Twelve weeks of smoking cessation therapy with varenicline increases the serum levels of apolipoprotein A-I only in the success group

Masahiko Iwaoka (MD, PhD)a,∗, Hiromasa Shimamura (MD)a, Takeshi Tsuji (MD)a, Kiyotaka Kugiyama (MD, PhD, FJCC)b

aDivision of Cardiology, Tokyo Kita Social Insurance Hospital, Tokyo, Japan
bDepartment of Internal Medicine II, Interdisciplinary Graduate School of Medicine and Engineering, University of Yamanashi, Yamanashi, Japan

Keywords:
Apolipoproteins
Lipoproteins
Smoking

Abstract

Background: Cigarette smoking adversely affects lipid profiles, and smoking cessation should improve lipid profiles in the long term. However, it remains unclear whether intensive, medication-based smoking cessation therapy can affect lipid profiles in the short term. Thus, we evaluated the short-term effects of smoking cessation therapy with varenicline on lipid profiles.

Methods: Participants included 86 consecutive subjects who received 12 weeks of smoking cessation therapy. All subjects were treated with varenicline, and no changes were made to their current lipotropic and antidiabetic medications during treatment. At first and last visits, lipid profiles and fasting blood glucose and hemoglobin A1c levels were evaluated and physical examination was performed. The success group, comprising subjects who attained exhaled carbon monoxide-confirmed 4-week continuous abstinence, included 69 subjects, whereas the failure group, comprising those who did not achieve complete smoking cessation, included 17 subjects. The number of cigarettes consumed per day was reduced in all subjects in the failure group.

Results: Serum apolipoprotein A-I (apoA-I) and high-density lipoprotein cholesterol (HDL-C) levels significantly increased from baseline to 12 weeks in the success group (apoA-I: 151.7 ± 28.0 vs. 158.6 ± 27.3 mg/dL, respectively, p < 0.01; HDL-C: 54.6 ± 15.7 vs. 57.9 ± 14.3 mg/dL, respectively, p < 0.01); however, there were no statistically significant differences observed in the failure group (apoA-I, 145.9 ± 33.4 vs. 146.8 ± 34.2 mg/dL, respectively, p = 0.87; HDL-C, 52.6 ± 15.7 vs. 53.3 ± 16.3 mg/dL, respectively, p = 0.80). The effect sizes (Cohen’s d) of apoA-I and HDL-C in the success group were 0.42 and 0.46, respectively. The post hoc statistical power values of apoA-I and HDL-C in the success group were 0.94 and 0.96, respectively.

Conclusion: These findings suggest that successful smoking cessation therapy with varenicline improves serum apoA-I and HDL-C levels in the short term.

© 2014 Japanese College of Cardiology. Published by Elsevier Ltd. All rights reserved.

Introduction

High-density lipoprotein (HDL) has diverse antiatherogenic functions that include not only cholesterol efflux and reverse cholesterol transport, but also antioxidative, anti-inflammatory, and antithrombotic activities [1–4]. There is extensive evidence that high serum HDL cholesterol (HDL-C) levels, particularly high levels of apolipoprotein A-I (apoA-I), which is a major protein in HDL, are associated with a reduced risk of coronary heart disease (CHD) [5,6].

Cigarette smoking is the most important preventable cause of cardiovascular disease [7]; it causes inflammation and oxidative stress and leads to vasomotor dysfunction and altered blood coagulation [8]. Moreover, a meta-analysis by Craig et al. previously showed that smokers had significantly lower serum HDL-C and apoA-I levels than non-smokers and that this association was dose dependent [9]. It has also been reported that smoking cessation with the help of a counseling program increased the levels of apoA-I and HDL-C [10,11]. However, it remains unclear whether intensive smoking cessation therapy can affect lipid profiles in the short term. It also remains unknown whether for subjects who do not achieve complete cessation, simply reducing the number of cigarettes smoked can improve lipid profiles in the short term.
We believe it is important to examine whether in comparison with simply reducing the number of cigarettes smoked per day, complete smoking cessation can contribute to a greater short-term improvement in lipid profiles. Therefore, we evaluated the short-term effects of smoking cessation therapy with varenicline on lipid profiles, comparing the results between subjects who succeeded in complete smoking cessation and those who simply reduced the number of cigarettes smoked per day.

Methods

Subjects

One-hundred and eighty-seven subjects presented to Tokyo Kita Social Insurance Hospital and expressed a desire to quit smoking from August 2010 to April 2013. Of these, 133 actually completed 12 weeks of smoking cessation therapy with varenicline. All subjects fulfilled the following criteria: (1) 20 years of age or older, (2) Brinkman index (number of cigarettes per day times smoking years) score of $\geq 200$, (3) Tobacco Dependence Screener score of $\geq 5$ [12], and (4) stated motivation to immediately quit smoking. These criteria were established by the Japanese medical insurance system for nicotine-dependent outpatients. Exclusion criteria included undergoing an operation, receiving chemotherapy, receiving hemodialysis, contracting an infectious disease, and experiencing an injury at any time from 3 months before the first visit to completion of smoking cessation therapy. Subjects whose lipotrophic or antidiabetic medications were changed within the same 3-month period before or during the study were also excluded to preclude the influence of these changes on lipid profiles and glycometabolism during the therapy. Finally, 86 consecutive subjects who received 12 weeks of smoking cessation therapy with varenicline and fulfilled the aforementioned criteria were included in this study. Patient selection is summarized in Fig. 1. Written informed consent was obtained from all subjects, and this study was approved by the ethics committee at Tokyo Kita Social Insurance Hospital.

Smoking cessation therapy with varenicline

All subjects were prescribed varenicline titrated up to 1.0 mg twice daily (0.5 mg once daily for 3 days, then 0.5 mg twice daily for 4 days, then 1.0 mg twice daily for 11 weeks). The target smoking cessation date was planned to be 7 days after varenicline initiation. After the first visit on day 1, follow-up visits were scheduled on days 15, 29, 57, and 85. Self-reported smoking status and exhaled carbon monoxide concentrations were assessed at each visit. The criterion for inclusion in the success group was exhaled carbon monoxide–confirmed 4-week continuous abstinence during weeks 9–12. This was defined as the proportion of subjects who reported

---

Fig. 1. Flow chart for the selection of study participants.
no smoking during this period; this was confirmed by an exhaled carbon monoxide measurement of ≤10 parts per million at the final visit [13].

**Blood sampling**

Venous blood samples were obtained after overnight fasting from all subjects at the first and last visits. All serum samples were analyzed for total cholesterol (TC), triglycerides (TG), HDL-C, C-reactive protein (CRP), fasting glucose, hemoglobin A1c (HbA1c), and apoA-I. TC, TG, HDL-C, and CRP levels were measured within 1 h after sampling. Further, serum samples collected in a tube containing sodium fluoride were analyzed for fasting glucose and HbA1c levels within 1 h after sampling. HbA1c values were determined using National Glycohemoglobin Standardization Program. Levels of apoA-I were measured using samples refrigerated at 4°C and analyzed within 2 days after sampling with a turbidimetric immunoassay kit at a commercially available laboratory (Mitsubishi Chemical Medience Co., Tokyo, Japan). Serum low-density lipoprotein cholesterol (LDL-C) levels were calculated using the Friedewald formula: LDL-C = TC − HDL-C − TG/5 [14].

**Statistical analysis**

Mean values in the success and failure groups were compared using unpaired t-test, whereas those for the first (baseline) and last visits in both groups were compared using paired t-test. The frequencies between the groups were compared using chi-square test or Fisher’s exact test. CRP levels were analyzed using a log transformation, because the distributions of CRP levels were not Gaussian. The correlation between apoA-I and loge-CRP levels from the first to the last visit was analyzed using correlation analysis. All tests were two-tailed, and p < 0.05 was used to indicate statistical significance. Analyses were performed using PASW Statistics software version 18.0 (SPSS Inc., Chicago, IL, USA). The G* power 3 program (Erdfelder, Faul, & Buchner, Mannheim, Germany) was used to calculate effect sizes and perform post hoc power analyses [15].

**Results**

Of the 86 study subjects, 69 met the criteria for the success group. Their baseline clinical characteristics are shown in Table 1. There were no significant differences between the success and failure groups with regard to clinical characteristics including age, gender, body weight, body mass index, smoking status, lipid profile, fasting glucose, HbA1c, loge-CRP, and use of statins and antidiabetic agents. As shown in Table 2, all the subjects in the failure group reported a reduction in the number of cigarettes smoked per day from the first to the last visit. Two subjects in the failure group reported no smoking during the final 4 weeks of therapy; however, their exhaled carbon monoxide levels were >10 parts per million at the last visit.

As shown in Table 3, despite a significant mean gain in body weight over 12 weeks (62.1 ± 11.4 vs. 63.4 ± 11.2 kg, respectively, p < 0.01), serum apoA-I and HDL-C levels significantly increased after smoking cessation therapy in the success group (apoA-I: 151.7 ± 28.0 vs. 158.6 ± 27.3 mg/dL, respectively, p = 0.01; HDL-C: 54.6 ± 15.7 vs. 57.9 ± 14.3 mg/dL, respectively, p < 0.01). loge-CRP levels also significantly decreased after smoking cessation therapy in the success group (−2.91 ± 1.22 vs. −3.17 ± 0.97 mg/dL, respectively, p = 0.04). There were no significant differences in the levels of LDL-C, fasting glucose, and HbA1c in the success group. The effect sizes (Cohen’s d) of apoA-I, HDL-C, and body weight were greater than those of LDL-C, fasting glucose, HbA1c, and loge-CRP, and the post hoc statistical power of apoA-I, HDL-C, and body weight were >0.9 (Table 3).

In the failure group, there were no significant differences in body weight or in the levels of HDL-C, LDL-C, apoA-I, fasting glucose, and HbA1c between baseline and last visit (Table 4). Moreover, loge-CRP levels actually significantly increased after smoking cessation therapy in the failure group (−3.01 ± 1.73 vs. −2.06 ± 1.52 mg/dL, respectively, p = 0.04). The effect sizes of apoA-I and HDL-C in the failure group were <0.1 (Table 4).

Serum levels of apoA-I of all study subjects had a significant inverse correlation with loge-CRP levels on both the first and the last visits (Fig. 2).

**Discussion**

Significant body weight gain is usually associated with a decrease in serum HDL-C and apoA-I levels, which are
Table 3
Change of variables and post hoc power analysis in the success group.

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Last visit</th>
<th>p value</th>
<th>Correlation coefficient between baseline and last visit</th>
<th>Cohen’ s d</th>
<th>Post hoc power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (kg)</td>
<td>62.1 ± 11.4</td>
<td>63.9 ± 11.2</td>
<td>&lt;0.01</td>
<td>0.99</td>
<td>0.81</td>
<td>0.99999998</td>
</tr>
<tr>
<td>Triglycerides (mg/dL)</td>
<td>134.6 ± 107.9</td>
<td>128.3 ± 97.1</td>
<td>0.40</td>
<td>0.82</td>
<td>0.10</td>
<td>0.13</td>
</tr>
<tr>
<td>HDL-C (mg/dL)</td>
<td>54.6 ± 15.7</td>
<td>57.9 ± 14.3</td>
<td>&lt;0.01</td>
<td>0.89</td>
<td>0.46</td>
<td>0.96</td>
</tr>
<tr>
<td>LDL-C (mg/dL)</td>
<td>112.2 ± 31.9</td>
<td>117.9 ± 32.0</td>
<td>0.05</td>
<td>0.73</td>
<td>0.24</td>
<td>0.51</td>
</tr>
<tr>
<td>ApoA-I (mg/dL)</td>
<td>110.5 ± 35.2</td>
<td>114.4 ± 41.3</td>
<td>0.13</td>
<td>0.86</td>
<td>0.19</td>
<td>0.34</td>
</tr>
<tr>
<td>Hemoglobin A1c (%)</td>
<td>6.3 ± 1.1</td>
<td>6.3 ± 1.2</td>
<td>0.44</td>
<td>0.93</td>
<td>0.00</td>
<td>0.05</td>
</tr>
<tr>
<td>loge-CRP (mg/dL)</td>
<td>−2.91 ± 1.22</td>
<td>−3.17 ± 0.97</td>
<td>0.04</td>
<td>0.60</td>
<td>0.26</td>
<td>0.56</td>
</tr>
</tbody>
</table>

HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA-I, apolipoprotein A-I; CRP, C-reactive protein.

Table 4
Change of variables and post hoc power analysis in the failure group.

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Last visit</th>
<th>p value</th>
<th>Correlation coefficient between baseline and last visit</th>
<th>Cohen’ s d</th>
<th>Post hoc power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (kg)</td>
<td>61.3 ± 13.0</td>
<td>62.3 ± 12.1</td>
<td>0.05</td>
<td>0.99</td>
<td>0.50</td>
<td>0.50</td>
</tr>
<tr>
<td>Triglycerides (mg/dL)</td>
<td>117.5 ± 60.6</td>
<td>127.4 ± 58.0</td>
<td>0.51</td>
<td>0.49</td>
<td>0.16</td>
<td>0.10</td>
</tr>
<tr>
<td>HDL-C (mg/dL)</td>
<td>52.6 ± 15.7</td>
<td>53.3 ± 16.3</td>
<td>0.80</td>
<td>0.74</td>
<td>0.06</td>
<td>0.06</td>
</tr>
<tr>
<td>LDL-C (mg/dL)</td>
<td>120.7 ± 43.1</td>
<td>115.6 ± 49.1</td>
<td>0.49</td>
<td>0.80</td>
<td>0.17</td>
<td>0.10</td>
</tr>
<tr>
<td>ApoA-I (mg/dL)</td>
<td>145.9 ± 33.4</td>
<td>146.8 ± 34.2</td>
<td>0.87</td>
<td>0.78</td>
<td>0.04</td>
<td>0.05</td>
</tr>
<tr>
<td>Fasting glucose (mg/dL)</td>
<td>104.8 ± 20.0</td>
<td>104.4 ± 26.8</td>
<td>0.94</td>
<td>0.65</td>
<td>0.02</td>
<td>0.05</td>
</tr>
<tr>
<td>Hemoglobin A1c (%)</td>
<td>6.1 ± 0.6</td>
<td>6.0 ± 0.6</td>
<td>0.34</td>
<td>0.93</td>
<td>0.45</td>
<td>0.40</td>
</tr>
<tr>
<td>loge-CRP (mg/dL)</td>
<td>−3.01 ± 1.73</td>
<td>−2.06 ± 1.52</td>
<td>0.04</td>
<td>0.46</td>
<td>0.56</td>
<td>0.58</td>
</tr>
</tbody>
</table>

HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA-I, apolipoprotein A-I; CRP, C-reactive protein.

cardioprotective lipids [16,17]. The association between obesity and low-grade inflammation has also been established [18], and CRP is a well-known marker of inflammation. However, in this study, despite the fact that subjects gained a significant amount of body weight during the smoking cessation process, they nonetheless experienced a significant increase in apoA-I and HDL-C levels and a significant decrease in loge-CRP levels. This finding suggests that the positive antiatherogenic effects of smoking cessation far outweigh the negative effects of any weight gain that may occur during the process.

Further, the present study demonstrated a significant inverse correlation between apoA-I and CRP levels, and our findings are compatible with those of previous reports showing the anti-inflammatory properties of apoA-I [1–4]. One possible explanation for this phenomenon is that resolving the inflammation induced by cigarette smoking may have contributed to the increase in apoA-I levels and reduction in CRP levels, as observed in the success group. However, the precise mechanisms by which smoking cessation leads to increased apoA-I levels are unclear; further, there may be other mechanisms at play besides those involved in resolving the inflammation induced by smoking. This seems particularly likely given that in comparison with the medium effect size of apoA-I levels in the success group, the effect size of loge-CRP levels was small. Previous studies have reported decreased adiponectin levels in current smokers that can be reversed upon smoking cessation [19], and Oku et al. found that adiponectin deficiency suppressed apoA-I synthesis in the liver in animal experiments [20]. These findings suggest that an increase in adiponectin levels also contributes to an increase in apoA-I levels after smoking cessation.

Another notable finding of our study was that reducing the number of cigarettes smoked per day did not lead to any significant short-term improvements in lipid profiles or loge-CRP levels. Further, negligible effect sizes of apoA-I and HDL-C levels were observed in the failure group, despite the meaningful effect sizes of body weight and loge-CRP levels in this group and meaningful effect size of apoA-I levels in the success group. The number of subjects in the failure group was small; thus, it is possible that apoA-I levels could have statistically increased given a larger sample size. However, even with a larger sample size, the increase in apoA-I levels may not have been substantially meaningful, considering the negligible effect size of apoA-I levels in the failure group.

Fig. 2. Correlation between serum apolipoprotein A-I (apoA-I) and loge-CRP (CRP) levels.
It is known that apoA-I plays an essential role in the cardioprotective action of HDL [21] and that apoA-I levels provide a good estimate of HDL concentration [22]. Low apoA-I levels have been shown to be more accurate than low HDL-C levels as a risk marker for atherosclerosis and cardiovascular events [23,24]. HDL-C levels are influenced not only by the number of HDL particles but also by other factors including cholesterol transfer protein activity [1,21]. This may account for the superior predictive value of apoA-I than HDL-C levels. Furthermore, HDL-C levels are still not considered as a primary target of therapy in the National Cholesterol Education Program guidelines and European Society of Cardiology/European Atherosclerosis Society guidelines, because an increase in circulating HDL-C levels does not necessarily decrease the risk of CHD events, CHD deaths, or mortality [25,26]. In contrast, if major interest is focused on apoA-I mimetic peptides, which not only are active in cellular cholesterol efflux but also exert antioxidative, anti-inflammatory, and anti-thrombotic effects [27].

Several clinical trials of smoking cessation have revealed substantial reductions in the risk for cardiovascular events in those who quit smoking [28–30]. Observational data suggest that smoking cessation reduces the risk for cardiovascular events and that the risk decline in risk begins within months after quitting [29]. To the best of our knowledge, the present study represents the first report demonstrating that successful smoking cessation is significantly associated with an increase in serum apoA-I levels in the short term—just 12 weeks after initiating treatment and as few as 4 weeks after achieving complete abstinence from smoking. We previously reported that low apoA-I levels predict adverse outcomes in patients with nonischemic heart failure [31]. Therefore, a rapid increase in apoA-I levels after smoking cessation is a promising finding that may contribute to positive outcomes in those with cardiovascular disease.

There are several limitations to our study. First, this study was limited by the small number of subjects, particularly in the failure group. We did not have sufficient study subjects to stratify each group (i.e. by duration of smoking cessation in the success group and by the number of cigarettes smoked per day during smoking cessation therapy in the failure group). Thus, we could not perform multivariate analysis on the effect of smoking cessation therapy on apoA-I levels. Second, the criteria for successful smoking cessation were mainly dependent upon subjects’ self-reports of their behavior; therefore, there is a possibility that subjects who did not exactly fulfill the success criteria were treated as part of the success group. Third, in an outpatient clinic, we could not assess the role of environmental confounders, such as nutrition intake and physical activity, on lipid profiles. Further studies involving a larger sample size and considering the effect of potential confounders should help in better elucidating the precise effects of smoking cessation on apoA-I levels.

**Conclusion**

Our findings suggest that successful smoking cessation therapy with varenicline improves serum apoA-I and HDL-C levels in the short term. Simply reducing the number of cigarettes smoked per day appears to be insufficient to achieve an improvement in lipid profiles and a reduction in the risk for cardiovascular events.

**References**