

# Effects of Traditional Way of Fish Consumption in Sri Lanka on Cardiovascular Risk Profiles

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## Abstract

The influence of type of fish consumption on cardiovascular risk profiles was studied in Sri Lankan community. A challenge experiment was set up to do a research on the influence of omega-3 fatty acid content in fish on cardiovascular risk profiles of healthy people. Hundred healthy undergraduates (subjects) who were between the ages of 23 to 30 years and full time resident in the hostels in Sri Lanka, were randomly selected for this study. The students were fed with fish curry and fried fish for the amount of  $80.41 \pm 9.43$  g of fish curry or  $62.50 \pm 11.04$  g of fried fish for five days per week. Initial and after 24 weeks and after one year the lipid profiles of the subjects [total cholesterol (TC), triglyceride (TG), low density lipoprotein (LDL-C), high density lipoprotein (HDL-C), and very low density lipoprotein (VLDL-C)] were estimated in serum samples collected at commencement and end of the experiment, using automatic biochemical analyzer and the turbidometric method respectively. The fish consumption showed direct effect on the cardiovascular risk profiles by decreasing LDL-C and increasing HDL-C and decreasing LDL: HDL ratio and TC: HDL ratio and revealed the decrease of TG in serum.

**Keywords:** Fish consumption, Lipid profiles, Lipoproteins, omega3 fatty acid, cardiovascular disease.

## Introduction

The beneficial health effects of fish and seafood consumption on cardiovascular risk factors have mainly been attributed to long chain omega-3 polyunsaturated fatty acids (LC omega-3 PUFAs)<sup>1</sup>. The dietary omega-3 PUFAs of fish exert beneficial effects by reducing platelet aggregation and improving blood lipoprotein profiles and have been consistently associated with triglyceride-lowering effects<sup>2</sup>. The most consistent effects of omega-3 PUFA are the reduction of serum cholesterol<sup>3</sup>, triglycerides, very-low density lipoprotein cholesterol (VLDL-C) and low density lipoprotein cholesterol (LDL-C)<sup>4</sup>. Further, intake of omega-3 PUFAs increases HDL-C<sup>5</sup>. Low HDL-cholesterol (HDL-C) as well as high LDL-C

is associated with the development of coronary heart disease (CHD)<sup>6</sup>. The fish consuming population had a lower atherogenic risk as opposed to the non-fish consuming population<sup>7</sup>. Nutritional studies have shown that the intake of omega-3 fatty acids from fish averages 1.3 gram per day in Japan as compared to 0.2 gram per day in the US<sup>8</sup>.

Two trials of 4 weeks and 8 weeks in duration showed that consumption of 125-150 g/ day (3.4-5.4g/day of omega-3 PUFA) of fish reduced of LDL-C by 14-15%<sup>9</sup>. Potential mechanisms for the cardioprotective effects of omega-3 fatty acids include: *anti-atherogenic effects such as* reduction in non-HDL-C levels, TG and VLDL-C levels, chylomicrons, VLDL-C and chylomicron remnants,

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increase in HDL-C levels, improvement in LDL-C and HDL-C particle size and plaque stabilization and *antithrombotic effects, decreased systolic and diastolic blood pressure* <sup>10</sup>.

In addition, the total cholesterol, TG, LDL-C are low in Eskimos whereas high HDL-C is raised. <sup>11</sup> The reduction of total SFAs, is one of the main targets of dietary recommendations in order to lower morbidity and mortality due to CVD <sup>12</sup>.

The fish consumption showed direct effect on the cardio vascular risk profiles by decreasing LDL-C and increasing HDL-C and decreasing LDL: HDL ratio and TC: HDL ratio <sup>9</sup>. Research revealed the decrease of TG in serum with fish consumption <sup>13</sup>.

The main aims of the present study were to investigate whether high omega-3 fatty acid fish consumption influences Cardiovascular risk factors in a dose-dependent manner among healthy people between 23 to 60 years of age.

## Materials and Methods

### Selection of Subjects and Approach

A challenge experiment was set up to do a research on the influence of omega-3 fatty acid content on cardiovascular risk profiles of healthy people. Hundred healthy undergraduates (subjects) who were between the ages of 23 to 30 years were randomly selected for this study.

### Consumption of Omega-3 fatty Acids Containing Fish

Fish cut and weighed ( $66.51 \pm 12.29$ g) into pieces of muscle. These pieces of fish muscle were made into a fish curry with coconut cream. The total weight of the fish curry including gravy was  $80.41 \pm 9.43$  g which was fed to subjects ( $n=39$ ). Another set of subjects ( $n=35$ ) were fed with  $62.50 \pm 11.04$  g fish fried in coconut oil (Vimal, Sri Lanka) daily. A six months trial commenced on December 2015 and ended on May 2016. . Subjects were fed  $80.41 \pm 9.43$  g of fish curry or  $62.50 \pm 11.04$  g of fried fish for five days per week. Initial, after 24 weeks and after one year the lipid profiles of the subjects [total cholesterol (TC), triglyceride(TG), low density lipoprotein (LDL-C), high density lipoprotein (HDL-C), very low density lipoprotein (VLDL-C)] and high sensitive C-reactive

proteins (hs-CRP) were estimated in serum samples collected at the commencement and end of the experiment, using automatic biochemical analyzer and the turbidometric method respectively.

### Determination of Cardio vascular risk lipid indices

Serum cholesterol was estimated using the cholesterol oxidase-phenol-4-aminophenazone method with a lipid clearing agent by enzymatic colorimetric assay (Spinreact®-Spain). The cholesterol in the sample oxidized by the action of cholesterol oxidase enzyme into 4-Cholestenone and hydrogen peroxide which in turn reacts with phenol and 4-aminophenazone in the presence of peroxidase enzyme to produce red color. HDL-C was determined after precipitation of other lipoproteins by sodium phosphotungstate with magnesium chloride reagent (Spinreact®-Spain) and read using analyzer (Indika Chembell, Thermoscientific, UK). The serum TG level was estimated using an enzymatic method (Indika Chembell, Thermoscientific, UK)<sup>12</sup>.

## Results and Discussion

This study was conducted to assess the effect of fish consumption (omega-3 fatty acid) on serum lipid profile in healthy young subjects

**Table 1: Polyunsaturated fatty acids (Omega-3 PUFA) in raw, fried and curried fish**

Fish Name (Common Name)	Raw Fish(mg/100g)	Fried Fish(mg/100g)	CurryFish (mg/100g)
	Omega-3FA	Omega-3FA	Omega-3FA
<i>Leiognathus bindus</i> (Pony fish)	23.65(0.73)	2.44(0.36)	11.27(0.59)
<i>Mugil cephalus</i> (Mullet)	15.53(0.42)	5.38(0.77)	13.20(1.01)
<i>Rasterlliger kanagurata</i> (Mackerel)	23.90(0.08)	5.40(1.01)	24.29(1.49)
<i>Chirocentrus dorab</i> (Wolf)	25.04(0.70)	6.96(0.92)	15.50(0.74)
<i>Selar crumenophththalmus</i> (Scad)	23.68(3.12)	4.09 (1.26)	4.35 (0.34)
<i>Katsuwonus pelamis</i> (Tuna)	33.91(2.25)	7.61 (0.63)	15.65(1.16)
<i>Sphyræna jello</i> (Baracuda)	22.82(0.16)	5.21(1.13)	22.13(1.00)
<i>Dussumieria acuta</i> (Herring)	26.10(0.26)	3.97(0.65)	8.03 (2.60)

Data presented as mean (standard deviation)

**Table 2: Cardiovascular risk profiles and lipid indices of the subjects of curry and fried fish consumers**

Lipid Profiles	Curry Fish Eaters				Fried Fish Eaters			
	End Profiles	Initial Profiles	R -value	P-value	End Profiles	Initial Profiles	R value	P-value
TC (mg/dl)	193.86±37.63	188.06±27.26	0.17	0.32	185.78±32.39	183.11±28.63	0.32	0.52
TG(mg/dl)	132.97±54.14	143.91±67.56	0.39	<b>0.03*</b>	113.99±46.13	124.49±60.14	0.38	0.14
LDL-C(mg/dl)	122.14±31.44	113.37±23.55	-0.06	0.73	116.74±26.93	111.76±26.07	0.16	0.22
HDL-C(mg/dl)	45.17±4.85	46.00±9.23	0.12	0.48	45.97±11.81	45.16±8.69	-0.03	0.64
VLDL-C(mg/dl)	26.59±10.83	28.79±13.51	0.38	<b>0.03*</b>	22.79±9.23	24.89±12.03	0.38	0.14
TC:HDL	4.28±0.60	4.19±0.71	-0.17	0.61	4.13±0.46	4.16±0.79	-0.05	0.81
LDL:HDL	2.67±0.53	2.52±0.56	-0.16	0.27	2.61±0.40	2.57±0.61	-0.08	0.66
NON-HDL-C	148.69±34.32	142.06±24.57	0.081	0.64	139.81±29.50	137.95±27.26	0.25	0.65
AC	3.28±0.58	3.18±0.71	-0.182	0.58	3.09±0.55	3.15±0.79	-0.09	0.65
API	0.05±0.00	0.05±0.00	-0.063	0.62	0.05±0.01	0.05±0.01	0.01	0.24
TGHDL	2.22±0.78	3.66±2.07	0.149	0.37	2.53±1.06	2.89±1.68	0.23	0.08
Hscrp	1.89±3.47	1.83±3.58	-0.019	0.95	1.73±3.05	1.34±2.69	-0.02	0.41

\*Significant, P < 0.05. AC: atherogenic coefficient, API: atherogenic index of plasma, HDL-C: high-density lipoprotein cholesterol, hs-CRP: high-sensitivity C-reactive protein, LDL: low-density lipoprotein, LDL-C: low-density lipoprotein cholesterol, TC: total cholesterol, TG: triglycerides, VLDL-C: very low-density

The omega-3 fattyacid in raw fish was significantly higher in almost all fishes than curry and fried fishes. By cooking fish with coconut cream and frying in coconut oil, the content of omega-3 fatty acid showed

decrease and which can be explained that the impact of omega-3 fattyacid in lipid profiles and atherogenesis did not reveal in the present result (Table 1).

**Table 3: Status of Lipid parameters and lipid indices during pre and post intervention (6 months) and 1 year post intervention**

Lipid Parameters	Pre	Post	Post	Significant (n=74) (2 tailed)
	Intervention	Intervention	Intervention (1 year)	
TC(mg/dl)	180.80(17.53)	175.20(27.42)	193.00(38.09)	0.01
TG(mg/dl)	96.40(28.47)	98.00(27.81)	86.93(21.68)	0.25
LDL-C(mg/dl)	110.53(29.89)	111.93(22.00)	131.44(33.19)	0.07
HDL-C(mg/dl)	44.27(6.79)	43.67(5.27)	44.13(3.93)	0.82
VLDL-C(mg/dl)	20.95(9.35)	19.60(5.56)	17.39(4.34)	0.38
NON-HDL-C (mg/dl)	136.53(15.42)	131.53(22.42)	148.87(34.85)	0.07
<b>Lipid Indices</b>				
TC:HDL	4.15(0.53)	4.00(0.21)	4.30(0.53)	0.01*
LDL:HDL	2.70(0.45)	2.54(0.27)	2.95(0.52)	0.08
AC	3.14(0.55)	3.00(0.22)	3.35(0.53)	0.04*
API	0.05(0.01)	0.05(0.01)	0.04(0.01)	0.55

\* Significant P < 0.05 AC: atherogenic coefficient, API: atherogenic index of plasma, HDL-C: high-density lipoprotein cholesterol, LDL: low-density lipoprotein, LDL-C: low-density lipoprotein cholesterol, TC: total cholesterol, TG: triglycerides, VLDL-C: very low-density.

The cardio vascular risk parameters and lipid indices showed differences in pre and post intervention of experiment as shown in Table 3 where in curry fish consumers had significant variation between pre and post intervention in TG and VLDL content ( $p < 0.05$ ). There was not significant difference in TC, LDL-C and HDL-C. However, the after another one year of post intervention showed that there was

no significant difference in the lipid profiles between the intervention while the lipid indices, TC: HDL and AC had significant different ( $p < 0.05$ ) between intervention as shown in Table 2. There was no significant different in cardio risk profiles between pre and post intervention in overall both curried and fried fish consumers ( $n=74$ ) (Table 3).

**Table 4: Comparison of pre and post intervention of fish consumption of all subjects**

	Pre-Intervention	Post -Intervention		
Lipid Parameters	Mean±SD	Mean ±SD	R value	P-value
TC(mg/dl)	183.10±28.63	185.78±32.39	0.324	0.520
TG(mg/dl)	124.49±60.14	113.99±46.13	0.379	0.139
LDL-C(mg/dl)	111.76±26.07	116.74±26.93	0.164	0.215
HDL-C(mg/dl)	45.16±8.68	45.97±11.81	-0.034	0.641
VLDL-C(mg/dl)	24.89±12.03	22.79±9.23	0.379	0.139
Non-HDL	137.95±27.26	139.81±29.50	-0.460	0.647
<b>Lipid indices</b>				
TC: HDL	4.16±0.79	4.13±0.46	-0.050	0.814
LDL: HDL	2.57±0.61	2.61±0.40	-0.084	0.661
AC	3.15±0.79	3.09±0.55	-0.091	0.649
API	0.05±0.01	0.04±0.01	0.006	0.244
<b>Defence Protein</b>				
HS-CRP(mg/L)	1.34±2.69	1.73±3.05	-0.018	0.412

\* Significant  $P < 0.05$  AC: atherogenic coefficient, API: atherogenic index of plasma, HDL-C: high-density lipoprotein cholesterol, hs-CRP: high-sensitivity C-reactiveprotein, LDL: low-density lipoprotein, LDL-C: low-density lipoprotein cholesterol, TC: total cholesterol, TG: triglycerides, VLDL-C: very low-density.

**Table 5: hs-CRP content in serum before and after fish consumption**

Parameter	Curry Fish Eaters			P	Fried Fish Eaters			P
	Initial	End	X <sup>2</sup>		Initial	End	X <sup>2</sup>	
Base line <1	65.70%	65.70%			74.40%	59%		
	n=23	n=23			n=29	n=23		
Inflammation	25.70%	17.10%			20.50%	25.60%		
	n=9	n=6			n=8)	n=10		
Acute	8.60%	17.10%			5.10%	15.40%		
Inflammation	n=3	n=6			n=2	n=6		
			1.6	0.45			2.92	0.23
hsCRP (mg/L)	0.92±0.82	0.98±0.95		0.44	0.72±0.72	0.88±0.74		0.40

As per the Table 5 the TC had not significantly changed due to curry or fried fish consumption ( $\chi^2 = 1.495$ ,  $p=0.474$ ). TG lower risk level increased in

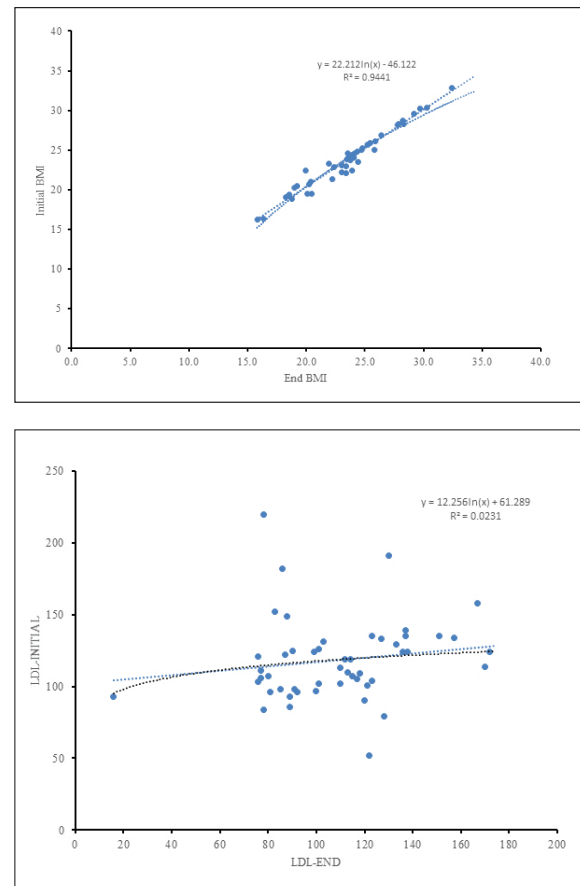
curry fish consumers from 11.4% to 22.9% whereas it had decreased in fried fish eaters from 5.1% to 2.6%. Overall, high risk level of TG showed

decrease not significantly in both curry fish and fried fish consumers (20% to 14.3% and 5.1% to 0.0%) respectively. Curry fish consumers had an increase of lower risk of LDL-C from 8.6% to 20% whereas in fried fish consumers decreased from 17.9% to 12.8%, but higher risk level of LDL-C had no change in curry fish consumers whereas in fried fish eaters the high risk level of LDL-C had decreased from 5.1% to 2.6%. HDL-C lower risk level had increased from 48.6% to 80% in curry fish consumers and decrease from 12.8% to 5.1% in fried fish consumers and risk level was considered as per the lipid profile risk levels stated by American Heart Association ([www.heart.org](http://www.heart.org)). However, TC: HDL ratio lower risk level had increase from 80% to 88.6% in curry fish consumers and increase from 66.7% to 88.6% in fried fish eaters. TC/ HDL-C and LDL/HDL-C ratio are risk indicators with greater predictive value than isolated parameters used independently. A study of <sup>14</sup>revealed that coconut cream supplementation was responsible for the reduction in the LDL-C and increase of HDL-C. As observed by <sup>14</sup>who observed rise of HDL-C with eating SFA rich coconut cream, this study agreed the rise in HDL-C while ingesting a saturated fat rich coconut cream cooked fish.

TG is found in high amount in both coconut cream and coconut oil as stated by<sup>14</sup>. Because of the reason, TG showed an increase in curry fish. It was observed that the hs - CRP has no significant difference before and after fish consumption and it reveals it could be a general inflammatory protein and it is subjected to vary with the inflammatory effects on body not only in blood vessels but also in other tissues (Table 5). The % of subjects in hs-CRP high risk group (>3) showed increase from 8.6% to 17.15% in curry eaters whereas it increased from 5.1% to 15.4% fried fish eaters. Since hs-CRP is subjected to change with general inflammation, it could not be used to predict the coronary artery inflammation. It was observed that hs-CRP had the significant difference between the males and females, and the males (0.98mg/L) had a significantly higher amount of hs-CRP than females (0.88mg/L).<sup>3</sup>stated that TC, TG and VLDL-C reduction due to the consumption of food with omega-3 PUFA and reached normal. But the present study did not agree with the above finding and omega-3 PUFA effects on cardio risk profiles is modulated by high intake of SFA with fish. <sup>4</sup> reported LDL-C reduction with omega-3 PUFA in animal food while our study showed increase of LDL-C with consumption of fish. However, increase of HDL-C with omega -3 PUFA

containing fish intake observed by <sup>6</sup>which is similar to the present study too. As stated by<sup>7</sup> results of some studies have shown contradictory results regarding atherogenic effects of omega-3 PUFAs from fish.

However, the LDL-C showed no significant increase due to fish consumption. Among both fish curry and fried fish consumers, TC, LDL-C, TG and VLDL-C showed significantly higher upper quintiles than lower quintiles. Because of rise of stated cardio risk profiles showed higher than optimum reference level.



**Figure 1: Correlation between pre and post intervention of BMI and LDL-C and VLDL-C**

In the present study, there was no significant difference between curried and fried fish eating population except in TG and VLDL-C.

## Conclusion

At the end of the trial experiment, due to feeding of omega-3 PUFA content fish, noticeable differences were observed on concentration of lipid cholesterol fractions and lipid indices among the subjects. Concentration of TC, TG, VLDL-C, Non-HDL and

LDL-C showed an increase among fish curry eaters than fried fish eaters whereas HDL-C and lipid indices such TC/HDL, LDL/HDL, AC and API were higher in fried fish eaters than fish curry eaters. However, the significant differences were observed in TG, VLDL-C and non-HDL-C in curry eaters. Significantly higher TG, VLDL-C and hs-CRP in male than female was observed in this study where males ate fish curry. Overall, non-HDL showed increase due to feeding of both type of fish consumption. Although, fish flesh consist of high amount of essential fatty acids, these role on cardiovascular profiles was prevented due to the style of consumption of fish.

### Conflict of Interest

There is no conflict of interest in this paper

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### Ethical Clearance:

Approval was obtained from the Ethical Clearance Board of the Faculty of Health Care Sciences, Eastern University, Sri Lanka.

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