

# Effects of Polyunsaturated Omega-3 Fatty Acids on Responsiveness to Dual Antiplatelet Therapy in Patients Undergoing Percutaneous Coronary Intervention

## The OMEGA-PCI (OMEGA-3 Fatty Acids After PCI to Modify Responsiveness to Dual Antiplatelet Therapy) Study

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- Objectives** The purpose of this study was to investigate whether omega-3 polyunsaturated fatty acids (PUFAs) are able to modify platelet responsiveness to dual antiplatelet therapy in stable coronary artery disease patients undergoing percutaneous coronary intervention (PCI).
- Background** Although previous studies have suggested antiplatelet properties of omega-3 polyunsaturated fatty acids, it is unknown whether they can enhance platelet inhibition on standard aspirin and clopidogrel treatment.
- Methods** The OMEGA-PCI (OMEGA-3 Fatty Acids After PCI to Modify Responsiveness to Dual Antiplatelet Therapy) study was an investigator-initiated, prospective, single-center, double-blind, placebo-controlled, randomized study. Patients receiving standard dual antiplatelet therapy (aspirin 75 mg/day and clopidogrel 600 mg loading dose followed by 75 mg/day) were randomly assigned to receive the addition of 1 g of omega-3 ethyl esters (n = 33) or placebo (n = 30) for 1 month. Platelet function was measured serially by light transmission aggregometry (adenosine diphosphate and arachidonic acid [AA] were used as agonists) and assessment of the phosphorylation status of the vasodilator-stimulated phosphoprotein at baseline, 12 h, 3 to 5 days, and 30 days after randomization.
- Results** The P2Y<sub>12</sub> reactivity index was significantly lower, by 22.2%, after 1 month of treatment with omega-3 polyunsaturated fatty acids compared with placebo when used in addition to dual antiplatelet therapy (p = 0.020). Maximal platelet aggregation induced by 5 and 20 μmol/l adenosine diphosphate was lower by 13.3% (p = 0.026) and 9.8% (p = 0.029), respectively, after 1 month of treatment with omega-3 polyunsaturated fatty acids compared with placebo. Platelet aggregation after AA stimulation was low and did not change significantly throughout the study. There were no cases of aspirin resistance during follow-up that was suggestive of good compliance with the medication.
- Conclusions** The addition of omega-3 ethyl esters to the combination of aspirin and clopidogrel significantly potentiates platelet response to clopidogrel after percutaneous coronary intervention. (J Am Coll Cardiol 2010;55:1671-8)  
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Acetylsalicylic acid has a relatively predictable effect on platelet aggregation with a moderate incidence of aspirin

resistance, partially associated with inadequate compliance (1). In contrast to aspirin, response to clopidogrel is more variable and the incidence of low responsiveness to the drug is much higher (2,3). Clopidogrel hyporesponsiveness is associated with poor compliance, gene polymorphisms, intrinsic high platelet reactivity, variability of the drug metabolism, and various drug interactions (4,5). Patients who are nonresponders to clopidogrel, particularly those having persistent high on-treatment platelet reactivity to adenosine diphosphate (ADP), have been shown to be at

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**Abbreviations  
and Acronyms**

<b>AA</b> = arachidonic acid
<b>ADP</b> = adenosine diphosphate
<b>PRI</b> = P2Y <sub>12</sub> reactivity index
<b>PCI</b> = percutaneous coronary intervention
<b>PGE<sub>1</sub></b> = prostaglandin E <sub>1</sub>
<b>PUFA</b> = polyunsaturated fatty acid
<b>VASP</b> = vasodilator-stimulated phosphoprotein

higher risk of post-percutaneous coronary intervention (PCI) ischemic events (1,4,6,7).

There is no reliably documented method for overcoming clopidogrel hyporesponsiveness when noncompliance or drug interactions have been excluded (3,8–10). Because platelet inhibition by clopidogrel is dose dependent, an increase in either the loading or the maintenance dose results in enhanced responsiveness to the drug (3,7,8). Recently, triple antiplatelet therapy with the addition of cilostazol

has also been reported to improve the biological effects of clopidogrel (9,10). The advantages of such therapies must be weighed against the potential to increase the risk of bleeding.

Increased consumption of oily fish or supplementation with ethyl esters of omega-3 polyunsaturated fatty acids (PUFAs) is currently recommended for secondary prevention of cardiovascular events, especially after acute myocardial infarction (11). The mechanisms of the beneficial effect of omega-3 PUFAs on cardiovascular disease are multifactorial and remain unclear. Both anticoagulant and antiplatelet actions of omega-3 PUFAs have been described. The antiplatelet effect of omega-3 PUFAs was explored in the 1980s and 1990s with inconsistent findings (12,13). Currently, it is not clear whether administration of omega-3 PUFAs affects platelet reactivity in patients already treated with dual antiplatelet therapy after PCI or myocardial PUFAs are able to modify platelet responsiveness to dual antiplatelet therapy in stable coronary artery disease patients undergoing PCI.

## Methods

**Subjects.** Consecutive patients with stable coronary artery disease (age range 30 to 80 years) undergoing PCI with stent implantation were eligible for the study. Exclusion criteria were as follows: acute coronary syndrome, bleeding, concomitant chronic anticoagulant therapy, thienopyridine use before enrollment, platelet count  $<100 \times 10^9/l$ , serum creatinine  $>177 \mu\text{mol/l}$  (2 mg/dl), and liver injury (alanine transaminase level  $>1.5$  times above the upper limit of the reference range). Patients were advised against the use of nonsteroidal anti-inflammatory drugs.

The OMEGA-PCI (OMEGA-3 Fatty Acids After PCI to Modify Responsiveness to Dual Antiplatelet Therapy) study was an investigator-initiated, prospective, single-center, double-blind, placebo-controlled, randomized study. Patients were eligible after elective coronary angiography (Fig. 1). All subjects self-reported regular intake of aspirin for at least 14 days. Clopidogrel in a 600-mg loading dose,

was given after blood collection for baseline measurements 12 h before PCI. Patients were then randomized in the 1:1 ratio using computerized random-number generation by an independent investigator on a double-blind basis to receive 75 mg clopidogrel once daily, 75 mg aspirin once daily, and 1,000 mg omega-3 PUFAs once daily (Omacor, Pronova Biocare AS/Solvay Pharma, Warsaw, Poland) or 75 mg clopidogrel once daily, 75 mg aspirin once daily, and placebo for 4 weeks (Fig. 1). Study medication was started together with the clopidogrel loading dose and continued daily thereafter. Omega-3 PUFAs were administered in a preparation containing ethyl esters of 460 mg eicosapentaenoic acid and 380 mg docosahexaenoic acid. Compliance with medication use was assessed from the number of tablets returned at each visit in both groups. Patients were provided with aspirin and clopidogrel at discharge to ensure compliance. The last tablets were taken in the presence of an investigator 2 to 4 h before blood sampling. Patients were also asked to comply with a diet recommended by European Society of Cardiology that encouraged increased consumption of the oily fish (11).

Platelet function assessment was performed at 4 time points: before clopidogrel and study medication loading (I); 12 h after the loading dose and immediately before PCI (II); 3 to 5 days after PCI (III), and 1 month after randomization (IV) (Fig. 1). Laboratory assays were combined with clinical evaluation. The occurrence of both ischemic and bleeding events was recorded.

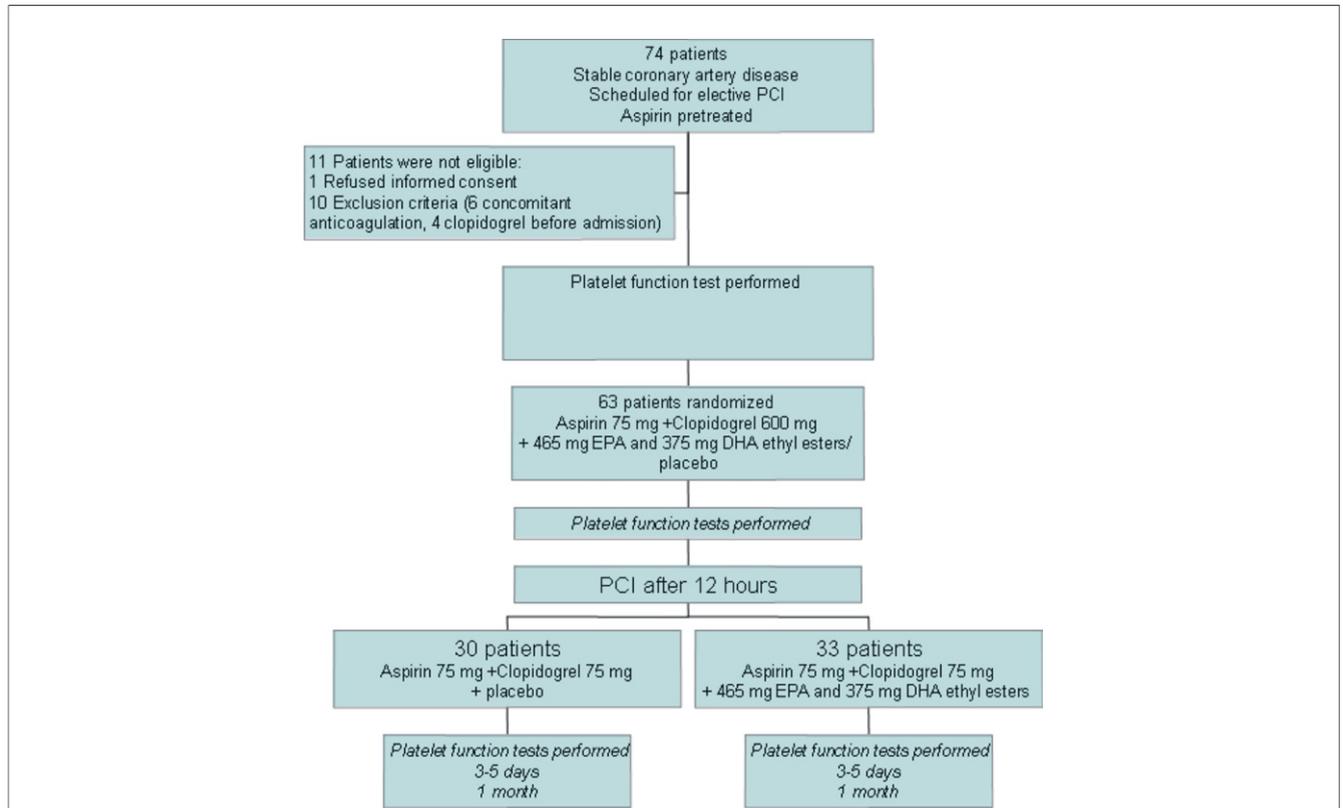
The study complied with the Guidelines of Good Clinical Practice (GCP) of the International Conference on Harmonisation (ICH) and was approved by the Ethics Committee of the Jagiellonian University. Each patient provided written informed consent before entering the study.

**End points of the study and sample size calculation.** The primary end point of the study was P2Y<sub>12</sub> reactivity index (PRI) assessed 30 days after randomization (at study time point IV). Secondary end points of the study included PRI at study time points II and III and maximal platelet aggregation induced by 5 and 20  $\mu\text{mol/l}$  ADP and by 0.5 mmol/l AA throughout the study.

We hypothesized that the addition of omega-3 PUFAs to dual antiplatelet therapy compared with placebo would result in an absolute 10% reduction of the primary end point of the study with an SD of the differences between the 2 groups of 16%. Choosing a power of 90% and a 2-sided alpha level of 0.05, at least 29 patients in each group were required.

**Sample collection.** Blood samples obtained from an antecubital vein were immediately drawn to the tubes containing 3.2% trisodium citrate. Samples were processed 30 to 60 min after blood collection.

**Light transmission aggregometry.** Blood-citrate Vacutainer tubes were centrifuged at 120 *g* for 10 min to recover platelet-rich plasma and subsequently centrifuged for another 10 min at 850 *g* to obtain platelet-poor plasma.



**Figure 1** Flow Diagram of the OMEGA-PCI Study

A prospective, randomized, double-blind, placebo-controlled study was undertaken in 63 patients undergoing elective percutaneous coronary intervention (PCI). DHA = docosahexaenoic acid; EPA = eicosapentaenoic acid.

Platelets were stimulated with 0.5 mmol/l AA, and 5 and 20  $\mu$ mol/l ADP. Aggregation was measured with the 2-channel Chronolog Aggregometer (Chrono-Log 490; Chrono-Log Corp., Haverton, Pennsylvania). Light transmission was adjusted using platelet-rich plasma (PRP) to represent 0% and platelet-poor plasma (PPP) to represent 100% transmission for each measurement. Curves were recorded for 6 min. Platelet aggregation was expressed as the maximal percentage of change in light transmission from baseline using platelet-poor plasma as reference.

**PRI.** The PRI was calculated by assessment of the phosphorylated status of vasodilator-stimulated phosphoprotein (VASP) according to standard protocols and a standardized assay (Platelet VASP, Biocytex Inc., Asnieres, France). The analyses were performed within 6 h after blood sampling. A citrated blood sample was incubated with prostaglandin E<sub>1</sub> (PGE<sub>1</sub>) and PGE<sub>1</sub> + ADP for 10 min. Afterward, the sample was fixed with paraformaldehyde and nonionic detergent.

Quantitative flow cytometry (FACSCalibur System, BD Bioscience, Warsaw, Poland) was used with labeled monoclonal antibodies against serine 239-phosphorylated VASP, followed by a secondary fluorescein isothiocyanate-conjugated polyclonal goat anti-mouse antibody. The mean fluorescence intensity of phosphorylated VASP levels after

stimulation with PGE<sub>1</sub> and PGE<sub>1</sub> + ADP was measured. The PRI expressed as a percentage was then calculated according to the formula:  $[(MFI_{PGE_1}) - (MFI_{PGE_1+ADP})] / (MFI_{PGE_1}) \times 100\%$ .

**Definition of nonresponders to aspirin and clopidogrel.** Aspirin resistance was defined based on light transmission aggregometry results (AA-induced aggregation >20%) (1,8). Clopidogrel responsiveness was defined as pre-treatment minus post-treatment aggregation. Nonresponsiveness was characterized as having an absolute difference between baseline and post-treatment maximal aggregation of <10% reduction with 5  $\mu$ mol/l ADP as the agonist (2).

When PRI was used to assess reactivity of the P2Y<sub>12</sub> receptor, patients with a PRI  $\geq$ 69% were considered as low responders to clopidogrel (3).

**Genotyping.** CYP2C19\*2 (681G>A, rs4244285) was genotyped using a commercially available, validated Drug Metabolism Genotyping Assay (TaqMan MGB probes, FAM and VIC dye-labeled) and TaqMan Universal PCR Master Mix with the Fast Real Time System 7900HT and SDS 2.3 software (all supplied by Applied Biosystems Inc., Foster City, California).

**Statistical analysis.** All continuous variables were expressed as mean  $\pm$  SD, and categorical variables were expressed as percentages. The Kolmogorov-Smirnov test

was used to determine normal distribution. The nonparametric Mann-Whitney *U* test was applied to compare both normally and non-normally distributed continuous variables between groups. The Wilcoxon signed rank test was used to assess continuous variables in each group separately at all time points of the study. The chi-square test, with or without the Yates continuity correction, was used to evaluate the differences in categorical variables between the respective study group when appropriate. All statistical tests were 2-sided. Statistical significance was accepted at  $p < 0.05$ . Statistical analysis was performed using the STATISTICA version 6.0 PL software package (StatSoft, Inc., Tulsa, Oklahoma).

## Results

**Demographics and clinical characteristics.** Sixty-three consecutive patients (48 men and 15 women; age  $63.1 \pm 10$  years) with stable coronary artery disease undergoing successful PCI with stent implantation were randomized (Fig. 1). Baseline demographics, clinical characteristics, and concomitant medications of the randomized subjects are shown in Table 1. There were no significant differences among baseline variables, including genetic polymorphism of cytochrome CYP2C19. During the study period, there were no changes in concomitant medications. During follow-up, no cases of aspirin resistance were detected in both patient groups based on light transmission aggregometry results. One patient in the placebo group and 1 patient in the omega-3 PUFA group were not available for the final platelet assessment, but no patients were lost to follow-up in either group.

**Adverse events.** The adverse events that occurred during the study were rated as mild, and their frequency did not differ significantly between groups. The number of patients reporting minor bruising was similar in both groups (6 patients in the omega-3 PUFA group and 5 patients in the placebo group), and they did not require medical intervention. There were no major cardiac adverse events during 1-month follow-up in both groups.

**Platelet investigations.** Platelet count was not different between groups at baseline and during follow-up (Table 2).

**PRI.** The primary end point of the study—the PRI—was significantly lower after 1 month of treatment with omega-3 ethyl esters compared with placebo by 22.2% (95% confidence interval: 2.93 to 21.87) (Fig. 2). Compared with baseline, platelet aggregation assessed with PRI was decreased in both treatment arms at all time points of the study. The primary end point of the study was significantly decreased compared with baseline values by 26.2% ( $p < 0.0005$ ) and 40.6% ( $p < 0.0005$ ) in the placebo and omega-3 PUFA groups, respectively. At the end of follow-up, there were 26.7% patients in the placebo group and 6.1% in the omega-3 group who were determined to be low responders to clopidogrel based on the PRI ( $p = 0.059$ ).

**Platelet aggregometry.** Compared with placebo, the addition of omega-3 PUFAs to standard post-PCI dual anti-

platelet therapy was associated with decreased maximal platelet aggregation induced by  $5 \mu\text{mol/l}$  ADP after 1 month of treatment by 13.3% (95% confidence interval: 1.24 to 12.16) (Table 2). Similarly, in the omega-3 PUFA group compared with the placebo group, the maximal platelet aggregation induced by  $20 \mu\text{mol/l}$  ADP was lower after 1 month by 9.8% (95% confidence interval: 0.67 to 10.33) (Fig. 3). Based on the results of light transmission aggregometry after challenge with ADP, clopidogrel resistance could be detected in 43.3% patients in the placebo group and 18.2% in the omega-3 PUFA group at the end of the study ( $p = 0.030$ ).

Platelet aggregation after stimulation with AA was markedly inhibited in both groups. There was 1 case of aspirin resistance in the omega-3 PUFA group at baseline. After introduction of the study treatment, that patient was no longer resistant to aspirin. No cases of aspirin resistance were detected during follow-up in both groups of patients, which confirmed that patients were compliant with the medication. At the end of the study, there was a trend toward higher platelet inhibition after omega-3 PUFA versus placebo treatment that reached borderline significance ( $p = 0.094$ ) (Table 2).

## Discussion

Our study indicates that omega-3 PUFAs added to standard dual antiplatelet therapy after PCI can influence platelet response to clopidogrel based on the results of 2 assays (platelet aggregometry in PRP and VASP phosphorylation) that assess various pathways of platelet activation. Triple therapy with aspirin, clopidogrel, and omega-3 PUFAs compared with standard dual antiplatelet therapy impaired platelet aggregation when stimulated by ADP and decreased intracellular signaling from the P2Y<sub>12</sub> receptor as assessed by VASP phosphorylation. The relatively high on-treatment platelet aggregation induced by ADP or assessed by PRI observed in our study could be associated with a high prevalence of diabetes, hyperlipidemia, and abdominal obesity in our patients. In the cyclooxygenase pathway of platelet activation, we found inhibited platelet aggregation after AA stimulation with a borderline effect of omega-3 PUFAs.

We used 2 separate assays for platelet function assessment because there is no consensually recommended test to quantify platelet inhibition by clopidogrel. Lordkipanidzé et al. (14) demonstrated recently that there is no strong correlation among various platelet function assays (aggregometry, PFA-100, and VerifyNow P2Y<sub>12</sub> [Accumetrics, San Diego, California]). Light transmission aggregometry is considered by many as the current gold standard in platelet function testing (14). Assessment of the phosphorylated status of VASP has also been shown to correlate with clinical outcomes (3,6,7).

There have been several clinical studies in which it was possible to demonstrate an augmented effect of thienopyri-

<b>Table 1 Demographics, Clinical Characteristics, and Concomitant Medications</b>			
	<b>Placebo (n = 30)</b>	<b>Omega-3 (n = 33)</b>	<b>p Value</b>
Age, yrs	63.8 ± 9.4	62.4 ± 9.7	0.550
Male	24 (80.0)	24 (72.7)	0.499
<b>Risk factors</b>			
Hypertension	29 (96.7)	32 (97.0)	0.515
Hyperlipidemia	29 (96.7)	32 (97.0)	0.515
Diabetes	9 (30.0)	9 (27.3)	0.811
Metabolic syndrome	22 (73.3)	24 (72.7)	0.957
Waist circumference, cm	102.4 ± 8.5	98.6 ± 11.0	0.112
Body mass index, kg/m <sup>2</sup>	29.3 ± 3.8	28.9 ± 4.2	0.524
History of cigarette smoking	16 (53.3)	15 (45.5)	0.532
Active smoking	6 (20.0)	5 (15.2)	0.613
Family history	18 (60.0)	23 (69.7)	0.420
<b>Medical history</b>			
Previous MI	12 (40.0)	9 (27.3)	0.285
Multivessel CAD	18 (60.0)	27 (81.8)	0.056
Previous PCI	7 (23.3)	5 (15.2)	0.409
Previous CABG	1 (3.3)	6 (18.2)	0.141
Previous stroke or PAD	11 (36.7)	8 (24.2)	0.283
Chronic kidney disease	10 (33.3)	5 (15.2)	0.091
Multivessel PCI	1 (3.3)	4 (12.1)	0.411
<b>Lesion location</b>			
LAD	13 (43.3)	16 (48.5)	0.682
LCx	12 (40.0)	8 (24.2)	0.180
RCA	5 (16.7)	12 (36.4)	0.079
Reference vessel diameter, mm	3.1 ± 0.5	3.2 ± 0.5	0.367
<b>Stent type</b>			
Bare-metal	23 (76.7)	23 (69.7)	0.534
Drug-eluting	7 (23.3)	10 (30.3)	0.534
No. of stents	1.3 ± 0.6	1.3 ± 0.5	0.912
Total stent length, mm	23.4 ± 15.3	24.7 ± 9.1	0.182
<b>Periprocedural MI</b>			
Maximal troponin level, ng/ml	2.83 ± 3.82	1.37 ± 0.67	0.724
<b>Medications</b>			
GP IIb/IIIa inhibitors	0	0	—
Beta-blocker	25 (83.3)	29 (87.9)	0.877
ACE inhibitors	27 (90.0)	28 (84.8)	0.815
ARB	2 (6.7)	7 (21.2)	0.198
Nitrates	12 (40.0)	14 (42.4)	0.845
Calcium antagonists	16 (53.3)	13 (39.4)	0.268
CYP3A4-metabolized statins	27 (90.0)	31 (93.9)	0.912
Non-CYP3A4-metabolized statins	0	0	—
Fibrates	2 (6.7)	0	0.431
Insulin	3 (10.0)	2 (6.1)	0.912
Proton pump inhibitors	8 (26.7)	8 (24.2)	0.825
<b>Genotype distribution</b>			
<b>G681A CYP2C19 polymorphism</b>			
GG	21 (70)	23 (69.7)	0.979
GA	9 (30)	9 (27.3)	0.811
AA	0	1 (3)	0.962
G allele frequency	51 (85)	55 (83.3)	0.798
<b>Laboratory data</b>			
Glucose, mmol/l	5.7 ± 1.8	5.1 ± 0.7	0.186
Hemoglobin, g/dl,	14.3 ± 1.3	14.1 ± 1.3	0.882
WBCs, ×10 <sup>9</sup> /l	7.15 ± 1.62	7.36 ± 2.02	0.563
Platelet count, ×10 <sup>9</sup> /l	313.9 ± 78.3	331.6 ± 89.1	0.280
C-reactive protein, mg/l	2.06 ± 1.56	2.09 ± 1.52	0.337

Values are expressed as mean ± SD or n (%).

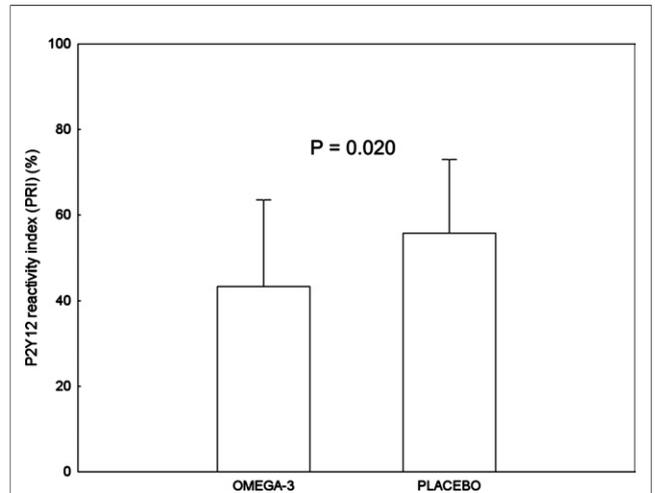
ACE = angiotensin-converting enzyme; ARB = angiotensin II receptor blocker; CABG = coronary artery bypass graft; CAD = coronary artery disease; LAD = left anterior descending artery; LCx = left circumflex artery; MI = myocardial infarction; PAD = peripheral artery disease; PCI = percutaneous coronary intervention; RCA = right coronary artery; WBC = white blood cell.

**Table 2** Changes in Platelet Count and Aggregation Over Time in Placebo and Omega-3 PUFA Groups

Platelet Studies	Group		p Value
	Placebo	Omega-3 PUFAs	
<b>Platelet count, %</b>			
Baseline	313.8 ± 78.0	331.6 ± 88.5	0.275
12 h	317.6 ± 108.9	353.1 ± 105.3	0.098
3-5 days	314.5 ± 98.4	294.8 ± 71.3	0.561
1 month	327.1 ± 110.6	357.1 ± 86.5	0.371
<b>PRI, %</b>			
Baseline	75.6 ± 20.0	73.1 ± 19.0	0.784
12 h	61.4 ± 20.4	50.2 ± 18.8	0.022
3-5 days	49.4 ± 14.5	46.7 ± 13.9	0.379
1 month	55.8 ± 17.2	43.4 ± 20.1	0.020
<b>LTA ADP, 5 μmol/l</b>			
Baseline	62.2 ± 9.6	62.4 ± 9.0	0.893
12 h	47.3 ± 10.4	44.8 ± 11.4	0.268
3-5 days	47.7 ± 13.4	42.2 ± 11.9	0.068
1 month	50.2 ± 12.2	43.5 ± 9.4	0.026
<b>LTA ADP, 20 μmol/l</b>			
Baseline	70.1 ± 8.7	68.8 ± 9.7	0.502
12 h	55.5 ± 12.2	52.3 ± 11.7	0.293
3-5 days	56.4 ± 16.9	50.7 ± 11.1	0.023
1 month	56.3 ± 11.5	50.8 ± 7.8	0.029
<b>LTA AA, 5 mmol/l</b>			
Baseline	1.1 ± 1.1	1.3 ± 1.2	0.510
12 h	1.1 ± 0.9	1.4 ± 1.7	0.783
3-5 days	1.5 ± 2.4	1.5 ± 1.3	0.639
1 month	2.1 ± 2.3	1.3 ± 1.4	0.094

AA = arachidonic acid; ADP = adenosine diphosphate; LTA = light transmission aggregometry; PRI = P2Y<sub>12</sub> reactivity index; PUFAs = polyunsaturated fatty acids.

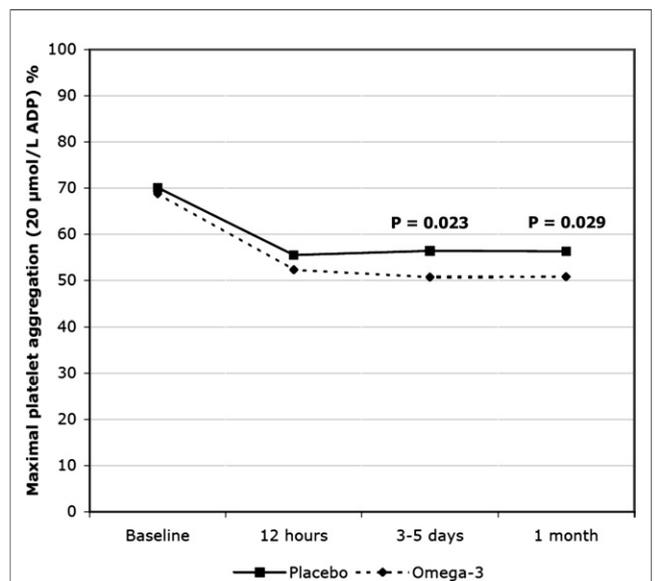
dine treatment. It was demonstrated that adjusting the clopidogrel loading dose, even above 600 mg according to platelet monitoring using the PRI, might significantly improve the clinical outcome after PCI in clopidogrel-resistant patients (7). Optimization of clopidogrel therapy by increasing the clopidogrel maintenance dose was also proven feasible (3,8). Recently, a concept of add-on oral antiplatelet therapy has been presented (9,11,15). Angiolillo et al. (9) showed in 25 diabetic patients with coronary artery disease receiving dual antiplatelet therapy that the PRI was significantly lower by 39% (p = 0.0002) after cilostazol treatment compared with placebo. Shim et al. (10) demonstrated in 400 patients undergoing PCI with drug-eluting stent implantation that adding cilostazol to aspirin and clopidogrel therapy decreased the prevalence of clopidogrel resistance (VerifyNow System, Accumetrics) from 41.4% in the placebo group to 26.5% in the triple antiplatelet therapy group (p < 0.001). Recently, Chen et al. (16) demonstrated that adding cilostazol to standard dual antiplatelet therapy resulted in the lower incidence of major adverse cardiac events in 8 months of follow-up in patients with acute myocardial infarction undergoing primary PCI. In our study, we demonstrated similar, although milder, effects on platelet function profiles with omega-3 ethyl esters. Based on these results, it could be estimated that adding low-dose



**Figure 2** Benefit of Omega-3 PUFAs on the Primary End Point of the OMEGA-PCI Study

Patients in whom omega-3 polyunsaturated fatty acids (PUFAs) were added to standard therapy with aspirin and clopidogrel had greater platelet inhibition when assessed by the P2Y<sub>12</sub> reactivity index (PRI) compared with placebo after 1 month of treatment. Error bars indicate SDs of the mean.

omega-3 PUFAs was responsible for 55% to 60% of the effect achieved with cilostazol demonstrated in the studies by Angiolillo et al. (9) and Shim et al. (10). Conversely, the effectiveness of triple therapy with omega-3 PUFAs was only slightly weaker compared with aspirin/high-dose



**Figure 3** Maximal Platelet Aggregation Induced by 20 μmol/l ADP Over Time in the Placebo and Omega-3 Groups

Potentiated platelet response to clopidogrel after percutaneous coronary intervention was demonstrated in patients receiving triple therapy with aspirin, clopidogrel, and omega-3 polyunsaturated fatty acids compared with standard dual antiplatelet therapy. Statistically significant maximal platelet aggregation between placebo and omega-3 groups at each time point is indicated. ADP = adenosine diphosphate.

clopidogrel maintenance therapy in the VASP-02 study (relative PRI reductions of 22.2% and 25.1%, respectively) (3).

Omega-3 PUFAs have been known for their antiplatelet and anticoagulant effects, although in higher doses than were used in our study (12,13). Recently, Larson et al. (15) demonstrated in 10 healthy volunteers using whole-blood impedance aggregometry that although omega-3 PUFAs alone were not able to change platelet aggregation, they facilitated aspirin-induced platelet inhibition. The mechanisms of favorable antithrombotic effects of omega-3 PUFAs are complex. It has been shown that alteration of fatty acid composition by omega-3 PUFA incorporation into platelet membranes can alter not only membrane permeability, but also modulate function and activity of membrane receptors and transporters (12,13). Omega-3 PUFAs may also lower thromboxane A<sub>2</sub> production because epoxidation and hydroxylation of both AA and PUFAs are catalyzed by cytochrome P-450 (17). Moreover, Engström et al. (18) found that fish oil administered with aspirin augments its effect by further decreasing thromboxane A<sub>2</sub> production and by counteracting the aspirin-induced unfavorable decrease in circulating prostacyclin (prostaglandin I<sub>2</sub>) and increase in leukotriene levels. Although the concept of more aggressive antiplatelet treatment based on the results of platelet function tests is feasible, the optimal degree of platelet inhibition is unclear and must be confirmed in trials evaluating cardiovascular outcomes and should be balanced with the excessive risk of bleeding (19). Omega-3 ethyl esters at a dose that was tested in the OMEGA-PCI study have previously shown efficacy and a good safety profile in patients after acute myocardial infarction (20) and patients with chronic heart failure (21). A 1-g dose of omega-3 PUFAs has been previously considered not to affect blood coagulation (12,13,15). The present study is the first to address the concept of modifying antiplatelet response with the addition of omega-3 ethyl esters to aspirin and clopidogrel therapy in patients with stable coronary artery disease undergoing PCI.

**Study limitations.** Because the present study was designed only to assess the effect on platelet responsiveness of adding omega-3 PUFAs to standard post-PCI therapy, the sample size lacked statistical power to detect differences in clinical outcomes. This issue was beyond the scope of the current study.

It would be useful to perform concomitant platelet membrane fluidity studies because lipophilic membranes can be targeted by omega-3 PUFAs, thus improving response to the antiplatelet drugs.

The dose of omega-3 PUFAs selected for the study was based on the applicability because it is recommended in cardiovascular disease secondary prevention. Higher platelet inhibition might be achieved with increased doses of omega-3 PUFAs.

## Conclusions

The addition of omega-3 ethyl esters to the standard dual antiplatelet therapy with aspirin and clopidogrel significantly potentiates platelet response to clopidogrel after PCI.

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**Key Words:** aspirin ■ clopidogrel ■ coronary stenting ■ omega-3 fatty acids.