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EFFECT OF DIETARY FROG OIL ON GROWING RATS

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ABSTRACT
Isocaloric and isonitrogenous diet formulated with palm oil (control, A) and frog oil (test B) were fed to growing albino rats of 5-6 weeks. Weight was monitored for 31 days and fat deposit, total cholesterol, triglyceride, HDL- cholesterol and LDL-cholesterol were determined according to standard procedures. Weight gain by animals fed diet B was higher but not significant ($p>0.05$), fat deposit was significantly higher ($p< 0.05$) in animals fed diet A compared to those who received diet B. Serum cholesterol (217± 0.01 mg/dl). Triglyceride (506± 0.01 mg/dl) were significantly higher ($p< 0.05$) in animals fed diet A compared with serum cholesterol (104± 0.04mg/dl) and triglyceride(113± 0.01mg/dl) of animals fed diet B, cholesterol levels (1.44± 0.0MG/G) in the heart of animals that received diet A was significantly higher ($p< 0.05$) compared to the value in animals that received diet B (1.08± 0.02 mg) both serum HDL-cholesterol and LDL- cholesterol 46.03± 0.01 and 69.77± 0.01 mg/dl respectively were significantly higher ($p<0.05$) in animals fed diet A compared with those who received diet B (33.04± 0.01 and 48.36 ± 0.02mg/dl) for HD and LDL- cholesterol levels respectively. The biochemical parameters analyzed in animals fed frog oil diet are comparable to values documented for oils of marine origin. Frog oil may therefore be superior to palm oil as a sourced of dietary oil to reducing physiological problems arising from increased serum cholesterol and triglyceride.

KEYWORDS: Isocaloric, isonitrogenous diet, frog oil, albino rats, Triglyceride.

INTRODUCTION

During the past several decades, reduction in fat intake has been the main focus of national dietary recommendation. In the public minds, the words “dietary fat” has become synonymous with obesity and heart diseases, where as the word “low fat” and “fat free” have become synonymous with heart health (Mokdad *et al*., 2000).

Fats and oils referred to as lipids, (simplest and most abundant) are the natural fats, which are also called triacylglycerol or triglycerides. These compounds are ethers of glycerol and three fatty acids. They are the main form of fat storage in plants and in the adipose cells (or fat cell) of animals, an average man’s body is 21 percent fat (26% for woman), This enough to supply the body’s energy need for 2-3 months (Roberts *et al*., 1995). Lipids may be of vegetable, animal and marine origin examples of vegetables fats includes solid fats as cocoa, butter and liquid oils as corn oil, olive oil.(forrest et al.,2004) marine oil in fish (Forest *et al*., 2004).

The fatty acids found in plant and animal lipids are of three kinds: saturated (no double bonds), mono-unsaturated (one double bonds), and polyunsaturated (two or more double bonds). They are presents in variable amounts: - (a) lipids form terrestrial animal sources beef, mutton and poultry are usually solid fats, having a high content of saturated fatty acids mainly palmitic acids and stearic acids. (b) Lipids from marine animal sources fish, seal and whales, are usually lipids oils having a high content of polyunsaturated fatty acids, oleic and linoleic acids. (c) Plant lipids with exception of coconut and palm oils are all lipid oils and have a high content of polyunsaturated fatty acids (Wijendran *et al*., 2009).

Several lines of evidence however have indicated that types of fats have a more important role in determining risk of coronary heart disease than total amount of fat in the diet (Baylin *et al*., 2003). In contrary, it has been proposed that population that can eat large amount of fish or marine mammals may be less prone to coronary heart disease due to their high content of polyunsaturated fatty acids (Baylin *et al*., 2003).the consumption of these polyunsaturated fatty acids, which are derived from marine oils, lower plama triglyceride concentrations and prolong template bleeding time (Diniz *et al*.,2004).

As the world’s population grows and more people adopt the eating habits of industrialized countries, both domestic and industrial demand for fat and oil has continued to rise. This deficiency has remained despite the present level of sophistication and advancement. Several schemes to increase the amount of fat available have been proposed including the growing of groundnut in east Africa etc (Diniz *et al*.,2004).

In view of this reason, the need to investigate other sourses of fats and oil is essential (Feinman 2010). Gras frog (*Rana pipiens*) an aquatic and terrestrial dweller is a popular food source among Chinese and Indians, and is gradually becoming popular in some part of Niger delta regions of Nigeria. It is also readily available in large quantity in nigeria particularly in the raining season which last from april-september each year.the oil obtained from frog is known as frog oil’,it has an iodine no of 74s.5% (Padmanaban and Sarkar,2010).

Though, frog oil has been very useful in biological, biochemical and physiological studies particularly in gonadotropin and ovulatory activities, however, no work has been done on the dietary effect of frog oil or it’s implication in animal diets since oil obtained form marine animals such as fish oil are known to reduce coronary heart disease (Forrest, 2004) and that consumption of poly

unsaturated fatty acids mainly dervide form marine oil, lowers plasma triglyceride concentration (Onyeneke *et al*., 2007).

In view of this, the study was therefore conceived to investigate the dietary implication of frog oil (Rana pipiens) which partly lives in water and on land in animal diets in comparism with plant oil which are polyunsaturated.

MATERIAL AND METHODS

The basic raw materials for this research work are the common grass frog (Rana, Pipiens) oil and albino rats. The grass frog oil was obtained from Bida in Niger state where it was processed and used for edible oil, around the locals in badegi where local rice farmers process it to edible oil. Growing albino rats of 5-6weeks were obtained from the animal house of science laboratory technology department, federal Polytechnic Bida. The rats were divided into two groups of 5 rats respectively and housed in standard metabolic cages.

Determination of energy value

Ballistic bomb calorimeter (gallenkamp, Germany) was used in determining the energy value of the basal diet and the experimental diet.

Animal sacrifice

At the end of the experimental period (31 days) the animals were fasted at least 8 hours and were later sacrificed by jugular dislocation, blood was collected and prevented from clotting using EDTA. The rats were quickly dissected and the organs (kidney, brain, liver and heart) were removed and weighed respectively. All the organs were homogenized with 0.25m sucrose solution 5:1v/w. The subcutaneous fat which extended along the front of the thigh across the abdomen were removed from both sides of the animals and weighed accordingly.

Analysis

Total protein total, cholesterol, HDL-cholesterol, LDL-cholesterol and triglycerides concentration of the homogenate organs and serum of rats were determined using the randox automated spectrophotometric kits.

Statistical analysis

The student T-test was used to analyze the result obtained for the organs weight, fat, deposit total protein, total triglycerides, total cholesterol, HDL and LDL cholesterol as reported by (Saunder *et al*., 1971).

RESULT AND DISCUSSION

The energy value of diet A was 531.85kcal/g and the energy value of diet B was also 531.85callg.

TABLE 1. Fat deposit 1100g body weight of rates fed with diets A and B(g)+

A	B
1.96±0.66*	1.35±0.20

+ Each value represents a mean of at least 4 rats ± S.D.

* Figure in the same row with asterisk are significantly different (P<0.05).

TABLE 2: Organ weight 1100g body weight of rats fed with diet A and B(g)+

Organs	Diet A	Diet B
Kidney	0.91±0.09	0.96±0.11
Brian	1.13±0.27*	0.96±0.24
Liver	4.98±0.51	6.11±0.55*
Heart	0.39±0.0.6	0.45±0.66*

+ Each value represents a mean of at least 4 rats + S.D

* Figure in the same row with asterisk are significantly different (P<0.05)

TABLE 3. Total protein content of rats fed with diets A and B in ml/g wet organ weight +

Organ	A	B
Kidney	0.052±0.01	0.053±0.02*
Brain	0.79±0.04*	0.072±0.02
Liver	0.149±0.02	0.201±0.01*
Heart	0.012±0.03	0.012±0.01
Serum (g/dl)	13.750±0.07*	6.750±0.07

+ Each value represents a mean of at least 4 determination s+S.D

* Figures in the same row with asterisk are significantly different (P<0.05).

TABLE 4. Total cholesterol content in organs of rats fed with diets A and B in mg/g wet organ weight +

Organ	A	B
Kidney	0.052±0.01	0.053±0.02*
Brain	0.79±0.04*	0.072±0.02
Liver	0.149±0.02	0.201±0.01*
Heart	0.012±0.03*	0.012±0.01
Serum (g/dl)	13.750±0.07*	6.750±0.07

+ Each value represents a mean of at least 4 determination s+S.D

* Figures in the same row with asterisk are significantly different (P<0.05).

TABLE 5. Total cholesterol content in organs of rats feel with diets A and B in mg/g wet

Organ	A	B
Kidney	4.87±0.01	8.06±0.03*
Brain	94±0.03	9.55±0.02*
Liver	24.15±0.02	26.58±0.01*
Heart	1.44±0.04*	1.08±0.02
Serum (g/dl)	217.00±0.01*	104.00±0.04

+ Each value represents a mean of at least 4 determination s+S.D
* Figures in the same row with asterisk are significantly different (P<0.05).

TABLE 6. HDL – Cholesterol content in the serum of rats fed with diets A and B mg/dl ±.

	A	B
Serum HDL – Cholesterol mg/dl	46.03±0.01	33.04±0.01
Serum LDL –Cholesterol mg/dl	69.77±0.11	48.36±0.02

+ Each value represents a mean of at least 4 determination s+S.D
* Figures in the same row with asterisk are significantly different (P<0.05).

TABLE 7. Total triglyceride content in organ of rats fed with diets A and B in mg/g wet organ weight +

Organ	A	B
Kidney	22.02±0.02	29.57±0.03
Brain	6.05±0.01*	3.65±0.01
Liver	26.64±0.03	46.74±0.01
Heart	2.11±0.02	2.16±0.03*
Serum (g/dl)	506.0±0.01*	1113.00±0.01

+ Each value represents a mean of at least 4 determination s+S.D n = 10
* Figures in the same row with asterisk are significantly different (P<0.05).

RESULTS AND DISCUSSION

The pattern of feed consumption by the groups of rats was almost the same and weight gain in weight as shown in figure 1 was not significantly different (p<0.05). The extra weight gain may have been in part due to a pathological accumulation of fat in the rats (Forest, 2004).

The fat deposit 100g body weight as shown in table 1 indicated that rats fed with diet A had significantly higher (p<0.05) fat deposit than rats fed with diet B which could mean that rats fed with diet A stored their fatty acids in their adipose tissues as rats are born in a foetal state with very little adipose tissue (Forest,2004). Rats fed with diet B may have stored their fatty acid around other organs in the body.

Table 2 shows organ/body weight ratio in which the liver and heart weight of rats fed with diet B were significantly higher (p<0.05) than those fed with diet A while the brain weight of rats fed with diet A significantly higher (p<0.05) than rats fed with diet B but the kidney weight was not significantly different (p<0.05). the increased in liver weight is similar to what was obtained by (Onyeneka *et al*.,2007) feeding rats with fat diets supplemented with methionine and lysine hydrochloride, this increase could may be mainly due to triglycerides similarly, the increased in heart weight may also imply a chance of coronary heart disease as lipid could accumulate around the organ though moderate amount is required for protective role.

The total protein concentration as shown in table 3 was significantly higher (p<0.05) in liver and kidney of rats fed with diet B than rats fed with diet A which agrees with the findings of (Jackson et al, 1988) when fish oil was fed to growing rats with a change in both protein synthesis and degradation and the resultant net rate of protein deposition remain the same. The serum and brain total protein concentration of rats fed with diet A were significantly higher (p<0.05) than rats fed diet B while that of heart was not significantly different (p<0.05).

Table 4,5 shows total cholesterol concentration in which the concentration of kidney, brain and liver of rats fed with diet B were significantly higher (p<0.05) than diet B. the increased in liver cholesterol levels may be due to increase rate of cholesterol synthesis as a result of high fat feeding (Sacks,1994) and decreased cholesterol metabolism as evidenced by a reduction in bile acid production and turn over after high fat feeding (Nicolosi et al, 1976). The low serum cholesterol of rats fed with diet B corresponds not only to a reduction of the concentration of serum cholesterol in low- density lipoproteins (LDL) of rats fed with diet B as shown in table 5,6 as reported by Shepherd et al, (1981), and Schaefer et al, (1981).

The reduction of LDL-cholesterol is presumably beneficial but the decrease of the HDL-Cholesterol is probably less desirable as judged from epidemiological findings indicating that a high HDL-Cholesterol concentration is associated with a low incidence of coronary heart disease (Gordon et al, 1997). This implies that the rats fed with diet B with both low concentration of LDL and HDL could cause coronary heart disease.

The triglyceride concentration of kidney, liver and heart of rats fed with diet B as shown in table 6,7 were significantly higher (p<0.05) than rats fed with diet A while the concentration of brain and serum of rats fed with

diet A were significantly higher than rats fed with diet B. the increase in triglyceride concentration in liver is a common feature when rats develop fatty liver (Creasey et al, 1961, Ugozio et al, 1995 and Aoyama et al, 1975) and this may be the reason why the liver weight of rats fed with diet A s which implies that rats fed with diet B has fatty liver. The increase in triglyceride concentration in the heart of rats fed with diet B could also support the evidence that low HDL-Cholesterol concentration is associated with coronary heart disease as reported by Gordon et al, (1977). Likewise, the increase in triglyceride concentration in serum of rats fed with diet A may be due to endogenous synthesis and decreased hepatic oxidation (Williams et al, 1985) resulting from the high fat content of diet A.

CONCLUSION

Base on the result of this study, it is concluded that frog oil with an iodine value of 74.5s, could cause coronary heart disease due to low concentration of HDL-cholesterol. It is also concluded, that frog oil can cause fatty liver due to increase triglyceride concentration in the liver of rats.

RECOMMENDATION

It is recommended from this study, that frog oil consumption must be carefully reduced by people and can be substituted with polyunsaturated oils from marine origin and vegetables sources to reduce the risk of coronary heart disease and developing fatty liver.

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