Review

Molecular Mechanisms of HDL-Cholesterol Elevation by Statins and Its Effects on HDL Functions

Shizuya Yamashita¹, Kazumi Tsubakio-Yamamoto¹, Tohru Ohama¹, Yumiko Nakagawa-Toyama², and Makoto Nishida²

¹Department of Cardiovascular Medicine, Osaka University Graduate School of Medicine, Osaka, Japan
²Health Care Center, Osaka University, Osaka, Japan

Numerous large-scale clinical studies have revealed that the low-density lipoprotein cholesterol (LDL-C)-lowering effect of 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) reductase inhibitors (statins) prevents coronary heart disease (CHD). Statins have not only LDL-C-lowering effects but also high-density lipoprotein cholesterol (HDL-C)-elevating effects, which differ among statins. In this article, we discuss the molecular mechanisms of HDL-C elevation by statins and its effect on HDL functions. We summarize the reports to date on the effects of statins on various proteins, enzymes and receptors involved in reverse cholesterol transport (RCT), which is one of the protective systems against atherosclerosis. Since statins increase the synthesis of apolipoprotein A-I (ApoA-I) and HDL neogenesis in the liver, the HDL-C-increasing effect of statins may reflect RCT activation. Moreover, HDL has pleiotropic effects, including anti-inflammatory and anti-oxidative effects, as well as RCT. In the future, it may be necessary to assess the functions of HDL elevated by statins, and select statins based on differences in their effects in clinical practice.


Key words; Reverse cholesterol transport, Apolipoprotein A-I, ATP-binding cassette transporter A1, Adiponectin, Cholesteryl ester transfer protein

Introduction

HDL is a lipoprotein that plays a central role in reverse cholesterol transport (RCT). Hypo-HDL-cholesterolemia has long been recognized as a strong and independent risk factor for coronary heart disease (CHD). Epidemiologically, a low level of high-density lipoprotein cholesterol (HDL-C) is associated with an increased risk for CHD, even when the low-density lipoprotein cholesterol (LDL-C) level is low. In addition, a low level of HDL-C is reportedly related to increased mortality.

The main roles of HDL are to collect excess cholesterol stored in peripheral tissues, such as lipid-laden foam cells, and transport it to the liver for excretion into bile. Apolipoprotein A-I (ApoA-I) produced in the liver and the small intestine, and free ApoA-I released during hydrolysis of triglyceride (TG)-rich lipoproteins bind cholesterols and phospholipids through ATP-binding cassette transporter A1 (ABCA1) to form pre-β HDL. Cholesterol extracted from peripheral cells are converted to cholesterol esters (CEs) by the action of lecithin:cholesterol acyltransferase (LCAT) to enter HDL particles and form HDL. Mature HDL produced after repeated cholesterol extraction and conversion to CE is returned to the liver through LDL receptors while CE is transported to VLDL (very low density lipoprotein) and LDL by cholesteryl ester transfer protein (CETP); however, HDL has recently been speculated to have various properties, including anti-inflammatory, anti-oxidative, anti-coagulation and vascular endothelial function-improving effects in addition to RCT (Fig. 1). These effects may comprehensively be involved in the prevention of atherosclerosis progression.
among the drugs clinically available at present and are used for the prevention of CHD worldwide; however, since the incidence of cardiovascular events has been shown to be relatively high in patients with low HDL-C levels even during statin treatment, the levels of HDL-C must also be paid attention during statin treatment. Concerning the effects of statins on levels of HDL-C, although statins clinically increase HDL-C levels, there is little evidence regarding their mechanism and the significance of their actions as compared to those pertaining to LDL-C. In this article, we discuss the usefulness of the statin-induced rise in HDL-C levels and review the findings to date on the effects on HDL functions and mechanisms of action, as well as the differences in effects among statins.

**Clinical Significance of Statin-Induced HDL-Cholesterol-Elevating Effect**

HDL levels during statin treatment are also important for predicting cardiovascular events. In the J-LIT study in which simvastatin was used, HDL levels below 40 mg/dL were associated with an increased risk for cardiovascular events. Moreover, sub-analysis of the TNT study showed that the incidence of cardiovascular events tended to increase even in patients with HDL-C levels below 40 mg/dL, who had LDL-C levels below 70 mg/dL while receiving statin treatment. These findings merit close investigation because it was unclear whether the changes were induced by low HDL-C levels in the subjects or by an insufficient HDL-C-elevating effect of the statins; however, several statin intervention studies have revealed the regression of coronary artery plaque volume to be related to the HDL-C-elevating effect of statins. The study of pravastatin showed that the decrease in plaque volumes correlated significantly with increased HDL-C levels. The COSMOS study, in which rosuvastatin was administered to patients with stable CHD, showed that plaque regression occurred in 60% of patients, regardless of their LDL-C levels and that there was a significant correlation between changes in HDL-C levels, and changes in plaque volumes, suggesting that a rise in HDL-C level leads to regression of coronary artery plaques. In the J-LIT study, the incidence of coronary events was decreased by 37.5% for primary prevention and 28.3% for secondary prevention, respectively, with each HDL-C elevation of 10 mg/dL. Therefore, the HDL-C-elevating effect of statins is expected to lead to the regression of coronary artery plaque volumes and prevention of coronary events.

**HDL-C-Elevating Effects Differ Among Statins**

**Relationships between Statin doses and HDL-C-Elevating Effects**

Fig. 2 summarizes the statin doses and HDL-C-elevating effects. The LDL-C-lowering effect, the main action of statins, is dose-dependent. Strong statins, including atorvastatin, pitavastatin and rosuvastatin, can reduce LDL-C levels by 40% or more. On the other hand, statins that produce a greater percent change in LDL-C do not necessarily show a greater increase in HDL-C levels. Moreover, the dose-dependent effect is not apparent in the rise in HDL-C
levels. There is a report suggesting that atorvastatin at a higher dose has a less potent HDL-C-elevating effect\(^{19, 20}\). The degrees of HDL-C-elevating effects differ among the types and doses of statins.

**Changes Over Time in HDL-C- and ApoA-I-Elevating Effects with Long-Term Statin Treatment**

Fig. 3 and 4 summarize the percent changes in HDL-C and ApoA-I levels with long-term statin treatment. A study that compared rosuvastatin, pravastatin and simvastatin showed no significant differences in the effects of these statins on HDL-C and ApoA-I levels\(^{21}\). In contrast, rosuvastatin 10 mg/day showed a greater HDL-C-elevating effect than atorvastatin 10 mg/day at 52 weeks\(^{22}\). Pitavastatin 2 mg/day had significantly greater HDL-C- and ApoA-I-elevating effects than atorvastatin 10 mg/day\(^{23}\). Pitavastatin, even as a replacement for other statins, significantly increases HDL-C levels and maintains the effect for a long time\(^{24, 25}\). These differences may be associated with the differing effects of statins on various factors involved in HDL metabolism.

**HDL Metabolism and the Influences of Statins**

**Production of HDL in the Liver**

1) **ApoA-I**

ApoA-I, a main component protein of HDL, is synthesized in the liver and intestine and by the catabolism of mature HDL, and serves as the starting point of RCT. Administration of statins increases ApoA-I levels\(^{23, 26}\). Statins raise the amount of ApoA-I mRNA via suppression of the Rho signalling pathway and activation of peroxisome proliferator-activated receptor alpha (PPAR \(\alpha\)) in HepG2 cells\(^{27}\) (Fig. 5). Comparisons among simvastatin, atorvastatin and pitavas...
tatin showed the lowest dose of pitavastatin to increase ApoA-I secretion from HepG2 cells\(^{28}\). The estimated distributions of each statin in the liver were 6.4 \(\mu M\) for simvastatin, 2.5 to 13.1 \(\mu M\) for atorvastatin and 5.3 \(\mu M\) for pitavastatin (Table 1). Pitavastatin was the only statin affecting ApoA-I secretion at a clinically used dose in the study by Maejima et al.\(^{28}\). This difference is assumed to be one of the explanations for HDL-C-elevating effects differing among statins.

### 2) ABCA1 in the Liver

ABCA1 in the liver transports cholesterol within cells to ApoA-I to form pre-\(\beta\) HDL. Statins increase the amount of ABCA1 mRNA in HepG2 cells\(^{28, 29}\). Although the mechanisms by which ABCA1 increases are not completely understood, pravastatin may activate ABCA1 gene expression in the liver via sterol regulatory element-binding protein 2 (SREBP-2) and the liver X receptor (LXR)\(^{30}\) (Fig. 5).

### 3) LCAT

LCAT is involved in the maturation of pre-\(\beta\) HDL to HDL via the esterification of free cholesterol of pre-\(\beta\) HDL\(^{31}\). The influences of statins on LCAT activities are still controversial\(^{32-40}\).

### Cholesterol Efflux from Peripheral Cells

#### 1) ABCA1 in Macrophages

ABCA1 in peripheral cells such as macrophages not only transports cholesterol within cells to ApoA-I to form pre-\(\beta\) HDL, but is also involved in cholesterol efflux from peripheral cells. A study showed that statins elevate the expressions of ABCA1 and ATP-binding cassette transporter G1 (ABCG1)\(^ {31}\), while other studies have found different effects\(^ {42-45}\). The effects of statins on ABC transporters are assumed to change depending on the cholesterol content stored in cells\(^ {45}\).

#### 2) ABCG1

ABCG1 in peripheral cells, such as macrophages, binds to HDL and pumps out cholesterol. Similar to ABCA1, the influences of statins on ABCG1 are controversial\(^ {41-45}\).

#### 3) Peripheral Cell-Type Scavenger Receptor Class B Type I (SR-BI)

Although SR-BI expressed in peripheral cells, such as macrophages, recognizes ApoA-I to form HDL particles, it is unclear whether this is clinically involved in cholesterol efflux; however, the amount of SR-BI expression reportedly correlates with the amount of cholesterol efflux\(^ {46}\). SR-BI may be one of the molecules determining HDL-C levels. Statins stimulate cholesterol efflux by increasing SR-BI expression in peripheral cells\(^ {47-49}\). Since the regulation of SR-BI expression involves SREBP-1\(^ {50}\), statins are assumed to regulate SR-BI expression via SREBP-1.

Few clinical results have been obtained on cholesterol efflux from peripheral cells before versus after administration of statins. It has only been reported that CE efflux capacity from macrophages was stable in an investigation of serum before and after simvastatin administration in patients with type 1 diabetes mellitus. In that study, HDL-C levels were elevated by simvastatin, pre-\(\beta\) HDL levels tended to decrease and CETP activity was reduced\(^ {51}\). Further evaluation of CE efflux with statin treatment is therefore needed.

### Transport of CE in HDL to the Liver

#### 1) CETP

CETP facilitates the operation of the atherosclerosis prevention system via HDL and ApoA-I. Mature HDL produced by repeated cholesterol efflux and

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**Table 1. Estimated distributions of statins in the liver**

<table>
<thead>
<tr>
<th></th>
<th>Lovastatin</th>
<th>Rosuvastatin</th>
<th>Cerivastatin</th>
<th>Pravastatin</th>
<th>Atorvastatin</th>
<th>Pitavastatin</th>
<th>Simvastatin</th>
<th>Fluvastatin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cmax (ng/mL)</td>
<td>2.7</td>
<td>19</td>
<td>3.2</td>
<td>45–66</td>
<td>13–67</td>
<td>41</td>
<td>6.9</td>
<td>200–440</td>
</tr>
<tr>
<td>Statin dose (mg)</td>
<td>40</td>
<td>40</td>
<td>0.3</td>
<td>40</td>
<td>40</td>
<td>2</td>
<td>40</td>
<td>40</td>
</tr>
<tr>
<td>Estimated Cmax (nM)(^ {8})</td>
<td>6.7</td>
<td>39.5</td>
<td>6.9</td>
<td>106.2–155.8</td>
<td>23.3–120.1</td>
<td>97.6</td>
<td>164.8</td>
<td>487.2–1,072.0</td>
</tr>
<tr>
<td>Liver/plasma Distribution(^ {88})</td>
<td>-</td>
<td>25.0</td>
<td>173.7</td>
<td>21.5</td>
<td>108.7</td>
<td>53.9</td>
<td>38.9</td>
<td>29.5</td>
</tr>
<tr>
<td>Estimated human liver distribution ((\mu M))(^ {888})</td>
<td>-</td>
<td>1.0</td>
<td>1.2</td>
<td>2.3–3.3</td>
<td>2.5–13.1</td>
<td>5.3</td>
<td>6.4</td>
<td>14.4–31.6</td>
</tr>
<tr>
<td>T1/2 (hour)</td>
<td>-</td>
<td>20</td>
<td>3.2</td>
<td>1.8–2.0</td>
<td>7.8–21</td>
<td>13</td>
<td>3.5</td>
<td>0.8–2.4</td>
</tr>
</tbody>
</table>

Abbreviations: Cmax, human maximum concentration; T1/2, half-life.

\(^ {8}\): Estimated Cmax (nM) was calculated from Cmax (ng/mL) and statin dose in reference 136.

\(^ {88}\): Liver/plasma distribution are calculated from reference 137–143.

\(^ {888}\): Estimated human liver distributions were calculated from human blood levels and using liver/plasma ratios in rats.
conversion to CE is indirectly returned to the liver by LDL receptors after CE in HDL is transported to ApoB-containing particles (VLDL, IDL and LDL) by CETP. Many studies have shown that statins inhibit CETP mass and activities [51-55]. CETP gene expression is regulated by LXR [56] and reacts to changes in the cholesterol amount in cells along with plasma cholesterol levels [57]; therefore, suppression of cholesterol synthesis and decreased cholesterol amounts in cells may reduce LXR expression, leading to decreased gene expression of CETP; however, another study showed decreased CETP activity without reduced CETP mass [58], suggesting that statins have a direct inhibitory action on CETP activity. On the other hand, statins significantly decrease ApoB-containing lipoproteins and TG. Thus, decreased CETP activities may only reflect decreased lipoproteins that are substrates of CETP.

2) SR-BI in the Liver
CE in HDL is directly returned to the liver via SR-BI in the liver that recognizes ApoA-I to selectively take up CE. Atorvastatin did not significantly change SR-BI expression in the livers of dogs and mice [63, 69] and there are no reports on the effects of other statins on SR-BI expression.

Enzymes Involved in Catabolism of TG-Rich Lipoproteins and HDL
1) Lipoprotein Lipase (LPL)
LPL is produced in adipose tissue, skeletal muscles and the heart, and hydrolyzes TG-rich lipoproteins in blood on the surfaces of vascular endothelial cells to produce HDL [60]. A rise in LPL activities due to genetic factors is known to raise HDL-C levels [61, 62]. A clinical study showed that atorvastatin, simvastatin and pravastatin increased LPL mass or activities in patients with type II diabetes mellitus [53-65]. Basic research using 3T3-L1 preadipocytes revealed that simvastatin and pitavastatin elevated LPL activities [66]. It has been proposed, as a mechanism, that enhanced expressions of PPAR α and peroxisome proliferator-activated receptor gamma (PPAR γ) by statins increase the expressions of LPL mRNA and proteins [63].

2) Hepatic Lipase (HL)
HL hydrolyzes phospholipids and TGs to convert HDL2 into more dense HDL3 [67, 68]. Atorvastatin dose-dependently shows HL activity-lowering effects exerted via an unknown mechanism (−11% at a dose of 10 mg/day and −22% at a dose of 80 mg/day) [69]. Thus, cholesterol in the HDL2 fraction may rise.

3) Endothelial Lipase (EL)
EL has phospholipase activity [70, 71] and hydrolyzes phospholipids in HDL particles [72]. HDL with decreased phospholipids is prone to decomposition and is metabolized, resulting in decreased HDL via the action of EL. EL is one of the factors promoting HDL catabolism. Pitavastatin decreases the serum EL mass through isoprenylation and suppression of RhoA [73]. Atorvastatin and simvastatin also suppress EL expression in macrophages [74, 75].

Other Factors Involved in RCT
1) Adiponectin
Adiponectin is one of the important molecules, secreted from adipocytes, that inhibits the progression of atherosclerosis. Adiponectin levels correlate positively with HDL-C levels [76]. Adiponectin promotes HDL production via ApoA-I production and increased ABCA1 expression in the liver [77, 78], and also in macrophages to activate RCT [79]. Pitavastatin and pravastatin elevate adiponectin expression in adipocytes [80]. Clinically, the adiponectin-elevating effects differ among statins [81-84], and pitavastatin reportedly increases adiponectin levels in hyperlipidemic patients with diabetes mellitus. Suppression of reactive oxygen species (ROS) production, and activation of SREBP1c and PPAR γ have been proposed as possible mechanisms [85].

Taken together, statins influence a variety of molecules involved in HDL metabolism and RCT. Thus, the effects of statins are illustrated and summarized in Fig. 6.

Effects of Statins on Properties and Functions of HDL Particles

Functional HDL and Dysfunctional HDL
As described above, HDL-C elevation by statins provides good clinical results; however, for some drugs, HDL-C elevation does not necessarily lead to a clinical benefit. Large-scale clinical studies, such as ILLUMINATE [86] and ILLUSTRATE [87], were conducted to prevent the occurrence and progression of atherosclerosis by raising HDL-C levels with the use of a CETP inhibitor, torcetrapib. Neither study showed inhibitory effects on atherosclerosis progression in coronary and carotid arteries. In fact, these studies were discontinued after only 1 year due to a significant increase in deaths from any cause, including cardiovascular death. It has been proposed that elevated blood pressure caused by increased aldosterone, induced during torcetrapib administration, was responsible for this excess mortality [88]. Other CETP
inhibitors under development are reported to have no blood pressure-elevating effect\(^8\)). It is unclear whether this result was attributable to a class effect of CETP inhibitors or an adverse effect specific to torcetrapib\(^8, 87\); however, these studies revealed not only a significant decrease in LDL-C levels but also a marked increase in HDL-C levels, suggesting that HDL-C elevation does not necessarily provide an anti-atherosclerotic effect. Recently, the prospective Framingham survey showed that the low plasma CETP activity group had a greater increased risk for cardiovascular disease than the high CETP activity group\(^8\))\(^9\); therefore, it cannot be ruled out that the mechanism of CETP inhibition may raise the risk for cardiovascular events, despite increasing HDL-C levels\(^9\)\(^0\)\(^-\)\(^2\).

Probucol\(^9\)\(^3\), known as a lipid-lowering drug, is also a potent antioxidant and possesses antiatherogenic capability, despite decreasing the plasma HDL-C level. We first reported that probucol treatment resulted in the regression of Achilles tendon xanthoma in patients with familial hypercholesterolemia (FH)\(^9\)\(^4\). We also demonstrated a positive correlation between the decrease in the plasma HDL-C level and the reduction rate of Achilles tendon xanthoma in patients with heterozygous FH. Probucol treatment reduced HDL particle size, inducing the promotion of cholesterol efflux from cells\(^9\)\(^5\), and enhanced hepatic SR-BI expression in rabbits and a human hepatoma cell line\(^9\)\(^6\), and

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**Fig. 6. Effects of statins on reverse cholesterol transport.**

ApoA-I, a main component protein of HDL, is produced mainly in the liver. ApoA-I receives cholestrols and phospholipids through ABCA1 on the surfaces of hepatocytes and macrophages to form pre-\(\beta\)-HDL. Adiponectin is secreted from adipocytes and promotes HDL production by increasing ApoA-I production and ABCA1 expression. Cholesterol present on pre-\(\beta\)-HDL is esterified by LCAT to form a spherical HDL particle (HDL3). HDL3 pumps cholesterols via ABCG1 and SR-BI on macrophages, resulting in a large particle (HDL2). HDL2 is returned to HDL3, undergoing hydrolysis of TG and phospholipids by HL and EL. A spherical HDL particle occasionally supplies ApoA-I through catabolism by LPL. Cholesterol ester in HDL is transported to VLDL and LDL by CETP, and VLDL and LDL are decomposed by LPL and HL to finally be transported to the liver via LDL receptors. HDL2 is also taken up by the liver directly via liver-type SR-BI. Red arrows represent factors increased by statins and blue arrows factors reduced by statins.

Abbreviations: ABCA1, ATP-binding cassette transporter A1; ABCG1, ATP-binding cassette transporter G1; ApoA-I, apolipoprotein A-I; CETP, cholesteryl ester transfer protein; EL, endothelial lipase; HL, hepatic lipase; LPL, lipoprotein lipase; SR-BI, scavenger receptor class B type I; TG, triglycerides.
increased CETP activity\textsuperscript{95}, both leading to the acceleration of reverse cholesterol transport. To further assess the clinical efficacy of probucol treatment in clinical settings, the POSITIVE study (Probucol observational study illuminating therapeutic impact on vascular events) was performed, assessing whether long-term probucol treatment was associated with a lowered risk of cardiovascular events in a very high-risk population of patients with FH in Japan\textsuperscript{97}. The study cohort included 410 patients with heterozygous FH. Probucol significantly lowered the event risk in the secondary prevention group, although not significant in primary prevention. These results suggest that long-term probucol treatment may prevent secondary cardiovascular events in a higher cardiovascular risk population of heterozygous FH, despite the reduction of HDL-C levels.

HDL particles exert various effects depending on their properties\textsuperscript{6, 7, 98-100}. Ordinary HDL is “functional HDL” which exerts an anti-atherosclerotic effect through the inhibition of phospholipid oxidation in LDL\textsuperscript{7} and down-regulates the expression of inflammatory cytokines and vascular adhesion molecules\textsuperscript{101} in addition to enhancing RCT\textsuperscript{4}; however, various factors change the properties of HDL particles. When the content of inflammatory serum amyloid A (SAA) and ApoJ increases in HDL particles, while ApoA-I and ApoA-II regulating RCT and paraoxonase with anti-oxidative action decrease, HDL particles are modified from “functional HDL” to “dysfunctional HDL” (Fig. 7). The factors involved in this transformation are speculated to be various complications, including infections\textsuperscript{102}, heart disease\textsuperscript{103}, diabetes mellitus\textsuperscript{104}, metabolic syndrome\textsuperscript{105} and chronic renal disease\textsuperscript{106}. Unlike CETP inhibitors, if the HDL elevating effect of statins prevents CHD occurrence, this suggests that statins affect HDL to function in an anti-atherosclerotic manner.

**Effect of Statins on HDL Functions**

1) **RCT**

The ability of HDL to efflux cholesterol from foam cells and transport CE back to the liver is the main function of HDL in the RCT system. Excessive expression of SR-BI in the liver suppresses atherosclerosis as well as reducing HDL-C levels\textsuperscript{107}, whereas
Atherosclerosis is enhanced in SR-BI knock-out mice, suggesting that inhibition of SR-BI in the liver results in RCT inactivation despite a marked increase in HDL-C levels. Debate about RCT functionality has focused mainly on CETP. HDL in patients with CETP deficiency has a decreased ability to extract CE from macrophages; however, it was reported that a system containing whole serum from CETP-deficient patients for incubation showed an increased ability to extract CE. Another study showed that macrophages with excessive ABCG1 expression had an increased ability to efflux cholesterol, even in HDL of CETP-deficient patients. Patients with low CETP activity have an increased incidence of cardiovascular events, while another study found that the higher the serum CETP levels in patients, the more frequently they experienced calcification of coronary arteries and IMT thickening of the carotid arteries; therefore, the relationship between CETP and RCT functionality is still controversial.

Because statins generally inhibit CETP activity, the ability to produce pre-β HDL by ApoA-I and ABCA1 is important for RCT activation. Clinical results demonstrated that atorvastatin did not change the production rate (PR) or fractional catabolic rate (FCR) of ApoA-I. Administration of atorvastatin to mice did not significantly change ApoA-I expression in the liver and reduced ABCA1 and CETP expressions in the liver, indicating that the HDL-elevating effect of atorvastatin may be mediated mainly by CETP inhibition. Administration of rosvastatin did not change ApoA-I, but significantly reduced the PR of ApoA-I and the FCR of ApoA-I and HDL particles that contain ApoA-I. This result suggested that rosvastatin also raises HDL-C levels via CETP inhibition. Rosuvastatin reportedly did not change pre-β HDL concentrations and reduced the activity and mass of CETP by decreasing the ability to efflux cholesterol from macrophages to plasma. On the other hand, pravastatin elevated ApoA-I concentrations by increasing FCR and PR, although it inhibited CETP. The ApoA-I synthesis rate and FCR by pitavastatin have not been clinically investigated; however, pitavastatin even at a low concentration (equivalent to a clinical blood concentration) promoted ApoA-I secretion and ABCA1 expression in HepG2 cells, and also clinically promoted maturation from pre-β HDL to HDL. Therefore, pitavastatin may be expected to exhibit an anti-atherosclerotic effect via RCT activation.

2) Anti-Inflammatory Action

As to the anti-inflammatory function of HDL, simvastatin treatment slowly improved the anti-inflammatory ability of HDL, as assessed by cell-free assay (CFA) in patients with CHD. Administration of atorvastatin to patients with rheumatoid arthritis significantly reduced the anti-inflammatory ability of HDL. Additionally, atorvastatin was reported to have a more potent blood SAA-lowering effect than simvastatin.
3) Anti-Oxidative Action

Statins may improve the anti-oxidative action of HDL via paraoxonase 1 (PON1). PON1 is an enzyme hydrolyzing peroxidized lipids, which is synthesized in the liver and is present in blood bound to ApoA-I of HDL particles. PON1 exerts its anti-oxidative action by inhibiting the oxidation of LDL and even HDL itself, depending on the degree of its activity. Pitavastatin was shown to increase the promoter activity and protein expression of PON1 in vitro. As for the mechanism, pitavastatin was revealed to promote PON1 expression via p44/42 MAP kinase-mediated phosphorylation of SREBP-2 and binding of Sp1 to PON1 DNA. The increased PON1 activity was clinically confirmed with the administration of fluvastatin, simvastatin, atorvastatin, and rosuvastatin. In a comparative study of simvastatin and atorvastatin, atorvastatin was superior in terms of increasing PON1 activity. A study comparing atorvastatin with rosuvastatin showed no significant difference in changes in PON1 activity between the two, but rosuvastatin significantly increased PON1 activity after treatment.

4) Vascular Endothelial Function-Improving Action

Many sphingosine 1-phosphates (S1Ps) that activate endothelial nitric oxide synthase (eNOS) in vascular endothelial cells are also present in HDLS. S1P is thought to play an important role in the reactions of HDL to endothelial functions because S1P activates eNOS in vascular endothelial cells via S1P1 receptors. Pitavastatin reportedly increased S1P1 receptor expression in cultured bovine vascular endothelial cells and also promoted eNOS phosphorylation induced by HDL. Thus, statins may enhance vascular endothelial function through the activation of eNOS by S1Ps bound to HDLs.

Table 2 summarizes the effects of statins on HDL functions, including RCT, anti-inflammatory, anti-oxidative, and improving actions of vascular endothelial functions.

Conclusions

The HDL-C-elevating effects of statins differ among statins, due to differences in statin effects on various factors involved in HDL metabolism. On the other hand, studies should focus on HDL functionality as well as HDL-C levels. There are both functional and dysfunctional HDL molecules and it is important to promote functional HDL activities, including RCT, anti-oxidative and anti-inflammatory actions. It is possible that CETP inhibition may inactivate RCT and increase dysfunctional HDL. Whether HDL pro-
duced by statins is functional is determined by a balance between the ability of HDL neogenesis and CE transport activity into the liver. Although statins generally inhibit CETP, some statins that have a high ability to produce ApoA-I and ABCA1 in the liver are expected to exhibit RCT-mediated anti-atherosclerotic actions. Furthermore, statins may improve anti-inflammatory and anti-oxidative actions of HDL. Elevation of HDL-C by statin treatment may simultaneously improve HDL functionality, resulting in the prevention of coronary artery plaque progression and the occurrence of CHD. In the future, the effects of each statin treatment on HDL functions should be investigated in more detail to allow statin selection based on differences in their effects in clinical practice.

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References

1) Castelli WP: Cholesterol and lipids in the risk of coronary artery disease—the Framingham Heart Study. Can J Cardiol, 1988; 4 Suppl A: 5A-10A
2) Okamura T, Hayakawa T, Kadowaki T, Kita Y, Okayama A, Ueshima H: The inverse relationship between serum high-density lipoprotein cholesterol level and all-cause mortality in a 9.6-year follow-up study in the Japanese general population. Atherosclerosis, 2006; 184: 143-150
41) Argmann CA, Edwards JY, Sawyez CG, O’Neil CH,


51) de Vries R, Kerstens MN, Sluiter WJ, Groen AK, van Tol A, Dullaart RP: Cellular cholesterol efflux to plasma from moderately hypercholesterolaemic type 1 diabetic patients is enhanced, and is unaffected by simvastatin treatment. Diabetologia, 2005; 48: 1105-1113


Matsuzawa Y: Genetic cholesteryl ester transfer protein deficiency is extremely frequent in the Omagari area of Japan. Marked hyperalphalipoproteinemia caused by CETP gene mutation is not associated with longevity. Arterioscler Thromb Vasc Biol, 1997; 17: 1053-1059


93) Yamashita S, Matsuzawa Y: Where are we with probucol; a new life for an old drug? Atherosclerosis, 2009; 207: 16-23


98) Meyers CD, Kashyap ML: Disorders in high-density metabolism with insulin resistance and chronic kidney disease. J Ren Nutr, 2007; 17: 4-8


104) Gowri MS, Van der Westhuizen DR, Bridges SR, Anderson JW: Decreased protection by HDL from poorly controlled type 2 diabetic subjects against LDL oxidation may be due to the abnormal composition of HDL. Arterioscler Thromb Vasc Biol, 1999; 19: 2226-2233

105) Watson KE, Hama S, Fonarow GC, Ansell BJ, Navab M, Fogelman AM: Metabolic syndrome patients have higher plasma lipid hydroperoxides and more pro-inflammatory HDL than dyslipidemic control subjects, even with comparable levels of HDL, hs-CRP and paraoxonase activity. Circulation, 2004; 110: III-52

106) Kaysen GA: Disorders in high-density metabolism with insulin resistance and chronic kidney disease. J Ren Nutr, 2007; 17: 4-8


113) Bilz S, Wagner S, Schmitz M, Bedynck A, Keller U,

Mackness MI, Arrol S, Durrington PN: Protection of low-density lipoprotein against oxidative modification by high-density lipoprotein associated paraoxonase. Atherosclerosis, 1993; 104: 129-135


Okajima F: Plasma lipoproteins behave as carriers of extracellular sphingosine 1-phosphate: is this an atherogenic mediator or an anti-atherogenic mediator? Biochim Biophys Acta, 2002; 1582: 132-137

Igarashi J, Miyoshi M, Hashimoto T, Kubota Y, Kosaka H: Statins induce S1P1 receptors and enhance endothelial nitric oxide production in response to high-density lipoproteins. Br J Pharmacol, 2007; 150: 470-479

Nakatani N: Efficacy of statins. Prog Med, 2004; 24: 9-14


143) Matsuda N, Akasaka I, Ohtawa M: Metabolic fate of fluvastatin, an inhibitor of HMG-CoA reductase (1); absorption, distribution and excretion of [14C] fluvas-tatin after single administration in rats. Xenobio Metabol And Dispos, 1995; 10: 513-528


156) Tomlinson B, Mak TW, Tsui JY, Woo J, Shek CC, Critchley JA, Masarei JR: Effects of fluvastatin on lipid profile and apolipoproteins in Chinese patients with hypercholesterolemia. Am J Cardiol, 1995; 76: 36A-139A