Effects of atorvastatin and atorvastatin withdrawal on soluble CD40L and adipocytokines in patients with hypercholesterolaemia

Chih-Sheng CHU, Kun-Tai LEE, Ming-Yi LEE, Ho-Ming SU, Wen-Choi VOON, Sheng-Hsiung SHEU, Wen-Ter LAI

Division of Cardiology, Department of Internal Medicine, Kaohsiung Medical University Hospital, Kaohsiung Medical University, Kaohsiung, Taiwan, Republic of China.

Objective — Beyond lipid lowering, statins have pleiotropic effects with favourable benefits against atherogenesis. Withdrawal of statin therapy has been demonstrated to abrogate vascular protective activity and even increase the incidence of thrombotic vascular events. The purpose of this study is to investigate the serial changes of soluble CD40 ligand (sCD40L) and two adipocytokines, adiponectin and resistin, after short-term atorvastatin therapy and withdrawal in patients with hypercholesterolaemia.

Methods and results — Thirty-two patients with hypercholesterolaemia received atorvastatin 10 mg/day for 3 months. Serum lipid profiles, and levels of sCD40L, adiponectin and resistin, were assessed before and immediately after 3 months' statin therapy. Serum levels of sCD40L and adiponectin were also measured on the 3 consecutive days after statin withdrawal. After 3 months' statin therapy, levels of sCD40L (1.93 ± 1.13 vs. 1.30 ± 0.97 ng/mL), total cholesterol and low-density lipoprotein cholesterol (LDL-C) were all reduced significantly ($p < 0.05$). However, sCD40L level tended to increase towards baseline on the first and second days after statin withdrawal, but was not significantly elevated until the third day after withdrawal (1.89 ± 1.28 vs. 1.30 ± 0.97 ng/mL, $p < 0.05$). Total cholesterol and LDL-C levels did not increase during the 3 days of statin withdrawal. No significant changes of adiponectin and resistin levels were shown after statin therapy.

Conclusions — These results indicate that the effect of statin on sCD40L level was abrogated after therapy withdrawal, and was independent of serum cholesterol level. Statin therapy did not significantly alter levels of adiponectin and resistin. *(Acta Cardiol 2006; 61(3): 263-269)*

Keywords: *hypercholesterolaemia – atorvastatin – soluble CD40 ligand – adipocytokines.*

Introduction

Atherosclerosis is the underlying systemic disorder in most patients with cardiovascular or cerebrovascular disease. Vascular biologic studies of atherosclerosis have recently focused on inflammation in vascular walls, and the pathophysiology of atherosclerosis is now considered more complex than mere lipid storage problems[1,2]. From basic research and clinical evidence, the overall clinical benefits of 3-hydroxy-3-methylglutaryl-conenzyme A reductase inhibitors, or statins, appear to be greater than may be expected from changes in lipid profile alone[3]. Beyond lipid lowering, the pleiotropic effects of statins have been well demonstrated and include the following: improved endothelial function; enhanced plaque stability; reduced oxidative stress and vascular inflammation; and reduced platelet activity[3,4]. Furthermore, *in vitro* and animal studies have shown that withdrawal of statin therapy might abrogate vascular protection because of a negative influence on endothelial function[5-10]. Clinical studies also reported that statin withdrawal might impair endothelial function and increase thrombotic vascular events[11,12].

Soluble CD40 ligand (sCD40L) has recently been proposed as a new pro-inflammatory cytokine in atherosclerosis[13]. Structurally, CD40L is a transmembrane

Address for correspondence: Wen-Ter Lai, MD, Division of Cardiology, Department of Internal Medicine, Kaohsiung Medical University Hospital, 100 Tzyou 1st Rd, Kaohsiung, 80708, Taiwan. E-mail: wtlai@cc.kmu.edu.tw

Received 23 June 2005; revision accepted for publication 31 October 2005.

surface protein with homology to tumour necrosis factor-α; it can be shed and measured in the circulation. Both membrane-bound and soluble forms of this ligand interact with CD40, which is constitutively expressed on B cells, macrophages, endothelial cells, and vascular smooth muscle cells. Besides mediating crosstalk between T- and B-cells in the immune system, the CD40 signalling pathway also plays a central role in plaque stability, with the rupture of vulnerable plaques resulting in thrombotic vascular events[14]. Previous reports showed that sCD40L levels were elevated in patients with hypercholesterolaemia, unstable angina or acute coronary syndrome[14,15]. Risk prediction by assessment of sCD40L might therefore identify high-risk patients and provide prognostic information about the risks of recurrent myocardial infarction and death[15]. Statins interfered with CD40 expression in human endothelial-cell cultures, and attenuated the CD40L-induced proinflammatory response[16–18]. Increased levels of sCD40L have also been suggested as the molecular link between hypercholesterolaemia and the prothrombotic state, and statins may alleviate both of these conditions[19,20]. The effect of statins to downregulate sCD40L has been reported previously; however, the effects of statin withdrawal on sCD40L remain unknown.

Interactions between adipocytokines and endothelial cells in atherosclerosis have recently been advocated[24,25].Two novel adipocyte-derived cytokines, adiponectin and resistin, have been reported to have direct vascular effects[26]. For example, in apolipoprotein E-deficient mice, adiponectin reduced atherosclerosis[23], and in humans, low serum levels of adiponectin were associated with increased risks of coronary artery disease or myocardial infarction[27,28]. Statins were recently reported to have beneficial effects on insulin sensitivity in non-diabetic patients with dyslipidaemia[29]; and one paper suggested statin therapy had no influence on serum levels of adiponectin or resistin in patients with diabetics[30]. However, the effect of statin therapy on these two adipocytokines in dyslipidaemic patients remained controversial, and the effects of statin withdrawal on adiponectin also remain unknown.

The aims of this study were therefore to determine the effects of statin withdrawal on sCD40L levels and adiponectin, and to observe the effects of statin therapy on levels of adiponectin and resistin in patients with hypercholesterolaemia.

Methods

Study population

Thirty-two consecutive outpatients with hypercholesterolaemia (total cholesterol > 200 mg/dL and/or low-density lipoprotein cholesterol [LDL-C] > 130 mg/dL) were enrolled in the study. An initial questionnaire was used to identify cardiovascular risk factors, case histories, and concomitant medications. Patients with any acute illness, leukocytosis (> 10,000 white blood cells per mm^3), thrombocytosis (> 450,000 platelets per mm^3), chronic inflammatory disease, or any type of connective-tissue disease, were excluded. Patients with acute coronary syndrome within 6 months of enrolment were also excluded. The study protocol was approved by the local institutional review board, and all patients provided written informed consent.

Study protocol

After dietary control for 4 weeks, all patients started treatment with atorvastatin 10 mg/day. Concomitant therapies for other illnesses were administered and unchanged, if needed. After 3 months, if total cholesterol had been reduced to < 200 mg/dL, patients were asked to stop atorvastatin. Samples of venous blood (10 mL) were obtained before atorvastatin therapy, the day after completion of 3 months' atorvastatin therapy, and on each of 3 consecutive days after stopping atorvastatin therapy. All blood samples were taken in the morning after a 12-hour fast. Samples were frozen and stored at –76ºC until analysis.

Measurement of study parameters

Total cholesterol and triglyceride levels were determined by an enzymatic method, high-density lipoprotein cholesterol was measured after phosphotungstic acid/magnesium chloride precipitation of fresh plasma, and LDL-C was calculated by the Friedenwald formula. Levels of sCD40L were measured in duplicate by enzyme-linked immunosorbent assay (Alpha Diagnostic International, Inc., San Antonio, TX), as were plasma levels of adiponectin (B-Bridge International, Inc. Otsuka Pharmaceutical Co., Tokyo, Japan) and resistin (LINCO Research, Inc., St Charles, Missouri, USA). Intra-assay and inter-assay of coefficients of variation were 3.9% and 5.7%, respectively, for sCD40L; 3.6% and 5.8%, respectively, for adiponectin; and 3.7% and 5.9%, respectively, for resistin.

Statistical analysis

Data are presented as the mean ± SD. The values of lipid profiles were displayed in mg/dL. The levels of plasma adiponectin are displayed in μg/mL. The levels of serum sCD40L and resistin were displayed in ng/mL, respectively. The changes of study parameters before and after statin therapy were analysed by paired t-test. The changes of lipid profiles, sCD40L and

adiponectin during statin withdrawal periods were evaluated by mixed-model analysis or generalized-estimate equation for repeated-measurement datasets. All p values are 2-tailed, and all confidence intervals are computed at the 95% level. A p value < 0.05 was considered statistically significant. Statistical analysis was performed with SAS software version 8.12.

Results

Study population

Baseline characteristics for the study population are summarized in table 1. The mean age of patients (9 men and 23 women) was 60.3 ± 13.2 years; 18 patients had hypertension, 2 had diabetes mellitus, 5 had coronary artery disease, and 1 had peripheral vascular disease.

Table 1. – *Baseline characteristics for the study population (n = 32)*

Characteristic	Values
Age (yrs)	60.3 ± 13.2
Sex (M/F)	9/23
Body mass index (kg/m^2)	26.6 ± 2.8
Smokers [n (%)]	5 (15.6)
Past history [n (%)]	
Coronary artery disease	5 (15.6)
Diabetes mellitus	2 (6.2)
Hypertension	18 (56.3)
Peripheral vascular disease	1 (3.1)
Transient ischaemic attack/stroke	0 (0)
Concomitant medication [n (%)]	
Aspirin	4 (12.5)
Antihypertensive therapy	
ACE inhibitor/angiotensin-receptor blocker	8 (25.0)
β-blocker	9 (28.1)
Calcium-channel blocker	12 (37.5)
Diuretic	3 (9.4)
Other	4 (12.5)
Clopidogrel	0 (0)
Fibrate or niacin	0 (0)
Oral hypoglycaemic agent	2 (6.2)
PPAR-γ-agonist (glitazone)	0 (0)

ACE = angiotensin-converting enzyme; PPAR-γ = peroxisome proliferator-activated receptor-γ.

Daily aspirin was being taken by 4 patients, but no patients were taking clopidogrel, fibrates, niacin, or peroxisome proliferator-activated receptor-γ-agonists. Patients' concomitant oral medication was unchanged throughout the study.

Lipid-profile changes

Lipid parameters measured before and after 3 months' atorvastatin therapy, and on the first, second and third days after atorvastatin withdrawal, are shown in table 2 and figure 1. After 3 months' treatment, significantly lower values (vs. baseline) were evident for total cholesterol (184.4 ± 23.2 vs. 255.4 ± 32.1 mg/dL; p < 0.001) and LDL-C (122.1 ± 23.2 vs. 178.1 ± 32.1 mg/dL; p < 0.001). However, no significant changes were noted in these parameters during the 3 consecutive days after atorvastatin withdrawal. Levels of triglycerides and high-density lipoprotein cholesterol were not significantly altered during the study period.

Changes in sCD40L

Serial measurements of sCD40L levels during and after withdrawal of atorvastatin therapy are shown in table 3 and figure 2. At baseline, no correlations were identified between cholesterol profiles and sCD40L levels. Serum sCD40L level was significantly reduced from 1.93 ± 1.13 ng/mL to 1.30 ± 0.97 ng/mL after 3 months' atorvastatin therapy (p < 0.05). However, no linear association was identified between changes in sCD40L and total cholesterol (p = 0.259) and between changes in sCD40L and LDL-C (p = 0.330). After atorvastatin withdrawal, serum sCD40L was elevated on day 1 (1.79 ± 1.40 ng/mL vs. 1.30 ± 0.97 ng/mL; p = 0.098) and day 2 (1.75 ± 1.12 ng/mL vs. 1.30 ± 0.97 ng/mL; p = 0.077), but these increases were not statistically significant. Conversely, sCD40L was significantly elevated on day 3 after atorvastatin withdrawal (1.89 ± 1.28 ng/mL; p < 0.05).

Table 2. – *Lipid parameters before and after 3 months of atorvastatin 10 mg/day, and during first 3 days after atorvastatin withdrawal.*

Values	Before treatment	After treatment	After withdrawal		
			Day 1	Day 2	Day 3
Total cholesterol (mg/dL)	255.4 ± 32.1	184.4 ± 23.1*	182.8 ± 24.3*	186.4 ± 28.4*	192.3 ± 32.5*
LDL cholesterol (mg/dL)	178.1 ± 32.1	122.1 ± 23.2*	122.7 ± 20.7*	125.5 ± 28.4*	128.2 ± 32.5*
HDL cholesterol (mg/dL)	42.2 ± 35.8	37.5 ± 19.8	37.4 ± 22.2	38.2 ± 22.2	37.1 ± 22.8
Triglycerides (mg/dL)	155.6 ± 125.9	122.9 ± 72.3	109.0 ± 54.0	112.1 ± 54.5	125.9 ± 55.3

*p < 0.05 vs. pretreatment.

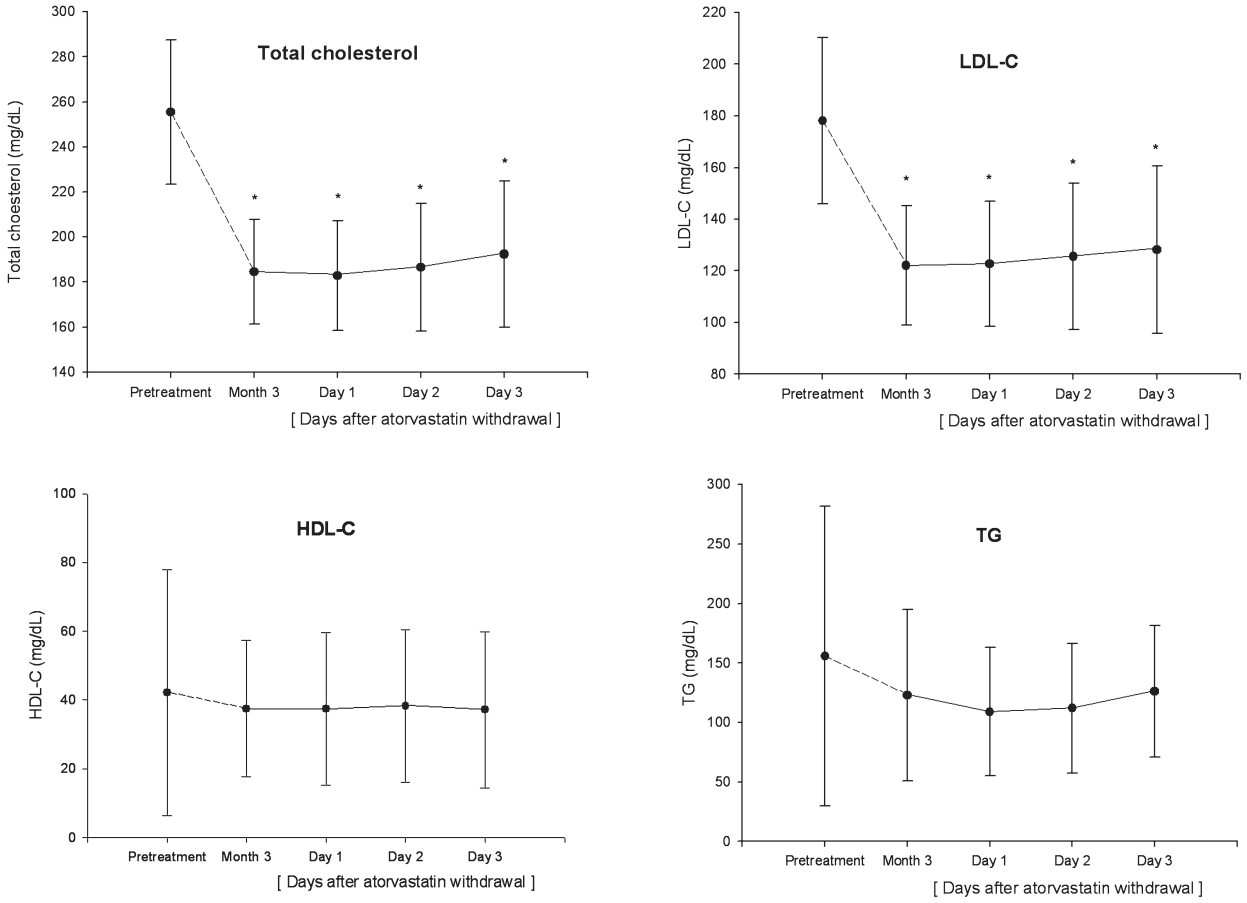

Fig. 1. – Lipid levels before and after 3 months' atorvastatin therapy, and during the 3 consecutive days after atorvastatin withdrawal. HDL-C = high-density lipoprotein cholesterol; LDL-C = low-density lipoprotein cholesterol; TG = triglycerides. *$p < 0.001$ vs. pretreatment.

Table 3. – *Serum levels of sCD40L, adiponectin and resistin before and after 3 months of atorvastatin 10 mg/day, and during first 3 days after atorvastatin withdrawal.*

Values	Before treatment	After treatment	After withdrawal		
			Day 1	Day 2	Day 3
sCD40L (ng/mL)	1.93 ± 1.13	1.30 ± 0.97*	1.79 ± 1.40	1.75 ± 1.12	1.89 ± 1.28†
Adiponectin (μg/mL)	4.48 ± 2.16	4.43 ± 2.37	5.02 ± 3.09‡	4.90 ± 2.69‡	4.68 ± 2.50‡
Resistin (ng/mL)	15.8 ± 8.03	15.5 ± 8.26	–	–	–

*$p < 0.05$ vs. pretreatment; †$p < 0.05$ vs. after treatment (month 3); ‡adiponectin levels after atorvastatin withdrawal were only available for 16 study participants.

Changes in adipocytokines

Serum levels of adiponectin and resistin before and after atorvastatin therapy are shown in table 3. No significant changes in serum levels of adiponectin (4.43 ± 2.37 μg/mL vs. 4.48 ± 2.16 μg/mL; $p = 0.434$) and resistin (15.5 ± 8.26 ng/mL vs. 15.8 ± 8.03 ng/mL; $p = 0.742$) were identified between measurements before and after atorvastatin therapy. In addition, in 16 randomly selected patients, no significant changes in serum levels of adiponectin were noted during atorvastatin withdrawal (figure 3).

Discussion

The major findings in this study were as follows:

- After 3 months' atorvastatin therapy in patients with hypercholesterolaemia, mean serum levels of sCD40L, total cholesterol, and LDL-C were significantly reduced; however, no linear relationship was demonstrated between changes in sCD40L and total cholesterol or LDL-C after 3 months' treatment.
- The serum sCD40L value was rapidly restored to its pretreatment level on treatment-withdrawal day 1,

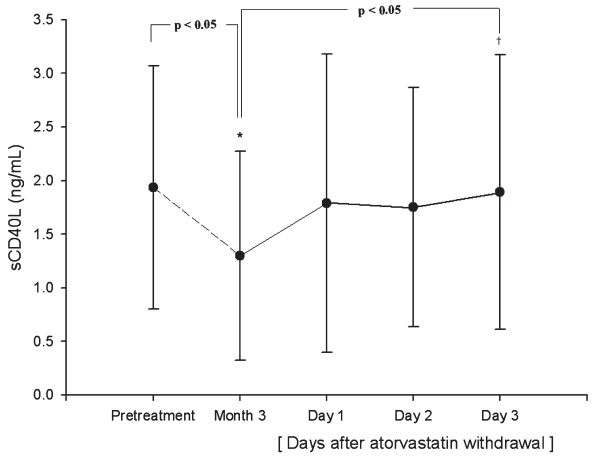

Fig. 2. – Serial levels of sCD40L in patients before and after atorvastatin therapy, and during 3 consecutive days of statin withdrawal. sCD40L = soluble CD40 ligand; $^{*}p < 0.05$ vs. pretreatment; $^{\dagger}p < 0.05$ vs. month 3.

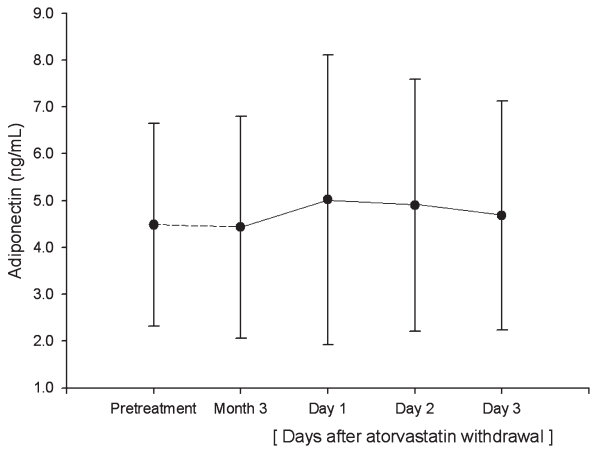

Fig. 3. – Serial levels of adiponectin before and after atorvastatin therapy, and on 3 consecutive days after atorvastatin withdrawal.

and increased significantly on withdrawal day 3; however, lipid parameters were not significantly altered during the 3-day withdrawal period.

- Atorvastatin therapy had no significant influence on serum levels of adiponectin or resistin.

The widespread use of statins for primary or secondary prevention in patients with high cardiovascular risk has become evidence-based practice. Meta-analyses of large cholesterol-lowering trials suggest that the overall clinical benefits of statins appear to be greater than might be expected from lipid-profile changes alone[3]. The pleiotropic effects of statins, beyond inhibition of cholesterol biosynthesis, involve various vascular biologic mechanisms and include the following: improved endothelial function and blood flow; decreased LDL oxidation; enhanced stability of atherosclerotic plaques; inhibition of vascular smooth muscle-cell proliferation and platelet aggregation; and reduced vascular inflammation[3,4].

The CD40 signal pathway is considered to be involved in the pathophysiologic link between hypercholesterolaemia, prothrombotic status, and plaque instability[1,13,33]. Increased levels of soluble and membrane-bound CD40 ligand have also been found in patients with unstable angina, and have been used as predictors of recurrent myocardial infarction and mortality[14,15]. In patients with moderate hypercholesterolaemia, upregulation of CD40 and CD40L may contribute to prothrombotic and proatherogenic status, whereas statin therapy may significantly reduce such status[19,20]. In this study, after 3 months' atorvastatin treatment, significant decreases in sCD40L were demonstrated in patients with hypercholesterolaemia. This result was comparable with previous reports and indicates that statins, besides lowering LDL-C, can also suppress sCD40L expression[20]. Comparison between the changes in sCD40L and those in total cholesterol or LDL-C revealed no significant linear associations. This may indicate that the sCD40L reduction was not totally dependent on lipid-profile decreases *per se*. The anti-inflammatory effects of atorvastatin may have contributed to the reduced level of sCD40L.

Withdrawal of statin therapy has been reported to have some adverse effects[5-12]. For example, increased thrombotic vascular events were observed in patients with established atherosclerosis after a switch from simvastatin to fluvastatin, an agent with less potency[11]. Furthermore, in a subgroup analysis of the Platelet Receptor Inhibition in Ischemic Syndrome Management (PRISM) study, withdrawal of statin therapy in patients with acute coronary syndrome increased cardiac risk 3-fold compared with continuation of statin therapy[12]. Laufs *et al.*[9] also demonstrated that statin withdrawal impaired vascular protection and induced a rebound-like decrease in the availability of nitric oxide. Suppression of endothelial NO production after statin withdrawal may be mediated by negative feedback regulation of Rho GTPase gene transcription[5], and an effect of statins against plaque instability was proposed to be mediated via RhoA GTPase[3]. Aukrust *et al.*[14] also reported that elevated sCD40L was related to endothelial adhesion molecules in patients with acute coronary syndrome.

In the present study, after 3 months' atorvastatin therapy, serum levels of sCD40L, total cholesterol, and LDL-C in patients with hypercholesterolaemia were significantly reduced. These findings were consistent with previous reports[19,20]. During atorvastatin withdrawal, serum sCD40L level began to rise on day 1, although not becoming significantly elevated until day 3. Of note, serum levels of total cholesterol and LDL-C were not significantly altered during the 3-day period of atorvastatin withdrawal. Our findings of increased sCD40L during atorvastatin withdrawal may indicate that (1) the beneficial effects of statin therapy

against plaque instability would be abolished earlier than statin effects on lipid profile, and may explain some of the adverse effects in the abovementioned reports of statin withdrawal; (2) the serum sCD40L level within 3 days of atorvastatin withdrawal was not greater than the baseline sCD40L value, which indicated no rebound increase in sCD40L; (3) after atorvastatin withdrawal, the dissociation of increased serum sCD40L level from increased LDL-C level further supported the notion that atorvastatin had a pleiotropic, anti-inflammatory effect that was unrelated to serum cholesterol level; (4) in addition, this result suggested that the biologic half-life of the effect of statin therapy on LDL-C was longer than that on sCD40L, and further suggests that statins may have more prominent effects on cholesterol than on sCD40L.

The adipocytokines adiponectin and resistin were recently identified as modulators in atherogenesis and metabolic syndrome[21,24]. Recent studies demonstrated direct effects of adiponectin, resistin and C-reactive protein on vascular endothelial cells[25,26]. Furthermore, the plasma concentration of adiponectin was significantly reduced in patients with coronary artery disease, and hypoadiponectinaemia was reported to be associated with myocardial infarction in men[22,28]. In an *in vivo* study, adiponectin reduced atherosclerosis in apolipoprotein E-deficient mice[23]. Resistin is expressed in adipose tissue, and its expression is strongly correlated with insulin resistance[24]. In addition, induction of vascular cellular adhesion molecule-1 by resistin was reported to be inhibited partially by adiponectin, and partially by pitavastatin[25], and Jove *et al.*[31] reported that the cholesterol-lowering effect of fenofibrate therapy correlated negatively with resistin expression in human adipose tissue. However, there are no data in the literature that focus directly on the lipid-lowering effects of statin therapy in relation to adiponectin and resistin changes.

Results from the present study reveal that 3 months' atorvastatin therapy had no significant effect on adiponectin and resistin, while total cholesterol and LDL-C decreased significantly. The pleiotropic, antiatherosclerotic effects of statins are believed to be mediated by reduced production of farnesyl- and geranylgeranyl-pyrophosphates[5]. Meanwhile, another drug class, thiazolidinediones (peroxisome proliferators-activated receptor [PPAR]-γ-agonists), was reported to influence the expression and release of adiponectin and resistin originating from adipose tissue, and with the net result of improved insulin activity[32]. The absence of change in adiponectin and resistin levels after statin therapy suggests that statins exhibit pleiotropic effects on atherosclerotic process independent of PPAR-γ pathway. Several questions remain unanswered. For examples, are changes in adiponectin and resistin likely to reflect cardiovascular risk only in specific patient groups? Are there some missing molecular links between statin therapy and these 2 adipocytokines? And is combination therapy with statins and PPAR-γ-agonists likely to have additive benefits on vascular inflammation via different pathways?

This study had several limitations. First, the number of subjects was relatively small, and some of the sCD40L changes did not parallel lipid-profile changes. A larger number of subjects may therefore be needed to confirm our findings. Second, the clinical condition was not correlated with sCD40L changes. Most patients enrolled in this study had no evidence of coronary artery disease, peripheral artery disease, or stroke, and the follow-up period after atorvastatin withdrawal was short. It may therefore be difficult to evaluate the correlation between increased sCD40L level and clinical outcome. Third, most of our patients with hypercholesterolaemia were outpatients without high cardiovascular risk. Further analysis clarifying the interaction between statin therapy and adiponectin and resistin levels in diabetics and/or individuals with high body mass index is warranted.

In summary, 3 months' atorvastatin therapy significantly reduced serum levels of sCD40L, total cholesterol and LDL-C. During a 3-day period immediately after statin withdrawal, the serum level of sCD40L increased towards the pretreatment value, while lipid profiles remained unchanged. These findings indicate that the pleiotropic effect of statin therapy on sCD40L was abolished after statin withdrawal and was independent of serum cholesterol level. In addition, statin therapy had no significant effect on serum levels of adiponectin and resistin.

Acknowledgement

This study was supported by research grant 93-KMUH-049 from Kaohsiung Medical University Hospital. The authors are grateful to Mon-Chang Wu and Nai-Won Kuo for data collection, and to Hung-Yi Chuang for assistance with statistical analysis.

References

1. Libby P. Vascular biology of atherosclerosis: Overview and state of the art. *Am J Cardiol* 2003; **91**: 3A-6A.
2. Libby P. Inflammation in atherosclerosis. *Nature* 2002; **420**: 868-74.
3. Takemoto M, Liao JK. Pleiotropic effects of 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors. *Arterioscler Thromb Vasc Biol* 2001; **21**: 1712-9.
4. Laufs U, Liao JK. Targeting Rho in cardiovascular disease. *Circ Res* 2000; **87**: 526-8.
5. Laufs U, Endres M, Custodis F, Gertz K, Nickenig G, Liao JK, Bohm M. Suppression of endothelial nitric oxide

production after withdrawal of statin treatment is mediated by negative feedback regulation of rho GTPase gene transcription. *Circulation* 2000; **102**: 3104-10.
6. Brandes RP, Beer S, Ha T, Busse R. Withdrawal of cerivastatin induces monocyte chemoattractant protein 1 and tissue factor expression in cultured vascular smooth muscle cells. *Arterioscler Thromb Vasc Biol* 2003; **23**: 1794-1800.
7. Gertz K, Laufs U, Lindauer U, Nickenig G, Bohm M, Dirnagl U, Endres M. Withdrawal of statin treatment abrogates stroke protection in mice. *Stroke* 2003; **34**: 551-7.
8. Vecchione C, Brandes RP. Withdrawal of 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors elicits oxidative stress and induces endothelial dysfunction in mice. *Circ Res* 2002; **91**: 173-9.
9. Laufs U, Wassmann S, Hilgers S, Ribaudo N, Bohm M, Nickenig G. Rapid effects on vascular function after initiation and withdrawal of atorvastatin in healthy, normocholesterolemic men. *Am J Cardiol* 2001; **88**: 1306-7.
10. Puccetti L, Pasqui AL, Pastorelli M, Bova G, Di Renzo M, Leo A, Cercignani M, Palazzuoli A, Auteri A, Bruni F. Platelet hyperactivity after statin treatment discontinuation. *Thromb Haemost* 2003; **90**: 476-82.
11. Thomas M, Mann J. Increased thrombotic vascular events after change of statin. *Lancet* 1998; **352**: 1830-1.
12. Heeschen C, Hamm CW, Laufs U, Snapinn S, Bohm M, White HD. Platelet Receptor Inhibition in Ischemic Syndrome Management (PRISM) investigators. Withdrawal of statins increases event rates in patients with acute coronary syndromes. *Circulation* 2002; **105**: 1446-52.
13. Schonbeck U, Libby P. CD40 signaling and plaque instability. *Circ Res* 2001; **89**: 1092-103.
14. Aukrust P, Muller F, Ueland T, Berget T, Aaser E, Brunsvig A, Solum NO, Forfang K, Froland SS, Gullestad L. Enhanced levels of soluble and membrane-bound CD40 ligand in patients with unstable angina. Possible reflection of T lymphocyte and platelet involvement in the pathogenesis of acute coronary syndromes. *Circulation* 1999; **100**: 614-20.
15. Varo N, de Lemos JA, Libby P, Morrow DA, Murphy SA, Nuzzo R, Gibson CM, Cannon CP, Braunwald E, Schonbeck U. Soluble CD40L: risk prediction after acute coronary syndromes. *Circulation* 2003; **108**: 1049-52.
16. Mulhaupt F, Matter CM, Kwak BR, Pelli G, Veillard NR, Burger F, Graber P, Luscher TF, Mach F. Statins (HMG-CoA reductase inhibitors) reduce CD40 expression in human vascular cells. *Cardiovasc Res* 2003; **59**: 755-66.
17. Wagner AH, Gebauer M, Guldenzoph B, Hecker M. 3-hydroxy-3-methylglutaryl coenzyme A reductase-independent inhibition of CD40 expression by atorvastatin in human endothelial cells. *Arterioscler Thromb Vasc Biol* 2002; **22**: 1784-9.
18. Schonbeck U, Gerdes N, Varo N, Reynolds RS, Horton DB, Bavendiek U, Robbie L, Ganz P, Kinlay S, Libby P. Oxidized low-density lipoprotein augments and 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors limit CD40 and CD40L expression in human vascular cells. *Circulation* 2002; **106**: 2888-93.
19. Cipollone F, Mezzetti A, Porreca E, Di Febbo C, Nutini M, Fazia M, Falco A, Cuccurullo F, Davi G. Association between enhanced soluble CD40L and prothrombotic state in hypercholesterolemia: effects of statin therapy. *Circulation* 2002; **106**: 399-402.
20. Semb AG, van Wissen S, Ueland T, Smilde T, Waehre T, Tripp MD, Froland SS, Kastelein JJ, Gullestad L, Pedersen TR, Aukrust P, Stalenhoef AF**.** Raised serum levels of soluble CD40 ligand in patients with familial hypercholesterolemia: downregulatory effect of statin therapy. *J Am Coll Cardiol* 2003; **41**: 275-9.
21. Ouchi N, Kihara S, Arita Y, Maeda K, Kuriyama H, Okamoto Y, Hotta K, Nishida M, Takahashi M, Nakamura T, Yamashita S, Funahashi T, Matsuzawa Y. Novel modulator for endothelial adhesion molecules: adipocyte-derived plasma protein adiponectin. *Circulation* 1999; **100**: 2473-6.
22. Ouchi N, Kihara S, Arita Y, Okamoto Y, Maeda K, Kuriyama H, Hotta K, Nishida M, Takahashi M, Muraguchi M, Ohmoto Y, Nakamura T, Yamashita S, Funahashi T, Matsuzawa Y. Adiponectin, an adipocyte-derived plasma protein, inhibits endothelial NF-kappaB signaling through a cAMP-dependent pathway. *Circulation* 2000; **102**: 1296-301.
23. Okamoto Y, Kihara S, Ouchi N, Nishida M, Arita Y, Kumada M, Ohashi K, Sakai N, Shimomura I, Kobayashi H, Terasaka N, Inaba T, Funahashi T, Matsuzawa Y. Adiponectin reduces atherosclerosis in apolipoprotein E-deficient mice. *Circulation* 2002; **106**: 2767-70.
24. Verma S, Li SH, Wang CH, Fedak PW, Li RK, Weisel RD, Mickle DA. Resistin promotes endothelial cell activation: further evidence of adipokine-endothelial interaction. *Circulation* 2003; **108**: 736-40.
25. Kawanami D, Maemura K, Takeda N, Harada T, Nojiri T, Imai Y, Manabe I, Utsunomiya K, Nagai R. Direct reciprocal effects of resistin and adiponectin on vascular endothelial cells: a new insight into adipocytokine-endothelial cell interactions. *Biochem Biophy Res Commun* 2004; **314**: 415-9.
26. Ouchi N, Kihara S, Funahashi T, Nakamura T, Nishida M, Kumada M, Okamoto Y, Ohashi K, Nagaretani H, Kishida K, Nishizawa H, Maeda N, Kobayashi H, Hiraoka H, Matsuzawa Y. Reciprocal association of C-reactive protein with adiponectin in blood stream and adipose tissue. *Circulation* 2003; **107**: 671-4.
27. Kumada M, Kihara S, Sumitsuji S, Kawamoto T, Matsumoto S, Ouchi N, Arita Y, Okamoto Y, Shimomura I, Hiraoka H, Nakamura T, Funahashi T, Matsuzawa Y; Osaka CAD Study Group. Coronary artery disease. Association of hypoadiponectinemia with coronary artery disease in men. *Arterioscler Thromb Vasc Biol* 2003; **23**: 85-9.
28. Pischon T, Girman CJ, Hotamisligil GS, Rifai N, Hu FB, Rimm EB. Plasma adiponectin levels and risk of myocardial infarction in men. *JAMA* 2004; **291**: 1730-7.
29. Sonmez A, Baykal Y, Kilic M, Yilmaz MI, Saglam K, Bulucu F, Kocar IH. Fluvastatin improves insulin resistance in non-diabetic dyslipidemic patients. *Endocrine* 2003; **22**: 151-4.
30. Shetty GK, Economides PA, Horton ES, Mantzoros CS, Veves A. Circulating adiponectin and resistin levels in relation to metabolic factors, inflammatory markers, and vascular reactivity in diabetic patients and subjects at risk of diabetes. *Diabetes Care* 2004; **10**: 2450-7.
31. Jove M, Planavila A, Cabrero A, Novell F, Ros E, Zambon D, Laguna JC, Carrera MV. Reductions in plasma cholesterol levels after fenofibrate treatment are negatively correlated with resistin expression in human adipose tissue. *Metabolism* 2003; **52**: 351-5.
32. Stumvoll M, Haring HU. Glitazones. Clinical effects and molecular mechanism. *Ann Med* 2002; **34**: 217-24.
33. Sanguigni V, Pignatelli P, Lenti L, Ferro D, Bellia A, Carnevale R, Tesauro M, Sorge R, Lauro R, Violi F. Short-term treatment with atorvastatin reduces platelet CD40 ligand and thrombin generation in hypercholesterolemia patients. *Circulation* 2005; **111**: 412-9.