Atherosclerosis 225 (2012) 388–396

Contents lists available at SciVerse ScienceDirect

Atherosclerosis

journal homepage: www.elsevier.com/locate/atherosclerosis

Effects of ezetimibe added to statin therapy on markers of cholesterol absorption and synthesis and LDL-C lowering in hyperlipidemic patients

Nuntakorn Thongtang [a,b], Jianxin Lin [c], Ernst J. Schaefer [a], Robert S. Lowe [c], Joanne E. Tomassini [c], Arvind K. Shah [c], Andrew M. Tershakovec [c,*]

[a] Lipid Metabolism Laboratory, Human Nutrition Research Center on Aging at Tufts University, 711 Washington Street, Boston, MA 02111, USA
[b] Faculty of Medicine, Siriraj Hospital, Mahidol University, Bangkok, Thailand
[c] Merck Sharp & Dohme Corp., Whitehouse Station, NJ 08889, USA

ARTICLE INFO

Article history:
Received 15 March 2012
Received in revised form
10 August 2012
Accepted 2 September 2012
Available online 13 September 2012

Keywords:
Non-cholesterol sterol
Lathosterol
β-sitosterol
Statin potency
Dyslipidemia

ABSTRACT

Objective: Statins inhibit cholesterol synthesis but can upregulate cholesterol absorption, with higher doses producing larger effects. Ezetimibe inhibits cholesterol absorption but also upregulates synthesis. We tested whether ezetimibe added to on-going statin therapy would be most effective in lowering LDL-cholesterol (LDL-C) in subjects on high-potency statins and whether these effects would be related to alterations in cholesterol absorption (β-sitosterol) and synthesis (lathosterol) markers.
Methods: Hypercholesterolemic subjects ($n = 874$) on statins received ezetimibe 10 mg/day. Plasma lipids, lathosterol, and β-sitosterol were measured at baseline and on treatment. Subjects were divided into low- ($n = 133$), medium- ($n = 582$), and high- ($n = 159$) statin potency groups defined by predicted LDL-C—lowering effects of each ongoing statin type and dose (reductions of ~20–30%, ~31–45%, or ~46–55%, respectively).
Results: The high-potency group had significantly lower baseline lathosterol (1.93 vs. 2.58 vs. 3.17 μmol/l; $p < 0.001$) and higher baseline β-sitosterol values (6.21 vs. 4.58 vs. 4.51 μmol/l, $p < 0.001$) than medium-/low-potency groups. Ezetimibe treatment in the high-potency group produced significantly greater reductions from baseline in LDL-C than medium-/low-potency groups (−29.1% vs. −25.0% vs. −22.7%; $p < 0.001$) when evaluating unadjusted data. These effects and group differences were significantly ($p < 0.05$) related to greater β-sitosterol reductions and smaller lathosterol increases. However, LDL-C reduction differences between groups were no longer significant after controlling for placebo effects, due mainly to modest LDL-C lowering by placebo in the high-potency group.
Conclusion: Patients on high-potency statins have the lowest levels of cholesterol synthesis markers and the highest levels of cholesterol absorption markers at baseline, and the greatest reduction in absorption markers and the smallest increases in synthesis markers with ezetimibe addition. Therefore, such patients may be good candidates for ezetimibe therapy if additional LDL-C lowering is needed.

1. Introduction

Statins play a central role in the treatment of atherogenic dyslipidemia and reduction of cardiovascular disease (CVD) risk. The cholesterol-lowering response to statin therapy, however, can vary widely between individuals [1,2] and contributes to the substantial number of patients with LDL-C levels above guideline-recommended targets [3–6]. Recent studies suggest that statin efficacy may be determined not only by their direct inhibitory effect on cholesterol synthesis but also by compensatory downstream changes in cholesterol metabolism. Statins reduce markers of cholesterol synthesis (e.g., lathosterol, desmosterol), which can elicit subsequent increases in markers of cholesterol absorption (e.g., campesterol, β-sitosterol) [7–11]. The magnitude of change in these sterol markers has been reported to vary by statin dose, with lower doses having smaller effects [8,10]. Differences between statins have also been observed. Atorvastatin was found to reduce serum lathosterol/cholesterol ratios more than simvastatin, while it increased plant sterol/cholesterol ratios more than simvastatin in

* Corresponding author. Merck Sharp & Dohme Corp., 351 N. Sumneytown Pike, North Wales, PA 19454, USA. Tel.: +1 267 305 5454; fax: +1 267 305 6427.
E-mail addresses: nuntakorn.thongtang@tufts.edu (N. Thongtang), jianxin.lin@merck.com (J. Lin), ernst.schaefer@tufts.edu (E.J. Schaefer), robert.lowe@merck.com (R.S. Lowe), joanne.tomassini@merck.com (J.E. Tomassini), arvind.shah@merck.com (A.K. Shah), andrew.tershakovec@merck.com (A.M. Tershakovec).

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http://dx.doi.org/10.1016/j.atherosclerosis.2012.09.001

patients with coronary heart disease [12]. We previously reported that atorvastatin 80 mg/day and rosuvastatin 40 mg/day caused similar reduction in markers of cholesterol synthesis, but rosuvastatin increased markers of cholesterol absorption significantly less than atorvastatin [13]. This study also suggested that the combined effect of statins on cholesterol synthesis and absorption may influence treatment efficacy, since the greatest reduction in total cholesterol and LDL-C was seen in subjects with the largest reduction in lathosterol and no compensatory increase in campesterol while treatment efficacy was the lowest in subjects where the converse was true.

Ezetimibe is a selective cholesterol absorption inhibitor that blocks the transport of cholesterol and phytosterols across the intestinal wall and significantly reduces LDL-C levels by 15–20% [14,15]. Ezetimibe decreases markers of cholesterol absorption but also produces a compensatory increase in markers of cholesterol synthesis [16]. Co-administration of ezetimibe with a statin has been shown to inhibit cholesterol absorption as well as synthesis [17] and these complementary effects produce significantly greater reductions in LDL-C than either drug alone [15,18,19].

The effects of ezetimibe added to different statins on cholesterol-lowering and cholesterol homeostasis has not been well studied, especially not in a large head-to-head comparison study. The goals of this *post hoc* analysis of the EASE (Ezetimibe Add-on to Statin for Effectiveness) study were to compare the effects of adding ezetimibe 10 mg to different statins and doses on plasma lipid-lowering effects and non-cholesterol sterol levels. Subjects enrolled in the EASE study had LDL-C levels above National Cholesterol Education Program Adult Treatment Panel III (NCEP ATP III) recommended targets while on statin therapy, and were randomized to receive either placebo or ezetimibe in addition to their ongoing statin [20]. We used lipid and non-cholesterol sterol data from the ezetimibe arm of this study to test the hypothesis that ezetimibe, when added to statin therapy, would be most effective in LDL-C lowering in subjects on high-potency statins and that these effects would be related to alterations in markers of cholesterol absorption (β-sitosterol, β-sitosterol/cholesterol) and synthesis (lathosterol, lathosterol/cholesterol).

2. Methods

2.1. Subjects and study design

This study included subjects from the ezetimibe add-on to statin arm of the EASE study (http://clinicaltrials.gov identifier NCT00092586; Study Protocol 040). Details of the study design and outcomes have been published previously [20,21]. Briefly, the EASE study was a multicenter, randomized, double-blind, placebo-controlled, 6-week parallel-group study. Participants with hypercholesterolemia were recruited from community based practices across the United States. Inclusion criteria were: 1) age $\geq$ 18 years, 2) on a stable, approved dose of any statin, 3) following a cholesterol-lowering diet for $\geq$6 weeks before study entry, and 4) LDL-C levels above risk-based NCEP ATP III targets. Subjects receiving lipid-altering agents other than statins during the 6 weeks before screening were excluded. Patients were randomized to receive ezetimibe 10 mg/day or placebo plus their current statin therapy and dose for 6 weeks. Statin type and dose were maintained throughout the study. The research protocol was approved by the investigational review boards at each site, and all participants provided written informed consent prior to study start.

For this current *post hoc* analysis, we assessed subjects who were randomized into the ezetimibe 10 mg plus statin arm of the EASE study ($n = 1940$). Of the 1124 subjects with samples available for measurement, only those who had complete sterol and lipid data at baseline and at the end of the 6-week study were included in this analysis ($n = 874$). The mean age and other baseline characteristics of those included and excluded from this analysis were similar.

Comparison of subjects was based on statin type or potency (low, medium, high) subgroups. The low-potency statin group (predicted LDL-C reduction of ~20–30%) included subjects receiving simvastatin $\leq$ 10 mg/day, lovastatin $\leq$ 20 mg/day, pravastatin $\leq$ 20 mg/day, and fluvastatin $\leq$ 40 mg/day. The medium-potency statin group (predicted LDL-C reduction of ~31–45%) included subjects receiving simvastatin $>$ 10– $\leq$ 40 mg/day, atorvastatin $\leq$ 20 mg/day, lovastatin $>$ 20–80 mg/day, pravastatin $>$ 20–80 mg/day, and fluvastatin $>$ 40–80 mg/day. The high-potency statin group (predicted LDL-C reduction of ~46–55%) included subjects receiving simvastatin $>$ 40–80 mg/day, and atorvastatin $>$ 20–80 mg/day.

2.2. Measurement of lipoproteins and non-cholesterol sterols

Plasma total cholesterol (total C), high-density lipoprotein cholesterol (HDL-C), and triglycerides (TG) were analyzed using standardized methods at the central laboratory of the trial (PPD Global Central Labs, Highland Heights, Kentucky, USA). LDL-C was calculated using the Friedewald formula [22]. Non-HDL-C was calculated by subtracting HDL-C from total C. Apolipoprotein (Apo) A-I, Apo B, and high-sensitivity C-reactive protein (hs-CRP) were measured by automated immunoassays at the central laboratory. Within and between coefficients for all assays were <10%. Plasma lathosterol and β-sitosterol were quantified by gas chromatography mass spectrometry after lipid extraction as previously described [16]. Since these plasma sterols are mainly carried in the LDL fraction [23], it is common practice to adjust them for total plasma cholesterol by expressing them as a ratio to cholesterol. An alternative is to express them as a ratio representing both absorption and synthesis (i.e., sitosterol/lathosterol). Plasma sterols were therefore expressed either in absolute terms, as a ratio to cholesterol, or as a ratio of β-sitosterol to lathosterol.

2.3. Statistical analysis

All continuous variables were expressed as means ± standard deviation (SD) for normally distributed data, or medians ± robust SD if non-normally distributed. Subjects receiving lovastatin and fluvastatin were grouped together as “other statins” when evaluating statin types due to the small number of subjects. For samples with sterol levels below the limit of detection (0.5 μg/ml) either at the time of randomization or at study end, a value of 0.25 μg/ml was assigned to prevent any bias of excluding subjects with very low sterol levels. All subjects were receiving statin therapy at study entry and baseline values represent levels while on treatment. Baseline cholesterol synthesis and absorption markers among the different statins were compared using an ANOVA model with terms for statin type and statin dose within statin type. Changes from baseline in plasma lipids, apolipoproteins, and hs-CRP after ezetimibe treatment were assessed using an ANOVA model with terms for statin type or statin potency. Data were presented as mean and 95% confidence interval (CI) or median and 95% CI for non-normally distributed data. Since baseline LDL-C levels can influence the lipid-lowering effect of hypolipidemic agents, data were also calculated as percent changes from baseline. Lipid data were also calculated by adjustment for lipid values from the placebo arm of the original EASE study. ANOVA with terms for statin type or statin potency were used to compare changes from baseline of plasma lipid values among groups as well as changes in lathosterol and β-sitosterol.