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ARTICLE

Add-on ezetimibe treatment to low-dose statins vs medium-intensity statin monotherapy in coronary artery disease patients with poorly controlled dyslipidemia

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Abstract

Although ezetimibe has potential value as an add-on therapy to statins, it is not established whether the addition of ezetimibe to statin therapy is more effective than double-dose statin monotherapy. We conducted a crossover design study. Twenty-one coronary artery disease (CAD) patients whose lipid profiles had not achieved Japanese guideline recommendations (JAS 2017), despite receiving low-dose statin therapy, were divided into two groups. Group A received ezetimibe 10 mg in addition to the baseline dose of statin for the first 3 months and was then switched to monotherapy with a double dose of statin for the next 3 months. Group B first received a double dose of statin for 3 months and was then switched to ezetimibe 10 mg in addition to a baseline dose of statin for the next 3 months. Compared with the baseline, double-dose statin therapy reduced low-density lipoprotein (LDL)-cholesterol (from 118 ± 22 to 104 ± 15 mg/dL, $P < 0.05$) and malondialdehyde-modified LDL (MDA-LDL) (from 142 ± 35 to 126 ± 24 U/L, $P < 0.05$) but did not lower high-sensitivity C-reactive protein (hsCRP) (3.02 ± 0.47 and 2.98 ± 0.41 log [ng/ml]). The addition of ezetimibe to a baseline dose of statin further reduced LDL-cholesterol (to 89 ± 15, $P < 0.0001$) and MDA-LDL (to 114 ± 22 U/L, $P < 0.001$) and reduced hsCRP (to 2.78 ± 0.38 log (ng/ml), $P < 0.05$). The changes in the levels of MDA-LDL ($R = 0.548$, $P = 0.010$) and hsCRP ($R = 0.473$, $P < 0.05$) were significantly correlated with the change in the LDL-cholesterol level after the addition of ezetimibe. Add-on ezetimibe treatment appears superior to double-dose statin therapy in CAD patients with poorly controlled dyslipidemia in terms of reductions in LDL-cholesterol level, lipid peroxidation, and inflammation.

Keywords Ezetimibe · Low-density lipoprotein-cholesterol · Malondialdehyde-modified-low-density lipoprotein · Oxidative stress · Inflammation

Introduction

The 3-hydroxy-3-methylglutaryl-coenzyme A reductase inhibitors (statins) reduce cardiovascular events in terms of both primary and secondary prevention [1, 2]. The reduction in cardiovascular risk by statins depends not only on strong lipid-lowering effects but also on direct cardiovascular protective effects, including improvement in vascular endothelial functions, anti-inflammatory actions, and antioxidant effects [3]. Intensive statin therapy lowers low-density lipoprotein (LDL)-cholesterol levels and cardiovascular events more effectively than does standard statin therapy [4–6]. On the other hand, ezetimibe, targeting Niemann-Pick C1-like 1 protein and thereby reducing cholesterol absorption from the intestine, also achieves a further reduction in LDL-cholesterol when added to statins [7, 8]. The Improved Reduction of Outcomes: Vytorin Efficacy International Trial (IMPROVE-IT) showed that ezetimibe 10 mg/day combined with simvastatin 40 mg/day improved cardiovascular outcomes compared with the

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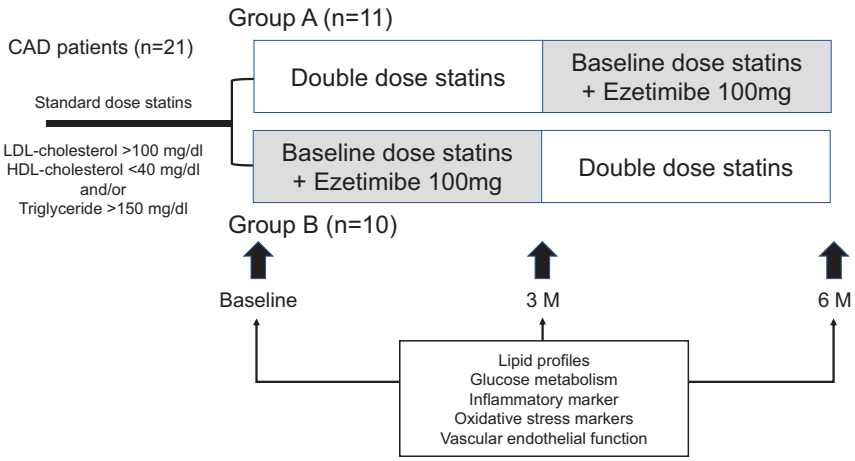

Fig. 1 Study protocol

effects of simvastatin 40 mg/day monotherapy in stable patients, who had acute coronary syndrome and whose LDL-cholesterol levels were within guideline recommendations [9]. Recently, a Japanese trial, HIJ-PROPER [10], demonstrated that pitavastatin 2 mg/day plus ezetimibe 10 mg/day targeting LDL-cholesterol of 70 mg/dl showed no more cardiovascular benefit than pitavastatin monotherapy (1–4 mg/day) targeting LDL-cholesterol of 90–100 mg/dl in acute coronary syndrome patients with dyslipidemia. In this trial, however, pitavastatin plus ezetimibe might have been more effective than pitavastatin monotherapy in patients with higher cholesterol absorption. Although the addition of ezetimibe to statin therapy has a potential advantage over statin monotherapy, there is little information regarding the effects of add-on ezetimibe.

This study was designed to compare the effects on lipid profiles, inflammatory status, oxidative stress status, and endothelial function of add-on therapy with ezetimibe to double-dose statin monotherapy in coronary artery disease (CAD) patients with poorly controlled dyslipidemia despite low-dose statin monotherapy.

Methods

Subjects and study design

We conducted a crossover study. Twenty-one patients with chronic CAD whose lipid profiles had not achieved Japanese guideline recommendations (JAS 2017) (i.e., LDL-cholesterol >100 mg/dl, high-density lipoprotein (HDL)-cholesterol <40 mg/dl and/or triglyceride >150 mg/dl), despite receiving low-dose statin therapy, were randomly divided into two groups. Group A received ezetimibe 10 mg/day in addition to a baseline dose of statins for the first 3 months and was then switched to a double-dose of statins for the next 3 months. Group B initially received a double-dose of statins for 3 months and was then switched to ezetimibe 10 mg/day in addition to a baseline dose of statins for the next 3 months. The study protocol is summarized in Fig. 1. Lipid profiles, glucose metabolism, inflammatory and oxidative stress status, and vascular endothelial function were assessed at baseline before the treatment and at the end of each treatment period. The study protocols were approved by the Ethics Committees of Dokkyo Medical University Hospital. Written informed consent was obtained from all participants.

Assessment of lipid profiles, glucose metabolism, inflammatory and oxidative stress status, and vascular endothelial function

Serum levels of LDL-cholesterol and high-density lipoprotein (HDL)-cholesterol were determined by a homogenous assay. Triglyceride levels were measured using an enzymatic technique. Apolipoprotein (apo) A-I and apo B were determined by validated electroimmunoassays [11, 12], and the apo B/apo A-I ratio was calculated. Malondialdehyde-modified LDL (MDA-LDL) was measured using a sandwich enzyme-linked immunosorbent assay [13]. Glycosylated hemoglobin A1c (HbA1c) was measured by high-performance liquid chromatography. Fasting blood glucose was assayed using the glucose oxidase method and serum insulin was measured by radioimmunoassay. Insulin resistance was assessed using homeostasis model assessment (HOMA-R) according to the following formula: fasting plasma glucose (mg/dl) × fasting plasma insulin (μU/ml)/405 [14].

As an inflammatory biomarker, high sensitivity C-reactive protein (hsCRP) was measured by particle-enhanced technology on a Behring BN II nephelometer (Dade Behring Inc., Newark, DE, USA) using monoclonal anti-CRP antibodies and a calibrator that was traceable to WHO reference material [15]. To evaluate the oxidative

stress status, we measured the levels of thiobarbituric acid reactive substances (TBARS) using a Colorimetric TBARS Microplate Assay Kit (Oxford Biomedical Research, Rochester Hills, MI, USA) in accordance with the manufacturer's instructions [16]. We assessed oxidative stress by measuring the derivative of reactive oxygen metabolites (d-ROMs) level [17]. The d-ROM test evaluates free radical activity by measuring the serum hydroperoxide levels (Diacron, Grosseto, Italy). The results of the d-ROM test are expressed in arbitrary Caratelli units (U. CARR), where 1 U. CARR corresponds to 0.08 mg/100 ml hydroperoxides [18]. A cholesterol synthesis marker, lathosterol, and cholesterol absorption markers, campesterol and sitosterol, were measured by gas–liquid chromatography [19]. The circulating level of proprotein convertase subtilisin/kexin type 9 (PCSK9) was measured using a commercial enzyme-linked immunosorbent assay kit (Medical & Biological Laboratories Co., Ltd., Nagoya, Japan) according to the manufacturer's instructions [20].

We assessed vascular endothelial function using simultaneous flow-mediated dilatation (FMD) and reactive hyperemia-peripheral arterial tonometry (RH-PAT), according to the method described by Tomiyama et al. [21]. FMD measurements were performed using UNEXEF18G (UNEX, Co, Nagoya, Japan), an ultrasound instrument specialized for FMD measurement. For the RH-PAT procedure, we used EndoPAT-2000 (Itamar Medical Ltd., Caesarea, Israel) and calculated the reactive hyperemia index (RHI).

Statistical analysis

Data were expressed as the mean ± standard deviation or median and interquartile range. Normality for the distribution of continuous variables was assessed using the Shapiro–Wilk test. Values with a positively skewed distribution were logarithmically transformed before analysis. Then, intra- and intergroup comparisons for continuous variables were performed using paired and unpaired *t* tests, respectively. The intergroup comparison of the categorical variables was performed using the chi-square test. The correlation between two variables was determined by Pearson's correlation coefficient. $P < 0.05$ was considered significant.

Results

The baseline characteristics of groups A ($n = 11$) and B ($n = 10$) are compared in Table 1. Age, gender, body mass index, other risk factors, and basal coronary artery disease were similar in the two groups. Baseline statins were atorvastatin 5 mg/day in two patients (10%), rosuvastatin 2.5 mg/day in four (19%), and pitavastatin 1 mg/day in 15 (71%), and the proportions of each treatment were similar in both groups. The proportion of concomitant medications, such as angiotensin converting enzyme inhibitors or angiotensin receptor blockers, calcium channel blockers, beta blockers, aspirin, P2Y12 inhibitors and antidiabetic agents, were also similar in both groups, and these medications were not changed during the study period in all

Table 1 Baseline characteristics

	Total ($n = 21$)	Group A ($n = 11$)	Group B ($n = 10$)	*P* value
Age; years	66 ± 11	63 ± 12	69 ± 9	0.516
Male gender; *n* (%)	20 (95)	10 (91)	10 (100)	0.329
BMI; kg/m^2	24 ± 4	24 ± 3	24 ± 5	0.916
Other risk factors; *n* (%)				
Hypertension	18 (86)	9 (82)	9 (90)	0.593
Diabetes	8 (38)	5 (45)	3 (30)	0.729
Smoking	15 (71)	7 (64)	8 (80)	0.407
Family history	5 (24)	2 (18)	3 (30)	0.525
Basal CAD; *n* (%)				0.22
AP	4 (19)	3 (27)	1 (10)	
OMI	15 (71)	8 (73)	7 (70)	
CSA	2 (10)	0 (0)	2 (20)	
Baseline statins; *n* (%)				0.22
Atorvastatin 5 mg/day	2 (10)	0 (0)	2 (20)	
Rosuvastatin 2.5 mg/day	4 (19)	3 (27)	1 (10)	
Pitavastatin 1 mg/day	15 (71)	8 (73)	7 (70)	
Concomitant medications; *n* (%)				
ACE inhibitors/ARBs	16 (76)	9 (82)	7 (70)	0.525
Calcium channel blockers	10 (48)	4 (36)	6 (60)	0.174
Beta blockers	9 (43)	6 (55)	3 (30)	0.253
Aspirin	19 (90)	10 (91)	9 (90)	0.447
P2Y12 inhibitors	5 (24)	4 (36)	1 (10)	0.144
Antidiabetic agents	7 (33)	3 (27)	4 (40)	0.536
LDL-cholesterol; mg/dl	118 ± 22	121 ± 20	115 ± 25	0.553
HDL-cholesterol; mg/dl	56 ± 13	56 ± 12	57 ± 14	0.863
Triglyceride; mg/dl	159 ± 96	173 ± 117	145 ± 69	0.516
Apo B/A-I	0.71 ± 0.17	0.71 ± 0.18	0.71 ± 0.17	0.991
MDA-LDL; U/l	142 ± 35	143 ± 41	141 ± 29	0.862
Hemoblobin A1c; %	6.15 ± 0.75	6.03 ± 0.83	6.28 ± 0.66	0.47
HOMA-R	3.65 ± 7.41	5.25 ± 10.13	1.90 ± 1.18	0.313
hsCRP; log (ng/ml)	3.02 ± 0.47	3.07 ± 0.57	2.97 ± 0.36	0.647
TBARS; μmol/l	10.9 ± 4.2	11.9 ± 4.9	9.9 ± 3.4	0.328
d-ROM; U.CARR	333 ± 76	369 ± 57	293 ± 78	0.025
Lathosterol; ng/ml	1.77 ± 1.39	1.44 ± 0.63	2.03 ± 1.85	0.308
Campesterol; ng/ml	5.98 ± 3.72	5.42 ± 2.37	6.53 ± 4.79	0.519
Sitosterol; ng/ml	3.56 ± 2.42	3.06 ± 1.12	4.06 ± 3.25	0.37
PCSK9; ng/ml	251 ± 80	285 ± 90	210 ± 44	0.123
FMD; %	4.07 ± 1.76	4.66 ± 1.82	3.47 ± 1.54	0.133
RHI	2.11 ± 0.57	1.93 ± 0.45	2.32 ± 0.65	0.188

BMI body mass index, *CAD* coronary artery disease, *AP* angina pectoris, *OMI* old myocardial infarction, *CSA* coronary spastic angina, *ACE* angiotensin converting enzyme, *ARB* angiotensin receptor blocker, *LDL* low density lipoprotein, *HDL* high density lipoprotein, *MDA-LDL* malondialdehide-modified low density lipoprotein, apo apolipoprotein, *HOMA-R* homeostasis model assessment insulin resistance, *hsCRP* high sensitivity C-reactive protein, *TBARS* thiobarbituric acid reactive substance, *d-ROM* derivative of reactive oxygen metabolites, *PCSK* proprotein convertase subtilisin/kexin, *FMD* flow-mediated dilatation, *RHI* reactive hyperemia index

patients. The parameters for lipid profiles, glucose metabolism, inflammatory and oxidative stress markers, and vascular endothelial function were also similar in both groups, except that the d-ROM level was higher in group A than in group B.

Body weight, blood pressure, heart rate, values of hepatorenal function tests did not change significantly at 3 months for the crossover point and at 6 months for the end of study period compared with baseline before the study, and these values were similar in both groups A and B at baseline, 3 months and 6 months (Table 2).

Table 3 exhibits the change in each parameter after each treatment with a double dose of statin or additional ezetimibe. The LDL-cholesterol level was reduced significantly after double-dose statin treatment ($P < 0.05$) and more significantly after additional ezetimibe treatment ($P < 0.0001$). The value was significantly lower after treatment with additional ezetimibe than with double-dose statin treatment ($P < 0.01$). MDA-LDL was also reduced significantly after double-dose statin treatment ($P < 0.05$) and more significantly reduced after additional ezetimibe treatment ($P < 0.001$). The value after treatment was significantly lower with additional ezetimibe treatment than with double-dose statin treatment ($P < 0.05$). The Apo B/A-I ratio was reduced significantly after double-dose statin treatment ($P < 0.001$) but less significantly after additional ezetimibe treatment ($P < 0.05$). The triglyceride level was significantly reduced after additional ezetimibe treatment ($P < 0.05$) but not after double-dose statin treatment. The level of hsCRP was significantly reduced after additional ezetimibe treatment ($P < 0.05$) but not after double-dose statin treatment. The cholesterol absorption markers campesterol and sitosterol were significantly reduced after additional ezetimibe treatment ($P < 0.0001$ and $P < 0.001$, respectively) but not after double-dose statin treatment. The value of campesterol after treatment was significantly lower after additional ezetimibe treatment than after double-dose statin treatment ($P < 0.05$). The other parameters did not change after treatment, and the values after treatment were compatible in both the double-dose statin and additional ezetimibe groups.

Table 2 Baseline parameters at each point of baseline, 3 months (cross-over point) and 6 months (end of study period)

	Total ($n = 21$)	Group A ($n = 11$)	Group B ($n = 10$)	P value
Body weight; kg				
Baseline	67 ± 10	67 ± 8	67 ± 12	0.960
3 months	67 ± 10	67 ± 8	67 ± 12	0.998
6 months	67 ± 10	67 ± 8	67 ± 12	0.924
Systolic blood pressure; mmHg				
Baseline	130 ± 15	130 ± 18	130 ± 10	0.952
3 months	128 ± 17	133 ± 16	123 ± 17	0.191
6 months	129 ± 16	134 ± 18	122 ± 14	0.647
Heart rate; /min				
Baseline	64 ± 12	64 ± 14	63 ± 9	0.857
3 months	66 ± 17	62 ± 16	70 ± 17	0.285
6 months	66 ± 20	65 ± 19	67 ± 21	0.771
AST; IU/l				
Baseline	26 ± 13	24 ± 8	28 ± 16	0.501
3 months	27 ± 10	25 ± 8	29 ± 12	0.312
6 months	27 ± 12	26 ± 13	27 ± 12	0.978
ALT; IU/l				
Baseline	27 ± 15	25 ± 15	30 ± 16	0.456
3 months	28 ± 13	25 ± 13	31 ± 12	0.256
6 months	28 ± 19	28 ± 24	29 ± 13	0.971
CK; U/l				
Baseline	123 ± 78	122 ± 69	123 ± 91	0.983
3 months	133 ± 67	138 ± 64	128 ± 74	0.740
6 months	131 ± 73	124 ± 51	138 ± 94	0.670
Creatinine; mg/dl				
Baseline	0.82 ± 0.16	0.77 ± 0.14	0.88 ± 0.17	0.120
3 months	0.83 ± 0.16	0.78 ± 0.14	0.88 ± 0.18	0.157
6 months	0.84 ± 0.17	0.78 ± 0.14	0.90 ± 0.19	0.100

AST aspartate transamiase, *ALT* alanine transaminase, *CK* creatine kinase

In each parameter, the values at 3 months and 6 months did not change significantly, compared to baseline values

Table 4 compares the changes in each parameter (baseline value minus after treatment value) between double-dose statin and additional ezetimibe treatments. The reduction in LDL-cholesterol was significantly greater after additional ezetimibe treatment than after double-dose statin treatment ($P < 0.05$). The reductions in campesterol and sitosterol were also significantly greater after additional ezetimibe than after double-dose statins ($P < 0.001$ and $P < 0.01$, respectively). The changes in the other parameters were comparable between double dose-statin and additional ezetimibe treatments.

Figure 2 shows the relationship between the change in MDA-LDL and those in LDL-cholesterol, TBARS and d-ROMs, separately assessed in each treatment with double-dose statins and additional ezetimibe. The change in MDA-LDL level was not correlated with the changes in LDL-cholesterol, TBARS, or d-ROMs after double-dose statin treatment. In contrast, the change in MDA-LDL level was significantly correlated with that in LDL-cholesterol ($R = 0.548$, $P = 0.010$) but not with that in either TBARS or d-ROMs after additional ezetimibe treatment.

Finally, we assessed the relationship between changes in hsCRP levels and LDL-cholesterol levels. The change in hsCRP was significantly correlated with that in LDL-cholesterol after additional ezetimibe treatment ($R = 0.473$, $P < 0.05$) but not after double-dose statin treatment (Fig. 3).

Table 3 Change in each parameter after each treatment of double-dose statin or additional ezetimibe

	Baseline ($n = 21$)	Statin ($n = 21$)	P value	Ezetimibe ($n = 21$)	*P* value	Statin vs ezetimibe *P* value
LDL-cholesterol; mg/dl	118 ± 22	104 ± 15	0.017	89 ± 15	<0.0001	0.002
HDL-cholesterol; mg/dl	56 ± 13	58 ± 13	0.281	60 ± 14	0.07	0.709
Triglyceride; mg/dl	159 ± 96	128 ± 64	0.091	123 ± 59	0.033	0.784
Apo B/A-I	0.71 ± 0.17	0.62 ± 0.10	0.0007	0.58 ± 0.19	0.023	0.525
MDA-LDL; U/l	142 ± 35	126 ± 24	0.013	114 ± 22	0.0008	0.017
Hemoblobin A1c; %	6.15 ± 0.75	6.13 ± 0.88	0.758	6.23 ± 0.85	0.282	0.696
HOMA-R	3.65 ± 7.41	1.91 ± 1.42	0.217	3.86 ± 6.21	0.921	0.168
hsCRP; log (ng/ml)	3.02 ± 0.47	2.98 ± 0.41	0.693	2.78 ± 0.38	0.035	0.126
TBARS; μmol/l	10.9 ± 4.2	11.6 ± 4.7	0.834	9.2 ± 4.3	0.442	0.124
d-ROM; U.CARR	333 ± 76	321 ± 43	0.31	309 ± 59	0.176	0.513
Lathosterol; ng/ml	1.77 ± 1.39	1.54 ± 0.95	0.152	1.96 ± 0.89	0.391	0.156
Campesterol; ng/ml	5.98 ± 3.72	5.63 ± 3.90	0.525	3.19 ± 2.48	<0.0001	0.027
Sitosterol; ng/ml	3.56 ± 2.42	3.39 ± 2.36	0.487	2.22 ± 1.49	0.0002	0.075
PCSK9; ng/ml	251 ± 80	259 ± 91	0.791	242 ± 91	0.771	0.648
FMD; %	4.06 ± 1.76	4.07 ± 1.58	0.999	3.73 ± 1.70	0.256	0.416
RHI	2.11 ± 0.57	1.97 ± 0.51	0.463	2.01 ± 0.63	0.346	0.855

Table 4 Comparison of change in each parameter (baseline minus after treatment) between double-dose statin and additional ezetimibe

	Statin ($n = 21$)	Ezetimibe ($n = 21$)	*P* value
LDL-cholesterol; mg/dl	13.4 ± 23.7	29.0 ± 22.6	0.036
HDL-cholesterol; mg/dl	−1.67 ± 6.89	−3.23 ± 7.77	0.492
Triglyceride; mg/dl	31.0 ± 80.1	36.3 ± 72.8	0.826
Apo B/A-I	0.13 ± 0.24	0.10 ± 0.11	0.592
MDA-LDL; U/l	16.4 ± 27.3	27.8 ± 32.1	0.224
Hemoblobin A1c; %	−0.08 ± 0.34	0.02 ± 0.35	0.328
HOMA-R	1.75 ± 6.28	−0.21 ± 9.41	0.433
hsCRP; log (ng/ml)	0.04 ± 0.50	0.24 ± 0.43	0.189
TBARS; μmol/l	−1.11 ± 6.14	0.16 ± 6.68	0.552
d-ROM; U.CARR	17.9 ± 68.3	27.8 ± 78.2	0.706
Lathosterol; ng/ml	0.27 ± 0.77	−0.17 ± 0.84	0.106
Campesterol; ng/ml	0.32 ± 2.12	2.75 ± 2.00	0.0008
Sitosterol; ng/ml	0.20 ± 1.20	1.37 ± 1.28	0.006
PCSK9; ng/ml	−7.91 ± 96.44	9.18 ± 101.70	0.69
FMD; %	0.14 ± 0.74	0.11 ± 0.42	0.861
RHI	0.26 ± 5.14	−0.59 ± 6.58	0.675

Discussion

In the present study, we compared the effects on lipid profiles, inflammatory and oxidative stress status, and endothelial function between two treatment arms, additional ezetimibe and double-dose statin, in CAD patients with uncontrolled dyslipidemia undergoing low-dose statin therapy by using a crossover design and a small sample size. The major findings were that the levels of LDL-cholesterol and MDA-LDL were significantly reduced after both treatment changes, but the reductions in LDL-cholesterol and MDA-LDL were greater after add-on ezetimibe than after double-dose statin treatment.

The addition of ezetimibe to statins has been shown to further reduce LDL-cholesterol, and its potential benefit to further improve long-term prognosis has been demonstrated in CAD patients receiving statin therapy [9, 10]. However, there has been little information regarding the incremental benefit of add-on ezetimibe to statins. Torimoto et al. [22] demonstrated in a randomized trial that rosuvastatin 2.5 mg/day plus ezetimibe 10 mg/day reduced the levels of LDL-cholesterol, MDA-LDL, and small, dense LDL in patients with hypercholesterolemia and type 2 diabetes more significantly than did rosuvastatin 5 mg/day. Uemura et al. [23] compared atorvastatin 10 mg/day plus ezetimibe 10 mg/day with atorvastatin 20 mg/day in Japanese CAD patients with abnormal glucose tolerance by a crossover design. These authors demonstrated similar results to ours, showing that the levels of LDL-cholesterol and MDA-LDL were reduced more prominently by treatment with add-on ezetimibe than by double-dose statin monotherapy. However, these previous reports did not assess the relationship between the reduction in MDA-LDL and LDL-cholesterol. Compared to the studies by Torimoto et al. and Uemura et al., the baseline dose of statins was lower in our study (atorvastatin 5 mg/day, rosuvastatin 2.5 mg/day and pitavastatin 1 mg/day); thus, the double-dose of statins still remained medium-intensity. Kurobe et al. [24]

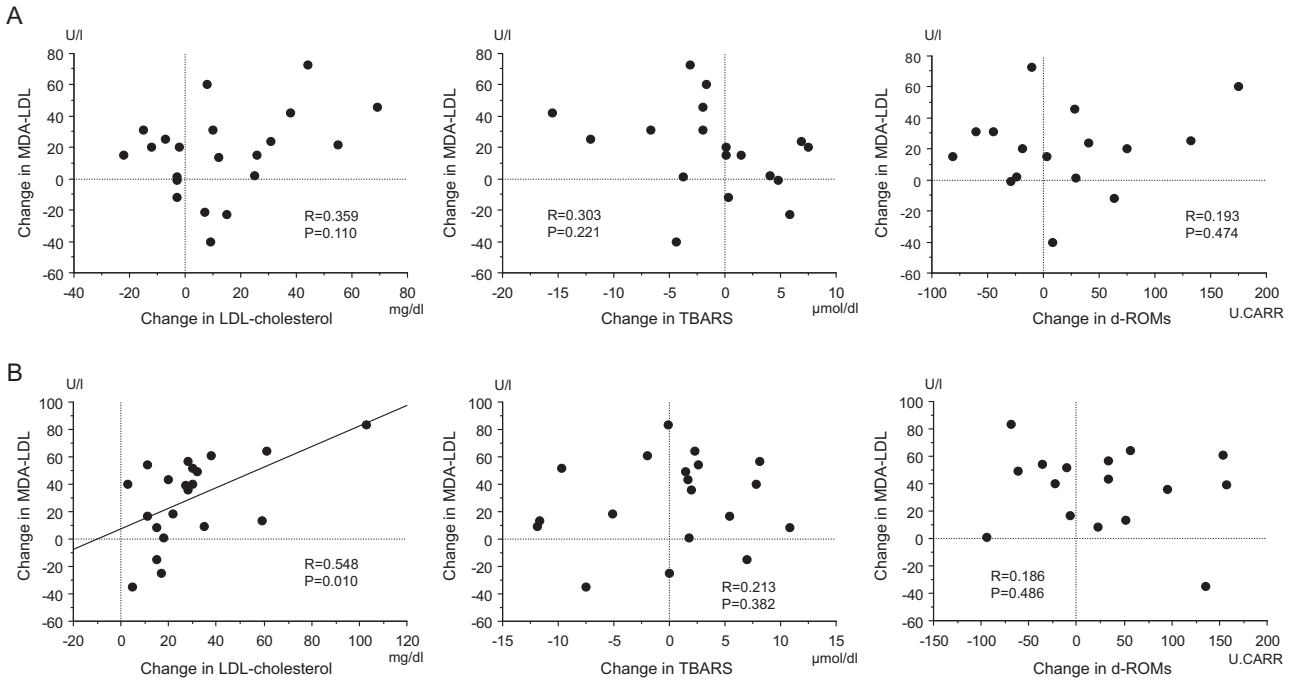

Fig. 2 Relationships between the change in MDA-LDL level and those in the levels of LDL-cholesterol, TBARS, and d-ROMs, separately assessed in each treatment period with double-dose statin and additional ezetimibe. **a** The change in MDA-LDL level was not correlated with the changes in LDL-cholesterol, TBARS or d-ROMs after double-dose statin treatment. **b** The change in MDA-LDL level was significantly correlated with the change in LDL-cholesterol level or with those in either TBARS or d-ROMs, after the addition of ezetimibe. LDL, low-density lipoprotein; MDA-LDL, malondialdehyde-modified LDL; TBARS, thiobarbituric acid reactive substances; d-ROMs, derivatives of reactive oxygen metabolites

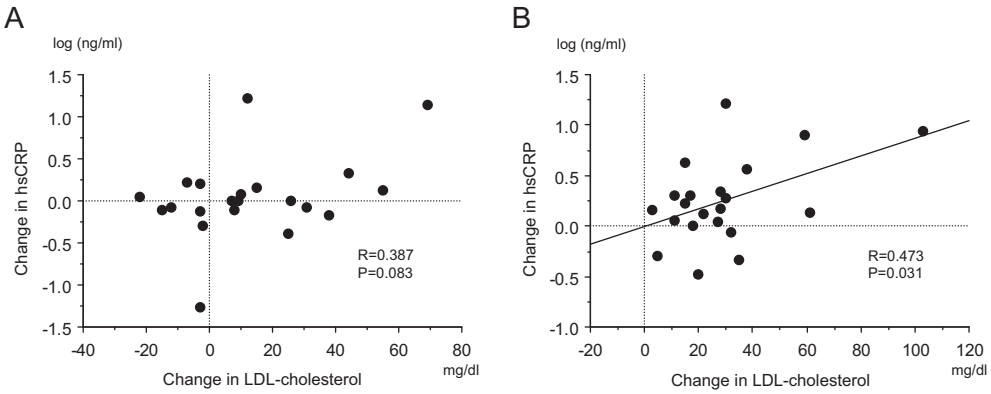

Fig. 3 Relationship between the change in hsCRP level and that in LDL-cholesterol. **a** The change in hsCRP level was not correlated with the change in LDL-cholesterol after double-dose statin treatment. **b** The change in the hsCRP was significantly correlated with that in LDL-cholesterol after the addition of ezetimibe treatment. hsCRP, high-sensitivity C-reactive protein

demonstrated that ezetimibe monotherapy (10 mg/day) for 3 months reduced LDL-cholesterol and MDA-LDL in patients with hypercholesterolemia. These authors also demonstrated that ezetimibe improved vascular endothelial function as demonstrated by increased FMD, although in our study FMD and RHI values did not change after the addition of ezetimibe. In addition, these authors observed that the increase in FMD value was correlated with the reduction in MDA-LDL level but not with that in the LDL-cholesterol level, suggesting that ezetimibe improved vascular endothelial function independently of the LDL-lowering effect via the reduction of oxidative stress. Qin et al. [25] demonstrated in vitro that ezetimibe protects endothelial cells against oxidative LDL-induced oxidative stress by restoring the mitochondrial membrane potential, which may be mediated by Akt-dependent glycogen synthase kinase-3 phosphorylation. Thus, ezetimibe may have a direct antioxidant effect as a pleiotropic action. In the

present study, however, the levels of oxidative stress markers, TBARS and d-ROMs, did not change after the addition of ezetimibe, and the change in the level of MDA-LDL after the addition of ezetimibe was correlated with that in LDL-cholesterol but not with that in the TBARS or d-ROM levels. This result suggests that the reduction in MAD-LDL by ezetimibe treatment depended on the LDL-cholesterol reduction but was independent of oxidative stress status.

The effect of ezetimibe on inflammation has also been widely investigated. Ezetimibe reduced vascular inflammation, as it significantly reduced vascular cell adhesion molecule-1 expression and vascular CD14 expression, a marker for mononuclear cell infiltration, in both apo E knockout and apo E/eNOS double knockout mouse atherosclerosis models [26]. In the IMPROVE-IT trial, the addition of ezetimibe 10 mg/day to simvastatin 40 mg/day resulted in a 14% reduction in the median hsCRP over the study duration compared with the effect of simvastatin monotherapy [27]. Kater et al. [28] demonstrated a synergic effect of simvastatin (20 mg/day) plus ezetimibe (10 mg/day) on hsCRP reduction compared with the effect of simvastatin (20 mg/day) monotherapy. In contrast, Wu et al. [29] demonstrated that the effect of atorvastatin 20 mg/day combined with ezetimibe 10 mg/day on hsCRP reduction was comparable to that of 40 mg/day atorvastatin. The anti-inflammatory effects of ezetimibe are controversial in the clinical setting. In the present study, the level of hsCRP was significantly reduced after the addition of ezetimibe but not after the double-dose statin treatment. In addition, the reduction in hsCRP level after the addition of ezetimibe was correlated with the reduction in LDL-cholesterol, suggesting that the reduced inflammation by ezetimibe was not a direct anti-inflammatory action of the drug but depended on LDL-cholesterol reduction. A similar correlation was found in pooled analyses of placebo-controlled trials of ezetimibe monotherapy or ezetimibe added to baseline statin therapy [30].

A pleiotropic anti-inflammatory effect of statins has been shown to contribute to benefits for cardiovascular disease, which were established in experimental studies and clinical trials [3, 31, 32]. A recent Japanese trial, the Randomized Evaluation of Aggressive or Moderate Lipid Lowering Therapy With Pitavastatin in Coronary Artery Disease, found that high-dose (4 mg/day) compared with low-dose pitavastatin (1 mg/day) therapy significantly reduced cardiovascular events in stable CAD patients. In this trial, an increase in the dose of pitavastatin from 1 mg/day to 4 mg/day significantly reduced the hsCRP level. The authors discussed the possibility that the anti-inflammatory action of pitavastatin might contribute to the event reduction independently of the effect attributable to the LDL-cholesterol reduction [33]. As the baseline statin treatment in the present study seems to have been low intensity, the double-dose treatment with each statin might still not have achieved a high intensity but might be medium intensity, possibly leading to failed hsCRP reduction after double-dose statin treatment. If the statin treatment was increased to quadruple dose, the reductions in LDL-cholesterol, MDA-LDL, and hsCRP by statin monotherapy might be similar to or greater than those with additional ezetimibe treatment. The reductions in LDL-cholesterol and hsCRP were identical between the two treatment arms of rosuvastatin 10 mg/day and rosuvastatin 2.5 mg/day combined with ezetimibe 10 mg/day [34]. In the present study, a cholesterol synthesis marker, lathosterol, did not change after double-dose statin treatment, suggesting that the increased dose effect of statins might be weak, while the cholesterol absorption markers, campesterol and sitosterol, were both significantly reduced after additional ezetimibe treatment, suggesting the additional effectiveness of ezetimibe.

Study limitations

The present study has several limitations. Although diminished to some degree by our choice of a crossover design, the biggest weakness is the small sample size. In addition, as we did not include a wash-out period in the switching arm, we cannot rule out a carry-over effect in the assessment of the results. Another weakness is that the baseline statin dose was low, and thus, this study actually compared add-on ezetimibe to low dose statins and medium-intensity statin monotherapy. A recent study established that high-intensity statin therapy could be more effective for secondary prevention in patients with CAD than low-intensity statin therapy. The efficacy of the addition of ezetimibe to high-dose statins should be discussed. In an era of intensive-statin therapy, it might be questionable to evaluate add-on ezetimibe therapy to low-dose statins in the real world clinical scene. Considering that the adverse effects of statins, including myalgia, fatigue or rhabdomyolysis and hepatic dysfunction, appear dose-dependently, the addition of ezetimibe to low-dose statins may be one of the therapeutic choices, even in the intensive-statin era. Thus, we believe that the present study provides some potentially important messages.

Conclusions

The results of this study suggest that the addition of ezetimibe to low-dose statin therapy might have advantages for LDL-cholesterol reduction, reduced lipid peroxidation, and anti-inflammatory effects over double dose statin treatment. The reduced lipid peroxidation and anti-inflammatory action might be due to the effect of ezetimibe on LDL-cholesterol reduction rather than the pleiotropic effects of this drug.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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