SESAME OIL ADMINISTRATION DOES NOT ALTER HEMATOLOGIC AND METABOLIC PARAMETERS IN FEMALE RATS

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ABSTRACT

Reports relating to effect of sesame oil intake on various hematologic and metabolic parameters in humans or animals are scanty. Hence we have decided us to undertake this study, investigating the effect of this edible oil on above-mentioned parameters in adult female rats Material and Methods: Adult female Sprague Dawley rats were given oral doses of 1 ml, 2 ml and 4 ml sesame oil twice per day in divided doses, for 30 days respectively. Control female rats were given normal tap water. Oral gavage of sesame oil was done continuously for the study period twice daily and at the end of the 30 day study period, animals were lightly anaesthetized with ether and sacrificed to collect blood samples for analysis. Hematologic parameters such as red blood cell (RBC) count, white blood cell (WBC) count, hemoglobin (Hg), platelets, lymphocytes and mean corpuscular hemoglobin concentration (MCHC) were analyzed by a Hematology Blood Analyzer. Metabolic parameters such as cholesterol, triglycerides, urea, uricacid, creatinine and protein were analyzed by specific analytical kits. Levels of antioxidant enzymes, superoxide dismutase (SOD), glutathione peroxidase (GPX) in blood were assessed by specific analytical kits. Statistical analysis of various data from control and study groups was done using a SPSS data analytical package.

Keywords: *Sesame Oil, Hematologic Parameters, Metabolic Parameters, Oral Feeding, Female Rats*

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INTRODUCTION

Sesame oil is extensively used in India as well as Asian countries as a cooking and seasoning medium for many centuries . This higher content of unsaturated fatty acids in this oil has been widely believed to be beneficial to human health compared to many other edible oils (Namiki ,2007), although no detailed study has been reported on effect of sesame oil on the various hematologic and metabolic parameters in humans or in experimental animals . Presence of natural antioxidants and polyunsaturated fatty acids in food products are considered useful for the prevention of oxidative damage and cardiovascular diseases (Das,2000). Polyunsaturated fatty acids are well established to be vital for normal growth and development and play important roles in the prevention and treatment of cardiac diseases, hypertension, diabetes, and inflammatory and autoimmune disorders. Many reports have established the protective effects of oils rich in polyunsaturated fatty acids (PUFA) (Morris,1994 ; Sacks et al, 1994) in preventing cardio-vascular disorders and there have been reports of their utility in prevention of development of hypertension as well (Mori et al, 1999; Prisco et al,1998).

Sesame oil is reported to consist of 43 percent of polyunsaturated fatty acids, and 40 percent monounsaturated fatty acids. Presence of lignans as well as vitamin E in this oil are thought to be responsible for many of its chemical and physiological properties as well as antioxidant and antihypertensive effects (Matsumura et al, 1995, 1998 ; Kita et al, 1995, Yamashita et al, 1995)

Previous research in our laboratory had investigated the effect of oral administration of graded amounts of coconut oil as well as olive oil on various hematologic and metabolic parameters in adult female rats as well as pregnant rats (Nandakumaran et al, 2009, 2011,2012,2014) .This study was also intended to establish a scientific basis for the wide-spread use of another edible oil, sesame oil in Indian sub-continent as well as middle-eastern countries and to assess the impact of graded doses of this edible oil administration on the hematologic and metabolic parameters in adult female rats, over a prolonged period.

There have been reports attributing beneficial role of sesame oil controlling hypertension and in reducing lipid peroxidation in patients undergoing anti-hypertensive treatment (Sankar et al, 2006, 2005) as well in experimentally induced diabetic rats (Ramesh et al, 2005)

However, studies examining the effect of administration of sesame oil on hematologic, metabolic and atherogenic parameters in experimental animals or in humans are scanty . Herein we report the effect of oral administration of this edible oil on hematologic (RBC,WBC,Platelets, Hemoglobin, MCV,MCHC) and metabolic (cholesterol, triglycerides, protein, urea, uric acid and creatinine) parameters as well as on status of anti-oxidant enzymes, namely superoxide dismutase and soluble glutathione peroxidase in adult female rats, after oral gavage of varying volumes of this edible oil for a period of 30 days.

MATERIAL AND METHODS

Adult female rats (Sprague Dawley Strain) weighing between 220-240 g were selected for this study. Animals were bred in our medical faculty animal house and housed in individual polypropylene cages , with room temperature controlled at 25±1 degree centigrade and with alternating 12 hour light 12 hour dark cycle.. 3 groups of 5 rats received 0.5 ml, 1 ml and 2 ml of olive oil(Idhayam Brand, Tamil Nadu, India) orally twice per day respectively for a continuous period of 30 days while control group animals were given normal drinking water during the 30 day study period. Food and standard animal feed were given ad libitum during the period of study At the end of the study period of 30 days , all animals were weighed and anesthetized lightly with ether and sacrificed and blood samples collected of directly by cardiac puncture. Hematologic parameters such as RBC,WBC, platelets, Hb, lymphocytes and MCHC were assessed in blood samples of all study and control groups, using a Hematology Analyzer(ERMA INC, PCE210, Japan).Concentrations of metabolic parameters such as ,protein, cholesterol, triglycerides, creatinine, urea and uric acid in various blood samples were determined using specific analytical kits(RandoxLabs,UK).Activity of antioxidant enzymes, superoxide dismutase(SOD), glutathione peroxidase (GPX) in blood samples of study and control groups was determined by spectrophotometry , using a widely used and specific analytical method (Randox Labs, UK)

Statistical Analysis

All Data are expressed as Means+SEM .Statistical analysis of data was done using SPSS statistical package. Analysis of variance (ANOVA) or Student's t-test were used where appropriate. Data were assessed statistically significant, if probability was <0.05

RESULTS

In control group, body weight of rats averaged 232 gm at the beginning of the 30 day treatment period while in the 1ml/day sesame oil, 2 ml/day and 4 ml/day sesame oil treated groups, weights of rats averaged 229, 235, and 236 gm before start of experiment After the 30 day study period, the corresponding weights averaged 229 g in control rats and 211 ,212 and 208 gm in corresponding groups of sesame oil treated rats. ANOVA Test showed the reduction in body weights of sesame oil-treated to be significantly different ($p<0.05$) than that of control group. Table I shows details of some hematological parameters, namely RBC, WBC, Platelets, Hb, lymphocytes and MCHC of the control and treated groups of rats after the 30 day period of oil administration. Although WBC and Hg appeared to be lower in rats receiving higher doses of sesame oil, statistical analysis by ANOVA Test did not show any significant difference ($p>0.05$) between control and treated groups. However, platelets were significantly lower (ANOVA Test; $p<0.05$) in rats receiving 1ml and 2 sesame oil as well as 4 ml sesame oil per day, compared to control rats. The

difference in lymphocyte concentration between control rats and rats receiving various dose of sesame oil was not statistically significant (ANOVA Test ; p>0.05).

Table II shows values of total protein , urea and uric acid in blood samples obtained from control and sesame oil-treated rats after 30 day study period . Though level of urea in blood of sesame oil treated rats appeared to be lower than that of control group, statistical analysis (ANOVA Test) did not show any significant difference(p>0.05) of total protein, urea and uric acid values of sesame oil-treated rats compared to corresponding values of control rats. Total cholesterol averaged 66 ±6 , 62±7 , 69 ±8 and 71±9 mg/dl in control, 1ml, 2ml, 4 ml olive oil per day treated groups respectively. Statistical analysis showed no significant difference (ANOVA Test ; p>0.05) in cholesterol level in control and treated groups of rats. Triglyceride values in blood averaged 79±9, 81±8, 77 ±10 and 76 ±9 mg/dl in control, 1ml, 2ml, 4 ml sesame oil per day treated groups respectively(Fig.1). ANOVA Test did not show significant difference (p>0.05)in triglyceride values in oil-treated groups compared to control group

The levels of creatinine in blood of control and oil treated rat groups are shown in Figure 2. Creatinine concentration averaged 0.70±0.15, 0.80±0.20 , 0.82±0.15 and 0.85±0.18 mg/dl in control, 1ml, 2ml, 4 ml sesame oil per day treated groups respectively. Although creatinine values were apparently higher in oil-treated rats, ANOVA Test showed no significant difference in creatinine values between control and study groups(p>0.05)

SOD Enzyme activity values averaged 1.25±0.12 , 1.58±0.09, 1.65±0.08 and 1.72±0.11 U/ml in control, 1ml, 2ml, 4 ml sesame oil per day treated groups respectively. The SOD levels in oil-treated rats were significantly higher than that of the control un-treated group (ANOVA Test, p<0.05). GPX enzyme activity in blood averaged 1.32±0.08 , 1.62±0.07, 1.69±0.09 and 1.71 ±0.10 Units/L in control, 1ml, 2ml, 4 ml sesame oil treated groups respectively. GPX values of sesame oil treated rats were significantly higher than that of control group as well (ANOVA Test ;p<0.05)

DISCUSSION

Our data did not show any harmful effect of sesame oil on the various hematologic and metabolic parameters in adult female rats, despite daily oral administration of moderate to high doses of this oil continuously for 30 days . Interestingly,the body weights of sesame oil treated rats were significantly lower than that of control untreated group. We are unable to speculate why sesame oil administration led to such a weight reduction in oil-treated rats and whether such a phenomenon could be replicated or extrapolated to humans as well. The amounts of sesame oil received by the three treated groups were equivalent to about 300 ml, 700 ml and 1200 ml daily of sesame oil for an average woman weighing 60 kg for a continuous duration of 30 days and the results indicating absence of any significance in hematological or

metabolic parameters were surprising and unexpected . Further since sesame oil was administered orally, twice daily and continuously for 30 days, we were able to ensure that all the animals received the exact graded dose of the edible oil during the full course of study period.

Our data did not indicate any deleterious effect of sesame oil administration, even in massive doses, on either cholesterol or triglyceride levels of treated rats . However, in a comparable study on adult female rats (Nandakumaran et al, 2009) administration of another widely-used edible oil, coconut oil in similar high doses , had in fact resulted in lowered cholesterol levels in treated rats. We are unable to explain the reason for the difference in effects of coconut and sesame oil on cholesterol disposition in treated rats and further concerted studies are warranted. Surprisingly, even after massive administration of sesame oil daily for 30 days, triglyceride, urea, uric acid and creatinine levels did not increase to significant or abnormal levels, implying that administration of this edible oil per se does not cause any damaging effects on either the liver or the kidney or the heart. In a previous study, we had reported that female rats receiving equivalent dose of coconut oil , a relatively more saturated edible oil, showed significantly lower urea level compared to control group in adult female rats (Nandakumaran et al, 2009) but though the urea blood level in sesame oil-treated rats were lower than that of control, no statistical significance could be established. The coconut oil administration in pregnant rats (Nandakumaran et al, 2011) however did not reduce urea level in blood of oil-treated rats compared to control un-treated rats Whether sesame oil administration or intake in humans could lead to a beneficial decrease in level of urea in blood is open to speculation. Absence of significant difference in uric acid and creatinine levels in blood of control and sesame oil treated rats implies that despite receiving massive amounts of the oil for a period of 30 days , intake of this edible oil did not cause any major defect in renal function in treated rats. This finding was also comparable to similar studies conducted with the more saturated edible oil, coconut oil in adult female (Nandakumaran et al,2009) as well as pregnant rats (Nandakumaran et al, 2011)

The absence of hypercholesterolemia and triglyceridemia in sesame oil- treated rats indicate that even after prolonged administration for a period of 30 days, this edible oil does not affect cholesterol and triglyceride metabolism negatively. Interestingly, platelet count in rats receiving massive amounts of olive oil was lower , in all groups of oil-treated rats than control rats receiving no oil and this finding along with the reported efficacy of sesame oil in increasing capacity of anti-oxidant enzymes and in reducing lipid peroxidation could be considered as another beneficial parameter preventing formation of thrombus or plaques in lining of coronary or other blood vessels in the body and could explain the beneficial effect of sesame oil in controlling blood

pressure and its utility in hypertensive patients reported by some research groups (Sankar et al, 2006, Matsumura et al, 1995). Interestingly a similar finding of reduced platelet count was reported by us in pregnant rats receiving graded comparable doses of olive oil as well (Nandakumaran et al, 2014)

Graded administration of relatively large dose of sesame oil for a period of 30 days did not show any significant undesirable alteration in various hemo-dynamic parameters investigated . We hasten to add that data from experimental animals cannot be extrapolated to humans and hence the data obtained from this study cannot be readily replicated to human situations as well . Sesame oil administration was shown to increase activity of antioxidant enzymes SOD and GPX in all three groups of oil-treated rats. Many research groups have established the antioxidant function of the above two enzymes , in providing protection from reactive oxygen species (Richardson et al 1975 ; Eppe et al, 1983; Muller et al, 2007) Our finding of higher anti-oxidant function in animals receiving sesame oil is comparable to a similar finding reported by us, of increased anti-oxidant activity in rats receiving another edible oil coconut oil in similar experimental conditions in female rates (Nandakumaran et al, 2009) as well as in pregnant rats, receiving high amount of coconut oil during pregnancy period (Nandakumaran et al,2011). We speculate that , by reducing oxidation of LDL moiety mediated through reactive oxygen species (Ide et al, 2009 ; Hsu et al, 2008) sesame oil could play a beneficial role in preventing formation of plaques and could be beneficial in lowering mortality rates from plaque formations and cardio-vascular disorders, etc in populations using sesame oil as major edible oil. .

Majority of beneficial effects of sesame oil in the various reported studies could be attributed mainly to the presence of furfural lignans, namely sesamin, sesamolin, and sesaminolglucosides(Sankar et al 2006 ; Ide et al, 2009 ; Kamal-eldin et al,2011) Sesame oil has also been reported to provide protection to kidneys as well as liver from endotoxins as well as drug induced toxicity(Hsu et al, 2008; Periasamy et al, 2010) . A beneficial protective effect of this edible oil on heavy metal toxicity has also been reported (Chandrasekharan et al,2004)

The current studies from our laboratory on rats and other studies from elsewhere (Sankar et al, 2005, 2006 ; Ramesh et al, 2006) strongly indicate absence of any significant noxious effect of consuming sesame oil in humans and considering the wide-spread beneficial effects reported in the various studies, it will be worth recommending the use of this oil along with coconut oil in the Indian population in line with their established traditional use for the past many centuries. However, considering reports of wide-spread contamination and unfair trade practices involving marketing of edible oils in poor and third world countries and considering the marketing and consumption of edible

oil business of billions of dollars every year , health authorities of developing and under-developed countries need to make extra efforts in ensuring delivery of high quality edible oils to the public with mimimum of contamination from pollutants, additives,etc in ensuring better quality of health for the community and public as a whole. More detailed studies both in animals as well as in humans are warranted to explore and corroborate the beneficial effects of above edible oils on human and community health.

Table 1:*Haematological parameters in control and sesame oil treated Female Rats*

	RBC (x 10 6/ul)	WBC (x 10 3/ul)	Platelets (x 10 3/ul)	Hb (g/dl)	MCHC (g/dl)	Lympho (x 10 3/ul)
Group I	7.22 ± 0.21	5.25± 0.65	828.0 ± 30.45	12.05± 0.32	26.90± 0.18	3.29± 0.06
Group II	6.72± 2.10	4.60 ± 0.80	505.8 ± 78.98	11.25± 2.56	24.95± 1.6	2.88± 0.43
GroupIII	6.19± 1.49	5.52± 0.59	445.0 ± 71.91	11.50 ± 2.15	27.92 ± 2.42	2.92± 0.52
Group IV	7.21± 0.30	5.02± 0.52	565.2 ± 118.7	12.15± 0.57	28.05± 0.16	3.20 ± 0.34

Values are means ± SEM of 5 rats in each group. Statistical analysis was done by ANOVA or Students' t-test where appropriate. RBC = Red Blood Cell, WBC= White blood cell, Hb= Hemoglobin, MCHC= Mean Corpuscular Hemoglobin Concentration. Group I =Control Rats ; Group II= 1ml dose sesame oil/ day Group III II= 2ml dose sesame oil /day Group IV= 4ml dose sesame oil/day

Table 2: *Some Metabolic Parameters in control and sesame oil treated Female Rats*

	Total protein (g/dl)	Urea (mg/dl)	Uric acid(mg/L)
Group I	5.79± 0.42	39.95± 6.62	22.25± 0.60
Group II	5.32± 0.21	24.45 ± 6.34	22.32 ± 0.21
Group III	5.35± 0.25	28.05± 9.10	21.95± 0.45
Group IV	5.91± 0.23	29.95± 7.62	22.52± 0.19

Values are means ± SEM of 5 rats in each group. Statistical analysis was done by ANOVA or Students' t-test where appropriate ; Group I =Control Rats ; Group II= 1ml dose sesame oil/ day Group III II= 2ml dose sesame oil /day Group IV= 4ml dose sesame oil/day

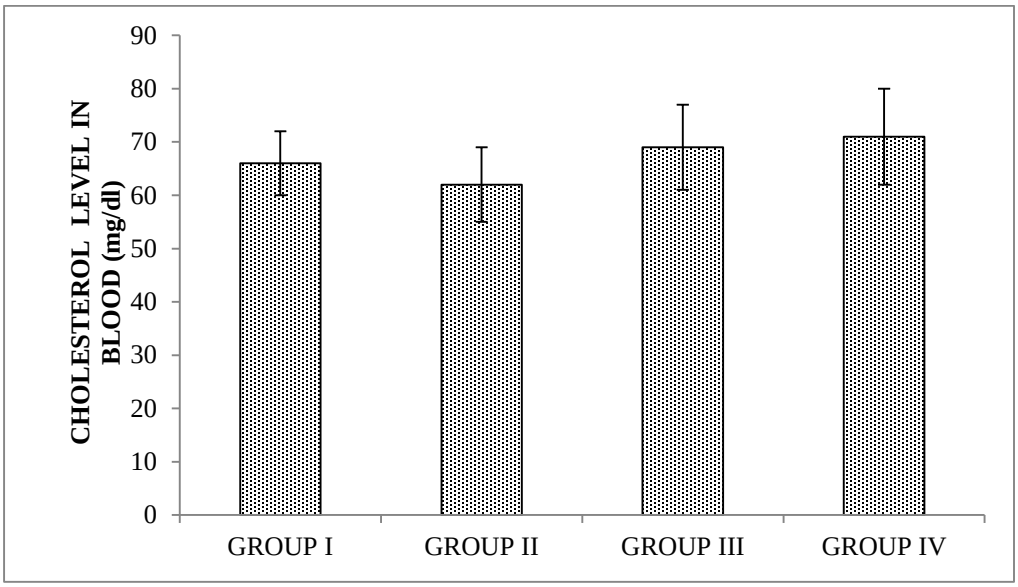

Fig I:*Total Cholesterol concentrations in bloodof femalerats.Values are Means± SEM of 5 animals in each group ;Group I=Control Group ; GroupII=1 ml/day of sesameoil, Group III=2 ml/day of sesameoil ; Group IV=4 ml/daysesameoil. Statisticalanalysiswasdone by ANOVA Test. Group I vs II p>0.05 ; Group I vs Group II p>0.05 ; Group I Vs Group IV p>0.05*

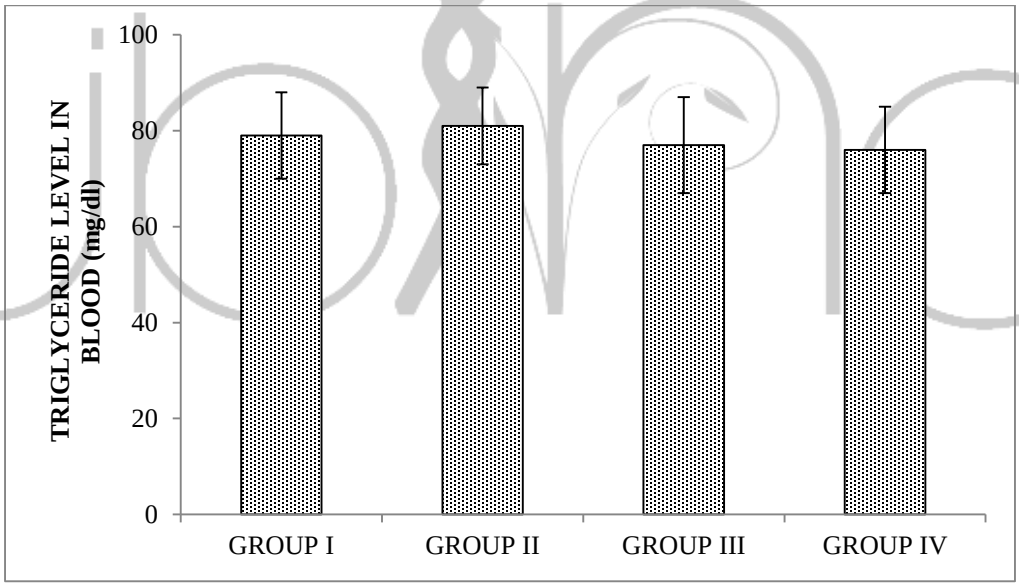

Fig 2:*Triglyceride concentrations in blood of femalerats.Values are Means± SEM of 5 animals in each group ;Group I=Control Group ; GroupII=1 ml/day of sesameoil, Group III=2 ml/day of sesameoil ; Group IV=4 ml/daysesameoil. Statisticalanalysiswasdone by ANOVA Test. Group I vs II p>0.05 ; Group I vs Group II p>0.05 ; Group I Vs Group IV p>0.05*

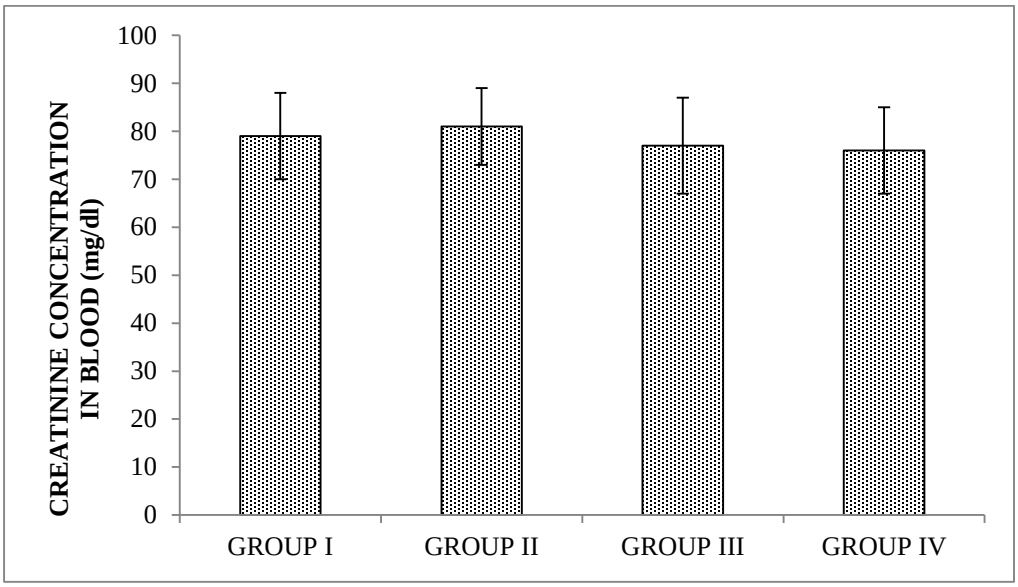

Fig 3: *Creatinine concentrations in blood of control and sesameoil-treatedfemale rats. Values are Means± SEM of 5 animals in each group ; Group I= Control Group II = 1 ml/day of sesameoil, Group III=2 ml/day of sesameoil ; Group IV=4 ml/daysesameoil. Statisticalanalysiswasdone by ANOVA test .Group I vs GroupII p>0.05 ; Group I vs Group III p>0.05 ; Group I Vs Group IV p>0.05*

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