Research Article

Impact of fish on biochemical indicators: A comparative analysis among female subjects of fish eaters and vegans

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ABSTRACT

Introduction and Aim: In India, cardiovascular diseases (CVD) contribute to a major cause of morbidity and mortality. Consuming more or significant quantity of fish has been linked to a lower the vulnerability of CVD. This is due to fish oils contains long-chain n-3 polyunsaturated fatty acids (PUFA), like docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). Frequent intake of these fatty acids has demonstrated to reduce the vulnerability of endothelial dysfunction, inflammation, arrhythmias, heart failure, myocardial infarction, and sudden death. The protective advantages of a diet high in fish seems to be at odds with the oxidation susceptibility of polyunsaturated fats (PUFAs). Therefore, the goal of the research was to assess the lipid profile, cardiac markers, and state of lipid peroxidation in adult females who regularly eat substantial quantities of fish.

Materials and Methods: Measurements were made of the following in healthy female individuals: total cholesterol (TC), HDL-cholesterol (HDL-C), triglycerides (TG), high sensitive C-reactive protein (hs-CRP), lipoprotein (a) [Lp(a)], malondialdehyde (MDA), and antioxidant activity (AOA). The atherogenic indices TC/ HDL-C, LDL-C/HDL-C and non- HDL-C/ HDL-C were determined. Applying Student's "t" test, the parameters of the two groups were compared.

Results: Among female individuals who consumed fish, HDL-C was the only cardiac marker that increased while all other cardiac markers and the lipid profile decreased. The atherogenic indices of fish eaters showed considerable declines, whereas the oxidant status between the study groups did not vary significantly ($p>0.05$).

Conclusion: The outcome of this study reveals that, without influencing an individual's oxidation status, a frequent consumption of fish reduces the lipid profile and cardiac markers.

Keywords: Fish eaters; vegetarians; lipid profile; hs-CRP; Lp(a); cardiovascular disease.

INTRODUCTION

Heart failure and stroke under cardiovascular diseases (CVD) are the world's leading cause of death and key elements which contributes to poor quality of life (1, 2). In India, age standardized death rate is 272 per 100000 populations, which is higher than global average of 235 per 100000 populations (3). Based to the WHO's 2014 "Global Status Report on Non-Communicable Diseases," the number of fatalities from non-communicable diseases is expected to rise from 38 million to 52 million by 2030, with approximately 23 million deaths possibly arising from cardiovascular disorders alone (4).

Obesity, hyperlipidemia, type 2 diabetes, hypertension, and cardiovascular disease (CVD) are among the non-communicable diet-related disorders which are prevalent in South Asians and are increasing in the Asian Indian population (5). In spite of the fact that the underlying pathophysiology of CVD is frequently complex, one of the main aetiological risk factors which can be altered is hyperlipidemia (6). Inflammation is another risk factor for coronary artery disease (CAD). An important part of the pathophysiology of atherosclerosis is vascular inflammation. C-reactive protein (CRP) inhibits the nitric oxide (NO) synthesis which triggers the events of atherosclerosis (7). Lipid peroxidation increases the risk of CVD by causing an imbalance between pro-oxidants and antioxidants, which leads to oxidative stress and a decrease in the body's anti-oxidant activity (AOA), which damages endothelium (8-11). The difference in CAD between premenopausal women and men of the same age suggests that endogenous sex hormones—androgens, progesterone, and/or estrogen—have an important impact on the processes leading to atherosclerosis. Oestrogen is important when it comes to preventing CAD before menopause and after menopause.

Protein, selenium, iodine, vitamin D, and n-3 polyunsaturated fatty acids (PUFAs), such as eicosapentaenoic acid (EPA; 20:5n-3) and docosahexaenoic acid (DHA; 22:6n-3), are all found in significant quantities in fish and shellfish (12). Because the benefits of the nutrients found in fish are well known, worldwide dietary regulations based on food have included the consumption of fish (13, 14). While earlier studies have emphasised on the nutritional qualities of fish, there is now evidence supporting the positive effects of eating fish as a whole, strengthen the immune system, improve cognitive function, and considerably decrease the risk of cardiovascular disease (15).

A higher prevalence of dyslipidemia is frequently linked to high chances of cardiovascular disease. This is due to the transition towards more and more westernised food, resulting in reduced consumption of fish (16, 17). Among main dietary sources of n-3 PUFAs, fish is related with an array of cardio protective benefits (18).

Hence, the current study's goal is to compare the results of consuming fish as a whole food, on the lipid profile and cardiac markers in fish eating and non fish eating females in the age group of 25-40 years.

MATERIALS AND METHODS

Selection of subjects

The study consists of 65 primarily fish-eating females and 73 vegetarian females of the age group 25-40 years. These participants were chosen from specific coastal areas in the Dakshina Kannada and Udupi districts of Karnataka.

Inclusion criteria

Vegetarians: Individuals who follow a strict lacto-vegetarian diet. Fish eaters: Individuals who eat fish at least five times a week, usually with vegetables and sometimes with other meats (the amount of fish consumed, as determined by dividing the total amount of fish cooked by the number of family members who ate it).

Exclusion criteria

Exclusions from the study were diabetes mellitus, hypertension, obesity, pregnancy, smoking, chronic renal disease, and chronic inflammatory diseases like tuberculosis and leprosy. An ethical clearance was granted by the institutional ethics committee. The investigator explained the study to the subjects and informed consent was obtained. Anthropometric data, such as height, weight, and waist circumference recorded, and assessed. To ascertain the subjects' dietary profile, a questionnaire was employed.

Sample collection

Following an 8 to 10 hour overnight fast, about 7ml of venous blood was collected from the subjects. In a plain vacutainer (red stoppered vacutainer tube without anticoagulant) about 3.5ml of whole blood was collected to measure the lipid profile, C-reactive protein (hs-CRP), lipoprotein (a) [Lp(a)], and antioxidant activity (AOA) parameters. About 3.5ml of whole blood was collected in the EDTA tube used to determine the oxidant status in RBC suspension.

Total cholesterol was estimated using the cholesterol-oxidase-peroxidase (-CHOD-PAP) end point method (19). HDL-C was measured directly by the selective inhibition end point oxidase (-CHOD-PAP) method (20). GPO-POD method was used for the estimation of triglycerides (TG) (21). VLDL-C=TG/5 formula used for the calculation of VLDL. Frieldwald' s formula: [Total cholesterol - HDL-C - (Triglycerides/5)] was used for the calculation of LDL-C (22). The cholesterol contained in the lipoprotein particles that are thought to be atherogenic is known as non-HDL cholesterol, is the difference between total cholesterol and HDL-C (19). Nephelometry method was used to determine the levels of high sensitive C-reactive protein (hs-CRP) (23). An immuno-turbidimetric method was used to measure lipoprotein (a) [Lp (a)] (24). Malondialdehyde (MDA) level was measured using a EDTA tube collected sample. The reaction product of TBA, or lipid peroxidation of red blood cells, was examined (25). The ability of the serum to prevent the synthesis of thiobarbituric acid reactive compounds (TBARS) was used to calculate total antioxidant activity (AOA) (26).

Ethical clearance for the study was obtained from the Institutional Ethics Committee. Subjects who were willing to sign the informed consent form were included in the study.

Significance of research

Intake of PUFA in the form of fish diet have the capacity to regulate atherosclerosis. The basic and clinical benefits of these supplements in the fish diet can be helpful in preventing the formation of atherosclerosis at the early age**.**

Statistical analysis

The evaluated parameters were stated as mean± SD. The differences between the parameters analysed with the help of SPSS program using Student's t test. p≤0.05 considered significant and p<0.01 is highly significant.

RESULTS

This study comprises of 65 fish eating and 73 vegetarian female subjects. In this comparative study, height and body weight were assessed. With the help of above anthropometric measurements BMI (Kg/m^2) was calculated. The BMI of female subjects those who consumed fish was 25.88 ± 4.30 kg/m^2 and in vegetarians BMI was 24.54 ± 4.57 kg/m^2 (p>0.05). The female participants' waist circumference measurements were compared using the student's 't' test. (p>0.05). The waist circumferences of female fish eaters were lesser than those of the female vegetarian participants.

All the markers had higher mean ±SD values among female individuals in the vegetarian group except HDL-C (which was decreased). The HDL-C level in female of fish eaters 54.29 ± 10.58mg/dL and 49.67 ± 8.56mg/dL in vegetarians, showed a very significant difference (p<0.01). Significant difference (p<0.05) observed in the Lp(a) parameter which was 11.38 ± 5.11 mg/dL in fish eaters and 13.34 ± 4.13 mg/dL in vegetarians. According to Table 2, Compared to female fish eaters, vegetarian female participants had significantly higher levels of all atherogenic indices. Significant increases were observed in the TC/HDL-C (p<0.01), non-HDL-C/HDL-C (p<0.01), and LDL-C/HDL-C (p<0.05) indices. The mean ±SD of the MDA value was calculated at the end of 0 hour, MDA was 3.91 ± 0.59 (μ mol/L) in fish eaters and 3.68 ± 0.52 (μ mol/L) in vegetarians (p>0.05). MDA was assessed using hydrogen peroxide as an oxidising agent after two hours and found to be 6.16 ± 0.78 (μ mol/L) in fish eaters and 5.84 ± 0.80 (μ mol/L) in vegetarians (p>0.05).

Table 1: Comparison of lipid profile and cardiac markers in female subjects of the study groups

Parameters in mg/dL	Females in the study groups	
	Fish-eaters (n=65)	Non fish-eaters (n=73)
TC	194.32 ± 35.38	200.03 ± 29.39
TG	104.26 ± 47.63	116.53 ± 46.71
HDL-C	54.29 ± 10.58	49.67 ± 8.56**
LDL-C	119.15 ± 32.75	126.41 ± 22.37
Non-HDL-C	140.03 ± 38.17	150.36 ± 28.38
Lp(a)	11.38 ± 5.11	13.34 ± 4.13*
hs-CRP^d	0.74 ± 0.39	0.85± 0.35

*n= number of subjects. All the values are Mean ± SD. Statistical test used: Student's 't' test. Level of statistical significance: *p≤0.05 was considered significant, **p<0.01 was considered highly significant, p>0.05 was considered non-significant. Abbreviations used TC-Total cholesterol; TG-Triglycerides; HDL-C High density lipoprotein cholesterol; LDL-C-Low* *density lipoprotein cholesterol, calculated using the formula: TC-(HDL-C+VLDL-C) where VLDL-C=TG/5; Non-HDL-C-Non high density lipoprotein cholesterol, calculated using the formula: TC-(HDL-C); hs-CRP[d] -high sensitive C-reactive protein in mg/L; Lp(a)-Lipoprotein (a).*

Table 2: Comparison of atherogenic indices in female subjects of the study groups

Atherogenic indices	Females in the study groups	
	Fish-eaters (n=65)	Non fish-eaters (n=73)
TC/HDL-C	3.72 ± 1.00	4.12 ± 0.79**
LDL-C/HDL-C	2.30 ± 0.85	2.62 ± 0.62*
Non-HDL-C/HDL-C	2.72 ± 1.00	3.12 ± 0.79**

*n= number of subjects. All the values are Mean ± SD. Statistical test used: Student's 't' test. Level of statistical significance: *p≤0.05 was considered significant, **p<0.01 was considered highly significant, p>0.05 was considered non-significant. Abbreviations used TC-Total cholesterol; TG-Triglycerides; HDL-C High density lipoprotein cholesterol; LDL-C-Low density lipoprotein cholesterol, calculated using the formula: TC-(HDL-C+VLDL-C) where VLDL-C=TG/5; Non-HDL-C-Non high density lipoprotein cholesterol, calculated using the formula: TC-(HDL-C)*

Table 3: **Comparison of pro-oxidant and antioxidant status in female subjects of the study groups**

Parameters	Females in the study groups	
	Fish eaters (n=65)	Non fish eaters (n=73)
MDA[a]	3.91 ± 0.59	3.68 ± 0.52
MDA[b]	6.16 ± 0.78	5.84 ± 0.80
AOA	1.39 ± 0.32	1.40 ± 0.23

*n= number of subjects. All the values are Mean ± SD. Statistical test used: Student's 't' test. Level of statistical significance: *p≤0.05 was considered significant, **p<0.01 was considered highly significant, p>0.05 was considered non-significant. MDA[a]-Malondialdehyde measured at the end of 0 h., MDA[b]-Malondialdehyde measured at the end of 2h., AOA-Antioxidant Activity.*

DISCUSSION

The current research was conducted to evaluated the effect of a moderate fish consumption on lipid profiles and cardiac markers in females in the age group of 25-40 years. The study's findings demonstrated that female fish eaters reported a lower level of cardiac markers and lipid profiles as compared to vegetarian subjects. Among the females aged 26-30 y, TC (p≤0.05), LDL-C (p<0.01), non-HDL-C(p<0.01) and Lp(a) (p<0.01) were significantly lower in fish eaters. In the age group of 31-35y, the female fish eaters reported increase in HDL-C level and decrease in all the atherogenic parameters except TG. In the age group of 36-40 y TG was significantly lower (p<0.01) and HDL-C was significantly higher r(p<0.01) among the fish eaters as compared to their counterparts.

The results of this study reflect the outcomes by Leslie *et al.*, who revealed that n-3 PUFAs, which are naturally present in seafood, significantly lower triglyceride level in both hyperlipidaemic and normolipidemic individuals (27).

The study conducted by Vuholm *et al*., (28) in healthy population demonstrated the use of fish or seafood to reduce the levels of TG. An intervention study by Zibaeenezhad *et al*., (29) reported that patients with hyperlipidemia exhibited a substantial decrease in their LDL cholesterol levels when they consumed 250 g of fatty fish twice a week, in contrast to the n-3 supplement group (p < 0.001). In contrast, the study conducted by Brown and Wahle (30) in young healthy men aged 21-35 y reported increased requirement for dietary antioxidants with high intake of fish oil concentrates. Review findings of McMullan *et al*., (31) on intervention studies revealed that beneficial role of fish rather than directly reducing LDL or total cholesterol, fish may prevent atherosclerosis by maintaining cholesterol level. The findings by Yanai *et al*., (32) that fish consumption possess the ability to raise HDL cholesterol, and fish by increasing the ApoA1, apolipoprotein of HDL, reduces the risk of atherosclerosis (33). Randomized controlled trial showed people at high risk of diabetes and cardiovascular disease showed decreased inflammation by consuming a diet rich in LCn3 from fatty fish (34).

Marcovina *et al.,* (35) study demonstrated the dietary modification of Lp(a) by comparing the Lp(a) levels. Compared to the nearby vegetarian Bantu people, the Lp (a) levels of the Bantu fisherman were significantly lower. The results of oxidative stress in red blood cells correlate with the findings of Cariappa *et al.,* (36) who reported that the concentration of MDA in plasma did not alter between the two groups. In addition, people with hypertriglyceridemia showed sustained erythrocyte resistance to oxidative stress, according to a study conducted by Mabile *et al.,* (37). Nevertheless, in vivo study, contradictory results on oxidative status after consuming eicosapentaenoic and docosahexaenoic acid were also reported (38). The results of this research not supported the contest that increased oxidation could result from a diet rich in fish.

CONCLUSION

The outcome of this study showed that females in the 25–40 year age group who consistently consume a moderate amount of fish had lower cardiac markers and lipid profiles, which reduces their risk of CVD. Additionally, this study implies that regular fish consumption does not raise lipid peroxidation in fish eaters. More comprehensive research could provide more precise conclusions about the function of fish in the treatment and prevention of cardiovascular disease.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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