ORIGINAL ARTICLE

Portal Triad Changes in Liver Associated with Rapeseed Oil Diet Supplemented with Atherogenic Element: an Experimental Study

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ABSTRACT

This experimental study was designed to see the possible effects of rapeseed oil on liver morphology. For this purpose, 60 albino rats of eight weeks age were selected and divided into five groups of twelve animals each with equal number of males and females. Group I, normal control, was fed on synthetic diet, Group II was on low rapeseed oil diet only and group III was on low rapeseed + hypercholesterolemic diet. Group IV was on high rapeseed oil diet only and Group V was on high rapeseed oil + hypercholesterolemic diet for the next 24 weeks. Histological examination was done on H&E, reticulin, trichrome and oil red O stains. Liver revealed mild to severe type of Portal triad fibrosis in groups II, III, IV and V. These findings were statistically significant when compared with control Group I. While bile duct proliferation(BDP) was mild in Group II and IV. Hence it is concluded that Rapeseed oil proved to be beneficial diet. It causes mild hepatic injury in low and high concentration. Hypercholesterolemic diet causes moderate to severe morphological changes in liver.

Key Words : Rapeseed Oil, Liver, athero sclerosis.

INTRODUCTION

Fats are composed of mostly long chain saturated fatty acids for example, palmitic acid, stearic acid and exist in solid or semi solid state at 37^0C. They are mostly derived from the animal sources. Oils contain fatty acids that are mostly unsaturated and exist in liquid form. Monounsaturated fats contain palmitoleic acid and oleic acid[1]. Oleic acid is present in olive oil and rapeseed oil (canola oil). Polyunsaturated fatty acids (PUFA) for example, linoleic acid, are present in edible oils like corn oil, soya oil, sunflower oil, cotton seed oil and palm oil[2,3].

Olive oil, comprising of high contents of monounsaturated fatty acids, significantly reduces serum cholesterol[4].

Polyunsaturated fatty acids are susceptible to oxidation. LDL oxidation appears to be necessary for LDL uptake by macrophages. Monounsaturated fatty acids (MUFA) prevent LDL oxidation[5]. Anti-oxidant supplements can protect cellular structure against oxidative stress and lipid peroxidation[6].

In an experimental model, increase in hepatic total cholesterol was observed when dietary lipids levels were increased from 12% to 20 % while protein levels were maintained at 30%[7,8]. High fat diet increases responsiveness of Ito cells (Hepatic stellate cells, fat storing cells) of the liver, leading to proliferation of Ito cells, hyperplasia of rough endoplasmic reticulum and increased collagen synthesis[9].

Hypertriglyceridemia appeared to be a cause of steatosis in man[9]. Free fatty acids are highly reactive and damage biological membranes. Hepatic peroxidation of lipids may result in generation of potentially toxic intermediates, that can induce an inflammatory response in liver[10]. Non-alcoholic steatohepatitis and fatty liver Cirrhosis occurs as a "two hit theory" in which hepatic steatosis is followed by lipid peroxidation, the production of cytokines and the induction of Fas ligand[11].

METHODOLOGY

Sixty albino rats of 8 weeks age, including equal number of males and females were taken for this study from Veterinary Research Institute, Ghazi Road, Lahore. The animals were randomly divided into three groups of twelve rats each. The animals were kept in separate cages according to sex, in the animal house of Postgraduate Medical Institute, 6 Abdul Rehman Chugtai Road, Lahore. They were numbered, acclimatized and kept in same room at 24 ± 2^0C, with food and water available all the time[12].

Group I animals were given synthetic diet for next 24 weeks. Rest of the rats (GroupsII, III, IV, and V) were weighed and fed on experimental diet for a total period of 24 weeks[13].

Animal Grouping: Group I, was given synthetic diet. This was the control group.

Group II was given low rapeseed oil diet only.

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Group III was given low rapeseed oil diet supplemented with atherogenic element.
Group IV was given high rapeseed oil diet only.
Group V was given high rapeseed oil diet supplemented with atherogenic element.

Specimen Collection: The rats of group II, III, IV, V and VI were scarified at the end of 24 weeks. They were dissected, and livers excised. The biopsy specimens were kept in the labelled jars for fixation with 10 % buffered formalin[14], processed routinely and stained with H&E, reticulin, trichrome and Oil red O stains.

RESULTS

Microscopic examination of rats livers regarding portal triad changes in Group II showed mild bile duct proliferation as compared to control group and difference was significant statistically (p=0.05).. Group III, IV, and showed non-significant bile duct changes when compared with control. Group II, III, IV and V showed significant portal triad fibrosis when compared with control group (P=0.00014, 0.0043, 0.00062 and p=0.00016).The detail of results are given in tables 1, 2 ,3 and 4.

Table 1: Comparison of portal triad changes in group I & II

Microscopic Feature	No. of animals with positive changes		P-values (I Vs II)
	Group I	Group II	
BDP	01	05	(0.05)
Portal triad fibrosis	00	08	0.00014
Portal triad inflammation	02	03	0.5 (NS)

Table 2: Comparison of portal triad changes in group I & III

Microscopic Feature	No. of animals with positive changes		P-values (I Vs III)
	Group I	GroupIII	
BDP	01	01	0.000 (NS)
Portal triad fibrosis	00	04	0.0043
Portal triad inflammation	02	00	0.5 (NS)

Table 3: Comparison of portal triad changes in group I & IV

Microscopic Feature	No. of animals with positive changes		P-values (I Vs IV)
	Group I	Group IV	
BDP	01	03	0.51 (NS)
Portal triad fibrosis	00	07	0.00062
Portal triad inflammation	02	02	0.5 (NS)

Table 4: Comparison of portal triad changes in group I & V

Microscopic Feature	No. of animals with positive changes		P-values (I Vs V)
	Group I	Group V	
BDP	01	00	0.51 (NS)
Portal triad fibrosis	00	12	0.00016
Portal triad inflammation	02	01	0.5 (NS)

DISCUSSION

Control Group I (Synthetic Diet): The animals of group I were sacrified after 24 week of Synthetic diet and livers were dissected out to see the morphological changes for comparison with experimental groups. The gross and microscopic examination revealed normal morphology.

Experimental group II (low rapeseed oil diet): The animals fed on experimental diet with low rapeseed oil were sacrified after 24 weeks. No gross changes were seen in the livers of albino rats. The microscopic findings regarding BDP and portal triad fibrosis were significant when compared with animals of control group (II). (p=0.05 and p=0.00014 respectively). These mild changes were consistent with the results of Burgos et al (1993) [7], Boshnakova et al (1994) [15], Cullen et al (1996) [16] and Fernnandez et al (1997) [17], who also observed significant morphological changes in liver of albino rats.

Experimental Group III (low rapeseed oil with Hypercholesterolemic Diet): Gross features of liver showed increase in size and weight with yellowish discoloration. Microscopic features revealed mild to moderate type of portal triad fibrosis (p= 0.0043). The difference was statistically significant in above mentioned morphological parameters. These findings are in agreement with the results of Boshnokova et al (1994)[15], Fernandez et al (1997)[17] and Puiggros et al (2002)[4].

Experimental group IV (high rapeseed oil diet): No gross morphological changes were seen in any of the albino rats liver fed on high rapeseed oil diet. Microscopic examination revealed that the number of lesions appeared in group IV are mild in nature and are statistically significant as compared to control group (I) (p=0.00062 for portal triad fibrosis). These findings are in agreement with the results of Husveth et al (2000)[18], Kratz et al (2002)[19] and Puiggros et al (2002)[4].

Experimental group V (high rapeseed oil with hypercholesterolemic element): Gross features showed marked increase in size and weight of liver with marked yellowish discoloration. Microscopic examination revealed that the number of lesions appeared in group V were moderate to severe in

nature and difference was highly significant as compared to control group (II) (p=0.00016 for portal triad fibrosis). These results are in favour of the findings of Husveth et al (2000)[18] and Puiggros et al (2002) [4], who also observed moderate to severe type of morphological changes in liver of albino rats.

CONCLUSION

Hence it is concluded that Rapeseed oil proved to be beneficial diet. It causes mild hepatic injury in low and high concentration. Hypercholesterolemic diet causes moderate to severe morphological changes in liver.

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