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HYPOCHOLESTEMIC ACTIVITY OF SEA SQUID FROM KARACHI COAST

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Studies have been carried out to determine the cholesterol content of sea squid and the effect of squid meat diet on the cholesterol level of rabbit serum. The cholesterol in five species of sea squid varied between 215 and 349 mg/100 g of raw tissue. The cholesterol in rabbit serum after feeding on diet containing sea squid meal ranged from 98.26-188.91 mg/100 ml of serum compared to control group with a value of 185.01-188.12 mg/100 ml of serum.

Key words: Squid, Hypocholestemic activity, Rabbit.

Introduction

Coronary heart disease (CHD), high cholesterol diet and high blood cholesterol level are some well known major problems of the world today [1]. Experiments in animals have clearly shown that high cholesterol cause damage to arteries and lead to CHD. There is an urgent need therefore, to give attention towards the therapeutic effects of marine foods which even though are reported to contain a high level of cholesterol in their lipid exert a hypocholestemic effect. Detailed information is required on the cholesterol of sea squid and other shellfish species. In Pakistan, people are generally unaware of the cholesterol contents of marine products and the effect of consumption of these products on the blood cholesterol. High values of cholesterol have been reported in shellfish i.e. 47 mg/100 g in oyster and 40 mg/100 g in cockers [2]. In squid *(Loligo duvucelli)* cholesterol has been reported to represent more than 90% of the total sterol content [3] ranging from 250-400 mg/100 g. This paper reports the cholesterol content of 5 species of sea squid found in Pakistan coastal waters and the effect of sea squid feeding on blood serum of rabbits.

Materials and Methods

Samples of sea squid were purchased from the commercial sources. Sampling of squid was carried out in collaboration of Marine Fisheries Department.

Extraction and estimation of cholesterol. Fresh squid (350 specimen) were dried in oven at 80° and ground upto 30 mesh size, this dry matter was stored at room temperature (20-30°) and used for the extraction of fat. The collective fat of each species was used for the extraction of cholesterol using (1:3) alcohol-ether mixture [4]. The extract was evaporated at 40-50° to dryness and the residue was dissolved in chloroform. The cholesterol was estimated using the Liberman-Burchard reaction [5] by standard graph prepared with known amounts of cholesterol.

Feeding experiments on rabbit. Healthy rabbits weighing 1-1.5 kg each, were purchased from a local animal supplier and housed in separate cages for a week and fed on basal diet (carrot, gram and leucine). Rabbits were divided into five groups comprised of two animals each. Three experimental group animals were fed with the test squid meal diet as follow: Squid meal was mixed with 50 kg basal diet (carrot, gram and leucine) at a range of 2, 4 and 6 g/kg of body weight of the experimental animal groups I, II and III respectively, Lopid (Gemfibrozil), Parke-Davis [6,7] (Dosage 1200 mg in two divided dosage/day for man) was mixed with basal diet at a range of 50 mg/kg of body weight of the standard animal group IV. The control group was maintained on the basal diet only (carrot, gram and leucine). The animals were fed daily for 6 weeks. Blood samples were taken from the ear veins of rabbits of each test, standard and control groups. The cholesterol level in the blood of experimental, standard and control animals was determined at weekly intervals.

Result and Discussion

The cholesterol content of five species of sea squid is given in Table 1 and those in blood serum of rabbits in Table 2, representing experimental, standard and control rabbits. It may be noted (Table 1) that large variation was found in the cholesterol content in different species examined. It varied from 215 ± 9 mg/100 g in *S. phoraonis* to 349 ± 20 in *L. duvucelli*. The other three species *Sepiella inermis, Sepia prashadi and Dosidicus gigas* were found to contain 260 ± 6 mg/100g, 279 ± 8 mg/100g and 300 ± 10 mg/100g wet tissue respectively. The large range of cholesterol found in squid meal will make it a difficult seafood to be categorized into low cholesterol diet particularly when the upper range is around 349 mg cholesterol/100g of wet tissue. (Table 1).

The feeding squid meal was comprised of a mixture of all the five species in equal proportions. The composite sample had an average cholesterol content of 280.60 mg/100 g. It is

evident from Table 2 that during a period of six weeks, feeding squid diet resulted in a gradual decrease in cholesterol in rabbit serum upto 2 weeks in group I and II and 3 weeks in group III reaching a level of 98.26 mg/100 g lower than the level of cholesterol (178.10 mg) obtained for standard (group IV). The cholesterol in squid diet fed animals increased thereafter gradually and sharply attaining more or less initial levels (0 hr). No relationship was obtained between the increase in squid meal in the diet and the cholesterol increase in the blood. Irrespective of cholesterol content of feed (having 215-349 mg/ 100 g dry matter), there was an initial hypercholesterolemic condition in all the test animals. No hypercholesterolemic condition has been reported in rabbits fed on plant material such as soyabean protein isolate having lipid up to 20.75% [8]. The standard group (IV) was fed with cholesterol reducing diet and as expected, the serum cholesterol decreased gradually throughout the feeding period. No difference in serum cholesterol levels was observed in control (group V), which remained almost constant throughout the experimental period. No initial decrease as noticed in squid fed groups was found in this case. No difference in the body weight of experimental animal was noted due to difference in the cholesterol content of diet used.

Cholesterol is necessary for the production of harmones in animals and perhaps there is great physiological demand for cholesterol during the period of the rapid maturation process. Rabbits fed on a low cholesterol semisynthetic diet have been reported to become hypercholesterolemic [9].

It has also been reported [10,11] that the ingestion of food having high cholesterol for a considerable time may result in an increase in blood cholesterol, a single high cholesterol diet does not result in such an increase. In rabbits, excessive feeding with cholesterol has been reported to cause arteriosclerosis [12]. The cholesterol was found to vary between 65-75% of the total serum or plasma sterol for which a normal value ranges from 80-300-400 mg/100 ml [13]. The decrease in the cholesterol level in animals on high cholesterol diet appears a unique phenomenon and needs further studies to confirm. A possible explanation may be as follow:

As may be expected, feeding squid meal having high cholesterol should result in the increase in blood cholesterol levels. However the cholesterol in the blood of test animals started decreasing upto 2-3 weeks of feeding and thereafter increased gradually. This may be attributed to the feed back mechanism within the body of rabbits.

Another factor should also be considered: squid lipid together with high cholesterol content is also rich in polyunsaturated fatty acids (PUFA) particularly the eicosapentaenoic acids (EPA) and docosahexaenoic acids (DHA) which have been shown to be an important link to reduce cholesterol and the risk of harmful fatty deposits from building up in the arteries.

TABLE 1. CHOLESTEROL CONTENT OF SQUID AND OTHER SHELLFISHERIES.

S. No.	Species	No. of sample	Cholesterol (mg/100g)
1.	Squid *(Loligo duvucelli)*	10	349 ± 20
2.	Squid *(Dosidicus gigas)*	8	300 ± 10
3.	Squid *(Sepiella inermis)*	5	260 ± 6
4.	Squid *(Sepia prashadi)*	3	279 ± 8
5.	Squid *(Sepia phoraonis)*	4	215 ± 9
6.	Lobster *(Panulirus polyphagus)*	8	371.25-553.5
7.	Shrimp *(Penaeus merguiensis)*	12	157.5-367.5
8.	Crab *(Syclla serrata)*	6	187.08-233.0

Species 6 to 8. (unpublished data).

TABLE 2. EFFECT OF SEA SQUID MEAL ON THE SERUM CHOLESTEROL (mg/100 ml) IN RABBITS.

	Weeks						
Groups	0	1	2	3	4	5	6
Group I	183.80	171.25	101.2	154.53	164.66	188.56	188.91
Group II	188.80	181.82	116.01	132.96	161.62	181.62	184.92
Group III	181.70	179.55	99.64	98.26	109.18	188.0	188.10
Group IV (Standard)	187.61	182.90	180.20	178.10	165.10	160.71	158.20
Control	185.01	188.0	186.71	188.12	187.19	188.10	187.9

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