African Journal of Biotechnology Vol. 6 (18), pp. 2158-2161, 19 September 2007
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2007 Academic Journals

Full Length Research Paper

Protective role of onion and garlic on physicochemical alterations and toxicity of heated soybean oil

Oyewole O. I.[1]* and Olayinka E. T.[2]

[1]Department of Biochemistry, College of Medicine and Allied Health Sciences, University of Sierra Leone, Freetown.
[2]Department of Chemistry and Biochemistry, Bowen University, Iwo, Nigeria.

Accepted 28 June, 2007

Fresh and soybean oil heated with or without onion or garlic were analyzed for their physicochemical and toxicological properties. Darkened appearance, off flavors, rancid taste and significant reduction of iodine value was obtained for the heated oil. Acid value, peroxide value, viscosity and concentration of malondialdehyde of the thermooxidized soybean oil were also significantly elevated (P<0.05) with considerable loss of ascorbic acid and tocopherols. Heating with onion and garlic considerably preserved the quality of the oil as indicated in the significant reduction in levels of lipid peroxidation indicators as well as reduction in loss of ascorbic acid and tocopherol. Rats fed with diet containing heated oil for a period of four weeks showed significant elevation (P<0.05) in the serum levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) starting from the second week to the end of the experimental period. This might be as a result of cellular damage caused by peroxides and other lipid oxidation products which allowed the enzymes to leak out from the tissues. The levels of these enzymes were maintained close to the control in rats fed with diets containing heated oil with garlic and onion indicating their protective role in lipid oxidation.

Key words: Physicochemical properties, soybean oil, lipid peroxidation, onion, garlic serum aminotransferases.

INTRODUCTION

Lipid oxidation produces free radicals which have high reactivity with oxygen leading to rapid conversion to peroxides or hydroperoxides. These products are quite unstable and thus undergo scission and dismutation reactions to form a wide spectrum of carbonyls, hydroxyl compounds and short chain fatty acids (Addis, 1986). Small amounts of some cholesterol oxidation products such as oxysterols are also presented in heated oil which could generate other reactive components (Maerker, 1987). This breakdown mechanism is responsible for the production of colour defects, rancid taste, off flavors as well as the myriad of other reactions which reduce shelf life and nutritional value of fat containing foods (White, 1991; Rodriguez–Estrada et al., 1997).

Several studies with rats have been carried out in which oil that had previously been used in cooking for a long time was fed as the sole source of dietary fat. General effects reported range from loss of appetite, poor food efficiency, reduced gut absorption, enlarged liver, kidney and lung, fatty hepatic necrosis, haemolysis, calciferous myocardial lesions and even death (Alexander, 1977, 1978; Miller and Long, 1990; Sanders, 1989). Consumption of diet containing thermally heated soybean oil was also reported to cause growth depression and disruption of skeletal muscle enzyme activities in rat (Odutuga and Oyewole, 2006). Heat is one of the chief accelerating factors of lipid oxidation. The extent of oxidation as well as the nature of degradation product depends on the heating temperature, duration of heating as well as the fatty acid chain involved (Jacobson, 1991).

Soybean oil is commonly used as cooking oil among people in Africa. It is rich in cholesterol and contains 80% linoleic acid which has been pointed out to be highly susceptible to oxidation (Muller, 1988). The bad economic situation in many homes in Africa often demands that oil previously used for cooking are reused again and this may constitute health risk to consumers of food prepare

*Corresponding author. E-mail: ioluoye@yahoo.com. Tel: +23233757782, +2348033727617.

Table 1. Diet composition fed to rats in this study.

Component	(g/100g)
Casein	25.0
D-L Methionine	0.4
D-L Lysine	0.4
Corn starch	25.0
Cellulose	25.5
Sucrose	3.7
Vitamin mix	1.0
Mineral mix	4.0
Soybean oil	15.0

with such heated oil. The role of spices such as onion, garlic, ginger and green pepper as antioxidant has long been realized (Cao et al., 1996; Borek, 2001). These spices contain substances that retard the rate of oxidation by scavenging free radicals or directing the break down of peroxides into stable substances that do not promote further oxidation (Fremont et al., 1998). This study intends to investigate the protective role on onion and garlic on physicochemical changes and toxicological effect of thermally heated soybean oil.

MATERIALS AND METHODS

Materials

The soybean oil used is a product of Unique Vegol SL Limited, Freetown Sierra Leone. Onion and garlic were obtained from Lumley Market, Freetown, Sierra Leone. They were cut into small pieces, dried and grinded into powder.

Thermal oxidation of oil

Fresh soybean oil was poured in stainless steel pot and heated in an open space at 200°C for 5 days at 2 h per day. The test samples were heated in the presence of onion or garlic (0.1 and 0.2 g per ml of oil) while another portion was heated without the spices. During heating, the oil was stirred continuously with a stirrer to ensure aeration and mixing and then allowed to cool down at room temperature overnight.

Measurement of lipid oxidation indicators

Acid value, peroxide value and iodine value were determined by titration method as described by Mehlenbacher (1960). Thiobarbituric acid method described by Moore and Robert (1998) was used for the determination of malondialdehyde content. Measurement of oil viscosity was carried out by the use of Ostwald viscometer.

Measurement of ascorbic acid and tocopherol concentration

The 2,4-dinitrophenylhydrazine method of Roe and Kuether (1943) was used for the determination of ascorbic acid concentration. The concentration of tocopherol in the oil samples was estimated using the method described by Waters et al. (1976).

Management of experimental animals

Eighty albino rats of the Wistar strain weighing between 80 – 100 g were used for this study. The rats were randomly divided into four experimental groups and fed with different diet as contained in Table 1. Group A was fed with fresh oil, B was fed with heated oil, C was fed with heated oil + 0.2 g onion/ml, and D was fed with heated oil + 0.2 g garlic/ml. Water was given to the rats *ad libitum* throughout the experimental period. Rats were withdrawn from each group on weekly basis for sacrifice and analyses.

Collection of blood and preparation of serum from rats

Rats were anaesthetized by putting them in jar containing cotton wool soaked in chloroform and allowed to go into unconscious state. They were sacrificed by cutting their jugular veins and their blood collected in plain bottles to allow for clotting. The clotted blood was centrifuged at 3000 rpm for 5 min and the clear supernatant (serum) separated from the pellet and kept frozen till required.

Assay for aminotransferases

The method of Reitman and Frankel (1957) was used for the determination of alanine aminotransferase and aspartate aminotransferase activities. All photometric measurements were done using the Jenway 6300 Spectrophotometer.

Statistical analysis

The data obtained were carefully subjected to statistical analysis using ANOVA with the aid of computer software package, SPSS version 12 and relevant conclusions were made.

RESULTS AND DISCUSSION

Table 2 shows the physicochemical properties of the fresh and thermooxidized soybean oil heated with or without onion or garlic. The colour of the oil was found to change from yellow to brick red after thermal processing while the oil also taste rancid. Acid value, peroxide value and concentration of malondialdehyde of the heated soybean oil were found to be significantly higher ($P<0.05$) than those obtained for fresh soybean oil. However these values were significantly reduced close to the control sample in oils heated with onion and garlic. The iodine number for the thermooxidized oil was significantly lower ($P<0.05$) than the fresh oil. Heating in the presence of onion and garlic increased the iodine number significantly ($P<0.05$) close to the control sample. There were no significant differences in values obtained for the two concentrations used (0.1 and 0.2 g/ml) and also between onion and garlic. Heated oil and oil heated with onion and garlic showed significant elevation in viscosity compared with fresh oil.

The effects of the diets on serum ALT and AST is shown in Figures 1 and 2, respectively. Significant ($P<0.05$) increase in the serum concentration of the two enzymes was recorded in rats fed with heated soybean oil diet starting from the second week to the end of the

Table 2. Physicochemical properties of soybean oil samples.

Parameters	Fresh oil	Heated oil	Heated oil +0.1 g onion/ml	Heated oil +0.2 g onion/ml	Heated oil +0.1 g garlic/ml	Heated oil +0.2 g garlic/ml
Colour	Yellow	Brick red	Brick red	Brick red	Brick red	Brick red
Taste	Tasteless	Rancid	Rancid	Rancid	Rancid	Rancid
Acid value (mg KOH/g)	$1.12^{a}\pm0.11$	$2.52^{b}\pm0.30$	$1.44^{ab}\pm0.22$	$1.38^{ab}\pm0.25$	$1.50^{ab}\pm0.26$	$1.47^{ab}\pm0.23$
Iodine value (g/100g)	$35.0^{a}\pm4.2$	$24.0^{b}\pm0.1$	$30.7^{ab}\pm3.4$	$31.8^{ab}\pm2.3$	$30.9^{ab}\pm2.8$	$31.6^{ab}\pm3.1$
Peroxide value (mmol 0_2/kg)	$20.0^{a}\pm7.1$	$35.0^{b}\pm8.7$	$27.6^{ab}\pm6.6$	$26.8^{ab}\pm5.9$	$28.8^{ab}\pm6.3$	$26.7^{ab}\pm5.8$
Viscosity (c.g.s)	$0.42^{a}\pm0.02$	$0.78^{b}\pm0.05$	$0.79^{b}\pm0.06$	$0.83^{b}\pm0.08$	$0.80^{b}\pm0.05$	$0.84^{b}\pm0.06$
Malondialdehyde (mg/kg)	$1.86^{a}\pm0.12$	$3.23^{b}\pm0.20$	$2.29^{ab}\pm0.15$	$2.22^{ab}\pm0.20$	$2.34^{ab}\pm0.18$	$2.30^{ab}\pm0.17$
Ascorbic acid (mg/100 g)	$19.8^{a}\pm2.9$	$12.7^{b}\pm1.8$	$17.5^{ab}\pm3.0$	$17.8^{ab}\pm2.8$	$17.5^{ab}\pm3.1$	$17.9^{ab}\pm2.9$
Tocopherol (mg/100 g)	$54.3^{a}\pm3.5$	$38.1^{b}\pm2.4$	$51.7^{ab}\pm2.7$	$52.0^{ab}\pm2.7$	$50.4^{ab}\pm3.0$	$50.8^{ab}\pm2.6$

*Values obtained are means of 5 analyses ±SD; *values with different letters are significantly different at $P<0.05$

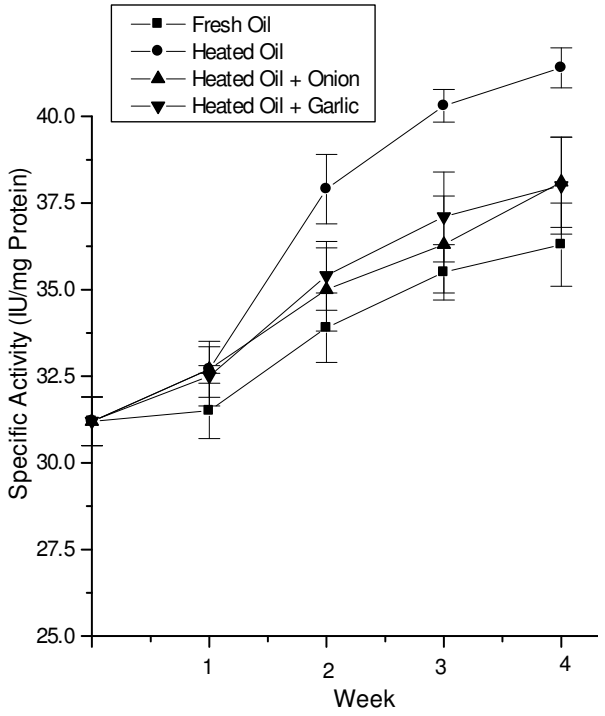

Figure 1. Serum alanine aminotransferase activity of rats fed with soybean oil diet.

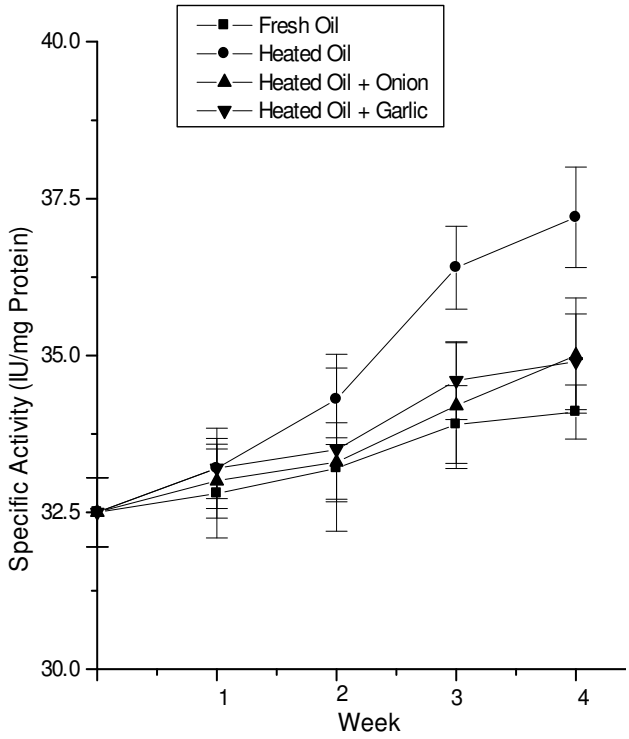

Figure 2. Serum aspartate aminotransferase activity of rats fed with soybean oil diet.

experimental period compared with the control. However there was no significant difference in the levels of the two enzymes in rats fed with diet containing heated oil with onion and garlic throughout the experimental period.

The darkened appearance obtained for the oil after thermal processing might arise from browning reaction taking place in the oxidized oil or due to bleaching which is brought about by destruction of carotene by oxidation products (Alexander, 1978). Color index of lipids has been reported to have significant correlations with total polar material although not a reliable indicator of oil quality (Takeoka et al., 1997). The rancid taste obtained might be as a result of decomposition to aldehydes, ketones and alcohol. Oils rich in linoleic acid have been found to go rancid easily when subjected to thermo-oxidation (Frankel, 1980).

The significantly high acid value, peroxide value and malonaldehyde content recorded for the heated soybean oil might be due to the liberation of free fatty acid and formation of peroxides and hydroperoxides from the lipid (Frankel, 1984). Peroxide value has been considered to

be the most common method of measuring hydroperoxides due to their transistory nature. They are the primary products of lipid oxidation and also intermediate in the formation of hydroxyl and carbonyl compounds (Kanner, 1994). Iodine number, which is a measure of unsaturation of the thermooxidized oil was significantly lower ($P<0.05$) than the fresh oil. The lower value obtained after heating might be because some of the double bond has been saturated during the process of oxidation as a result of attack by molecular oxygen (Chiba et al., 1981). Heating in the presence of onion and garlic significantly reduced these lipid oxidation indicators because they possess antioxidant properties.

The increase in viscosity obtained for the heated oil might be attributable to polymer formation among the degradation products upon cooling. Polymer formation has been found to increase with increase in unsaturation (Billek, 1985). Oils heated with garlic or onion also showed elevated viscosity probably because the spices were in powder (solid) form which definitely make the oil sample (liquid) more viscous. The significant loss in ascorbic acid and tocopherol in the heated oil occur probably as a result of their destruction by hydroperoxides generated during peroxidation. These losses were significantly reduced in the presence of onion and garlic because the substances possess antioxidant properties which prevent or retard the rate of lipid oxidation.

The observed significant increase in the levels of these enzymes in the rat serum caused by intake of heated soybean oil diet might be as a result of release of the enzymes from some tissues indicating tissue damage. ALT and AST are membrane bound enzymes associated with the mitochondria and cytoplasm. Their elevation in the serum in this study could be due to leakage of the enzymes through the damaged cell membrane caused by free radical attack (Kagan, 1988). Increase in serum ALT and AST activity has been reported in conditions involveing necrosis of hepatocytes, myocardial cells, erythrocyte and skeletal muscle cells (Macfarlane et al., 2000). The presence of onion or garlic in heated oil diet probably retard generation of appreciable level of free radical that could have resulted in membrane damage in tissues of rats fed with the diets.

Conclusion

It is evident from this study that there were gross changes in the physicochemical properties of soybean oil upon heat treatment which led to loss of quality and nutritional value. The study also indicated that consumption of food containing thermally heated soybean oil might be toxic. Heating in the presence of onion and garlic however markedly reduced the extent of peroxidation, nutritional loss and toxicity. Therefore, handling and processing techniques involving heat treatment which may bring about oil deterioration should be discouraged while the use of onion and garlic as spices in fatty food is recommended.

REFERENCES

Addis PB (1986). The Chemistry of polymerized oils. Food Chem. Toxicol. 24: 1021-1030.

Alexander JC (1977). Biological effects due to changes in fats during heating. J. Am. Oil. Chem. Soc. 55: 714-734.

Alexander JC (1978). Dietary fat and cancer N.Y Allan R. Liss Inc. pp. 185-209.

Billek G (1985). Heated fats in the diet. In: The role of fats in human nutrition. (Padey FB, Podmore J eds.) pp. 163-172.

Borek C (2001). Antioxidant health effects of aged garlic extract 1. J. Am. Soc. Nutr. 231: 43-54.

Cao G, Sofic E, Prior RL (1996). Antioxidant capacity of tea and common vegetables. J. Agric. Food Chem. 152: 33-37.

Chiba TK, Fuyimoto T, Kaneda S, Kubota K, Ikegani Y (1981). Decomposition products and peroxide intermediate in fats peroxidation, Ibid. 58: 587-590.

Frankel EN (1980). Lipid oxidation. Prog. Lipid Res. 19 (1-2): 1-22.

Frankel EN (1984). Lipid oxidation: mechanisms, products and biological significance. J. Am. Oil Chem. Soc. 61: 1908-1917.

Fremont L, Gozzelino MT, Franchi MP, Linard A (1998). Dietary flavonoids reduce lipid peroxidation in rats fed polyunsaturated or monounsaturated fat diets. J. Nutr. 128: 1495–1502.

Jacobson GA (1991). Quality control in deep fat frying operations. Food Technol. 45(2): 72-74.

Kagan VE (1988). Lipid peroxidation in biological membrane. Bocaraton CRC Press, pp. 52-59.

Kanner J (1994). Oxidative processes in meat and meat products: quality implications. Meat Sci. 36: 169–189.

Macfarlane I, Bomford A, Sherwood RA (2000). Liver diseases and Laboratory Medicine. ACB Ventures Publications London.

Maerker G (1987). Cholesterol autooxidation-current status. J. Am. Oil Chem. Soc. 64: 388-392.

Mehlenbacher VC (1960). Analysis of fats and oils. Garrard Press, Champaign, Illinois, p. 586.

Miller RW, Long PH (1990). A-91 days feeding study in rats with heated olestra/vegetable oil blends. Food Chem. Toxicol. 28: 307-315.

Moore K, Robert LJ (1998). Measurement of lipid peroxidation. Free Radical Res. 12: 18-22

Muller HG (1988). An introduction to tropical food science. Cambridge Univ. Press, p. 115.

Odutuga AA, Oyewole OI (2006). Effect of thermo-oxidized soybean oil on growth rate and skeletal muscle enzyme activities in rats. Biokemistri, 18(1): 9-13.

Reitman S, Frankel S (1957). A colorimetric method for the determination of serum glutamic oxaloaceytate aminotransferase. Am. J. Clin. Pathol. 28: 56-63.

Rodriguez–Estrada MT, Penazzi G, Coboni MF, Bertacco G, Lercker G (1997). Effect of different cooking methods on some lipids and protein components of hamburgers. Meat Sci. 45: 365–375.

Roe JH, Kuether CA (1943). The determination of ascorbic acid in whole blood and urine through the 2, 4 dinitrophenylhydrazine derivative of dehydroascorbic acid. J. Biol. Chem. 147: 399.

Sanders TAB (1989). Nutritional aspect of rancidity. In Allen J.C. and Hamilton RJ eds. Rancidity Food 7: 125-139.

Takeoka GR, Full GH, Dao LT (1997). Effect of heating on the characteristics and chemical composition of selected frying oils and fats. J. Agric. Food Chem. 45(8): 3244-3249.

Waters RD, Kesterson JW, Braddock RJ (1976). Method for determining the α-tocopherol content of citrus essential oils. J. Food Sci. 41(2): 370–371.

White TJ (1991). Methods of measuring change in deep fat frying oils. Food Technol. 45(2): 75-80.