Low dose of ROSuvastatin in combination with EZEtimibe effectively and permanently reduce low density lipoprotein cholesterol concentration independently of timing of administration (ROSEZE): A randomized, crossover study — preliminary results

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Abstract

Background: In an attempt to improve low density lipoprotein-cholesterol (LDL-C) level control in patients ineffectively treated with statins, we evaluated the effectiveness of a fixed-dose combination (FDC) of 10 mg rosuvastatin and ezetimibe and its relation to the timing of drug administration.

Methods: A randomized, open label, single center, crossover study involving 83 patients with coronary artery disease and hypercholesterolemia with baseline LDL-C ≥ 70 mg/dL. In arm I the FDC drug was administered in the morning for 6 weeks, then in the evening for the following 6 weeks and vice versa in arm II. The primary endpoint was the change in LDL-C after 6 and 12 weeks.

Results: The median LDL-C concentration at baseline, after 6 and 12 weeks respectively was: 98.10 mg/dL (Q1;Q3: 85.10;116.80), 63.14 mg/dL (50.70;77.10) and 59.40 mg/dL (49.00;73.30); p < 0.001. LDL-C levels were similar regardless of the timing of drug administration (morning 62.50 mg/dL [50.70;76.00] vs. evening 59.70 mg/dL [48.20;73.80]; p = 0.259), in both time points: 6 week: 63.15 mg/dL (50.75;80.65) vs. 63.40 mg/dL (50.60;74.00), p = 0.775; and 12 week: 62.00 mg/dL (50.20;74.40) vs. 59.05 mg/dL (47.65;66.05), p = 0.362. The absolute change in LDL-C concentration for the morning vs. evening drug administration was — 6 week: –34.6 mg/dL (–56.55; –19.85) (–34.87%) vs. –31.10 mg/dL (–44.20; –16.00) (–35.87%) (p not significant); 12. week: –34.20 mg/dL (–47.8; –19.0) (–37.12%) vs. –37.20 mg/dL (–65.55; –23.85) (–40.06%) (p not significant). The therapy was safe and well tolerated.

Conclusions: Fixed-dose combination of rosuvastatin and ezetimibe significantly and permanently decreases LDL-C regardless of the timing of drug administration. (Cardiol J 2021; 28, 1: xx–xx)

Key words: hypercholesterolemia, fixed-dose, secondary prevention, timing of administration, adherence, apolipoprotein B, lipoprotein(a)
Introduction

Coronary artery disease (CAD) remains the most common single cause of death worldwide [1]. Hypercholesterolemia constitutes one of its major risk factors [2]. According to the European Society of Cardiology (ESC) and European Atherosclerosis Society (EAS) 2016 guidelines for the management of dyslipidemias, the therapeutic target for low density lipoprotein-cholesterol (LDL-C) is < 70 mg/dL (< 1.8 mmol/L) [1, 3]. The first line treatment of hypercholesterolemia is statin therapy [3]. However, when the therapeutic target of LDL-C is not achieved, the addition of cholesterol absorption inhibitor — ezetimibe — to statin therapy is recommended [3, 4]. Unfortunately, lipid-lowering therapy is discontinued in a high percentage of patients with CAD [5]. One year after myocardial infarction (MI) only approximately 50% of patients report persistent use of statins [6, 7]. Furthermore, even when patients follow the recommendations and continue statin therapy, only a minority obtains optimal level of LDL-C [8, 9].

The aim of the present study was to evaluate the effectiveness of hypercholesterolemia treatment with rosuvastatin and ezetimibe in patients ineffectively treated with statin monotherapy. Also under investigation was whether the timing (morning vs. evening) of rosuvastatin and ezetimibe administration affects their efficacy.

Methods

Study design and population

The study was designed as a randomized, open-label, single-center, crossover study. It was conducted in accordance with the principles contained in the Declaration of Helsinki and Good Clinical Practice guidelines and aimed to evaluate: the effectiveness of combined therapy with rosuvastatin and ezetimibe in patients ineffectively treated with statin monotherapy. Also under investigation was whether the timing (morning vs. evening) of rosuvastatin and ezetimibe administration affects their efficacy.

Endpoints

The primary outcome was defined as change in LDL-C after 6 and 12 weeks of the investigated therapy, with respect to timing of the study drug administration. The secondary endpoints included: change in total cholesterol (TC) and triglycerides (TG) levels after 6 and 12 weeks (also with respect to timing of the study drug administration), concentration of high-sensitivity C-reactive protein (hs-CRP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), creatine kinase (CK), apolipoprotein B (apoB) and lipoprotein(a) (Lp(a)) at baseline and after 6 and 12 weeks of the therapy.
**Blood collection and laboratory measurements**

A detailed description of blood collection and laboratory measurements has been previously published [10]. Routine laboratory measurements were performed in fresh serum (basic lipid profile [TC, TG, LDL-C], AST, ALT, CK). The remaining serum was aliquoted and stored at –80°C until assayed for hs-CRP, apoB, Lp(a). All measurements (except for CRP) were performed using the Horiba ABX Pentra 400 analyzer (Horiba ABX, Montpellier, France). LDL-C was measured directly. CRP was measured using the Alinity c analyzer (Abbott Laboratories, Chicago, IL, USA) with the Alinity c CRP Vario High Sensitivity assay for the quantitative, immunoturbidimetric determination of CRP with a limit of detection of 0.4 mg/L. Laboratory measurements were performed at the Department of Laboratory Medicine, Nicolaus Copernicus University, Collegium Medicum, Bydgoszcz, Poland.

**Statistical analysis**

Statistical analysis was carried out using the Statistica 13.0 package (StatSoft, Tulsa, USA). The Shapiro-Wilk test demonstrated that the investigated continuous variables were not normally distributed. Therefore, continuous variables were presented as median and quartiles (lower and upper) and nonparametric tests (the Mann-Whitney unpaired rank sum test, the Wilcoxon signed rank test, and the Friedman ANOVA) were used for statistical analysis. The $\chi^2$ test was used for comparisons of qualitative variables. Differences were considered statistically significant at $p < 0.05$.

**Results**

Eighty-three patients were enrolled into the study. The mean age was 64.6 ± 8.7 years. The majority of included patients had a documented history of CAD (93.98%) and 62.7% had prior MI. There were no differences between patients in both study arms (Table 1). At the time of enrollment, the majority of patients were treated with atorvastatin (72.29%), 16.87% used rosuvastatin, and the rest were treated with simvastatin.

### Primary endpoint

After 6 weeks of therapy with the study drug, there was a significant reduction in LDL-C (median: 98.10 mg/dL; interquartile distribution [Q1;Q3]: 85.10;116.80 vs. 63.14 mg/dL; 50.70;77.10; $p < 0.001$). The decrease was constant over time after 12 weeks (63.14 mg/dL [50.70;77.10] vs. 59.40 mg/dL [49.00;73.30]; $p = 0.077$; Fig. 1). There was no significant difference between LDL-C with respect to the timing of the study drug administration (morning: 62.50 mg/dL [50.70;76.00] vs. evening: 59.70 mg/dL [48.20;73.80]; $p = 0.259$; Fig. 2), in both time points after 6 and 12 weeks, respectively (after 6 weeks: 63.15 mg/dL [50.75;80.65] vs. 63.40 mg/dL [50.60;74.00]; $p = 0.775$; after 12 weeks: 62.00 mg/dL [50.20;74.40] vs. 59.05 mg/dL [47.65;66.05]; $p = 0.362$). After 6 weeks the absolute change in LDL-C was –34.6 (–56.55; –19.85) (–34.87% [–46.83; –22.69]) for the morning administration of the study drug, and –31.10 (–44.20; –16.00) (–35.87% [–47.87; –17.96]) (p not significant) for the evening administration, respectively.

In a subgroup of 20 patients additional measurements were performed at 12 and 24 hours after the last dose of the study drug. In patients receiving the study drug in the morning, LDL-C measured in the evening (i.e. 12 h after the last dose) were significantly lower than the next morning (i.e. 24 h...
after the last dose of the study drug) [52 mg/dL (46.95;75.85) vs. 64.95 mg/dL (50.35;77.05); p=0.019] (Fig. 3). Nevertheless, both results were significantly lower compared with baseline LDL-C [12 h: 93.5 mg/dL (86.15;113.4) vs. 52 mg/dL (46.95;75.85); p=0.000089; 24 h: 93.5 mg/dL (86.15;113.4) vs. 64.95 mg/dL (50.35;77.05); p<0.000001]. Among patients taking the drug in the evening, LDL-C measured next morning (i.e. 12 h after the last dose of the study drug) was comparable with the LDL-C level measured in the evening the same day (i.e. 24 h after the last dose of the study drug) [61.05 mg/dL (45.85;74.05) vs. 63.35 mg/dL (44.75;71.00); p = 0.255] (Fig. 4).

**Secondary endpoints**

Total cholesterol was significantly lower after 6 weeks of therapy with the study drug and this effect was stable throughout the observational period (Fig. 5). Moreover, the effect was independent of the timing of study drug administration (Table 2). Similar results were achieved for TG. There was a significant reduction in TG concentration compared with baseline values (Table 2), and the outcome was again independent of the timing of the study drug administration (Table 2).

With regard to apoB, a significant reduction was found in its concentration after 6 weeks of treatment with the study drug (93.00 [77.00;
The reduction was persistent throughout the observational period, and was independent of the timing of the study drug administration (Table 2).

Lipoprotein(a) decreased after the study drug administration, however without statistical significance (Table 2).

There was no alteration in AST activity. An increase in ALT activity compared with baseline values was recorded, regardless of the timing of the study drug administration. Nevertheless, the morning and evening measurements showed no significant difference (Table 2). There were no cases of ALT activity increase $\geq 3 \times \text{URL}$.

There was a statistically significant, transient increase in CK activity after initiation of the treatment (Table 2), always $< 5 \times \text{URL}$. After 12 weeks of therapy CK activity showed no significant differences compared with baseline levels (73.0 [55.0;108.0] vs. 84.0 [60.0;125.0]; $p = 0.245$). Similar CK activity was noted regardless of the timing of the study drug administration (morning: 86.0 [56.0;116.0] vs. evening: 80.0 [63.0;128.0]; $p = 0.984$).

High-sensitivity C-reactive protein concentration was significantly reduced after 6 weeks of treatment (Table 2). The effect was stable throughout the observational period (Table 2).

**Discussion**

The main finding of the ROSEZE study is confirmation of the effectiveness and safety of combined therapy for hypercholesterolemia using an FDC of low dose rosuvastatin (10 mg)
and ezetimibe, regardless of the timing of drug administration, in patients unsuccessfully treated with statin monotherapy. According to available research, the current study is the first trial assessing the effectiveness of an FDC of rosuvastatin and ezetimibe in relation to the daily timing of drug administration.

As demonstrated in a meta-analysis of trials assessing statin therapy, each 40 mg/dL drop in LDL-C translates into a significant reduction in all-cause mortality (by 10%) [11] and major cardiovascular events (by 23%) [12]. More powerful statins compared with weaker ones, produce a highly significant 15% (95% confidence interval [CI] 11.1–18; p < 0.0001) further reduction in major vascular events [11]. More intensive LDL-C lowering therapies including potent statins alone or combinations of statins with ezetimibe or proprotein convertase subtilisin/kexin type 9 (PCSK-9) inhibitors are associated with a great reduction in risk of total and cardiovascular mortality, especially when the baseline LDL-C level exceeds 100 mg/dL [13].

Surveys and national databases of patients with hypercholesterolemia and CAD demonstrated that the LDL-C target levels recommended by 2016 ESC guidelines were achieved only in a small percentage of patients: 19.3% according to EUROSPIRE IV [5], 19–25% according to a large real-life German registry from the years 2011–2016 [8] and were only 5.8% according to a large Italian database published in 2019 [9]. Other lessons coming from available registries include (i) too low usage of high intensity statins and (ii) frequently premature discontinuation of lipid lowering therapy (LLT).

Several trials demonstrated the superiority of combined treatment of hypercholesterolemia with statins and ezetimibe compared with statin monotherapy, revealing that addition of ezetimibe to statin therapy provides more extensive reduction of LDL-C than doubling the statin dose, and thus allows more patients to achieve the LDL-C goal [14–16]. The IMPROVE-IT trial, demonstrated that adding ezetimibe to low intensity statin (simvastatin 40 mg) carries benefit (24% of additional reduction in LDL-C compared with statin monotherapy and lowering the risk of cardiovascular events compared with statin monotherapy with a 2.0-percentage-point lower rate of primary end point defined as a composite of death from cardiovascular disease, a major coronary event or nonfatal stroke [17]) independent of age [18] and sex [19] with a good safety profile, supporting the use of intensive, combined LLT to optimize cardiovascular outcomes [17–19].

In studies evaluating the effectiveness of more potent regimens, rosuvastatin enabled LDL-C reduction by 44–47% [20–22]. The PULSAR trial revealed that in high-risk patients with hypercholesterolemia, rosuvastatin 10 mg is more efficient at reducing the LDL-C level than the commonly used 20 mg dose of atorvastatin, enabling LDL-C goal achievement and improving other lipid parameters [22]. In the MRS-ROZE study, Kim et al. [23] demonstrated that FDCs of ezetimibe and rosuvastatin provided superior efficacy to rosuvastatin alone in lowering LDL-C, TC and TG levels (reduction by 56–63%, 37–43%, and 19–24%, respectively).

Similarly, the I-ROSETTE trial reported the LDL-C lowering efficacy of each ezetimibe/rosvu- vastatin combination to be superior to each of the respective doses of rosuvastatin [24]. The mean percent change in LDL-C in all ezetimibe/rosvu- vastatin combination groups exceeded 50% [24]. Moreover, the number of patients who achieved target LDL-C levels after 8 weeks of an observational period was significantly higher in the combined therapy group than in the rosuvastatin monotherapy group (92.3% vs. 79.9%, p < 0.001) [24]. Rosuvastatin alone or in combination with ezetimibe is very effective even in patients with familiar hypercholesterolemia. Mickiewicz et al. [25] demonstrated a reduction in LDL-C concentration by 45.9% and 55.4% depending whether it concerned monogenic or polygenic subjects with familiar hypercholesterolemia [25].

As expected, the present study also demonstrated that an FDC of rosuvastatin 10 mg/ezetimibe 10 mg significantly reduces LDL-C and other lipid fractions including TC and TG. The median percentage change in LDL-C after FDC drug in the current study was ~35–40%. It allowed achievement of the target LDL-C level < 70 mg/dL in 66.27% and 69.88% in patients receiving the FDC drug in the morning and in the evening, respectively. Nevertheless, this result cannot be regarded as fully satisfactory, considering that according to the newly published 2019 European Guidelines for the Management of Dyslipidemias, the therapeutic goal for LDL-C in very high-risk patients was further lowered to the level of < 55 mg/dL (< 1.4 mmol/L) [4], thus the combination with 10 mg of rosuvastatin might not be sufficient for every single patient.

Apolipoprotein B, which is known to be a more informative marker of adequacy of statin treatment than LDL-C [26], similar to other studies [27–30] it was reduced in the present study by 26.9%, achiev-
Although, in our study there was no significant difference between obtained LDL-C levels according to the timing of study drug intake (morning vs. evening), the discrepancies found in LDL-C reduction depending on the time of test performance: 12 or 24 hours after the last dose of the drug studied, are not completely clear and need further exploration. According to a research by Nishida et al. [35], rosuvastatin exposure decreases under the fed condition compared with strict fasting. However, it was only a pharmacokinetics study assessing potential food effect and the bioequivalence between co-administered ezetimibe and rosuvastatin and FDC tablets containing ezetimibe and rosuvastatin in healthy Japanese subjects under fasted and fed conditions [35].

According to available research, the present study is the first one to reveal the influence of the timing of an FDC of rosuvastatin and ezetimibe on the effectiveness of LLT. Observations herein, indicate similar potency of tested therapy, regardless of whether the drug was administered in the evening or in the morning. As a consequence of this, we encourage administration of the FDC of rosuvastatin with ezetimibe in the morning with the majority of other drugs. This timing modification may translate into better adherence to recommended LLT, compared with traditional statin dosing in the evening, which leads to its frequent omission. Simplifying the treatment strategies and reducing the number of tablets by using polypills are one of key factors for better cooperation between health care providers and patients, and better drug adherence [3].

Similar to previous studies in patients with CAD [36, 37], in the present study, the tested therapy provoked a further reduction in hs-CRP concentration, compared with baseline values. Considering the fact that our population of patients had been already treated with statins for secondary prevention prior to the study, it was the addition of ezetimibe that led to a further reduction of the inflammatory process. This finding supports the existence of a pleotropic anti-inflammatory, in addition to hypolipemic, effect of ezetimibe, both of which are responsible for residual atherosclerotic risk [38].

The safety and tolerability of the tested therapy were similar in both arms of the current study. There were no cases of rosuvastatin/ezetimibe-related adverse events including muscular, hepatic and gastrointestinal events.

Limitations of the study
This study has several limitations. The first one is a relatively small sample size. Due to the preliminary nature of the study, the number of patients included in the current analysis is lower than calculated earlier for the sample size. Second, the study was designed to assess changes in LDL-C concentration without addressing the relationship between LDL-C reduction and clinical outcomes. Third, the divergences between LDL-C levels assessed 12 and 24 hours after the last dose of rosuvastatin and ezetimibe may have had a multifactorial origin which is difficult to explain at this point and would likely require an adequate sample size for clarification.

Conclusions
The present study demonstrates that a combination of rosuvastatin and ezetimibe as a polypill is effective and well tolerated, showing similar efficacy whether administered in the morning or in the evening.

Conflict of interest: None declared

References


