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Omega-3 fatty acids alter lipoprotein subfraction distributions and the in vitro conversion of very low density lipoproteins to low density lipoproteins

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The purpose of this study was to determine the effects of a fish oil concentrate (FOC) on the in vitro conversion of very low density lipoproteins (VLDL) to intermediate (IDL) and low density lipoproteins (LDL). Six hypertriglyceridemic patients were randomly allocated to receive either placebo (olive oil) or FOC (1 g/14 kg body weight/day) for 4 weeks in a crossover study with a 4-week washout period. The FOC provided 3 g of eicosapentaenoic + docosahexaenoic acid per 70 kg of body weight, and it lowered plasma triglyceride and VLDL cholesterol levels by 35% and 42%, respectively. Decreases in the largest particles ($VLDL_1$) were primarily responsible, with no effect noted in smaller VLDL particles ($VLDL_2$ and $VLDL_3$). The FOC increased LDL cholesterol levels by 25% ($P < 0.06$) but did not affect LDL particle size. $VLDL_1$ and $VLDL_3$ were incubated in vitro with human postheparin lipases. Although triglycerides from both types of VLDL were hydrolyzed to the same extent with both treatments, particles isolated during the FOC phase were more readily converted into IDL and LDL than were control particles. These data suggest that the marine ω3 fatty acids may enhance the propensity of VLDL to be converted to LDL, partly explaining the decreased VLDL and increased LDL levels in FOC-treated patients. (J. Nutr. Biochem. 10:151–158, 1999)

Keywords: fish oil; lipoprotein lipase; hepatic triglyceride lipase; eicosapentaenoic acid; docosahexaenoic acid; very low density lipoproteins; low density lipoproteins

Introduction

The triglyceride- and very low density lipoprotein (VLDL)-lowering effects of fish oils have been demonstrated in both normal[1–5] and hypertriglyceridemic (HTG) subjects,[4,6–10] with a 25 to 35% reduction being normal.[11] Studies in both humans and animals have shown that fish oil concentrate (FOC) inhibits hepatic triglyceride synthesis and the secretion of VLDL from liver.[4,9,12] Because ω3 fatty acids treatment has failed consistently to stimulate postheparin lipoprotein lipase (LpL) or hepatic lipase (HL) activity,[3,13,14] or to accelerate Intralipid clearance,[15] it has been assumed that the primary mechanism by which ω3 fatty acids lower triglycerides is via reduced production, not enhanced clearance.

The effect of ω3 fatty acids on plasma low density lipoprotein (LDL) levels is less clear. Levels usually are not affected in normal subjects taking ω3 fatty acids but often increase in HTG patients.[16] The mechanism responsible for this is unknown. The enzymes that primarily control the production of LDL from VLDL are LpL and HL.[17–19] LpL acts on chylomicrons and larger VLDL whereas HL seems to prefer denser lipoproteins such as VLDL remnants.[17,20,21] Although ω3 fatty acids do not affect (postheparin) enzyme activities, it is possible that they could increase the susceptibility of VLDL to LpL and/or HL-mediated lipolysis and thereby increase the production of LDL from VLDL.

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Table 1 Description of patients

Gender	4M/2F
Age (yr)	42–67
Wt (kg)	85.5 ± 20.5
Body mass index (kg/m^2)	27.7 ± 3.7
Triglyceride*	5.53 ± 0.84
Cholesterol	5.88 ± 1.01
VLDL-C	1.99 ± 0.39
LDL-C	2.67 ± 0.75
HDL-C	1.04 ± 0.31

VLDL–very low density lipoprotein. C–cholesterol. LDL–low density lipoprotein. HDL–high density lipoprotein.
*Lipids given in mmol/L.

Methods and materials

Patients

We recruited six type IV HTG patients from the lipid clinic population. Their ages and other parameters are listed in *Table 1*. None was taking any medications known to affect lipid metabolism. Each patient gave informed consent before beginning the trial, which had been approved by the Human Subjects Committee of the Medical Center.

Protocol

Patients were randomly assigned to receive FOC rich in eicosapentaenoic acid (EPA) and docosahexaenoic (DHA) ethyl esters or olive oil ethyl esters (placebo) in a single blind, crossover trial. Dosages were body weight adjusted so that a 70-kg patient took 5 g of FOC containing 3 g of EPA + DHA or 5 g of placebo. Each 1.0-g FOC capsule provided 640 mg EPA + DHA, whereas the 1.0-g placebo capsules contained largely oleic acid (518 mg) and linoleic acid (154 mg) and were devoid of ω3 fatty acids.[22] Taking five capsules each day added 45 kcal to the diet. Both sets of capsules were obtained from the Fish Oils Test Materials Program of the National Institutes of Health and the Department of Commerce. Each contained 3 mg vitamin E and 3 mg cholesterol. There were two 4-week treatment phases separated by a 4-week washout period.

Plasma lipid and lipoprotein assays

Blood was drawn with minimal stasis into tubes containing 1 mg EDTA/mL. Whole plasma triglyceride and cholesterol levels were measured enzymatically using cholesterol/HP reagent (Boehringer-Mannheim Corp., Indianapolis, IN USA) and triglyceride (GPO-Trinder) reagent (Sigma Diagnostics, St. Louis, MO USA) on a Cobas Mira (Roche Analytical Instruments Inc., Nutley, NJ USA) following the manufacturer's instructions. In the in vitro analyses, glycerol-blanking was used to obtain true triglyceride levels. Plasma HDL cholesterol was measured following precipitation of the apoB containing lipoproteins with heparin and manganese chloride.[23] Screening VLDL and LDL cholesterol levels were estimated by the Freidewald equation.[24] VLDL and LDL were measured by β-quantitation using a TL-100 ultracentrifuge (Beckman Instruments, Brea, CA USA) and a TLA-100.3 rotor. The VLDL fraction was isolated from plasma by centrifugation at 100,000 rpm for 120 minutes and was removed by aspiration. Infranatant cholesterol (i.e., LDL + HDL) was measured, and the LDL cholesterol obtained by subtracting HDL cholesterol from LDL + HDL cholesterol. VLDL and LDL subfractions were isolated and analyzed as described by Thomas et al.[25] and Ziogas et al.,[26] respectively. Our laboratory participates in the Lipid Standardization Program of the NIH/CDC and in the Pacific Biometrics ALERT proficiency survey.

Fatty acid analysis

VLDL lipids were extracted with methylene chloride:methanol.[27] VLDL triglyceride (TG) and phospholipid (PL) were isolated by thin layer chromatography on Silica Gel G plate using hexane: ethyl ether:formic acid (70:30:1) as the developing solvent system. Saponification and methylation of TG and PL fatty acids in VLDL were carried out as described previously.[15] The fatty acid methyl esters were quantified with GC9A gas chromatograph (Shimadzu, Columbia, MD USA) equipped with a 30-m, 0.32-mm inner diameter SP2330 capillary column and were identified by comparison with a standard mixture of fatty acid methyl esters (Sigma, St. Louis, MO USA).

Preparation of total lipases from human postheparin plasma

Postheparin blood from one of the investigators was drawn 15 minutes after intravenous injection of heparin (100 U/kg), and the tubes were immediately placed on ice. Plasma was separated by centrifugation at 4°C and stored at −80°C. The method for preparation of postheparin lipases involved their isolation from plasma by heparin affinity chromatography[28–30] and their subsequent incubation with emulsified ^{3}H-triolein to determine activity.[22] Once isolated lipases were incubated with large and with small VLDL particles; resulting lipoproteins were isolated by density gradient ultracentrifugation.

Before application to the affinity column,[22] the frozen postheparin plasma (10 mL) was thawed and centrifuged in the cold at 3,000 rpm for 2 minutes to sediment precipitated proteins. The plasma supernatant was mixed 1:1 with buffer and placed on the column (6 mL of gel). The sample tube was rinsed with 2 mL of fresh buffer and this also was applied to the column. The column then was washed with 10 mL of buffer followed by 20 mL of 0.3 M NaCl in buffer, and then 0.6 mL of 1% heparin buffer. Three mL of 1% heparin buffer was put on column and all of the eluate collected. The enzymes thus isolated were diluted 50 or 100 times with 1% heparin buffer. The preparation of the substrate and the incubation procedures have been described previously.[22] Enzyme activity was expressed in units, with 1 unit equalling 1.0 μmol fatty acid released per milliliter of undiluted enzyme solution per hour at 37°C.

Conversion of VLDL subfractions to LDL in vitro

Only the $VLDL_1$ and $VLDL_3$ fractions were used in the incubation experiments because our objective was to compare the in vitro metabolic fact of large versus small VLDL particles obtained during the placebo and fish oil phases. $VLDL_1$ or $VLDL_3$ were concentrated centrifugally (Millipore, Bedford, MA USA) to 5 to 10 mg TG/mL. Concentrated fractions were dialyzed against buffer overnight at 4°C, and the cholesterol and triglyceride concentrations were re-measured. Incubation of VLDL subfractions with human postheparin lipases was carried out as follows: VLDL triglyceride (2.5 mg) was combined with 6.25 units of total lipase (which contained 46% LpL and 54% HL activity), 750 μL of the 20% bovine serum albumin (BSA) solution, and sufficient buffer to bring the total volume to 2.5 mL. Thus, all samples were adjusted to 1 mg TG per milliliter of incubation mixture. The samples were incubated at 20°C for 2 hours (free fatty acid release was linear to at least 3 hours; data not shown) after which time the samples were placed on ice and the density adjusted to 1.21 g/mL with solid NaBr. The samples were subjected to density gradient ultracentrifugation as described above to isolate all three VLDL

Table 2 Effects of the fish oil concentrate (FOC) on plasma lipid and lipoprotein levels in six hypertriglyceridemic patients (mmol/L)

	Placebo	FOC
Cholesterol	6.01 ± 0.75	5.78 ± 0.49
Triglyceride	6.34 ± 2.33	4.10 ± 0.62[a]
VLDL-Chol	2.36 ± 0.88	1.37 ± 0.49[a]
LDL-Chol	2.64 ± 0.80	3.23 ± 0.73
HDL-Chol	1.01 ± 0.26	1.09 ± 0.28

VLDL–very low density lipoprotein. Chol–cholesterol. LDL–low density lipoprotein. HDL–high density lipoprotein.
[a]$P < 0.05$.

subfractions, intermediate density lipoproteins (IDL) and LDL in a single spin. The cholesterol and triglyceride (glycerol-blanked) contents of each fraction were determined and compared between treatments. Free fatty acids released in the in vitro VLDL incubations were analyzed colorimetrically (NEFA kit, Wako Chemical Co, Japan).

Statistical analysis

Data are presented as means ± SD. Paired *t*-tests were used to evaluate treatment effects, with a *P*-value of less than 0.05 being required for statistical significance.

Results

Compliance with the supplements was greater than 95%. The mean intake of FOC was 5.8 g/d. When adjusted for body weights, the target dose of 3.0 g of ω3 fatty acid per day per 70 kg was achieved. There was no significant difference in body weights between the placebo and FOC periods.

The effects of ω3 fatty acids on plasma lipids and lipoproteins are give in *Table 2*. When compared with placebo values, FOC did not alter the plasma total cholesterol levels, but TG levels were lowered by 35% ($P < 0.05$). Corresponding to the change in plasma TG was a 42% ($P < 0.05$) decrease in VLDL cholesterol levels; LDL cholesterol levels increased by 25% ($P < 0.06$); and there was an nonsignificant rise in HDL cholesterol levels.

The VLDL cholesterol subfraction profile was altered by the FOC with the majority of the decrease seen in the largest subfraction ($VLDL_1$), which fell by 50% ($P = 0.025$; *Figure 1*). Other subfractions were unaffected. LDL cholesterol subfraction distribution was unaltered by FOC supplementation with only the amount (*Figure 1*), not the size (*Figure 2*) of LDL changing with treatment. As expected, patient LDL tended to be smaller and denser compared with LDL from a group of healthy controls.

All long chain ω3 fatty acids increased in both VLDL-TG and VLDL-PL with concomitant reductions of 18:2 ω6 in VLDL-PL and of 18:2 ω6 and 20:3 ω6 in VLDL-TG (*Table 3*). Although marine ω3 fatty acid levels increased seven times in VLDL-TG and three times in VLDL-PL, the absolute amounts present were still quite small, accounting for less than 3% and 8% of total fatty acids, respectively.

The total amount of free fatty acids released from $VLDL_1$ (60–65%) and $VLDL_3$ (approximately 45–50%) during in vitro incubation with postheparin lipases was not affected by treatment (*Figure 3*). Likewise, the total amount of VLDL TG remaining after incubation was the same in the control and FOC periods for both $VLDL_1$ (approximately 29%) and $VLDL_3$ (approximately 36%). Naturally, the total cholesterol recovered after incubation was virtually 100% for both substrates because cholesterol was only re-distributed, not metabolized. These data suggest that the TG carried in FOC-VLDL was not more susceptible to hydrolysis than was that carried in olive oil-VLDL.

On the other hand, FOC supplementation increased the in vitro conversion of VLDL to IDL and LDL. This effect was seen whether larger particles ($VLDL_1$; (*Figure 4*) or smaller particles ($VLDL_3$; *Figure 5*) were used as the substrate, and whether lipoprotein TG or cholesterol was used as a marker lipid. After incubation of $VLDL_1$ with plasma lipases, $VLDL_1$ cholesterol fell by 26% ($P < 0.005$) in the fish oil group compared with the placebo group. The $VLDL_2$,

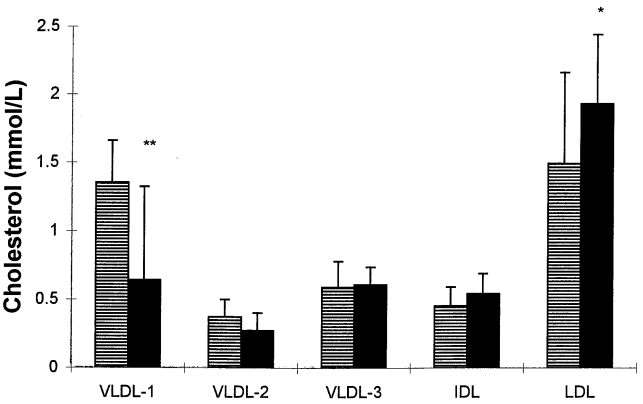

Figure 1 Distribution of apo-B containing lipoproteins in six hypertriglyceridemic patients treated with olive oil placebo (shaded bars) or fish oil concentrate (black bars) for 4 weeks. See Materials and methods for description of very low density lipoprotein (VLDL) subfractions. $^{**}P = 0.025$, $^{*}P = 0.058$. IDL–intermediate density lipoproteins. LDL–low density lipoproteins.

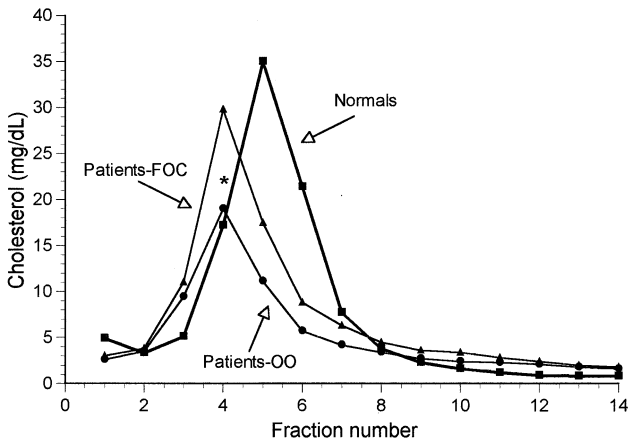

Figure 2 Distribution of low density lipoprotein particle size determined by density gradient ultracentrifugation in five patients during olive oil (OO) placebo and fish oil concentrate (FOC) treatment compared with the distribution in six healthy controls. $^{*}P = 0.052$ for fraction 4, FOC vs. OO.

Table 3 Fatty acid composition (mol %) of VLDL triglyceride and phospholipid in six hypertriglyceridemic patients supplemented with fish oil concentrate (FOC) or olive oil placebo

Fatty acid	VLDL Triglyceride		VLDL Phospholipid	
	Placebo	FOC	Placebo	FOC
14:0	3.4 ± 0.6	2.8 ± 0.7	1.5 ± 0.7	1.7 ± 0.8
16:0	34.4 ± 2.7	31.8 ± 4.4	34.3 ± 2.6	33.0 ± 1.9
16:1	5.0 ± 0.6	5.2 ± 0.6	1.0 ± 0.2	1.0 ± 0.2
18:0	3.7 ± 0.9	3.1 ± 0.6	14.5 ± 2.1	14.5 ± 1.7
18:1 ω7,9	35.5 ± 1.8	34.6 ± 2.9	10.8 ± 1.1	11.5 ± 3.8
18:2 ω6	16.6 ± 2.8	18.0 ± 2.7	21.7 ± 2.7	18.6 ± 1.7[a]
20:3 ω6	0.8 ± 0.3	0.4 ± 0.1[a]	3.7 ± 0.4	3.0 ± 0.4[b]
20:4 ω6	1.0 ± 0.2	1.0 ± 0.3	9.2 ± 1.42	8.2 ± 1.0
20:5 ω3	0.1 ± 0.1	1.0 ± 0.3[c]	0.4 ± 0.1	2.8 ± 1.0[c]
22:4 ω6	0.2 ± 0.1	0.2 ± 0.1	0.4 ± 0.3	0.4 ± 0.3
22:5 ω6	0.1 ± 0.1	0.1 ± 0.0	0.4 ± 0.1	0.2 ± 0.0[c]
22:5 ω3	0.2 ± 0.1	0.7 ± 0.1[c]	0.6 ± 0.1	1.1 ± 0.2[c]
22:6 ω3	0.2 ± 0.5	1.3 ± 0.4[c]	1.5 ± 0.4	4.0 ± 0.9[c]
EPA +DPA + DHA	0.4	2.9[c]	2.4	7.9[c]

VLDL–very low density lipoprotein. EPA–eicosapentaenoic acid. DPA–docosapentaenoic acid. DHA–docosahexaenoic acid.
[a]$P < 0.05$.
[b]$P < 0.005$.
[c]$P < 0.001$.

$VLDL_3$, IDL, and LDL cholesterol produced from $VLDL_1$ increased by 29% ($P < 0.05$), 39% ($P < 0.05$), 30% (NS), and 56% ($P < 0.02$), respectively, for the FOC treatment. Similar differences between FOC and control VLDL also were seen in the lipase-mediated conversion of $VLDL_3$ to LDL (*Figure 5*). When compared with placebo, $VLDL_3$ cholesterol in the FOC group decreased by 19% ($P < 0.02$), and the IDL and LDL cholesterol derived from $VLDL_3$ increased by 29% ($P < 0.05$) and 20% ($P < 0.01$), respectively. When TG was used as the marker, $VLDL_3$ and IDL were increased by FOC treatment, but there was no difference between treatments in LDL-TG postincubation.

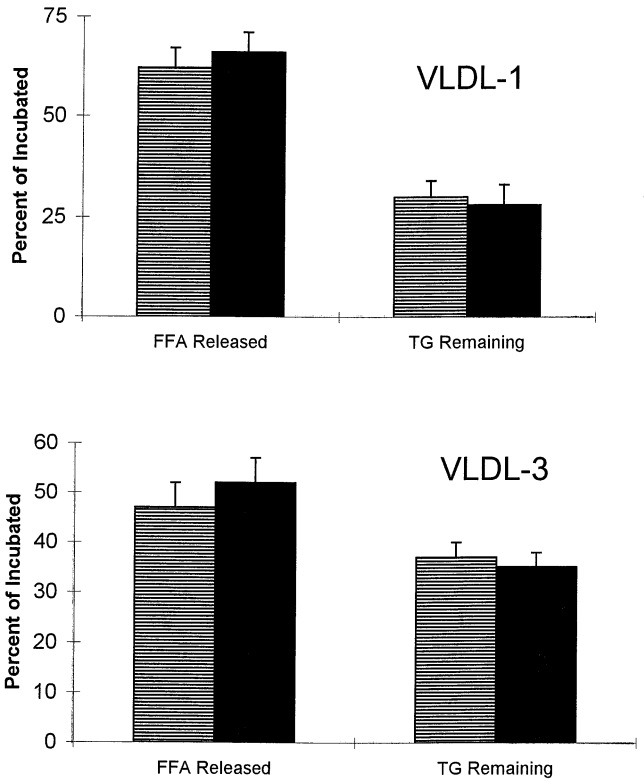

Figure 3 Large ($VLDL_1$) and small ($VLDL_3$) triglyceride (TG) rich lipoprotein particles were isolated from six hypertriglyceridemic patients treated for 4 weeks with olive oil placebo (shaded bars) or fish oil concentrate (black bars). They were incubated on an equal TG basis with postheparin lipases. The amount of free fatty acid (FFA) released and TG remaining were measured. VLDL–very low density lipoprotein.

Although incubations were based on lipoprotein TG levels rather than on particle numbers (i.e., apo B-100 content), it is likely that particle numbers were similar in the placebo and FOC treatments because VLDL TG:cholesterol preincubation ratios were similar (6.5 ± 0.9 and 6.1 ± 1.0 for $VLDL_1$, and 3.1 ± 0.5 and 2.7 ± 0.2 for $VLDL_3$ for placebo and FOC, respectively). Ratios for each subfraction were not significantly different from each other, as expected, because the subfractions were isolated under identical centrifugal conditions in both periods. Even though FOC lowered circulating levels of $VLDL_1$ (but not $VLDL_3$), the in vitro incubations were carried out on an equal TG mass basis so the individual susceptibility could be separated from concentration effects.

Discussion

Two significant observations regarding the effects of ω3 fatty acids on human lipoprotein metabolism were made in this study: They did not alter LDL size distributions, but they did produce VLDL particles which were more readily converted in vitro to IDL and LDL.

The role of LDL size distributions in the etiology of atherosclerosis has become an area of intense investigation.[31–34] There is growing consensus that in humans the smaller, denser LDL particles characteristic of hypertriglyceridemia are more atherogenic than the more buoyant

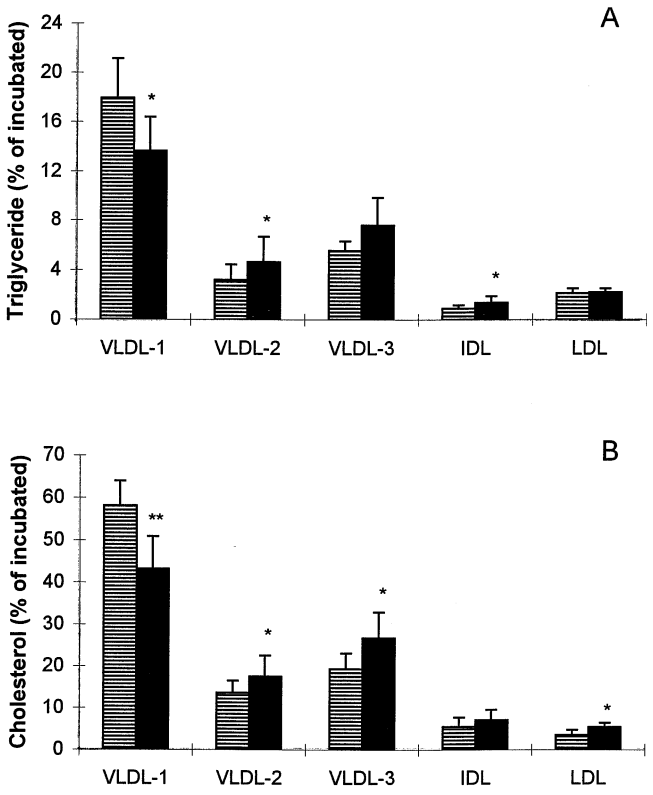

Figure 4 Incubation with $VLDL_1$. $VLDL_1$ was isolated from patients ($N = 6$) plasma after 4 weeks of supplementation with olive oil placebo (shaded bars) or fish oil concentrate (black bars). One milligram of $VLDL_1$-triglyceride was incubated with postheparin lipases and the resultant particle distribution determined. Both triglyceride (*A*) and cholesterol (*B*) were used as particle markers. $^*P < 0.05$. $^{**}P < 0.01$, both vs. placebo. VLDL–very low density lipoprotein. IDL–intermediate density lipoprotein. LDL–low density lipoprotein.

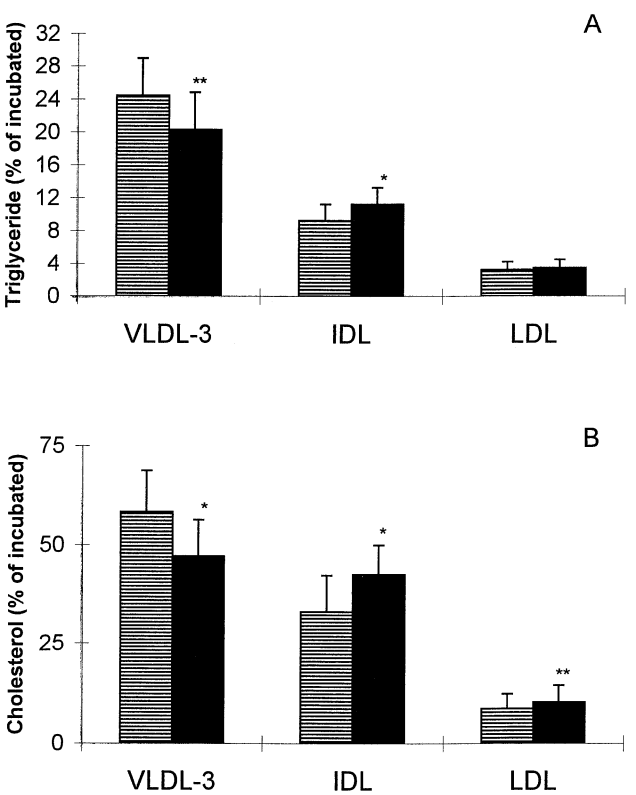

Figure 5 Incubation with $VLDL_3$. $VLDL_3$ was isolated from patients ($n = 6$) plasma after 4 weeks of supplementation with olive oil placebo (shaded bars) or fish oil concentrate (black bars). One milligram of $VLDL_3$-triglyceride was incubated with postheparin lipases and the resultant particle distribution determined. Both triglyceride (*A*) and cholesterol (*B*) were used as particle markers. $^*P < 0.05$. $^{**}P < 0.01$, both vs. placebo. VLDL–very low density lipoprotein. IDL–intermediate density lipoprotein. LDD–low density lipoprotein.

particles.[35,36] Nevertheless, the role of small, dense LDL as a causal factor in atherogenesis has been questioned, partly because of the high correlation between LDL size and elevated TG and reduced HDL cholesterol levels.[37,38] When TG levels are taken into account, LDL size usually ceases to be a significant predictor of disease,[32,39] and lowering TG levels has been reported to increase LDL size.[40] Interestingly, in normolipidemic patients with coronary heart disease (CHD) LDL particles tend to be larger than in controls,[34] and in non-human primates smaller LDL particles appear to be less atherogenic than larger ones.[41] Therefore, the role of LDL particle size in CHD risk stratification is not clear.

Because of the tight correlation between TG levels and LDL particle distributions, we anticipated that fish oil treatment would increase LDL size. However, we found no change in mean LDL density (a correlate size). Other studies using gradient gel electrophoresis[42,43] or apo B-100/cholesterol ratios[44] also have reported no effect of ω3 fatty acids on LDL size. Increases in LDL size from baseline have been reported with ω3 fatty acid supplementation but either these changes were not significantly different from the changes observed with placebo[45,46] or no placebo treatment was included.[47] Thus, the bulk of the literature suggests that ω3 fatty acids do not alter LDL size differently from ω6 fatty acids, and therefore, if the former do indeed have anti-atherogenic properties,[48,49] changing LDL size may not be one of them.

The increase in plasma LDL cholesterol has been noted repeatedly in fish oil fed patients.[16,50] Although the mechanism has been studied in human and animal experiments, a clear explanation has yet to emerge. Obviously, it must be due to either decreased clearance of LDL or increased production of LDL (either directly from the liver or indirectly from VLDL). Hepatic LDL receptor activity is decreased in Syrian hamsters[51–53] and miniature pigs[54] fed fish oil, and plasma LDL cholesterol levels have been negatively correlated with hepatic LDL receptor binding activity,[53] suggesting that ω3 fatty acids may depress LDL receptor activity. Others have reported that LDL receptor activity in rats actually is increased by fish oil feeding.[55] Whether ω3 fatty acids alter LDL receptor activity in humans is not known, but LDL kinetic studies showed no decrease in fractional catabolic rate relative to a high saturated fat diet[56] or a vegetable oil diet,[44] implying that in humans, ω3 fatty acids may not affect LDL removal mechanisms.

It has been reported that small VLDL (vs. large VLDL) are preferentially converted to LDL.[19,57,58] In accordance with previous studies, these fatty acids affected primarily

the largest VLDL particles, with little to no impact on the smaller VLDL subspecies.[57,59] If ω3 fatty acid feeding increased levels of small VLDL while decreasing levels of large ones, then the increase in LDL would not be unexpected. However, we did not find increases in smaller VLDL subspecies with fish oil feeding either here or in a previous study using gel chromatography.[59] Nevertheless, it is possible that ω3 fatty acids could enhance the conversion of VLDL to LDL via other mechanisms.[60–62] Accelerated conversion could be due to increased inherent susceptibility of VLDL particles to lipolysis (the mechanism under investigation here) or to increased in vivo lipolytic activity, or both. The latter is supported by observations that heparin injection (which markedly stimulates LpL activity) transiently raises LDL levels,[63] and acute inhibition of LpL[66] or HL[67] likewise reduces LDL levels. These data suggest that LDL production is controlled to some extent by lipolytic enzyme activity. Although several previous studies have not found any effect of ω3 fatty acids on postheparin LpL activity,[3,13,14] preheparin LpL activity was increased in the six HTG patients examined here.[22] Thus, it is possible that these fatty acids affect both particle susceptibility to lipolysis and lipolytic activity.

We found that VLDL derived from ω3 fatty acid-fed subjects was inherently more susceptible to conversion to LDL, at least in vitro. The mechanism responsible for accelerated conversion is unknown. One possibility is enhanced binding to LpL. This was suggested by a recent study in which increased margination (binding to LpL at the endothelium) was inferred from a kinetic analysis of chylomicron catabolism in ω3 fatty acid-fed rats.[68] If LpL bound more tightly to FOC VLDL than control VLDL, then it might stay with the particle longer, producing a smaller remnant. This also might explain why we found equivalent free fatty acid release during incubation of control and FOC VLDL. LpL may release smaller amounts of free fatty acids from a larger number of control particles and larger amounts of free fatty acids from a smaller number of FOC particles. Hypothetically, the modified VLDL fatty acid composition could affect binding of LpL to VLDL. We found that ω3 fatty acid levels in VLDL-TG and VLDL-PL increased by sevenfold and threefold, respectively, during the ω3 fatty acid period. Precisely how altered fatty acid composition could affect susceptibility to conversion to LDL is not known, but changes in apolipoprotein composition and other potential mechanisms are currently under investigation.

Not unexpectedly, fish oil lowered plasma TG and VLDL cholesterol levels consistent with many previous studies.[8,9,69,70] The mechanism of the hypotriglyceridemic effect has long been thought to be reduced hepatic VLDL secretion.[4,9,12,71–73] Contributing to the evidence for this mechanism were reports that fish oil treatment did not stimulate postheparin LpL activity in most but not all[74] human trials, and it either lowered,[54,75,76] raised,[77–79] or did not affect[80] LpL activity in animal studies. Recent data from our laboratory suggest that chylomicron TG clearance is accelerated in rats by pretreatment with ω3 fatty acids[68] and preheparin lipase activity is stimulated.[22] Thus, the effects of ω3 fatty acids on in vivo TG lipolysis remain unclear.

Conclusion

From these investigations that FOC VLDL, while not more susceptible to in vitro lipolysis, appears to be more readily converted to LDL than control VLDL. Although much data support the hypothesis that ω3 fatty acids reduce VLDL synthesis, these observations argue for an additional effect on catabolism. Studies of LDL and VLDL turnover in HTG patients treated with ω3 fatty acids will be needed to clearly demonstrate the mechanism by which these fatty acids alter lipoprotein levels.

Acknowledgments

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