EFFECT OF DIET OILS ON LIPID LEVELS OF THE BRAIN OF RATS

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ABSTRACT

The effects of sunflower oil, olive oil, margarine, soybean oil and butter on cholesterol and triglyceride levels were investigated in the brain of rats. Rats were fed with a diet containing 15 % of the oils for a period of 8 weeks. The rats were then decapitated, brain samples were removed, homogenized and cholesterol and TG levels were measured. Brain cholesterol and Triglycerides levels of all animals receiving different oils were significantly higher according to the control except TG level of animals receiving margarine. Brain cholesterol levels of the vegetable oils were statistically higher than those receiving saturated oil. Thus, cholesterol level of the rats fed with olive oil group was significantly higher than that of the other groups but those receiving sunflower oil group and Triglycerides level of the animals receiving olive oil was significantly higher than that of the other groups except animal supplemented with soybean oil. Thus, it is concluded that brain lipid levels of rats are differently effected by various oils and mechanism of which needs to be investigated.

KEY WORDS

Dietary oils, Brain, Cholesterol, Triglyceride

INTRODUCTION

Lipids are one of the main constituent of the diet. They are both a source of high energy and essential fatty acids (EFA) and are required for the absorption of fat-soluble vitamins (1). There is a substantial literature indicates that the chemistry and the function of both the developing and mature brain can be influenced by the diet (2). Indeed, dietary fatty acids has been reported to play an essential role in brain function (3). Also, although the mechanism is not clear, the fatty acid composition of cell membranes is determined by the fatty acid content of the diet (4,5).

Lipids are an important constituent of the brain, not only because of myelin, but also because of the large surface-to-volume ratio of neurons-neurons contain a higher proportion of lipid than other cells because lipid is the main constituent of the neuronal cell membrane (6). Lipids comprise 50-60 % of dry weight of the adult brain, of which approximately 35 %

are in the form of polyunsaturated fatty acids (PUFA), particularly arachidonic (AA, 20:4(n-6)) and docosahexaenoic (DHA, 22:6(n-3)) acids (7). These PUFA are derived through biosynthesis from their respective dietary EFA precursors, linoleic and linolenic acid, or they can be obtained directly from dietary sources such as eggs, fish and meat or, more recently, from single- cell oils (8). Some authors have speculated an association of the evolution of intelligent behaviour with the consumption of PUFA (9,10).

Lipids play both structural and functional roles, and these roles are particularly relevant in the brain which is the organ with the highest concentration of lipids after adipose tissue (1). It has long been argued that dietary fat, via its ability to alter brain membrane fatty acid composition, can alter membrane fluidity (6). Lipids have the potential to influence neuronal function in various ways. They can alter the physical properties of membrane, and in this way influence a variety of membrane functions, including effects on ion channels and transport, endo- and exocytosis, and the activities of membrane-bound proteins. Since neurotransmission depends on membrane receptors alterations in membrane phospholipid-fatty acid composition may also affect the nature of these interactions.

One of more common types diets recommended by physicians is designed to lower cholesterol levels, and, as shown in the

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nineteenth century, cholesterol is an important constituent of the brain. However, it has been reported that in case of lowered cholesterol levels, although mortality from cardiovascular disease tended to be lower, more patients were dying from accidents and suicide (6). Although, the effects of dietary fats on serum levels of lipids have been discussed, little is known about their effects on brain levels of these parameters.

In view of these facts, we believe that brain may also be effected from dietary oils. Therefore, it is important to show the role of dietary oils on lipid composition of the brain. Thus, the aim of this study was to assess the effect of most commonly consumed oils, namely sunflower oil, olive oil, margarine, soybean oil and butter on cholesterol and triglyceride (TG) levels of the brain.

MATERIALS AND METHODS

This study was performed on 72 Inbred male *Sprague-Dawley* rats, supplied by Center for Experimental and Applied Medical Research; University of Selcuk. The animals were 4 months old at the start of the experiment and housed in conventional wire-mesh cages in a room temperature regulated at $21 \pm 1^\circ\text{C}$, humidity 45-50 %, and light/dark cycles (12h). Environmental conditions (humidity, heat, light, ventilation etc.) were kept constant for 24 hours daily during the period of the study. The rats were separated into six groups of 12 each. Control group was fed regular laboratory chow consisting of 24 % protein, 3.62 % fat, 7 % cellulose, 10 % ash and 12 % water. The other five groups were named as sunflower oil (rats receiving sunflower oil), olive oil (rats receiving olive oil), margarine (rats receiving margarine oil), soybean oil (rats receiving soybean oil) and butter (rats receiving butter) groups. Each oil group was fed ad libitum a chow containing 15g oil/100g, for a period of 8 weeks. These oils were obtained from local supermarkets. The fatty acid composition of the oils, determined by gas chromatography, is given in Table-1. Margarine and butter were slightly melted and then mixed with the chow to obtain a homogeneous mixture. The mixtures were prepared daily and unused chow in a period of 24 hours was discarded in order to prevent spontaneous peroxidation. Oils were reported to have no significant peroxide content at room temperature for 24 hours (11).

After that period, the rats were decapitated between 9 and 10 am under ketamin anesthesia. The brain was quickly removed, washed in cooled 0.15 M NaCl. The brain tissue was homogenized in 2 ml of homogenizing buffer (100 mM KH_2PO_4 - K_2HPO_4 , pH 7.4, plus 0.1 % digitonin) by an ultrasonic homogenizer (12). Then the samples were

centrifuged at 5000 rpm for 15 min at 4°C . The supernatant was removed and the lipid parameters were studied. The study was approved by Ethic Committee of Meram Faculty of Medicine, University of Selcuk. Brain cholesterol and TG levels were measured by commercially available enzymatic kits. The results were calculated as mg/g protein. Tissue protein levels were determined by Biuret method (13).

Since, we have used enzymatic kits in the measurement of lipids we haven't performed extraction of the lipids. Because enzymes works properly in such homogenates.

Results are reported as mean $\pm$ standard deviation (SD). Data were statistically evaluated using one-way ANOVA computer program. Probability values (P) of < 0.05 were considered to be significant in the multiple range analysis. If ANOVA was significant, Post Hoc Tukey significant difference test was used for pair-wise comparisons between groups using the above computer program. All analyses were done with the Statistical Package for the Social Sciences (SPSS, Version 10.0).

RESULTS AND DISCUSSION

As seen from Table-2, Brain cholesterol and TG levels of all groups were higher than those of the control group but TG level of the margarine group. Thus, these changes in vegetable oils groups were statistically significant except TG level of the margarine group. Brain cholesterol levels of the vegetable oils groups were statistically higher than that of the saturated oil groups and control group. Cholesterol level of the olive oil group was significantly higher than that of the other groups but sunflower oil group and TG level of the olive oil group was significantly higher than that of the other groups except soybean oil group. Brain TG levels of all vegetable oil groups were significantly higher than the same parameter of the margarine group.

Cholesterol is used for membrane synthesis and for many other activities by cells throughout the body. Brain is a site of high lipid turnover. Approximately 25 % of the total amount of the cholesterol present in human is localized to this organ and most of its cholesterol is a product of local synthesis (14). Thus, there is a highly efficient recycling of cholesterol in the brain, with minimal losses to the circulation (15).

Although there is selective uptake of essential PUFA into brain, cholesterol and nonessential fatty acids do not enter the brain parenchyma. Also, the lipid content of the brain is considered to be relatively resistant to change induced by dietary intake (6). Kalman et al. (16) found that, after 10 weeks

of hypercholesterolemic diet the rabbits developed significant increases in plasma and liver cholesterol levels. In contrast, the cholesterol content of the brain cortex did not change in response to this diet. Adult rabbit brain remained resistant to dietary perturbations. Also, Haris et al. (17) showed that cholesterol content of all organs including brain is not affected by high-fat, high-cholesterol diet and genetics but that of the liver. However, our results show that brain cholesterol and TG levels are effected to various degree in rats fed with different oils.

Previously it was believed that the lipid content of the brain was considered relatively resistant to change induced by dietary intake. However, in the late 1980's, it became apparent that diet could alter the content and function of the rat brain in short period of time (6) as found in our study.

Also, brain cholesterol levels of rats fed with vegetable oils were increased significantly compared to butter and margarine fed rats. Since, vegetable oils do not contain cholesterol this is an interesting result which needs to be investigated. However, it can be argued that the effect of oils on brain cholesterol may be resulted from the influence of the oils on

Table-2: Brain cholesterol and triglyceride levels of control and oil groups (mg/g protein).

Groups	Cholesterol (mean $\pm$ SD)	Triglyceride (mean $\pm$ SD)
Control	22.58 $\pm$ 2.29	32.57 $\pm$ 3.71
Sunflower oil	30.51 $\pm$ 2.30	36.14 $\pm$ 4.59
Olive oil	31.80 $\pm$ 2.90	43.62 $\pm$ 4.92
Margarine	24.49 $\pm$ 2.00	30.68 $\pm$ 4.32
Soybean oil	27.81 $\pm$ 1.71	39.73 $\pm$ 4.32
Butter	24.43 $\pm$ 1.47	35.56 $\pm$ 2.52

de novo synthesis. Indeed, Poirier (18) has reported that brain cells, particularly astrocytes and neurons, cultured in vitro synthesized cholesterol at a rate, which was inversely proportional to the cholesterol content in the growth environment. Also, Angulo-Guerrero et al. (19) have observed lower brain cholesterol levels in animals fed with total PUFA deficient diet. These findings support our results.

Thus, our findings show that brain cholesterol and TG levels of rats fed with various oils are increased compared to those of controls but TG level of the margarine group. Thus, these changes in vegetable oil groups were statistically significant except TG level of the margarine group.

Table-1: The fatty acid composition of the oils

Fatty acid	Sunflower oil	Olive oil	Margarine	Butter	Soybean oil
C4:0				3.8	
C6:0				2.3	
C8:0			0.6	1.1	
C10:0				2.4	
C12:0			24.5	3.1	
C14:0	0.1		0.31	10.6	0.1
C14:1				1.4	
C15:0				1.6	
C16:0	6.6	12.3	9.33	26.6	10.5
C16:1	0.1	1.3		2.3	0.1
C18:0	4.6	2.1	4.89	12.6	3.9
C18:1	18.6	72.8		28.3	22.4
C18:1c			17.0		
C18:1t			11.3		
C18:2n-6			28.2		
C18:2	67.1	9.8		2.5	54.5
C18:3	1.1	0.8		0.8	7.7
C20:0	0.5	0.3			0.3
C20:1	0.4	0.3		0.2	0.2
C22:0	0.8	0.1			0.3

Our study is the first experiencing five different oils in respect to their effect on brain lipids. Brain being a highly protected organ, the fatty acid composition of the brain has been reported to be extensively modulated by dietary lipids (19). Dietary fatty acids also influence the fatty acid composition of brain membrane lipids (20-22). Thus, it can be postulated that fatty acid modulation may result in a change in brain TG levels as seen in our study.

Brain lipid levels of rats are differently effected by various oils, mechanism of which needs to be investigated, especially, the differences between the saturated and unsaturated oils and those of the olive oil group draws further attention. Further, the significance of these findings in respect to the functions and health of the brain remains to be evaluated.

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