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Short communication

Administration of natural astaxanthin increases serum HDL-cholesterol and adiponectin in subjects with mild hyperlipidemia

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ABSTRACT

Background: Astaxanthin has been reported to improve dyslipidemia and metabolic syndrome in animals, but such effects in humans are not well known.
Methods: Placebo-controlled astaxanthin administration at doses of 0, 6, 12, 18 mg/day for 12 weeks was randomly allocated to 61 non-obese subjects with fasting serum triglyceride of 120–200 mg/dl and without diabetes and hypertension, aged 25–60 years.
Results: In before and after tests, body mass index (BMI) and LDL-cholesterol were unaffected at all doses, however, triglyceride decreased, while HDL-cholesterol increased significantly. Multiple comparison tests showed that 12 and 18 mg/day doses significantly reduced triglyceride, and 6 and 12 mg doses significantly increased HDL-cholesterol. Serum adiponectin was increased by astaxanthin (12 and 18 mg/day), and changes of adiponectin correlated positively with HDL-cholesterol changes independent of age and BMI.
Conclusions: This first-ever randomized, placebo-controlled human study suggests that astaxanthin consumption ameliorates triglyceride and HDL-cholesterol in correlation with increased adiponectin in humans.

1. Introduction

Co-existence of hypertension, impaired glucose tolerance, and dyslipidemia has been termed metabolic syndrome which increases the risk of developing type 2 diabetes and cardiovascular diseases. Hypertriglyceridemia and low levels of high-density lipoprotein (HDL)-cholesterol are characteristics of metabolic syndrome [1,2]. The dysregulation of adipocytokines, secreted from adipose tissue, has been reported to affect insulin sensitivity involved in glucose and lipid metabolism [3]. Among these adipocytokines, adiponectin has been established to be one of key molecules for the underlying mechanisms of type 2 diabetes and metabolic syndrome [3,4]. Adiponectin has been reported to correlate positively with HDL-cholesterol, and inversely with triglyceride (TG) and very low density lipoprotein (VLDL)-cholesterol in non-diabetic subjects or in patients with type 2 diabetes [4,5]. Such associations persist when adjusted for potential confounders or obesity-related variables [6].

Astaxanthin is a xanthophyll carotenoid pigment found in marine animals, and in addition to inhibition of lipid peroxidation and low density lipoprotein (LDL) oxidation, astaxanthin has been reported to decrease serum TG and increase HDL-cholesterol and adiponectin in insulin resistant rats and in obese mice fed a high fat diet [7–10]. However, in a previous study with human subjects, intake of astaxanthin did not show an increase in HDL-cholesterol and a decrease in TG, but that study did not have a randomized design [8]. In addition, subjects in the previous study had normal levels of serum lipids at baseline and were administered astaxanthin for just 14 days. Moreover, the effect of astaxanthin consumption on serum adiponectin levels in humans is not well known.

We therefore performed a randomized, double-blind, placebo-controlled study in moderately hypertriglyceridemic subjects with 12-week astaxanthin administration to investigate whether astax-

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Table 1
Background of each group and parameters at week 0 and week 12.

	Group	*n*	Week 0	Week 12
Gender[a] (male/female)	18 mg/day	16	11/5	
	12 mg/day	15	10/5	
	6 mg/day	15	10/5	
	0 mg/day	15	10/5	
Age[b] (year)	18 mg/day	16	43.8 ± 10.4	
	12 mg/day	15	42.8 ± 8.8	
	6 mg/day	15	47.0 ± 7.0	
	0 mg/day	15	44.3 ± 7.0	
Body weight[b] (kg)	18 mg/day	16	67.3 ± 5.6	68.1 ± 12.4
	12 mg/day	15	64.2 ± 7.2	63.9 ± 7.0
	6 mg/day	15	64.6 ± 12.2	64.8 ± 11.7
	0 mg/day	15	69.4 ± 7.9	69.9 ± 8.0
BMI[b]	18 mg/day	16	23.9 ± 7.0	24.2 ± 3.3
	12 mg/day	15	23.0 ± 2.2	22.9 ± 2.1
	6 mg/day	15	23.6 ± 3.2	23.7 ± 3.0
	0 mg/day	15	25.1 ± 2.8	25.3 ± 2.7
Blood pressure (mmHg)				
Systolic	18 mg/day	16	126.3 ± 15.1	121.7 ± 17.3
	12 mg/day	15	129.9 ± 18.7	122.8 ± 14.6
	6 mg/day	15	118.1 ± 20.8	120.9 ± 9.7
	0 mg/day	15	127.9 ± 8.7	128.5 ± 13.9
Diastolic	18 mg/day	16	77.6 ± 11.9	76.7 ± 11.6
	12 mg/day	15	80.6 ± 10.1	77.5 ± 6.6
	6 mg/day	15	75.2 ± 11.3	75.0 ± 5.5
	0 mg/day	15	81.3 ± 7.1	81.6 ± 10.2
Total cholesterol[b] (mg/dl)	18 mg/day	16	234 ± 29	233 ± 35
	12 mg/day	15	215 ± 22	225 ± 29
	6 mg/day	15	219 ± 29	228 ± 30
	0 mg/day	15	209 ± 31	212 ± 25
Triglyceride[b] (mg/dl)	18 mg/day	16	151 ± 26	112 ± 40[**]
	12 mg/day	15	147 ± 21	110 ± 44[*]
	6 mg/day	15	151 ± 23	125 ± 41[*]
	0 mg/day	15	145 ± 21	140 ± 40
LDL-cholesterol[b] (mg/dl)	18 mg/day	16	157 ± 25	159 ± 31
	12 mg/day	15	136 ± 27	144 ± 32
	6 mg/day	15	141 ± 26	150 ± 28
	0 mg/day	15	135 ± 32	138 ± 32
HDL-cholesterol[b] (mg/dl)	18 mg/day	16	51 ± 6	55 ± 8[*]
	12 mg/day	15	55 ± 8	63 ± 8[**]
	6 mg/day	15	51 ± 11	56 ± 11[**]
	0 mg/day	15	52 ± 10	53 ± 11
Plasma glucose (mg/dl)	18 mg/day	16	98 ± 10	98 ± 11
	12 mg/day	15	98 ± 5	97 ± 6
	6 mg/day	15	95 ± 7	96 ± 9
	0 mg/day	15	98 ± 12	99 ± 11
Percentage changes of trigyceride (%)	18 mg/day	16		−23.8 ± 31.3[#]
	12 mg/day	15		−25.2 ± 31.0[#]
	6 mg/day	15		−17.7 ± 22.9
	0 mg/day	15		−2.0 ± 31.8
Percentage changes of HDL-cholesterol (%)	18 mg/day	16		6.7 ± 9.2
	12 mg/day	15		15.4 ± 12.2[##]
	6 mg/day	15		10.6 ± 10.8[#]
	0 mg/day	15		1.8 ± 12.0

The data represent the mean ± SD.
[a] Fisher's exact test (two-side test).
[b] ANOVA(two-side test).
[#] $p < 0.05$. Compared with control assessed by Dunnet multiple comparison method.
[##] $p < 0.01$. Compared with control assessed by Dunnet multiple comparison method.
[*] $p < 0.05$. Difference compared to week 0 (paired *t*-test); Significant difference was not shown in each group.
[**] $p < 0.01$. Difference compared to week 0 (paired *t*-test); Significant difference was not shown in each group.

anthin consumption ameliorates dyslipidemia and is associated with an increase in serum adiponectin levels.

2. Methods

Sixty-one healthy subjects (41 men and 20 women) with serum TG levels of 120–200 mg/dl were enrolled at Shiba Palace Clinic, Tokyo. Excluded were subjects with TG levels under 120 mg/dl or over 200 mg/dl, aged <20 years or >65 years, body mass index (BMI) >28 kg/m^2, diabetes, hypertension, cardiovascular diseases, liver dysfunction, renal dysfunction, endocrine diseases, or, using agents affecting lipid metabolism and lipid oxidation. The study was approved by the Ethics Committee of Institute of General Health Development, Shiba Palace Clinic, and all subjects provided written consent.

The enrolled subjects, aged 44 ± 8 years, were asked to maintain their habitual diet and lifestyle throughout the study. Three-day dietary records were obtained before and at end of the astaxanthin administration. Nutrient intake status [dietary energy (kcal/day), carbohydrate (g/day), fat (g/day), protein (g/day), and fiber (g/day)] was calculated from a database related to the composition of Japanese foods by national registered dieticians. The study subjects were randomized into four study groups with a 12-week treatment of 6, 12, and 18 mg/day of astaxanthin (AstaREAL® Astaxanthin, Fuji Chemical Industry, Toyama, Japan) or placebo prepared by Fuji Chemical Industry. After 12-h overnight fasting, venous blood was collected for biochemical measurements, and all subjects underwent anthropometric and blood pressure measurements at baseline and end of the study.

Fasting plasma glucose and serum total cholesterol, TG, LDL-cholesterol, and HDL-cholesterol were determined by standard enzymatic methods and homogenous methods (Sekisui Medical Co., Ltd., Tokyo, Japan). Serum adiponectin was measured by the ELISA method (Otsuka Pharmaceutical Co., Ltd., Tokyo).

Values are given as mean ± standard deviation. The significance of differences in parameters between before and after astaxanthin administration was assessed by paired Student's *t*-test analysis. Significant differences in percentage changes of parameters between groups were compared by one-way ANOVA and tested further by Fisher PLSD multiple comparison method. Correlations between two variables were evaluated by Pearson's simple linear regression analysis. A multiple stepwise regression analysis was performed to evaluate the independent contribution of age, BMI, and percentage changes in TG and adiponectin to percentage changes in HDL-cholesterol. A value of $p < 0.05$ was considered statistically significant.

3. Results

Baseline characteristics, plasma glucose and serum lipid data did not differ between the four groups (Table 1). Daily intake amounts of dietary energy, protein, fat, carbohydrate, and fiber did not differ between the four groups at baseline and at the end (Online supplemental table). Consumption of astaxanthin was well accepted by the study subjects, none of which dropped out from the study.

BMI, blood pressures and plasma glucose did not change in any group (Table 1). The 12-week administration of astaxanthin significantly decreased serum TG and increased HDL-cholesterol, although total cholesterol and LDL-cholesterol were not changed (Table 1). The decrease in TG was significantly greater in the groups receiving 12 and 18 mg/day of astaxanthin than the control, and the increase in HDL-cholesterol was significantly greater in the 6 and 12 mg/day groups than in the control (Table 1). The 12-week astaxanthin administration at doses of 12 and 18 mg/day significantly increased serum adiponectin levels (Fig. 1A).

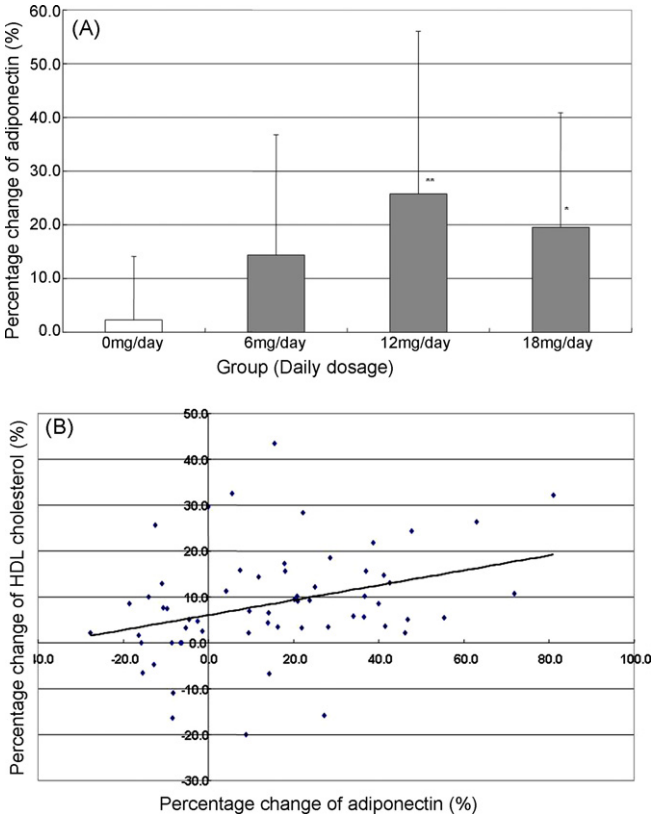

Fig. 1. (A) Percentage changes in adiponectin in response to astaxanthin administration $^*p < 0.05$; $^{**}p < 0.01$ as compared with control assessed by Fisher PLSD multiple comparison method. (B) Correlations between percentage changes in adiponectin and those in HDL-cholesterol $r = 0.331$, $p = 0.0089$; 95% confidence intervals: 0.086–0.538.

Fig. 1B shows the significant positive correlation between the percentage change in adiponectin and that in HDL-cholesterol ($r = 0.331$, $p = 0.0089$). However, there was less correlation between the percentage change in adiponectin and the percentage change in TG ($r = -0.227$, $p = 0.079$). The association of adiponectin changes with HDL-cholesterol changes was further assessed by a multiple stepwise regression analysis including age, BMI, and changes in TG as independent confounders. The percentage change in adiponectin was still positively associated with the percentage change in HDL-cholesterol independent of age and BMI ($b = 0.251$, $F = 4.49$) although TG changes also independently correlated with HDL-cholesterol changes ($b = -0.35$, $F = 8.73$).

4. Discussion

An astaxanthin-induced amelioration of TG and HDL-cholesterol has been observed in animal studies [9,10], but affirmative effects of astaxanthin in human studies are still unreported, because randomized clinical trials or well-defined clinical studies have not, to our knowledge, been performed. The present double-blind, placebo-controlled study could be the first to clearly demonstrate that the administration of astaxanthin at doses of 12 and 18 mg/day significantly decreased TG and increased HDL-cholesterol and adiponectin in humans.

The present study suggests that astaxanthin can improve the serum lipid profile in humans, including an increase in HDL-cholesterol, a robust negative risk factor for atherosclerotic cardiovascular disease through a variety of mechanisms, including reverse cholesterol transport from peripheral tissues to liver [11]. Torcetrapib, a cholesterol ester transfer protein (CETP) inhibitor,

has recently been reported to produce excess cardiovascular and non-cardiovascular mortality, probably because of off-target effects [12]. However, a variety of approaches may be expected to successfully treat low HDL and reduce the risk of atherosclerosis. The HDL-increasing effect of astaxanthin is of potentially great interest and deserves further investigation for elucidating the mechanisms of this observation, and for verifying astaxanthin's effect on cardiovascular disease prevention in larger-scaled studies.

An inverse relation of adiponectin with TG and VLDL-cholesterol, while a positive correlation of adiponectin with HDL-cholesterol has been observed [4,5]. Adiponectin reduces serum TG by increasing VLDL catabolism through enhanced lipoprotein lipase and VLDL receptor expression in skeletal muscles related, in part, to improved insulin resistance [7,9,13]. Along with improved TG metabolism, adiponectin could account for the increased HDL-cholesterol observed in the present study. Meanwhile, previous reports have shown the significant association between serum adiponectin and HDL-cholesterol is independent of obesity and insulin resistance, and that adiponectin may have a direct effect on HDL-catabolism [14]. Recent studies reported that adiponectin enhances apolipoprotein A-1-mediated cholesterol efflux from macrophages through ATP-binding cassette transporter A1-dependent pathway by activating liver X receptor alpha and peroxisome proliferators-activated receptor (PPAR)gamma, leading to the increased HDL-cholesterol [15]. Actually, the present study shows a markedly positive correlation between the percentage change in adiponectin and that seen in HDL-cholesterol level.

The mechanisms of astaxanthin-mediated adiponectin elevation are still poorly understood. Although serum adiponectin levels are low in obese subjects as reported previously [4], BMI values were not changed by astaxanthin administration in the present study. Adiponectin is regulated by PPARgamma pathway and is involved in close-inhibition relationships with inflammatory cytokines including tumor necrotic factor alpha [3,4]. The anti-inflammatory actions of astaxanthin might be assumed as one of the reasons for increasing adiponectin by astaxanthin consumption, but it remains to be confirmed.

In conclusion, the present randomized, placebo-controlled study shows, for probably the first time, that, in humans with serum TG levels of 120–200 mg/dl, astaxanthin administered at 12 and 18 mg/day for 12 weeks decreased TG levels, while HDL-cholesterol increased with 6 and 12 mg/day of astaxanthin, in concert with measured levels of adiponectin. Although the present study does not reveal the specific underlying mechanisms, astaxanthin may be expected to successfully treat impaired lipid metabolism and prevent atherosclerosis because of its possible therapeutic increase of HDL-cholesterol and adiponectin. However, well defined in vitro studies, long-term and large-scaled clinical trials will be needed to confirm such astaxanthin benefits.

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Conflict of interests

None declared.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.atherosclerosis.2009.10.012.

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