups (p = 0.0001) revealed statistically significant differences.

Blood cell counts were improved in high dose ASEO treated rats. The findings of blood cell counts by ASEO is a unique findign which was evaluated for the first time, hence it is incomparable as no previous studies are available.

Thebloodlipidsweredecreasedwithaconcomitant rise in HDLc cholesterol in the present study (p =0.0001). High dose ASEO showed a significant decrease in total triglycerides, total cholesterol and low density lipoprotein cholesterol (LDLc). High density lipoprotein cholesterol (HDLc) was increased which is of clinical importance for the CAD patients. The underlying mechanism was not evaluated in the present study because of study design and funding issues. The findings of present study are supported by studies^{21,22} Previous studies reported immune enhancing effects through NK cell and T-cell activity and IL-2.20,22 Other previous studies^{11,12} used raw Allium extract in hypercholesterolemia patients and reported a significant decrease. The findings of present study are in agreement with above studies. As regards mechanism of action of ASE induced Hypolipidemia, the previous studies²³⁻²⁵ postulated this happened through inhibition of HMG-CoA reductase enzyme in the liver. The findings of present study are in keeping with previous studies.11-14

The findings are of clinical importance as ASEO may help in inhibiting the initiation and progression of atherogenesis and atheroma plaque formation in arteries. The underlying mechanism on the inhibition of atherogenesis remains to be studied and elucidated by

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molecular studies in future. Serum triglycerides were lowered in high dose treated ASEO, the finding is in agreement with previous studies.^{22,23} One proposed mechanism of decrease in blood triglycerides is through stimulation of hormone sensitive lipase (HSL) of adipose tissue which mobilizes the trigyclerides.^{22,23} However, it needs further clarification through molecular studies in future. Both the findings of Hypolipidemia and hematopoietic stimulating effects of ASEO are in agreement with previous literature.^{24,25}

CONCLUSION

Allium sativum essential oil (ASEO) phytotherapy showed a rise in HDLc and a reduction in LDLc, triglycerides and total cholesterol. ASEO treatment improved the hemoglobin, hematocrit, red blood cell counts, white blood cell counts and blood platelet counts. Allium sativum essential oil (ASEO) may be used for hyperlipidemia in coronary artery disease patients.

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"The price of greatness is responsibility."

Winston Churchill

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