Technical

Nutritional Evaluation of Refined, Heated and Hydrogenated *Hibiscus sabdariffa* Seed Oil

G. SAROJINI, K. CHITTEMA RAO and **P. GEERVANI,** Department of Foods and Nutrition, College of Home Science, Andhra Pradesh Agricultural University, Hyderabad 500 004, India

ABSTRACT

Alkali refined *Hibiscus sabdariffa* seed oil (Mesta Oil) of the family *Malvales* was heated and hydrogenated to eliminate the cyclopropene fatty acids (CPFA). Such processed oils were fed to weanling rats at 10% level of the diet for 4, 8 and 12 weeks. The food intake and gain in weight were found to be less in the refined oil group than in the peanut oil control group. The digestibility was found normal with all the processed oils. Serum and liver lipid composition with respect to total lipids, cholesterol and phospholipids were comparable to those fed with peanut oil. The liver architecture did not show any abnormalities with *H. sabdariffa* oil feeding.

INTRODUCTION

There is a large gap between the supply of and demand for vegetable oils in India. To meet this situation a number of unusual sources of oil-bearing seeds have been examined and exploited, and still the search is going on. Among these unusual oilseeds, seeds of *Hibiscus cannabinus* and *Hibiscus sabdariffa* of the family *Malvales* have been identified as good sources of oil. Physicochemical characteristics of the oils are similar to those of most commonly used edible oils (1), but the fatty acid composition has been found to contain cyclopropene fatty acids (CPFA) and epoxy oleic acid (2). The process of refining the crude oil did not affect the levels of these two unusual fatty acids. Heating for 10 min at smoke temperature was found to inactivate the cyclopropene acids and not the epoxy acids. Partial hydrogenation of the refined oil for 1.5 hr using nickel catalyst also was found effective in activating CPFA, which was reported earlier (3). The effect of feeding such oils at 10% level in the diet to rats is reported here.

MATERIALS AND METHODS

Seventy-two male weanling Wistar rats (22-24 days old) weighing from 34-36.5 g were divided into four groups. One group served as control, and the other three as experimental groups. They were fed with one of the diets ad libitum under hygienic conditions (Table I), and had plenty of clean water to drink.

After 4 weeks of feeding, 6 animals from each group were picked randomly, killed under mild anesthesia and their blood and livers collected. The blood was allowed to clot for serum. The livers were homogenized with chloroform-methanol mixture (2:1) to a final volume 20 times the weight of the liver, filtered, and the volume recorded. At the end of 8 and 12 weeks the same was repeated. From the livers of the rats killed at the end of 12 weeks, liver tissue cuts of 1 cm were taken from the right lobe and fixed separately in 10% neutral buffered formalin for histapathological studies.

The digestibility of the oils both in the control and experimental groups was determined at the end of 7 weeks, after necessary corrections were made for metabolic fat excretion.

The serum and liver homogenates were analyzed for total lipids, total cholesterol and phospholipids. The methods followed for analysis were: total lipids in serum and feces by Henry (4); total cholesterol in serum and liver by Zak et al. (5); phospholipids in serum and liver by the modified method of Youngberg and Youngberg (6), and total lipids in liver by Folch (7).

TABLE I

Percentage Composition of Experimental Diets[a]

Ingredients	Groups I	II	III	IV
Corn starch	67.5	67.5	67.5	67.5
Casein	13.5	13.5	13.5	13.5
Peanut oil	10.0	–	–	–
H. sabdariffa seed oil				
Refined	–	10.0	–	–
Heated for 10 min	–	–	10.0	–
Partially hydrogenated	–	–	–	10.0
Cellulose powder	3.0	3.0	3.0	3.0
Salt mixture 1	4.0	4.0	4.0	4.0
Vitaminized starch 2	1.0	1.0	1.0	1.0
Vitaminized oil 3	1.0	1.0	1.0	1.0

[a]The salt mixture, vitaminized starch and vitaminized oil according to Indian Standard Institute specifications - 7481 (1974).
Salt mixture 1—NaCl, 139.3 g; KI, 0.79 g; KH_2PO_4, 389.0 g; $MgSO_4$ (anhydrous), 57.3 g; $CaCO_3$, 381.4 g; $FeSO_4 \cdot 7H20$, 27.0 g; $MnSO_4 \cdot H_2O$, 4.01 g; $ZnSO_4 \cdot 7H_2O$, 0.548 g; $CuSO_4 \cdot 5H_2O$, 0.477 g; $CoCl_2 \cdot 6H_2O$, 0.23 g.
Vitaminized starch 2—1 g of corn starch contained: Vitamin K (Menadione), 0.5 mg; thiamine, 0.5 mg; riboflavin, 1.0 mg; pyridoxine, 0.4 mg; calcium pantothenate, 4.0 mg; niacin, 4.0 mg; choline, 200 mg; inositol, 25 mg; paraminobenzoic acid, 10 mg; vitamin B_{12}, 2 mcg; biotin, 0.02 mg; folic acid, 0.2 mg.
Vitaminized oil 3—1 g of the oil contained: Retinol, 300 mcg; vitamin D_2, 2.5 mcg; α-tocopherol acetate, 10 mg.

For the detailed histopathological study the microtome procedure of Culling (8) modified by Singh and Sulochana (9) was followed. The results were subjected to analysis of variance (10).

RESULTS AND DISCUSSION

Growth and Digestibility

The results of the mean food intake, gain in body weight, feed efficiency ratio (FER) and digestibility for the rats fed peanut oil, refined, heated and partially hydrogenated *H. sabdariffa* oil at 10% for 4, 8 and 12 weeks are indicated in Table II.

In the first 4 weeks of feeding the gain in body weight and FER for the rats fed refined oil were significantly lower than for the groups fed peanut, heated and partially hydrogenated oils ($P<0.01$). After 8 and 12 weeks of feeding the differences were minimized. The differences among the

TABLE II

Mean Food Intake, Weight Gain, FER and Digestibility of Rats Fed Peanut Oil and *H. sabdariffa* Oils at 10% of Diet

Diet/period of feeding	Food intake (g) Weeks 4	8	12	Body weight (g) Weeks Initial	4	8	12	FER* (wt. gain/ food intake)	Digestibility %
Peanut oil	261 ± 7	296 ± 8	298 ±10	35.0 ±0.6	77.8[a] ±12.9	146.6 ±12.9	184.5 ± 6.2	0.27[a] ±0.03	96.9 ±0.1
H. sabdariffa seed oil									
Refined	202 ± 6	267 ± 9	294 ± 9	35.0 ±0.5	43.9[b] ±8.5	112.4 ± 9.4	145.8 ± 4.9	0.22[b] ±0.03	95.9 ±0.3
Heated	222 ± 7	272 ± 8	299 ±11	35.0 ±0.5	51.6[c] ±7.6	119.9 ±11.1	156.0 ± 9.0	0.24[c] ±0.02	96.1 ±0.2
Hydrogenated	217 ± 6	271 ± 7	297 ±10	35.0 ±0.6	48.5[c] ±5.8	105.9 ± 9.6	138.8 ± 4.9	0.24[c] ±0.02	96.5 ±0.2

*FER was calculated at the end of 4 weeks.
±Standard deviation (S.D.). Means carrying the same superscript are not significantly different at 1% level.

TABLE III

Liver Weight and Liver Weight/Body Weight for Control and Experimental Rats

Diet	Liver weight (g) Weeks 4	8	12	Liver weight as % of body weight Weeks 4	8	12
Peanut oil	4.4 ±1.2	5.6 ±0.8	6.7 ±0.9	3.8 ±0.5	3.2[a] ±0.4	3.0 ±0.3
H. sabdariffa seed oils						
Refined	3.0 ±0.4	5.1 ±0.6	5.9 ±0.9	3.9 ±0.6	3.7[b] ±0.3	3.1 ±0.5
Heated	3.6 ±0.6	5.9 ±0.2	6.7 ±1.1	4.2 ±0.7	3.9[b] ±0.2	3.5 ±0.7
Hydrogenated	3.7 ±0.1	5.9 ±0.2	6.6 ±0.9	4.5 ±0.1	4.2[c] ±0.2	3.7 ±0.2

Means carrying the same superscript are not significantly different at 1% level.
±S.D.

groups with respect to digestibility co-efficient were negligible, and the values were comparable to those for most commonly used edible oils.

Liver Weight

The mean liver weight and the ratio of liver weight to whole body weight of the control and experimental groups are presented in Table III.

The absolute liver weights of rats fed experimental diets were comparable to the peanut oil fed group except for the rats fed refined oil, which consistently recorded lower liver weights than the other experimental and control groups. However, the liver weight/whole body weight ratio was higher for the rats fed experimental oils than for those fed peanut oil.

Total Lipids in Serum and Liver

The mean values for the total lipids in serum and liver of rats fed peanut and processed *H. sabdariffa* oils are given in Table IV.

Slight differences were found between groups with respect to the total lipids in serum and liver, but they were not significant ($P<0.01$). The total lipids in serum and liver increased significantly ($P<0.01$) with the feeding period. The total lipid level in the livers of rats fed refined oil for 12 weeks in this study was less than that reported by Rukmini et al. (2) for the same feeding period.

Total Cholesterol in Serum and Liver

The mean values for the total cholesterol in serum and liver of rats fed control and experimental oils are shown in Table V.

The serum cholesterol level of rats fed groundnut and *H. sabdariffa* seed oils, both refined and processed, did not differ significantly up to 8 weeks of feeding. But after 12 weeks these were significantly different ($P<0.01$). Among the *H. sabdariffa* oil groups, rats fed refined oil were found to have very high levels of serum cholesterol. This might be due to the cyclopropene fatty acids present in the oil, which is known to affect the metabolism of fat by interfering with enzyme activity (11).

Phospholipids in Serum and Liver

The mean phospholipids in serum and liver of control and experimental animals are detailed in Table VI.

The differences in phospholipid levels in serum of rats fed peanut and *H. sabdariffa* seed oils were not significant ($P<0.01$). However, the rats fed peanut oil had low levels of phospholipids in serum during all the periods of feeding.

TABLE IV

Mean Total Lipid Concentration in Serum and Liver of Rats Fed Control and Experimental Oils

	Serum total lipids (mg/100 ml)			Liver total lipids (g/100 g liver wt)		
	Weeks			Weeks		
Diet	4	8	12	4	8	12
Peanut oil	182.5 ±21.7	220.0 ±14.1	292.5 ±23.4	4.2 ±0.7	4.2 ±0.3	6.0 ±0.9
H. sabdariffa seed oils						
Refined	212.5 ± 8.3	232.5 ± 8.3	322.5 ±64.1	4.4 ±1.0	4.5 ±0.3	6.1 ±1.2
Heated	212.5 ±12.9	237.5 ±25.7	306.3 ±50.2	4.2 ±0.5	4.6 ±0.6	6.2 ±1.5
Hydrogenated	207.5 ± 4.3	242.5 ±10.9	322.5 ±53.1	4.4 ±0.7	4.6 ±0.8	6.4 ±0.4

±S.D.

TABLE V

Mean Cholesterol Concentration in Serum and Liver of Rats Fed Control and Experimental Oils

	Serum cholesterol (mg/100 ml)			Liver cholesterol (mg/100 g liver wt)		
	Weeks			Weeks		
Diet	4	8	12	4	8	12
Peanut oil	60.2 ±1.5	66.8 ±2.1	84.5[a] ±12.1	151.8 ± 7.4	171.5 ±11.4	238.8 ±16.5
H. sabdariffa seed oil						
Refined	61.7 ±2.1	67.5 ±2.4	110.2[b] ± 6.0	158.9 ± 7.7	175.8 ±13.3	258.1 ±17.5
Heated	63.9 ±4.4	69.0 ±3.2	97.7[c] ± 4.9	156.9 ±12.0	177.5 ± 8.2	255.3 ±12.5
Hydrogenated	60.2 ±1.5	66.8 ±3.6	95.5[c] ± 6.4	158.1 ±14.0	172.9 ±17.8	254.3 ±20.2

Means with the same superscript are not significantly different at 1% level.
±S.D.

TABLE VI

Mean Phospholipid Levels in Serum and Liver of Rats Fed Control and Experimental Oils

	Serum phospholipids (mg/100 ml)			Liver phospholipids (g/100 g liver wt)		
	Weeks			Weeks		
Diet	4	8	12	4	8	12
Peanut oil	113.5 ± 4.8	123.5 ± 3.3	127.7 ±30.3	2.1 ±0.3	2.3 ±0.3	2.6 ±0.2
H. sabdariffa seed oil						
Refined	123.3 ± 4.7	128.0 ± 3.5	138.3 ± 7.4	2.3 ±0.7	2.4 ±0.2	2.8 ±0.1
Heated	133.8 ± 4.2	125.5 ± 4.3	135.3 ±12.5	2.3 ±0.7	2.2 ±0.3	2.3 ±0.2
Hydrogenated	120.0 ± 6.1	125.5 ± 5.5	135.1 ±16.1	2.3 ±0.4	2.2 ±0.2	2.8 ±0.3

±S.D.

Liver phospholipid content appeared to be unaltered by dietary treatment.

Histopathological Changes

Feeding for 12 weeks with refined, heated and partially hydrogenated *H. sabdariffa* seed oil at the 10% level did not alter the liver architecture of the experimental rats compared to that of rats fed peanut oil.

From the growth performance of the group of rats fed refined *H. sabdariffa* seed oil and also based on other observations in the study, it is concluded that refined oil is inferior to heated and partially hydrogenated oil. These observations indicate that *H. sabdariffa* seed oil may be considered an edible oil after suitable methods of processing such as refining, bleaching, deodorization and heating. Partial substitution or blending of this oil with other commonly used cooking oils will bring down the levels of unusual fatty acids and may make it suitable for human consumption.

ACKNOWLEDGMENTS

The A.P. Agricultural University provided the opportunity to do the Research, and I.C.A.R. provided financial assistance. P.G. Tulpule provided much help during the investigation.

REFERENCES

1. Ramakrishna, G., N. Krishna Reddy, G. Azeemoddin, D. Atchyutaramayya and S.D. Thirumala Rao, J. Oil Tech. Assn. India. 11:75 (1979).
2. Rukmini, C., M. Vijayaraghavan and P.G. Tulpule, JAOCS 59: 415 (1982).
3. Sarojini, G., Physicochemical and Biological Studies with Mesta Oil. Thesis submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Home Science to the A.P. Agricultural University, Hyderabad, India (1982).
4. Henry, R.J., Clinical Chemistry–Principles and Techniques. Harper and Row Publications, 2nd edition, 1964, p. 836.
5. Zak, B., R.C. Dickerman, E.G. White, H. Burnett and P.J. Charney, Am. J. Clin. Pathol. 24:1307 (1954).
6. Oser, B.L., Hawk's Physiological Chemistry, 14th edition, McGraw Hill Book Co., New York (1965), Chapters 23 and 29 and pp. 1060-1067.
7. Folch, J.M., K.M. Lees and G.M.S. Stanley, J. Biol. Chem. 226:497 (1957).
8. Culling, C.F.A., Handbook of Histopathological Techniques, London Butter Worth, 2nd edition, 1963.
9. Singh, U.B., and S. Sulochana, Laboratory Manual for Histological and Histochemical Methods (Monograph) (1976).
10. Snedecor, G.W., and W.G. Cochron, Statistical Methods, 6th edition, Oxford and IBH Publishing Co., 1967, Chapter 2.
11. Goodnight, K.C. Jr., and A.R. Kemmerer, J. Nutr. 91:174 (1967).

[Received October 1, 1984]

Lipid Content and Fatty Acid Profiles of Various Deep-Fat Fried Foods[1]

L.M. SMITH,* A.J. CLIFFORD, R.K. CREVELING and **C.L. HAMBLIN,** Departments of Food Science and Technology and Nutrition, University of California, Davis, CA 95616

ABSTRACT

Forty-one brands of nine different types of snack and convenience foods were purchased from food stores and fast service restaurants in the Sacramento area of California. All samples had been prepared by deep-fat frying. They included potato chips, corn chips, tortilla chips, cheese chips, cheese puffs, cake donuts, french fries, chicken pieces and fish pieces. These samples were analyzed in duplicate for total fat and fatty acid composition. The total lipid content of each type of food varied among different commercial sources; the average percentages were as follows: potato chips, 40; cheese puffs, 38; corn chips, 35; cheese chips, 25; tortilla chips, 24; cake donuts, 22; chicken thighs, 14; french fried potatoes, 14, and fish pieces, 10. The fatty acid profiles of the total lipids in several brands of potato chips were relatively constant. The fatty acid profiles of the total lipids in the corn and cheese snack foods varied widely. Fatty acid compositions of donuts, chicken and fish pieces and french fries were influenced by the amount and fatty acid profile of the lipids in each uncooked food, as well as by the composition of the cooking fat.

INTRODUCTION

Consumption of a wide variety of fast foods including convenience and snack foods continues to increase in the U.S.A. and other countries (1-3). Many of these foods, including french fried potatoes, chicken and fish pieces, potato chips, corn chips, tortilla chips, extruded snacks and donuts, are prepared by deep-fat frying. In this process, the cooking oil or fat often is kept hot for long periods of time at about 180 C and moisture and air are mixed into it. The fried foods absorb this heated fat and contribute substantially to the fat ingested by consumers. The lipid composition of deep-fat fried foods is of considerable interest to nutritionists concerned with nutrient intake of young people, who tend to consume large quantities of convenience and snack foods, and to individuals who wish to alter their fat intake for medical reasons. However, compositional data for these products are sparse and widely scattered in the scientific literature.

This paper provides information on the total lipid content, the fatty acid composition and the variability among selected deep-fat fried foods available in California.

EXPERIMENTAL

Samples

Forty-one samples of nine types of snack and convenience foods were obtained from food stores and fast service restaurants in the Davis and Sacramento areas of California. Potato chips, corn chips, tortilla chips, cheese chips and cheese puffs were sampled from brands including Albertson, Alpha Beta, Buffalo, Cheetos, Doritos, Granny Goose, Lady Lee, Laura Scudders, Lays, Party Pride and Tostidos. Plain cake donut brands included Fluffy, Hostess, Taylor's and Winchell's. French fries were collected from Carl's Junior, Fluffy Donut, Jack in the Box, Kentucky Fried Chicken, London Fish and Chips and McDonald's. Brands of chicken pieces (thighs) included Church's, Kentucky Fried, Pioneer and Swanson's. Fish pieces included samples from London Fish and Chips, H. Salt, Long John Silver's and Skipper's. All samples were stored under nitrogen at 4 C for 1-4 days before being analyzed at least in duplicate.

[1]Presented in part at the Sixth International Congress of Food Science and Technology, Dublin, Ireland, September 1983.
*To whom correspondence should be addressed.