

# ORIGINAL RESEARCH ARTICLE

## **A comparative study of the effect of heating on antioxidant potential and lipid profile of rats fed with two vegetable oils-sunflower oil and sesame oil (commonly used in culinary preparation)**

### **ABSTRACT**

**Aims:** Vegetable oils are fats, which are extracted from plant materials especially from seeds, which may change its chemical composition when heated. Therefore the present study investigated the effect of heating of sunflower oil and sesame oil on the *in-vivo* antioxidant status and the lipid profile in Wistar albino rats.

**Place and duration of study:** Department of Biological Sciences, University of Botswana, Botswana. The study was conducted from January 2012 - December 2012.

**Methodology:** The animals were divided into five groups of six rats each and maintained as follows: Normal control received the normal diet and two positive controls treated with heated sunflower oil and sesame oil, two experimental groups-treated sunflower oil and sesame oil after frying the potato chips. At the end of the experimental period of 60 days the rats were sacrificed and the blood was collected for biochemical estimations. The weights of the animals were recorded every week.

**Results:** The results indicated that sesame oil is much better in keeping the antioxidant status and cause lesser lipid peroxidation as well as hepatotoxicity when compared to sunflower oil. Again the cardioprotective effect of sesame oil is higher than that of the sunflower oil.

**Conclusion:** So in conclusion, sesame oil showed a higher level of protective effect than that of sunflower oil and is a better choice for culinary purposes.

**KEY WORDS:** *polyunsaturates, lipid peroxidation, oxidative stress, hepatotoxicity, cardioprotective, omega 3 fatty acids.*

## 28 INTRODUCTION

29 Cooking oil is plant, animal, or synthetic fat, which is edible. It is often used in frying, baking,  
30 seasoning and other types of cooking. Biochemically it is an edible triester of fatty acids and  
31 glycerol [1]. The appropriate amount of fat as a component of daily food consumption is a topic  
32 of controversy. The FDA recommends that 30% or fewer of calories consumed daily should  
33 come from fat. Other nutritionists recommend that not more than 10% of a person's daily  
34 calories should come from the fat [2]. Heating oil changes its characteristics. Oils that are healthy  
35 at room temperature can become unhealthy when heated above certain temperatures; this is due  
36 to the changes occurring in their chemical composition during the process of heating. When  
37 choosing cooking oil, it is important to match the oil's heat tolerance with the cooking methods  
38 used. The smoke point of any oil depends primarily on its free fatty acid content (FFA) and  
39 molecular weight. Through repeated use, as in deep frying, the oil accumulates food residues or  
40 by-products of the cooking process that lower its smoke point further [3]. During cooking, the  
41 oils and fats are exposed to high temperature which may result in changes in their chemical  
42 properties. Heating oil at a temperature above its smoke point results in breaking down its  
43 triglyceride to glycerol and fatty acids. Furthermore, heating breaks down the glycerol to  
44 acrolein which ends up in losing flavor and nutritional value of the specific oil. In addition, the  
45 heating of oils and fats without catalysts produces secondary lipid peroxidation abducts such as  
46 carbonyls and cyclic fatty acids which are toxic to human body [4]. Experts say that the  
47 consumption of dishes cooked in reheated oil may expose you to high levels of toxic aldehydes;  
48 chemicals that are known to cause neurodegenerative diseases and cancer. This finding does not  
49 exclude even popular cooking oils like soybean and sunflower oils that are also capable of  
50 generating these harmful chemicals when reheated. Researches on heated oils indicate the  
51 possibility of these to undergo thermal oxidation which results in the production of reactive  
52 oxygen species (ROS) which are harmful to the cells. In response to ROS production, the body  
53 activates its antioxidant system to defend against invasion of free radicals, which results in  
54 oxidative stress [5].

55 Sesame oil is obtained from seeds of *Sesamum indicum*, a plant that grows natively in India. It is  
56 an annual plant growing 50cm to 100 cm tall. It has oppositely arranged leaves which are 4cm to  
57 14cm long with an entire margin [6]. The oil from the nutrient-rich seed is popular in alternative

medicine, from traditional massages to the traditional Indian medical practice, Ayurveda. It has been used in the treatment of several chronic diseases, including hepatitis, diabetes and migraines. Research shows that sesame seed oil is a potent antioxidant [7]. It penetrates into the skin quickly and enters the blood stream through capillaries. Molecules of sesame seed oil maintain good cholesterol (HDL) and lower bad cholesterol (LDL). Sesame oil has a lowering effect on blood pressure and levels of sodium in the blood [8].

The sunflower, or *Helianthus annuus*, is a distinctive plant which produces large bright yellow flowers which resemble small suns, the flowers grow on tall stalks with simple leaves, and they have been known to reach the height of 3 m in ideal growing conditions. Sunflower oil is one of the most commonly and widely used cooking oil in the world [9]. It is a rich source of Vitamin E, A and D with low saturated fats and has the ability to withstand high temperatures. Nevertheless, it is not devoid of side effects and draw backs. In addition to vitamin E, the oil is also rich in Omega 6 fatty acid which is a causative agent of colon and prostate cancer. As the sunflower oil contains vitamin E, it can lead to decreased clotting of blood and this can elevate the risk of bleeding [10].

Heating cooking oil or fats changes its chemical composition and reuse of such oils which are exposed to high temperatures can cause harmful effects. So the main objectives of this study were to analyze the biochemical changes in the blood biomarkers and to establish the toxic effect of two different cooking oils commonly used in culinary preparations when exposed to high temperature during cooking. Hence to summarize, the main goals were to evaluate the antioxidant status in the blood samples of albino rats after feeding them with used cooking oils and next to compare the extent to which each of these oils can produce the changes in the lipid profile of these rats fed with these cooking oils.

## 2. MATERIALS AND METHODS

### 2.1 Plant Material

Oils were purchased from the local supermarkets (Spar and Pick 'n' Pay in South Africa)

## 2.2 Chemical

All the chemicals used were of the analytical grade and bought from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA).

## 2.3 Animals

Male albino Wistar rats weighing approximately 200–250 g were used for all the experiments. They were housed in colony cages at an ambient temperature of 25-28°C with a 12-hour cycle of light and dark. Animals had water *ad libitum* and were fed on commercial normal diet for rats bought from Nola Food Corp. (Cape Town, South Africa). The experiments were conducted following internationally accepted principles for laboratory animal care at the Department of Biology, University of Botswana, Botswana.

## 2.4 Oil samples preparation

The oils were heated or fried separately; there were two groups of rats for each of the oil. One group received heated oil and the other one received fried oil. The oils were prepared in the following manner: cooking oil was heated for four minutes in the absence of any food items. After that the oil was allowed to cool and then it was mixed with normal rat food and administered to the first group of rats used as positive control (PC) groups. The other groups of rats were fed with oils that were used to fry thin strips of potatoes for four minutes as well. The potatoes were discarded and the oils were mixed with normal rat food named as experimental groups (EX). Oils were mixed with feed at the rate of 10% of the feed weight.

## 2.5 Experimental design

The animals were divided into five groups of six rats each and maintained as follows:

Group 1- NC-Normal control- received commercial diet without any oil supplemented

Group 2- PC 1-Positive control- treated with heated sunflower oil (10mL/ Kg body weight)

Group 3- PC 2- Positive control- treated with heated sesame oil (10mL/ Kg body weight)

Group 4- EX 1-Experimental group- treated with sunflower oil used to fry the potatoes (10mL/ Kg body weight)

Group 5- EX 2-Experimental group- treated with sesame oil used to fry the potatoes (10mL/ Kg body weight)

The experiment was run for 60 days, at the end of the experiment the rats were fasted overnight and the rats were killed with mild ether anesthesia. Blood was collected in heparinized tubes and centrifuged at the rate of 6000 rotations per minute. Plasma samples were drawn after that and stored at  $-70^{\circ}\text{C}$  for various biochemical estimations.

## 2.6 Biochemical Analysis

Thiobarbituric acid reactive substances (TBARS) in plasma were estimated by method described by Niehaus and Samuelsson [11] and reduced glutathione (GSH) was estimated by the method of Ellman [12]. Superoxide dismutase (SOD) was assayed by the method of Kakkar *et al.* [13]. The assay of SOD activity was based on the principle of inhibitory effects of SOD on reduction of nitro blue tetrazolium dye by superoxide radicals. A single unit of enzyme was expressed as 50% inhibition of NBT (Nitro blue tetrazolium) reduction /min/mg protein. Catalase (CAT) was estimated by the method of Hans Bisswagner [14]. Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), Alkaline phosphatase (ALP), were estimated by using the kits from Sigma- Aldrich Chemical Company, (St. Louis, MO) USA. LDL, HDL Total cholesterol and triglycerides were estimated by using kits also from Sigma.

## 2.7 Statistical Analysis

All data were expressed as mean  $\pm$  S.E. mean with  $n=6$ . Analysis of variance was performed by one way ANOVA and significant difference between the means were determined by Holm-Sidak method.  $P\text{-value} \leq 0.05$  was regarded as statistically significant value. In all these cases, Statistical Software Sigma stat, 3.1 were used to analyze the data.

# 3. RESULTS

## 3.1 Percentage increase in weight

The average weights of these animals are shown in figure 1 and 2 which shows a gradual increase in weight from week one onwards, this may be due to lipids getting deposited in the adipose tissue. This was highest in PC 1 than in EX 1. There was no significant difference in weight gain in any of these groups.

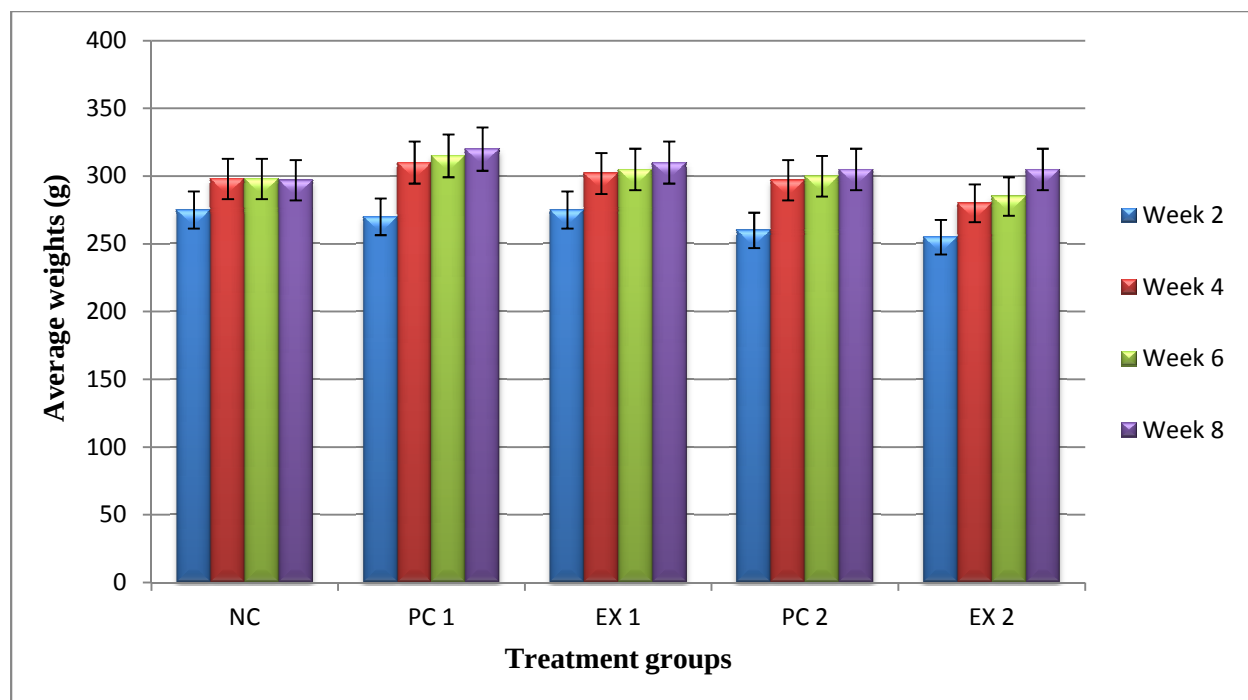


**Figure 1: Average weights of experimental rats taken at an interval of two weeks over a period of eight weeks.**

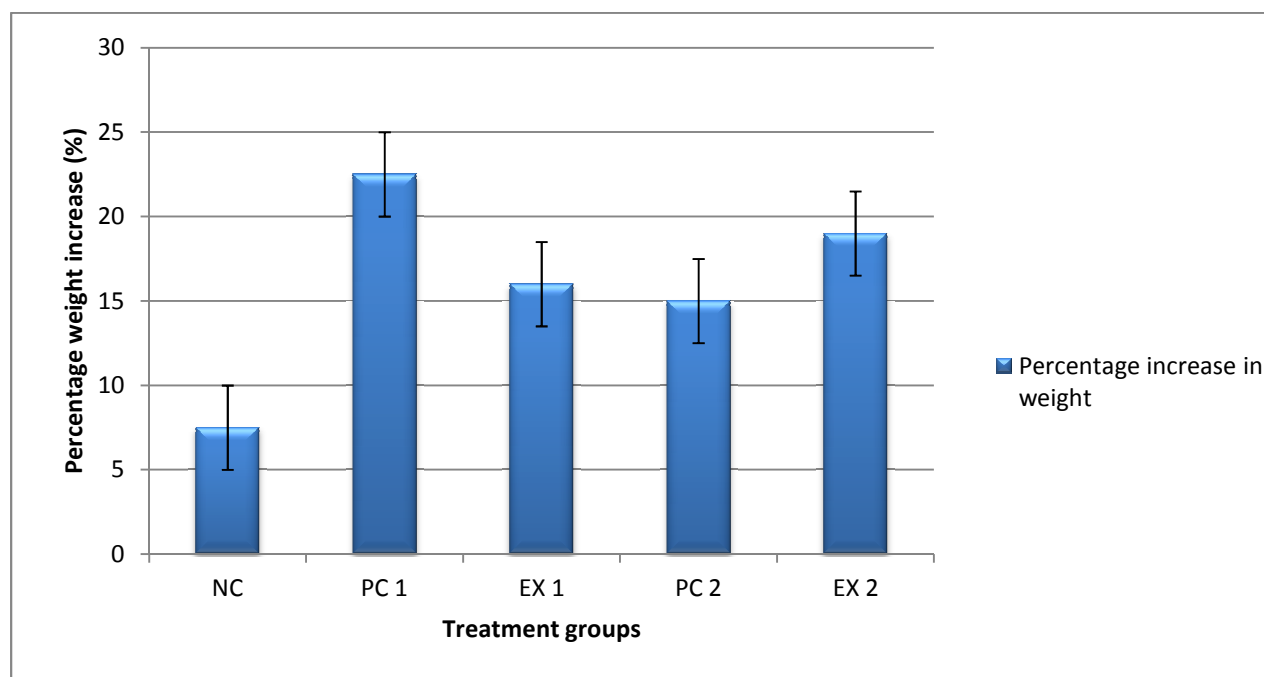


**Figure 2: % weight increase in weight of rats fed with different oils.**

### 3.2 Lipid peroxidative indices and liver markers

The results presented in Table 1 shows liver function indices of alanine aminotransferases (ALT), aspartate amino transferases (AST), alkaline phosphatases (ALP), concentrations in the serum after 60 days of oral administration of sunflower oil and sesame oil. There was a significant ( $P<0.05$ ) increase in the activities of all these liver marker enzymes (ALT, AST and ALP) in the EX groups when compared to control groups (NC). The PC groups did not show any significant difference in the liver marker enzymes when compared with the NC group. The levels of TBARS are a clear indication of the amount of lipid peroxidation in every group and it also showed the same type of effects as the liver markers.

**Table 1. Lipid peroxidative indices**

| Groups | TBARS<br>(nmol/L)<br>PLASMA | AST (U/L)<br>PLASMA | ALT (U/L)<br>PLASMA | ALP (U/L)<br>PLASMA |
|--------|-----------------------------|---------------------|---------------------|---------------------|
| NC     | 1.28 ± 0.08                 | 40.16 ± 0.23        | 38.98 ± 0.09        | 45.76 ± 0.04        |
| PC1    | 1.58 ± 0.21                 | 46.95 ± 0.56        | 39.04 ± 0.46        | 47.96 ± 0.34        |
| PC2    | 1.05 ± 0.34                 | 38.05 ± 0.91        | 34.08 ± 0.05        | 42.76 ± 0.72        |
| EX1    | 6.87 ± 0.18 *               | 78.95 ± 0.41*       | 76.88 ± 0.03*       | 88.56 ± 0.17*       |
| EX2    | 5.07 ± 0.11 *               | 67.15 ± 0.71*       | 71.67 ± 0.23*       | 83.76 ± 0.67*       |

TBARS: Thiobarbituric acid reactive substances, ALT: Alanine aminotransferase,  
AST: Aspartate aminotransferase and ALP: Alkaline phosphatase.

\* Significantly different

n= 6 in each group and the results were given as mean ±SEM.

### 3.3 The antioxidant status in the blood

The antioxidant status in the blood included the non-enzymatic antioxidants such as GSH as well as the enzymatic antioxidants such as CAT and SOD (Table 2). The results indicate that all the parameters checked showed a significant decrease in Ex groups when compared to NC groups ( $P \leq 0.05$ ). The PC groups did not show any significant difference between the NC groups but they showed a slight variation in different oils.

**Table 2. The antioxidant levels in the blood**

| Groups | GSH (mg/dl)<br>PLASMA | CAT (U/mg Hb)<br>HAEMOLYSATE | SOD (U/mg Hb)<br>HAEMOLYSATE |
|--------|-----------------------|------------------------------|------------------------------|
| NC     | 35.07± 1.09           | 48.75 ±1.32                  | 2.65 ± 1.17                  |
| PC1    | 34.13± 0.12           | 44.15 ±1.01                  | 2.15 ± 0.19                  |
| PC2    | 38.67± 1.11           | 49.78 ±0.92                  | 2.81 ± 0.13                  |
| EX1    | 17.88 ±0.34*          | 28.48 ±1.19*                 | 1.28 ±0.48*                  |
| EX2    | 19.09 ±0.84*          | 32.46 ±0.61*                 | 1.88 ±0.98*                  |

GSH: Reduced glutathione, SOD: Superoxide dismutase and CAT: Catalase.

### 3.4 Lipid profile in the plasma

The results of the lipid profile in the blood plasma are presented in Table 3. It shows that the consumption of the oils had significantly lowered HDL levels ( $p < 0.001$ ) in EX groups but the levels are different in sesame oil, it is much higher when compared to the sunflower oil. All other parameters checked showed significant increase in EX groups when compared to NC group, once more the levels are slightly different for both the oils used.

177 **Table 3. Levels of different types of lipids in the plasma**

| Groups | TOTAL<br>CHOLESTROL<br>(mg/dL) | HDL (mg/dL)   | LDL (mg/dL)  | TRIGLYCERIDES<br>(mg/dL) |
|--------|--------------------------------|---------------|--------------|--------------------------|
| NC     | 10.20 ± 1.25                   | 56.09 ± 1.32  | 15.00 ± 2.23 | 61.00 ± 4.21             |
| PC1    | 18.28 ± 1.09                   | 47.08 ± 1.52  | 36.08 ± 1.92 | 111.00 ± 0.28            |
| PC2    | 12.02 ± 0.28                   | 68.07 ± 0.39  | 16.68 ± 1.62 | 66.00 ± 1.53             |
| EX1    | 124.40± 9.10*                  | 12.80 ± 1.14* | 150.87±3.1*  | 230.00 ± 9.31*           |
| EX2    | 42.45± 1.18*                   | 21.87 ± 0.17* | 105.07±1.1*  | 201.80 ± 0.91*           |

178

179 n= 6 in each group

180 \* Shows significant difference and the results were given as mean ±SEM. P ≤0.05

181 Group 1- NC- Normal control. Fed with normal diet without any supplementation

182 Group 2- PC 1 –Positive control 1 –treated with heated sunflower oil

183 Group 3-PC 2- Positive control 2–treated with heated sesame oil

184 Group 4- EX 1-Experimental group 1-treated sunflower oil used to fry the potatoes

185 Group 5- EX 2-Experimental group 2- treated with sesame oil used to fry the potatoes

186

## 187 4.DISCUSSION

188 Most of the lipids are in the form of triglycerides and they are the major fuel source in the body.

189 Other lipids, including phospholipids, glycolipids and cholesterol are crucial constituents of the

190 biological membrane. Fats are part of a normal, balanced, healthy diet and the body needs them

191 for a variety of metabolic reactions. Fats provide a concentrated source of energy. One gram of

192 fat provides 37kj (9kcal), more than double that provided by either protein or carbohydrate

which provide 17kj/g (4kcal) and 16kj/g (3.75kcal), respectively [15]. Fat is a carrier for fat-soluble vitamins A, D, E and K. Some fats are essential to our well-being; these are known as the essential fatty acids (EFA's) often referred to as the omega fatty acids. Linoleic acid (omega 6) and alpha linoleic acid (omega 3) are the most common essential fatty acids found in vegetable oils. Individual fatty acids can either be saturated or unsaturated and the latter being further subdivided into mono and polyunsaturated fatty acids. Fatty acids are either saturated or unsaturated. Saturated fats have no double bonds while the unsaturated fatty acids have single or multiple double bonds. Saturated fatty acids are extremely stable, that is, they do not easily become rancid, and they can increase blood cholesterol levels, one of the major factors in heart disease [16].

Fig 1 and 2 showed the effect of these oils in the weight of these animals and result did not show any significant difference. The effects of sunflower oil and sesame oil on lipid peroxidation is presented in table 1 which indicate an increase in lipid peroxidation as measured by formation of TBARS or dyne conjugates. This shows the mitochondrial dysfunction in rats subjected to the oxidative stress created by reused oils. This was much restricted in the control groups. The data obtained showed that the reused oils administration in the experimental groups in albino rats caused acute oxidative stress along with necrosis in the liver, which corresponded to the elevation of serum ALP, ALT and AST activity [17]. These pathological changes were accompanied with structural alterations of the hepatocytes, including glycogen and fat accumulation, organelle abnormality and cytoplasmic membrane degeneration due to lipid peroxidation causing the leakage of liver enzymes in to the blood stream. These conditions were not occurred in the control groups. Again the liver markers levels were higher in sunflower oils treated when compared with sesame oil treated experimental groups [18].

The antioxidant status is given in Table 2 which shows that the positive control groups have almost the same antioxidant status as that of the normal control but the experimental groups were significantly different from the control groups. GSH is a non-enzymatic antioxidant which inhibits the generation of reactive oxygen species and hence the oxidative stress that damages the structural integrity of cell membrane and membrane organelles. The decrease in GSH levels in EX groups were due to the decreased availability of GSH caused by enhanced lipid peroxidation. In the present study, a significant decrease in the GSH content in the EX groups as compared to

the entire PC and NC groups were also observed. A decrease in the SOD and Catalase activities were observed in EX 1 and EX 2 where reused oils were administered. It appears that the entire control group takes over the work of scavenging the free radicals and hence the normal levels of both enzymatic and non-enzymatic anti-oxidant were maintained. SOD catalyzes the dismutation of superoxide into oxygen and hydrogen peroxide, and it is an important antioxidant defense in all cells exposed to oxygen. Hydrogen peroxide is then converted to oxygen and water by glutathione peroxidase and CAT [19]. It has been reported that SOD, CAT, and GSH constitute a mutually supportive team of defense against reactive oxygen species. One of the secondary effects of ROS formation is mitochondrial dysfunction which results in ATP depletion and oxidative stress. The results prove that the sesame oil worked in the way of scavenging the superoxide anion thus preventing the chain reactions that leads to the production of ROS.

Apart from oxidative stress generated in EX groups, there was also a significant reduction in the level plasma HDL along with an increment in the levels of triglyceride; LDL and total cholesterol were also noted. This may be due to the additional supplemented oils in the diet. Free radicals enhance the oxidation of light density lipoprotein (LDL) and oxidized LDL affects many biological processes involved in atherogenesis. A recent study found that a toxin called 4-hydroxy-trans-2-nonenal (HNE) forms when the oils are reheated. Once absorbed in the body, HNE reacts with DNA and proteins affecting basic cellular processes. Frying foods at or above 375oF can lead to the accumulation of 4-hydroxy-trans-2-nonenal (HNE) in the oil. HNE is a toxic substance that has been associated with an increased risk of stroke, atherosclerosis, elevated levels of LDL cholesterol, Alzheimer's, Parkinson's, various liver diseases and cancer [20]. Heating once can create the formation of HNE in the oil, and reusing oil at too high of a heat can cause even more HNE to build-up. Researchers found that HNE is more likely to build up in oils with high levels of linoleic acid. The oil with the highest percentage of linoleic acid is the sunflower oil compared to sesame oil [21].

There are two types of fatty acids that are termed 'essential' because the body cannot produce them. These are omega-3 and omega-6 fatty acids, when the balance of omega-6s and omega-3s in the cell is disturbed, it can cause physiological changes in the cellular functions. The more double bonds in a fatty acid, the more reactive it is. Polyunsaturated fats tend to react with

oxygen, which can cause chain reactions, damaging other structures and perhaps even vital structures like DNA [22].

According to our findings we recommend that sesame oil is more advisable to use in cooking than the sunflower oil due to its high smoking point. Sesame seed oil is a potent antioxidant which will neutralize oxygen radicals by scavenging the ROS. *In vitro*, sesame seed oil has inhibited the growth of malignant melanoma so this is used as an anticancer agent. Sesame seed oil is a cell growth regulator and slows down cell growth and replication. Sesame seed oil absorbs quickly and penetrates through the tissues to the very marrow of the bone. It enters into the blood stream through the capillaries and circulates faster. The liver does not sweep sesame seed oil molecules from the blood, accepting those molecules as friendly. Sesame oil has a lowering effect on blood pressure and levels of sodium in the blood acting as a diuretic [23]. On the other-hand sunflower oil is rich in Omega-6 fatty acids which are harmful in excess with many side effects [24].

#### 4. CONCLUSION

The results of this study demonstrated that sesame seed oil have many protective characteristics when compared to sunflower oil. This is due to its antioxidant potential and rich in Omega-3 fatty acids. Importantly instead of sticking to one type of oil, consumers should try to use different oils always in moderate amounts.

#### CONSENT

Not applicable.

#### ETHICAL APPROVAL

The author hereby declares "Principles of laboratory animal care" (published in 2005) were followed. All experiments have been examined and approved by the appropriate ethics committee.

#### 5. REFERENCES

1. Alfred Thomas. *Fats and Fatty Oils*. Ullmann's Encyclopedia of Industrial Chemistry. Weinheim.2002; Wiley-VCH.

2. The Culinary Institute of America *The Professional Chef*. New York: Wiley.2011; ISBN 0-470-42135-5.
3. Warner, K. *Impact of high-temperature food processing on fats and oil*. Advances in Experimental Medicine and Biology. 1999; 459: 67-77.
4. Abramovic, H. and Klofutar, C. *The Temperature Dependence of Dynamic Viscosity for Some Vegetable Oils*. Acta Chim. Slov. (Acta.chem-soc.si). 1998; 45 (1): 69-77.  
Received 2013-02-07
5. Che Man, YB, Liu, J.L., Jamilah, B., Abdul *Quality changes of RBD palm olein, soybean oil and their blends during deep-fat frying*. Journal of food lipids. 1999; 6 (3): 181-193.
6. Elleuch, M., Besbes, S., Roiseux, O., Blecker, C. and Attia, H. *Quality characteristics of sesame seeds and by-products*.Food Chemistry. 2007; 2: 641.
7. Suja, K. P., Jayalekshmy, A. and Arumugha, C. *Antioxidant activity of sesame cake extract..* Food Chemistry,2005; 91: 213.
8. Lim, JS; Adachi, Y; Takahashi, Y; Ide, T. "Comparative analysis of sesame lignans (sesamin and sesamol) in affecting hepatic fatty acid metabolism in rats".*Br J Nutr.*2007;**97** (1): 85–95.
9. Yanishlieva, N. V. I. and Marinova, E. M. Effects of antioxidants on the stability of triacylglycerols and methyl esters of fatty acids of sunflower oil.Food Chemistry. 1995; 54: 4, 377.
10. De Marco, Elena; Savarese, Maria; Parisini, Cristina; Battimo, Ilaria; Falco, Salvatore; Sacchi, Raffaele.*Frying performance of a sunflower/palm oil blend on comparison with pure palm oil*. European Journal of Lipid Science and Technology. 2007; 109 (3) 273.
11. Niehaus, W. G. & Samuelson, B. *Formation of MDA from phospholipid arachidonate during microsomal lipid peroxidation*. European Journal of Biochemistry.1986; 6: 126-130
12. Ellman, G. L. *Tissue sulhydryl groups..* Archives of Biochemistry and Biophysics.1959; 82: 70-77
13. Kakkar, P., Das, B., Viswanathan, P. N.*A modified spectrophotometric assay of superoxide dismutase*.Indian Journal of Biochemistry and Biophysics.1984; 21: 130-132.

14. Bisswanger, H. *Practical enzymotologist*. Wiley-VCH Verlag GmbH & Company, KGaA, Weinheim. 2004. pp 79
15. Alfred Thomas. "Fats and Fatty Oils". Ullmann's Encyclopedia of Industrial Chemistry. Weinheim: Wiley-VCH.2002. pp 88
16. Katan, Martjin B., Mozaffarian, Dariush, Micha, Renata, Wallace, Sarah. ed. "effects on Coronary Heart Disease of Increasing Polyunsaturated Fat in Place of Saturated Fat: A Systematic Review and Meta-Analysis of Randomized Controlled Trials". PLoS Medicine. 2010; 7 (3):100-25
17. G. Aldini, I. Dalle-Donne, R. Colombo, R. MaffeiFacino, A. Milzani, M. Carini *Lipoxidation-derived reactive carbonyl species as potential drug targets in preventing protein carbonylation and related cellular dysfunction*. ChemMedChem.2006; (1) 1045–1058
18. K.S. Fritz, D.R. Petersen *Exploring the biology of lipid peroxidation-derived protein carbonylation*. Chemical Research in Toxicology.2011; 24 pp. 1411–1419
19. Yanishlieva, N. V. I. and Marinova, E. M. Effects of antioxidants on the stability of triacylglycerols and methyl esters of fatty acids of sunflower oil. Food Chemistry. 1995; 54: 4, 377.
20. K. Uchida. 4-Hydroxy-2-nonenal: a product and mediator of oxidative stress. Progress in Lipid Research.2003; 42, . 318–343
21. Mensink, RP; Zock, PL; Kester, AD; Katan, MB. "Effects of dietary fatty acids and carbohydrates on the ratio of serum total to HDL cholesterol and on serum lipids and apolipoproteins: a meta-analysis of 60 controlled trials". The American Journal of clinical nutrition. 2003; 77 (5): 1146-55.
22. Mozaffarian, Dariush; Katan, Martijin B.; Ascherio, Alberto; Stampfer, Meir J.; Willett, Walter C. "Trans Fatty Acids and Cardiovascular Disease". New England Journal of Medicine. 2006; 354 (15) 1601-113
23. Namiki M. Nutraceutiactal functions of Sesame: .A review. Critical Reviews in Food Science and Nutrition. 2007. 47:651-673.
24. ArunaK ,Kalpana,C, Viswanathan, P and Menon V.P. Toxic effect of sunflower oil on ethanol treated rats. HepatologyReasearch .2002; Vol 26:12,125-135