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IMPACTS OF HIGH DIETARY FAT ON SERUM CHOLESTEROL AND DEVELOPMENT OF FATTY LIVER IN RATS

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ABSTRACT

The present study was designed to evaluate the impacts of high dietary fat on serum Total cholesterol and fatty liver syndrome in rats. Rats are fed on diets containing cholesterol; they develop fatty livers which are characterized by the presence in the liver of excessive amounts of cholesteryl esters, and glyceride. Increasement of glyceride content depend on a number of factors, such as the dietary contents of choline, While the nature of the "cholesterol" fatty liver and the effects on its composition of a number of dietary and other factors. In the present paper, we investigated the quantitative changes which occur in the "cholesterol" fatty liver, as a result of variations in the fat content of the diet, with particular reference to the deposition of cholesterol and of glyceride on diets of constant cholesterol content. Investigation was conducted on 90 day old Wister rats. It was observed that the serum TC values in rats of groups B and C were higher than control group. Furthermore, the serum TC and TG value was higher in rats of group C than group B. Grossly, the livers of rats of groups B and C were enlarged, pale in colour, soft in consistency and were having petechial haemorrhages with fat and fibrin deposits. Histopathologically, livers of groups B and C showed fatty infiltration, haemorrhages and mass of eosinophilic materials. The vacuoles coalesced to create clear space that displaced the nucleus to the periphery of the cell. The results suggested that addition of dietary fat from animal and vegetable sources in the diet of rats not only resulted in increase in serum TC and TG but also in marked macroscopic and microscopic changes in vital organ liver.

INTRODUCTION: Cholesterol is the most important sterol in the human body. Cholesterol is majorly synthesized by animals but small quantities are also synthesized in other eukaryotes such as plants and fungi. It is completely absent among prokaryotes[1]. Fats are important in the diet of mammals as concentrated source of energy and the essential nutrients, amount of saturated fatty acids (lauric, myristic, palmitic acid, linoleic acid and arachidonic acid), sucrose, fructose, increases the plasma lipids particularly, cholesterol and triglyceride level. Whatever the source of fat, it is a high-energy diet. Excessive consumption of high energy/ caloric diets combined with restricted activity is believed to result in fatty liver syndrome, a condition of disturbed metabolism leading to excessive fat deposition in the liver[2]. A strong association of reticulolysis with severity of liver haemorrhage has been described in experimental rats. Hepatic fibrosis was documented with cholesterol supplemented diet[3]. Rupture of intrahepatic portal veins associated with

degenerative changes in the veins is also described in the same rats. Focal necrosis of heaptocytes may lead to vascular injury and haemorrhage. Excessive lipid peroxidation of unsaturated fatty acids in the liver may overwhelm cell repair mechanisms and result in tissue damage3. The present study was designed to evaluate the effects of increased dietary fat source both from animal and vegetable origins on the deposition of fat in liver and serum cholesterol in rats.

MATERIALS AND METHODS:

Experimental animal and design: Eighteen adult male Wister albino rats weighing about 180-200g were used. The animals were given standard pellets and tap water *ad libitum* and were housed in separate cages (360 x 200 x 190 mm^3). All animals were kept under standard laboratory conditions at 22 ± 2^0C, relative humidity of 55%. The specimens were divided into three groups of six animals each and randomized (Latin Square Method). They were labeled through picric acid for their individual identification. The Body weight and food intake of experimental models were recorded at regular intervals.

Group A : Control group

Group B : Animals with animal fat

Group C : Animals with vegetables fat

Rations containing 6% of vegetable or animal fat were fed to groups B and C, respectively. Group A acted as control and was fed standard diet (Hindustan liver Ltd.).

SERUM TOTAL CHOLESTEROL, TRIGLYCERIDE AND PHOSPHOLIPIDS: Serum cholesterol values of rats from each group were determined on 42nd day of age through Libermann Burchard Reaction [4] and Triglycerides was examined by enzymatic Method, using a commercial kit [5]. Phospholipid was estimated by analytical method [6]. The body weight (initial and final) vital organs (liver and heart) were examined.

Blood Sample Collection: The animals in all groups were kept for 4-5h fasting periods before collections of blood samples by retro-orbital bleeding. The blood was collected through heparinised capillaries into heparinised vials and in EDTA (an anticoagulant, 2mg ml^{-1}) containing tubes and without EDTA tubes. The collected blood samples were centrifuged for 10min at 5000rpm and at 4°C. The serum were collected and stored at 4°C for further investigation.

Histopathological Examinations: Six rats from each group were slaughtered on day 42 and liver samples were collected. The livers were examined to record any gross changes. Organs showing significant gross lesions were processed and examined for histopathological changes [7].

RESULTS AND DISCUSSION: Our results indicated that serum total cholesterol values in rats fed on fat either from animal (165 ± 8.4 mg/100 ml) or vegetable origin 136±8.2mg/100 ml) were higher ($p<0.05$) than that of control 80±5.1mg/100ml). Triglyceride content was recorded in group B 178.10±4.11 whereas the group C animal was shown 149.13±4.41mg/100ml triglyceride concentration respectively (**Table 1**). Our findings are in line the kidney function test urea and cholesterol were increased in rats fed on 15% sheabutter cake or kernal cake.8-10 the serum cholesterol value were significantly higher ($p<0.05$) in rats fed on animal fat as compared to those fed on vegetable fat. The triglyceride concentrations were also increased with fats based diet administration [9, 11].

The body weights were increased significantly in rats fed with animal fat and vegetable fat as compared to normal control animal subjects. The Group B with animal fat gain higher weight 1.98±0.11 as compared to group C 1.83±0.13/kg (**Table 2**). The results indicate that fat based diet directly acts on the vital organs of the body to after the metabolism and play a role in enhancement of body weight. the vital organs liver and heart weight were also increased in group B and C.

Animal fat receiving group B gain higher weight of liver 4201±4.3 and heart 245 ±4.1mg/100gbwt, whereas no any significant result accounted in group C heart 198±3.8, liver 198 ±3.8mg/100ml (**Table 3**). Data were justified, these excess weight responsible for development of fatty liver and accumulation of fats bodies in heart muscles and liver tissues.

TABLE : 1 IMPACTS OF ANIMAL AND VEGETABLE FATS ON LIPID PROFILES.

Experimental groups	Total Cholesterol mg/100ml)	Triglyceride (mg/100ml)
Group A (Control group)	80 ± 8.4	62.12 ± 4.30
Group B (Animal fat fed)	165 ± 8.2	178.10 ± 4.11
Group C (Vegetable fat fed)	136 ± 5.1	149.13 ± 4.41

a *P ≤ 0.001* when group B compared with group C. The values represents Mean±SEM

TABLE 2. CHANGES IN THE BODY, LIVER AND HEART WEIGHT AFTER FEEDING OF ANIMAL AND VEGETABLE FATS.

	Body weight/kg		Organs weight mg/100gbwt.	
Experimental groups	Initial	Final	Liver	Heart
Group A (Control group)	1.60±0.09	1.62±0.84	2603±5.6	230 ±5.5
Group B(Animal fat fed)	1.66±0.01	1.98±0.11	4201±4.3	245 ±4.1
Group C (Vegetable fat fed)	1.49±0.01	1.83±0.13	2661±2.1	198 ±3.8

P: significant when Group B is compared with Group C. *b P* 0·01 when Group B is compared with Group C. *P* 0·001 when Group C is compared with Group B. The values represent Mean ± SEM.

TABLE 3: CHANGES IN TISSUE LIPID PROFILES AFTER FEEDING OF ANIMAL AND VEGETABLE FAT.

	Cholesterol (mg/gm)		Triglyceride (mg/gm)		Phospholipids (mg/gm)	
Experimental groups	Liver	Heart muscles	Liver	Heart muscles	Liver	Heart muscles
Group A (Control group)	11.93 ± 0.14	9.12 ± 0.24	4.96 ±0.05	4.37 ±0.13	9.32 ±0.06	12.0 ±0.03
Group B (Animal fat fed)	29.5 ± 0.21	24.25 ± 0.21	9.90 ±0.18	9.0 ±0.06	15.0 ±0.04	17.5 ±0.30
Group C (Vegetable fat fed)	19.34 ± 0.04	15.05 ± 0.04	6.34 ±0.04	7.2 ±0.07	11.25 ±0.03	11.71 ±0.41

p: is significant when group B compared with group C. c ≤ p 0.01, b ≤ p 0.001, The values represent Mean ± SEM.

The histopathological examination of liver marked macroscopic and microscopic lesions with both types of fats groups. Grossly, the livers of both groups B and C were enlarged in size, dark pale in colour, soft in consistency and were having petechial haemorrhages with deposition of fat and fibrin on the liver. Livers of the control group were grossly normal.the consumption of fish oil resulted yellowish pale and swollen [12]. Liver weight; liver fat content and haemorrhagic scores were higher in group fed high-energy low protein diet [13].

Histopathologically, liver of the groups B and C showed fatty infiltration, haemorrhages and mass of eosinophilic materials. The vacuoles coalesced to create clear space that displaced the nucleus to the periphery of the cell. There were many ruptured hepatocytes, with enclosed fat globules, which coalesced to produce so called fatty cysts [13]. Fatty liver syndrome induced by feeding of high energy-low protein diets and observed distinct histopathological fatty changes in the liver [14, 15].

Livers of the control group were microscopically normal. From the above discussion, it can be concluded that addition of dietary fat from animal and vegetable sources in the diet of rats not only resulted in increases body weight, serum total cholesterol and triglyceride concentration but also caused marked macroscopic and microscopic changes in liver as well as physiologically alter the heart muscles.

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