Part II

Pharmacological Interventions

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Effect of Intensive versus Moderate Lipid Lowering on Endothelial Function and Vascular Responsiveness to Angiotensin II in Stable Coronary Artery Disease

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Abstract

Recent evidence demonstrates that intensive lipid-lowering therapy with high dose statin provides significant clinical benefit beyond moderate lipid lowering therapy. However, dose-dependent effects of short-term statin therapy on vascular function have not been demonstrated. We studied endothelial function and, in addition, vascular responsiveness to angiotensin II in coronary artery diseased patients randomized to low or high dose atorvastatin (10 or 80 mg), or placebo. Internal thoracic artery segments were obtained during coronary bypass surgery and studied in vitro. Endothelium dependent vasodilatation was improved with atorvastatin therapy ($p=0.035$), but was significantly further improved with atorvastatin 80 mg as compared to 10 mg treated patients ($p=0.05$). Endothelium improvement was accompanied by reduced vascular response to angiotensin II ($p=0.039$). These findings suggest a mechanism for the clinical benefit of intensive lipid lowering treatment in coronary heart disease.

Introduction

Evidence is showing that intensive lipid-lowering therapy with 80 mg of atorvastatin per day in patients with stable coronary artery disease provides significant clinical benefit beyond that afforded by treatment with 10 mg per day.[1] The endothelium is recognized as an important target for therapy, particular for cholesterol-lowering drugs.[2] Recent studies have indicated that patients with elevated LDL-cholesterol may, in addition, also display increased vascular responses to angiotensin II.[3;4] Changes in vasomotor function are thought to precede atherosclerosis and its clinical consequences. To date, no evidence exists to support additional beneficial effects of high dose statin treatment on vascular function, providing a possible mechanism. In the present study, we assessed the short-term effects of high and low dose statin treatment on endothelial function and responsiveness to Angiotensin II in humans.

Table 1. Clinical backgrounds

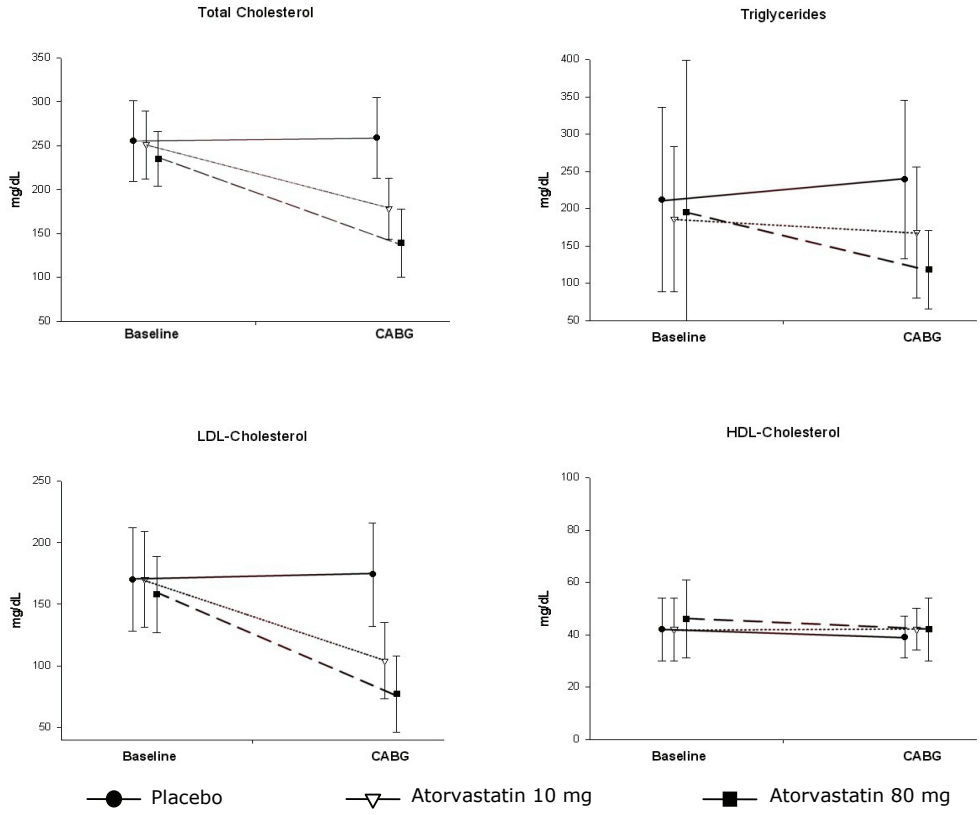
Variable	Total Population (N=156)	Vascular Measurements Obtained (N=46)
Age – (years)	62.8±8.6	61.9±8.5
Male/female	142/14	40/6
Hypertension	29 (19%)	10 (22%)
Cigarette Smoker	11 (7%)	6 (13%)
Previous myocardial infarction	52 (33%)	12 (26%)
Previous PCI/CABG	24/1	4/0
Blood pressure – (mmHg)		
Systolic	130±16	132±16
Diastolic	76±10	78±12
Concurrent Medication		
Beta-blockers	132 (85%)	41 (89%)
Calcium channel blockers	96 (62%)	27 (59%)
Nitrates	124 (79%)	43 (93%)
Anti coagulant	149 (96%)	45 (98%)
Days on study medication	28.4±10.9	27.7±7.4

PCI = percutanuous intervention ; CABG = coronary artery bypass grafting; Anti coagulants includes antiplatlet drugs.

Methods

This study was designed in a prospective, randomized, double-dummy, double-blinded, placebo-controlled manner with 3 parallel groups of patients scheduled for elective coronary artery bypass grafting. Treatment of any cholesterol-lowering agent, angiotensin converting enzyme inhibitors or angiotensin II receptor antagonists was not allowed within 4 weeks of enrolment up to completion of the study. Patients were randomized to 1 of the 3 treatment arms receiving (daily p.o.) 80 mg atorvastatin, 10 mg atorvastatin or matching placebo. The institutional review board approved the study and all patients provided written informed consent.

Figure 1.



Fasting lipids in each group. Total and LDL cholesterol levels decreased in atorvastatin treated groups (both, $p<0.001$). Triglycerides were significantly decreased only in atorvastatin 80 mg ($p<0.05$). Total and LDL cholesterol at CABG was lower in the atorvastatin 80 mg versus 10 mg treated group (both, $p<0.05$).

During coronary artery bypass grafting, when available, segments of internal thoracic arteries were collected as excess graft material and transported immediately to the laboratory, where in vitro vascular measurements were performed within 6 hours of harvesting as published previously.[5;6] In brief, rings were checked for viability by repeated stimulation (3-4 times) with 10 μmol/L phenylephrine. Rings that failed to produce a contractile response to phenylephrine of at least 100 μm displacement were discarded from further experiments. Endothelial dependent relaxation was studied by response to methacholine. Endothelium independent relaxation was studied by the response to a single high concentration of sodium nitrite ($NaNO_2$; 10 mmol/L; yields approximately 10 nmol/L nitric oxide).[7] Vascular responsiveness to Angiotensin II was studied as described previously.[5;6] A reference response was evoked by stimulation with 60 mmol/L potassium chloride to produce maximal constriction.

Changes in lipid profile within groups were analyzed with paired-samples0 T-tests and between groups with independent-sample T Tests. Complete dose-response curves were tested using repeated measures analysis of variance test. All analyses were 2-tailed, and a p-value of ≤ 0.05 was considered to indicate statistical significance. Data are presented as mean values ± SD, unless otherwise specified. All statistical calculations were made with SAS software package v8.2 by SAS institute.

Results

Baseline characteristics of patients studied are presented in table 1. A segment of internal thoracic artery was collected from 116 (74%) patients. Of the collected vessels, adequate control responses could be evoked in 46 (40%) patients for the endothelial dependent relaxation and in 40 (34%) patients for the responsiveness to angiotensin II. However, there were no essential differences in baseline characteristics of patients with adequate control responses compared to the total population (table 1). On average patients were treated for 28 days. Effect of high and low dose statin treatment was comparable with the recently published Treating to New Targets Study (figure 1).[1]

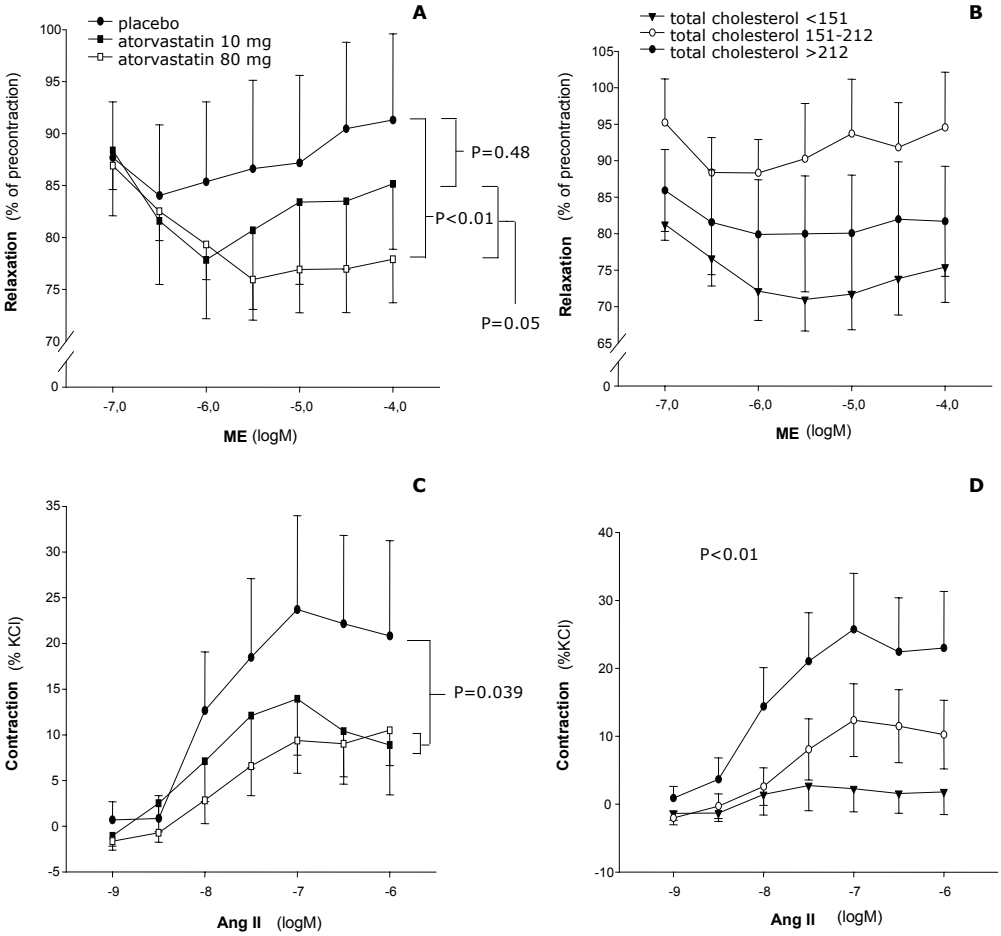
Endothelium measurements and vascular responses to angiotensin II are presented in figure 2. Endothelium dependent vasodilatation was improved with atorvastatin therapy (p=0.035), but significantly further improved with atorvastatin 80 mg as compared to 10 mg treated patients. Endothelial dependent vasodilatation was not associated with serum cholesterol levels 1 day before surgery. Endothelium-independent vasodilatation did not differ among the 3 groups (data not shown). Vascular responsiveness to angiotensin II was decreased by statin therapy, albeit not dose-dependently, and seemed to be strongly related to serum cholesterol levels of the day before surgery. Contractions to phenylephrine and KCl did not differ among groups (data not shown).

Discussion

Recently, the Treating to New Targets study established significant clinical benefit of intensive lipid-lowering therapy with 80 mg of atorvastatin per day as compared to treatment with 10 mg of atorvastatin.[1] Our findings suggest that this clinical benefit might be preceded with pathophysiologically relevant improvements of vascular function.

In good agreement with the current findings are previous experimental studies indicating that statins can dose-dependently increase the production of nitric oxide by activation of endothelial nitric oxide synthase and by increasing its mRNA half-life, independent of cholesterol levels.[8;9] Decreased endothelium

Figure 2.



Data are presented as Mean±SEM. A.) Endothelium dependent vasodilatation to methacholine (ME) of each group expressed as the percentage of displacement induced by phenelephrine precontraction. The endothelium dependent vasodilator response was improved in statin treated patients compared to placebo ($p<0.01$) and even further improved in patients on high dose atorvastatin ($p=0.05$), compared to placebo or low dose atorvastatin. B.) Endothelium dependent vasodilatation according to tertiles of cholesterol (non-significant). C.) Vascular responsiveness to angiotensin II (Ang II) of each group. Vasoconstrictor response to Ang II was reduced in statin treated patients as compared to placebo ($p<0.05$), but was not different between high and low dose atorvastatin treatment. D.) Vascular responsiveness to Ang II according to tertiles of cholesterol. Responses to Ang II were associated to serum cholesterol levels 1 day before surgery ($p<0.01$).

dependent relaxation in coronary and systemic arteries have been associated with cardiovascular risk factors and events.[10;11] Another novel finding of this study are the new insights on effects of statin treatment beyond endothelium function. Previously, we demonstrated in hypercholesterolemia a strong associated with upregulation of AT_1 receptor.[3;4] These findings support our notion that angiotensin II responsiveness is strongly associated with serum cholesterol levels. However, elucidation of the exact mechanism how statins, or

cholesterol, effects the responsiveness to angiotensin II and endothelium function is complicated by the functional interplay between nitric oxide and the renin-angiotensin system (for review see Yan[12]). Furthermore, the observed changes might only be one of many changes elicited by statin treatment resulting in reduction in clinical events.
A drawback of the present study was that vascular measurements were only obtained from 40% of the available vessels, which was lower than anticipated, based on our previous experiments.[5;6] We assume differences in current operation and preservation techniques of the arteries to be responsible for this difference, rather than differences in baseline characteristics (see table 1). Moreover, considering the principles of randomization and double-blinded treatments, and given that the decision of the surgeon not to obtain vascular material or the laboratory analyst judgement of control responses were not based on knowledge of the treatment patients received, the remaining analyzable vessels provided an unbiased comparison of the true effect of different dosage of atorvastatin versus placebo. This does not withstand that we are the first to report dose-dependent effects of atorvastatin with in vitro vascular measurements of human arteries.

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