HYPOCHOLESTEROLEMIC EFFECTS OF FISH AND VEGETABLE OILS ON THE SERUM LIPID PROFILE OF EXPERIMENTALLY INDUCED HYPERCHOLESTEROLEMIC RATS

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Abstract

Aims: This study was conducted to find out the effect of fish and vegetable oils on the serum lipid profile of experimentally induced hypercholesterolemic rats. **Methods:** Long Evan rats were fed with cholic acid and cholesterol for 14 days to make them hypercholesterolemic. The hypercholesterolemic rats were then supplemented with the oils of Pangsius pangasius fish, Soybean, Flaxseed, Sesame, Black seed and Mustard for 35 days. Serum lipid profile (total cholesterol-TC, low density lipoprotein-LDL, high density lipoprotein-HDL and triglyceride-TG) was determined with ELISA plate reader using commercial kits. **Results:** All of the oils tested were noted to reduce the serum TC, LDL and TG, but had increase the good cholesterol HDL level in the hypercholesterolemic rats. Among these oils, Pangsius pangasius fish oil showed the strongest anticholesterol property. In reduction of total cholesterol, after the Pangsius fish oil, the next prominent activity was given by black seed oil followed by flaxseed oil, sesame oil, mustard oil and soybean oil and in term of lowering of LDL level, the effect was in order of black seed, flaxseed, mustard, sesame and soybean oil. In giving the effect on good cholesterol HDL level, mustard oil showed the strongest increasing effect, which was followed by black seed oil, flaxseed oil, sesame oil, Pangsius oil and soybean oil. The drop of triglyceride level by the tested oils, it was in line of Pangsius oil, sesame oil, black seed oil, flaxseed oil, soybean oil and mustard oil. **Conclusion:** The present study showed that both the fish and vegetable oils have significant anticholesterol effects on hypercholesterolemic rats. However, compared to the vegetable oil, fish oil has stronger effect on the blood lipid profile. Regular dietary intake of fish and vegetable oils would reduce the risk of cardiovascular complication.

Keyword: Fish oil, Vegetable oil and Cardiac complications

INTRODUCTION

Cardiovascular disease is one of the major health problems in the world. It is dramatically increasing in the last 10 years (Yamada M et al., 1997). Blood lipid profile determines the risk of cardiac disease. Lipid profile includes total cholesterol (TC), high density lipoprotein cholesterol (HDL-C) often called good cholesterol, low density lipoprotein cholesterol (LDL-C) often called bad cholesterol and triglycerides (TG). An extended profile may also include very low density lipoprotein cholesterol (VLDL-C) and Non-HDL-C.

Cardiac complication is a lifestyle disease. The major cause of this disease is due to high intake of cholesterol containing foods. Dietary saturated fatty acid increase blood cholesterol level, while polyunsaturated fatty acids (PUFAs) reduce blood cholesterol. It has been documented that fish and vegetable oils containing polyunsaturated fatty acids (PUFAs) reduce blood lipid profile. Fish oil containing ω-3 PUFAs is much more effective than the vegetable oil (ω-6 PUFAs) in reduction of lipid profile in human (Endres S et al., 1995; Iwata T et al., 1992; Von Shacky C 1992; Simopoulos AP 1997; Harris WS 1989). Epidemiological studies show that eating fish or vegetable oil concurrently decreases blood cholesterol and LDL levels and increases HDL, and thus reduces the risk of coronary death (Yamori Y et al., 1985; Dyeberg J et al., 1975). It is also shown that high concentrations of HDL (over 60 mg/dl) have protective effect against cardiovascular diseases such as ischemic stroke and myocardial infarction but higher LDL promotes cardiovascular disease. Very low LDL increases the risk of cardiovascular disease, if their HDL level is not high (Barter et al., 2007). The present study has made an attempt to find out the effect of lipid lowering effect of some selected fish and vegetable oils on experimentally induced hypercholesterolemic rats.

MATERIALS AND METHODS

Collection of fish and vegetable oil:

Pangsius pangasius fish oil was collected from the fish abdomen and kept under nitrogen until it was used. Flaxseed, Mustard, Soybean, Sesameand Black seed oils were purchased from the “Bangladesh Standard Testing Institute (BSTI)” Dhaka.

Design of experimental animal model:

Thirty-five adult Long Evan rats (100-120g) were procured from the Animal House of International Centre for Diarrheal Disease and Research, Bangladesh (ICDDRB), Dhaka, Bangladesh. They were housed at standard environmental conditions of temperature and dark/light cycle, and were fed with commercial pellet diet and drinking water for 7 days. Baseline serum total cholesterol was measured by tail bleeding using commercial kit. All of the rats were then start feeding with 1.5% cholesterol and 0.5% cholic acid along with the pellet diet and water for 14 days to make them hypercholesterolemic. On the 15th day lipid profile (TC, LDL, HDL, TG) of the hypercholesterolemic rats were determined with ELISA plate reader using commercial kits.

The hypercholesterolemic rats were then divided into seven groups- 1-6 experimental groups for six test oils and the group 7 for hypercholesterolemic control. The animal of experimental group was fed orally with respective test oil for 35 days along with pellet diet and water. The control rats received the normal diet and water only. The supplemented oil dose was 50ml/kg body weight.

The experiment was designed as

Animal group	Test material	Dose	Diet	Duration
Group-1	Pangsius fish oil	50ml/kg body weight	Pellet diet and water	35 days
Group-2	Mustard oil			
Group-3	Flaxseed oil			
Group-4	Black seed oil			
Group-5	Sesame oil			
Group-6	Soybean oil			
Hypercholesterolemic control	-			

On the 35th day, rats of each group were individually anesthetized with diethyl ether and blood was collected from the heart and processed for extraction of serum. The sera were kept at -20^{o}C for analysis of lipid profile. Ethical guideline was maintained in animal handling during the study and permission was obtained from the concerned department.

Serum analysis

Serum lipid profile was determined with the use commercial kits. Total cholesterol was analyzed enzymatically with CHOD/PAP method(Kaplan L.A. et al. 1989, Norbert W. et al., 1987) and HDL was estimated with precipitation (Naito H K. 1984; Grove T H. 1979) using commercial kits (Cam Tech Medical, UK). Triglyceride was determined enzymatically also using by commercially available kit (Audit Diagnostics, Ireland). Serum LDL was determined according to the Friedewald formula with use of HDL and total cholesterol value. ELISA plate reader (Labsystems, MultiskanEX, Finland) was used to analyze the lipid profile.

Statistically Analysis

SPSS software package (version 20.0, IBM SPSS Inc. Chicago, USA) was used to analyze the data. Descriptive statistics were calculated for all variables. Independent sample t-test was performed to values of lipid profile for their different test oils. Level of significance was expressed as $p<0.05$.

RESULTS

Supplementing Pangsius fish oil and five vegetable oils to experimentally induced hypercholesteroemic rats (table 1); it was resulted in that all of the tested oils showed strong cholesterol lowering effect on the hypercholesteroemic rats. The TC, LDL, HDL and TG profile of the hypercholesteroemic rats were 236.34±16.35, 184.76±16.14, 24.54±1.04 and 135.18±2.21mg/dl respectively. Supplementing the oils of Pangsius fish, Mustard, Flaxseed, Blackseed, Sesame and Soybean for 35days to the hypercholesteroemic rats, serum total cholesterol (TC) was reduced to 95.28±2.07, 120.23±2.62, 109.86±2.99, 106.28±1.47, 116.57±1.61 and 129.12±1.57mg/dl respectively; low density lipoprotein (LDL) was decreased to 49.22±3.18, 56.28±3.41, 54.97±2.66, 51.92±2.07, 64.95±1.55 and 80.12±2.19mg/dl respectively; high density lipoprotein (HDL) was increased to 29.08±3.52, 34.98±1.18, 31.57±0.91, 32.27±0.63, 31.15±0.68 and 25.34±0.77mg/dl; and the triglyceride (TG) was lowered to 84.88±2.38, 144.82±1.67, 116.60±1.41, 110.41±4.85, 102.34±0.89 and 118.29±0.92mg/dl respectively.

DISCUSSION

High blood lipid profile is the risk of cardiovascular diseases (CVD). Hyperlipidemia remains the strongest risk factor for CVD. Prevention of CVD risk factors such as obesity and dyslipidemia has been an important challenge in developing countries. CVD is a major health problem in the world resulting in premature morbidity and mortality. Over 80% of CVD deaths take place every year in the developing countries like Bangladesh (BBS, 2009). By 2015, almost 20 million people may endure from death due to CVDs, mainly heart disease and stroke. High intake of cholesterol induces multiple cardiac complications including coronary heart disease and stroke (Haddad FH et al., 2002).

The Cardiovascular disease is a lifestyle disease. The major risk factors for CVD are high cholesterol diet, physical inactivity, and tobacco use obesity. The risk from high cholesterol is not immediate may not affect today or tomorrow. The damage accumulates over years even decades. If care is not taken, it can have a terrible cost resulting series chronic health disorders, predominantly cardiac complication. A total cholesterol reading can be used to assess an individual's risk for heart diseases; however, it should not be relied upon as the only indicator. The individual components that make up total cholesterol reading – LDL, HDL, TG, even VLDL are also important in measuring risk of CVD.

Currently a number of anticholesterol drugs like atorvastatin, fluvastatin, lovastatin, pravastatin, rosuvastatin, simvastatin are using to reduce the blood cholesterol level. But these drugs have wide range of toxic side effects including hepatotoxicity, mitotoxicicty. If these drugs can be replaced by natural products/foods that can reduce the cholesterol level, it will be better than the drugs. Natural products/foods, that have apparently no side effect, are the choice as anticholesterol agents. Eating healthy diet and doing physical activity or exercise can outsmart high cholesterol. It is fish and vegetable oils containing PUFAs that can reduce the blood cholesterol. This study has, therefore made an attempt to experiment the lipid lowering effect of selected fish and vegetable oils on hypercholesterolemic rat model.

All of the oils tested were noted to reduce the serum TC, LDL and TG, but had increase the good cholesterol HDL level in the hypercholesterolemic rats. Among these oils, Pangsius pangasius fish oil showed the strongest anticholesterol property. The stronger anticholesterol activity of fish oil is because of ω-3 PUFAs, while vegetable oil contains ω-6 PUFAs, which is less active in lowering of blood lipid profile. In reduction of total cholesterol, after the Pangsius fish oil, the next prominent activity was given by black seed oil followed by flaxseed oil, sesame oil, mustard oil and soybean oil and in term of lowering of LDL level, the effect was in order of black seed, flaxseed, mustard, sesame and soybean oil. In giving the effect on good cholesterol HDL level, mustard oil showed the strongest increasing effect, which was followed by black seed oil, flaxseed oil, sesame oil, Pangsius oil and soybean oil. The drop of triglyceride level by the tested oils, it was in line of Pangsius oil, sesame oil, black seed oil, flaxseed oil, soybean oil and mustard oil.

The finding of this study was found consistent as reported by others. Nicolaysen and Regard (Nicolaysen R and Regard R. 1961) reported that dietary marine fish oil reduced blood cholesterol in the experimentally induced hypercholesterolemic rats. Recently, anticholesterolemic effects of Hilsha fish oil were also reported in streptozotocin-treated diabetic rats (Mahmud I et al., 2004). Fish oil containing ω-3 PUFA is found to inhibit the activity of 3-hydroxy-3-methyl-glutaryl-CoA reductase (HMG-CoA reductase), which is the rate-limiting enzyme in cholesterol biosynthesis(Ide T et al., 1980). Kobatake et al. reported that docosahexaenoic acid (ω-6

PUFA) is more effective in lowering serum cholesterol level of experimentally induced hypercholesterolemic (Kobatake Y et al. 1984).

CONCLUSION

Based on the present experimental results, it can be suggested that both the fish and vegetable oils have strong and significant anticholesterol effects on hypercholesterolemic rats. However, compared to the vegetable oil, fish oil has stronger effect on the blood lipid profile. Regular dietary intake of fish and vegetable oils would reduce the risk of cardiovascular complication.

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Table-1: Effects of fish and vegetable oils on the serum lipid profile (mg/dl) of experimentally induced hypercholesterolemic rat*

Treatment	TC[a]	LDL[b]	HDL[c]	TG[d]
Pangash oil[1]	95.28±2.07	49.22±3.18	29.08±3.52	84.88±2.38
Mustard oil[2]	120.23±2.62	56.28±3.41	34.98±1.18	144.82±1.67
Flaxseed oil[3]	109.86±2.99	54.97±2.66	31.57±0.91	116.60±1.41
Black seed oil[4]	106.28±1.47	51.92±2.07	32.27±0.63	110.41±4.85
Sesame oil[5]	116.57±1.61	64.95±1.55	31.15±0.68	102.34±0.89
Soybean oil[6]	129.12±1.57	80.12±2.19	25.34±0.77	118.29±0.92
Highpercholesterolemic[7]	236.34±16.35	184.76±16.14	24.54±1.04	135.18±2.21

Level of significance: $p<0.05$ *Dose: ml/kg body weight

Data's are expressed as mean ± SD (n= 5). Values in the same column that do not share a common superscript are significantly different ($P < 0.05$). TC = total cholesterol; LDL-C = low density lipoprotein-cholesterol; HDL-C = high density lipoprotein-cholesterol; TG = Triglyceride; Highpercholesterolemic= Cholesterol 1.5% + 0.5% Cholic acid-fed rats.

a [1,7]:	t = 19.13	p = .000	b [1,7]:	t = 18.42	p = .000	c [1,7]:	t = 2.76	p = .025	d [1,7]:	t = 34.68	p = .000
a [2,7]:	t = 15.67	p = .000	b [2,7]:	t = 17.41	p = .000	c [2,7]:	t = 14.79	p = .000	d [2,7]:	t = 7.79	p = .000
a [3,7]:	t = 17.01	p = .000	b [3,7]:	t = 17.73	p = .000	c [3,7]:	t = 11.36	p = .000	d [3,7]:	t = 15.89	p = .000
a [4,7]:	t = 17.71	p = .000	b [4,7]:	t = 18.25	p = .000	c [4,7]:	t = 14.23	p = .000	d [4,7]:	t = 10.40	p = .000
a [5,7]:	t = 16.30	p = .000	b [5,7]:	t = 16.51	p = .000	c [5,7]:	t = 11.87	p = .000	d [5,7]:	t = 30.86	p = .000
a [6,7]:	t = 14.59	p = .000	b [6,7]:	t = 14.35	p = .000	c [6,7]:	t = 1.38	p = .205	d [6,7]:	t = 15.80	p = .000