Biochemical and Biophysical Research Communications 441 (2013) 321–326

Contents lists available at ScienceDirect

Biochemical and Biophysical Research Communications

journal homepage: www.elsevier.com/locate/ybbrc

Arctigenin promotes cholesterol efflux from THP-1 macrophages through PPAR-γ/LXR-α signaling pathway

Xiaolin Xu [a], Qian Li [b], Liewen Pang [a], Guoqian Huang [a], Jiechun Huang [a], Meng Shi [a], Xiaotian Sun [a], Yiqing Wang [a,*]

[a] Department of Cardiothoracic Surgery, Huashan Hospital, Fudan University, Shanghai 200032, China
[b] Department of Integrative Medicine and Neurobiology, School of Basic Medical Sciences, Shanghai Medical College, Fudan University, Shanghai, China

ARTICLE INFO

Article history:
Received 9 October 2013
Available online 17 October 2013

Keywords:
Cholesterol efflux
ATP binding cassette transporter A1 (ABCA1)
ATP binding cassette transporter G1 (ABCG1)
Atherosclerosis

ABSTRACT

Cholesterol efflux from macrophages is a critical mechanism to prevent the development of atherosclerosis. Here, we sought to investigate the effects of arctigenin, a bioactive component of *Arctium lappa*, on the cholesterol efflux in oxidized low-density lipoprotein (oxLDL)-loaded THP-1 macrophages. Our data showed that arctigenin significantly accelerated apolipoprotein A-I- and high-density lipoprotein-induced cholesterol efflux in both dose- and time-dependent manners. Moreover, arctigenin treatment enhanced the expression of ATP binding cassette transporter A1 (ABCA1), ABCG1, and apoE, all of which are key molecules in the initial step of cholesterol efflux, at both mRNA and protein levels. Arctigenin also caused a concentration-dependent elevation in the expression of peroxisome proliferator-activated receptor-gamma (PPAR-γ) and liver X receptor-alpha (LXR-α). The arctigenin-mediated induction of ABCA1, ABCG1, and apoE was abolished by specific inhibition of PPAR-γ or LXR-α using small interfering RNA technology. Our results collectively indicate that arctigenin promotes cholesterol efflux in oxLDL-loaded THP-1 macrophages through upregulation of ABCA1, ABCG1 and apoE, which is dependent on the enhanced expression of PPAR-γ and LXR-α.

1. Introduction

Atherosclerosis is the primary factor causing cardiovascular events. It is well known that macrophages, as professional scavengers, play a critical role in atherogenesis through uptake of modified low-density lipoprotein (LDL) and secretion of inflammatory modulators, cytokines, and matrix-degrading enzymes [1]. The internalization of various forms of modified LDL that accumulated in the artery results in the development of macrophages foam cells, whose accumulation in the arterial wall is a hallmark of the early atherosclerotic lesion [2]. Clearance of arterial cholesterol deposits by macrophages is beneficial during the early stages of atherosclerosis [3].

Accumulating evidence suggests that reverse cholesterol transport (RCT) is a good antiatherogenic strategy by which accumulated cholesterol is transport from the vessel wall to the liver for clearance into the bile and ultimately the feces [3]. Cholesterol efflux from macrophages is the first and potentially most important step in macrophage RCT, by which intracellular cholesterol from macrophage is transferred to extracellular acceptors, such as apolipoprotein A-I (apoA-I) and high-density lipoprotein (HDL) [3]. It consists of five independent pathways, including ATP-binding membrane cassette transporter A1 (ABCA1), scavenger receptor B1 (SR-B1), caveolin, Cyp27A1 and passive diffusion [4].

Arctigenin, a bioactive component of *Arctium lappa*, exerts many biological activities, including antioxidant, antitumor and anti-inflammatory properties [5–7]. Recently, it is shown that arctigenin inhibits the production of nitric oxide (NO) and the release of tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6) in lipopolysaccharide (LPS)-stimulated macrophages RAW 264.7 and THP-1 cells [8]. The inhibitory effects of arctigenin on NO production seem to be due to down-regulation of inducible nitric oxide synthase (iNOS) protein expression and suppression of the iNOS enzymatic activity [8]. Excess NO has been shown to decrease the efficacy of liver X receptor α (LXR-α)-ABCA1-dependent cholesterol efflux, thereby promotes oxLDL-mediated cholesterol accumulation in foam cells, and leads to atherosclerosis progression [9]. In the present study, we sought to investigate the effects of arctigenin on cholesterol efflux in oxLDL-loaded THP-1 macrophages.

2. Material and methods

2.1. Cell culture and treatment

The human monocyte-macrophage cell line (THP-1) was brought from American Type Culture Collection (Manassas, VA,

* Corresponding author. Fax: +86 21 52887075.
E-mail address: yiqingwangbiopaper@163.com (Y. Wang).

0006-291X/$ - see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.bbrc.2013.10.050

USA). THP-1 cells were maintained in RPMI 1640 (Invitrogen Life Technologies, Grand Island, NY, USA) containing 10% fetal bovine serum (FBS; Invitrogen) in a humidified atmosphere of 5% CO_2 at 37 °C.

THP-1 cells (5×10^5 cells per 35 mm dish) were differentiated into macrophages by stimulation with 160 nmol/ml of phorbol 12-myristate 13-acetate (PMA) for 72 h and then exposed to 50 mg/L of oxLDL for 24 h. THP-1-derived macrophages were treated with different concentrations of arctigenin (0, 10, 50, and 100 μM) for 12 h or exposed to 100 μM of arctigenin for varying durations. *Arctigenin (catalog No. SMB00075) was purchased from Sigma (St. Louis, MO, USA), with a purity of ⩾95%.* For lipid uptake, cells were incubated in RPMI 1640 supplemented with 0.2 % fatty acid-free bovine serum albumin (BSA).

2.2. Cholesterol efflux assay

Cholesterol efflux experiments were performed as before [10]. THP-1 cells were stimulated with 160 nmol/ml of PMA for 72 h to differentiated into macrophages and exposed to 50 mg/L oxLDL and [^{3}H]cholesterol (1.0 μCi/mL) for another 24 h in RPMI 1640 supplemented with 0.2% BSA. Cholesterol-loaded macrophages were treated with different concentrations of arctigenin for 12 h or exposed to 100 μM of arctigenin for varying durations. The cells were washed with PBS and incubated in RPMI 1640 containing 0.2% BSA in the presence or absence of apoA-I (10 μg/mL), HDL_2 (50 μg/mL) or HDL_3 (50 μg/mL), for another 24 h. The percentage of cholesterol efflux was calculated via dividing media-derived radioactivity by the sum of the radioactivity in the media and cells. The fold changes of the percentage of cholesterol efflux were calculated by dividing the percentages of cholesterol efflux in the cells with different treatment by the percentage of cholesterol efflux in control cells.

2.3. Semiquantitative reverse transcription-polymerase chain reaction (RT-PCR)

Total RNA in the cells with indicated treatment was extracted by a Trizol Plus RNA Purification kit (Invitrogen). Complementary DNA (cDNA) was synthesized by an Omniscript RT kit (Qiagen, Valencia, CA) and used for PCR. Target gene specific primers (provided if requested) were used to amplify cDNA fragments by PCR with Taq DNA polymerase (Invitrogen). The PCR cycles are performed in a thermal cycler (Sigma–Aldrich, St. Louis, MO, USA). The RT-PCR products were subjected to electrophoresis in 1.5% agarose gel. Bands intensities were quantified by the Quantity One image software (Bio-Rad, Hercules, CA, USA) and normalized to the internal control β-actin.

2.4. Western blot analysis

Cells were harvested and protein extracts prepared according to Instruction Manual (Bio-Rad). Extracts were then subjected to Western blot analysis. Primary antibodies against SR-B1 (Santa Cruz Biotechnology, Santa Cruz, USA), SR-A1 (Santa Cruz Biotechnology), CD36 (Santa Cruz Biotechnology), ABCA1 (Cell Signaling, Beverly, MA, USA), ABCG1 (Cell Signaling), apoE (Cell Signaling), PPAR-γ (Cell Signaling), LXR-α (Cell Signaling), and β-actin (Sigma). The proteins were visualized using a chemiluminescence method (ECL Plus Western Blot Detection System, Amersham Biosciences, Foster City, CA, USA) and analyzed with a GS-700 Imaging Densitometer (Bio-Rad).

2.5. Transfection of siRNA

The human small interfering RNA (siRNA) against PPAR-γ (5′-GGAUGCAAGGGUUUCUUCCtt-3′ and 5′-GGAAGAAACCCUUGCAUCCtt-3′ for PPAR-gamma (psiRNA)) and LXR-α (5′-GGAGUGUGUCCUGUCAGAAtt-3′ and 5′-UUCAGACAGGACACACUCCtc-3′) and scrambled control RNA oligonucleotides were purchased from Ambion Inc. (Austin, TX, USA). The transfection of siRNA was performed using Lipofectamine 2000 according to the manufacturer's instructions (Invitrogen). Briefly, after stimulation with PMA for 72 h, different concentrations of siRNA (30, 60 nM) were added to THP-1 cells. The cells were incubated for 24 h, washed and then 50 mg/L of oxLDL were added for another 24 h in 0.2 % fatty acid-free BSA. 100 μM of arctigenin was used to treat the cells for 12 h.

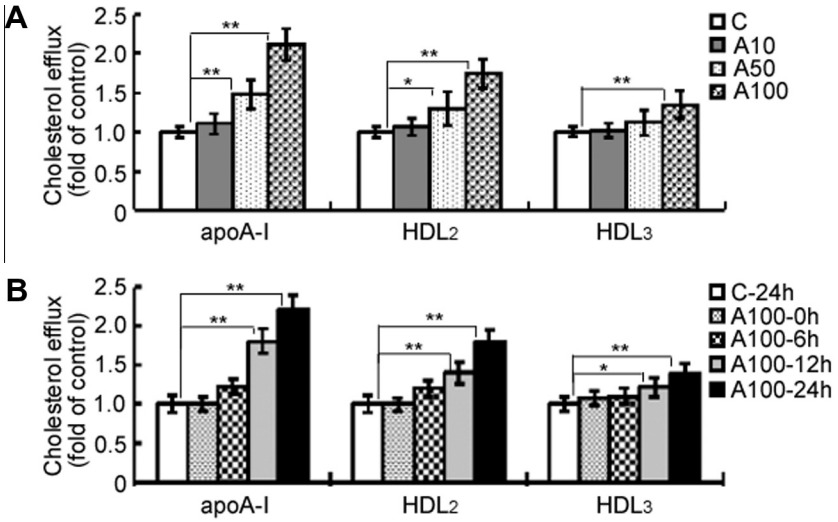

Fig. 1. Dose- and time-dependent effects of arctigenin on cholesterol efflux in oxLDL-stimulated THP-1 macrophages. Cholesterol efflux from oxLDL-stimulated THP-1 macrophages was measured as described in Section 2. The cells treated with different concentrations of arctigenin were incubated for12 h (A) or exposed to 100 μM of arctigenin for varying durations (B). Fold changes of cholesterol efflux efficacy were calculated by dividing the percentages of cholesterol efflux in the cells with different treatment by the percentage of cholesterol efflux in control cells. Data represents mean ± SD ($n = 6$). $^*P < 0.05$; $^{**}P < 0.01$ versus indicated controls.

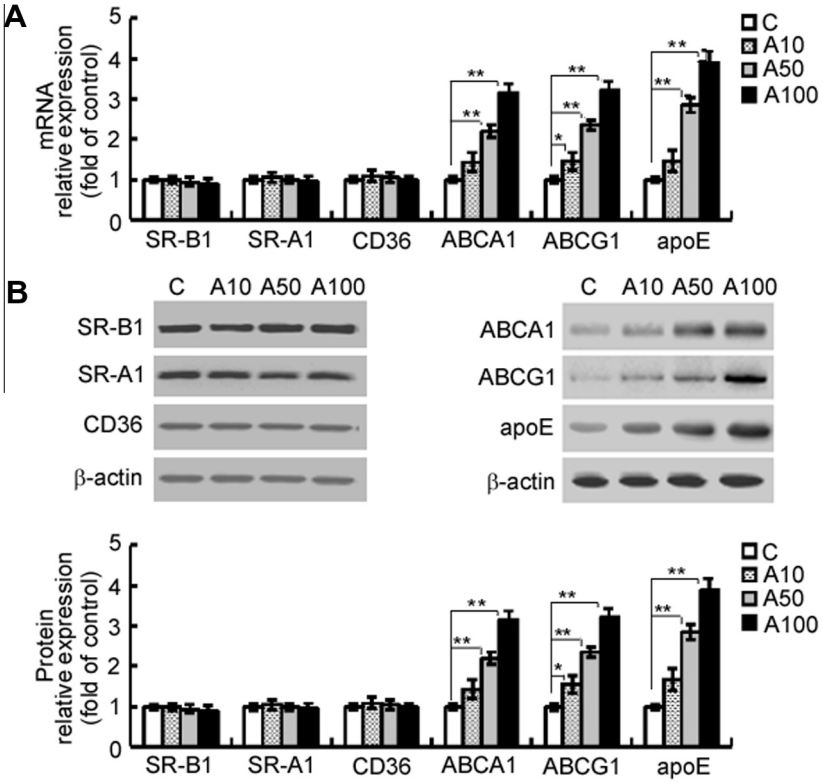

Fig. 2. The effects of arctigenin on the expression of SR-B1, SR-A1, CD36, ABCA1, ABCG1 and apoE in oxLDL-loaded THP-1 macrophages. THP-1 macrophages were stimulated with 50 mg/L oxLDL for 24 h in RPMI 1640 with 0.2% BSA. Then cells were treated with 100 μM of arctigenin for another 12 h. (A) RT-PCR and (B) Western blot analysis were performed for analyzing the expression of SR-B1, SR-A1, CD36, ABCA1, ABCG1 and apoE at mRNA and protein levels, respectively. Semiquantitative analysis of RT-PCR and Western blots was performed by densitometric analysis and normalized to β-actin levels. Fold changes of target gene expression were calculated by comparing their expression levels in cells with different treatments to that in control cells. Data represents mean ± SD ($n = 3$). $^{*}P < 0.05$; $^{**}P < 0.01$ versus indicated controls.

2.6. Statistical analysis

Data are expressed as mean ± standard deviation (SD). Statistical analysis was performed using the SPSS11.0 package software (SPSS Inc., Chicago, IL, USA). Differences in the means among groups were determined by one-way analysis of variance (ANOVA) followed by Tukey's post hoc test. A P value of less than 0.05 was considered to indicate statistical significance.

3. Results

3.1. Arctigenin accelerates cholesterol efflux from oxLDL-stimulated THP-1 macrophages

Twenty four hours after oxLDL loading in THP-1 macrophages, 100 μM of arctigenin clearly promoted the cholesterol efflux mediated by apoA-I, HDL_2, and HDL_3 by 2.1-($P < 0.01$), 1.75-($P < 0.01$) and 1.35-fold ($P < 0.01$), respectively, in oxLDL-loaded THP-1 macrophages (Fig. 1A). 50 μM of arctigenin significantly increased the cholesterol efflux induced by apoA-I and HDL_2 by 1.5-fold ($P < 0.01$) and 1.3-fold ($P < 0.05$), respectively (Fig. 1A). However, 10 μM of arctigenin did not show obvious effects on apoA-I and HDL-induced cholesterol efflux ($P > 0.05$; Fig. 1A).

In the time course experiments, 100 μM of arctigenin started to significantly enhance the cholesterol release-induced by apoA-1 ($P < 0.01$; Fig. 1B), HDL_2 ($P < 0.01$; Fig. 1B) and HDL_3 ($P < 0.05$; Fig. 1B) at 12 h. 24 h after the treatment with arctigenin, THP-1 cells showed 2.2-fold ($P < 0.01$), 1.8- ($P < 0.01$) and 1.4-fold ($P < 0.01$) increases in the cholesterol efflux-mediated by apoA-I, HDL_2 and HDL_3, respectively (Fig. 1B).

3.2. Arctigenin enhances the expression of ABCA1, ABCG1 and apoE in oxLDL-loaded THP-1 macrophages

Arctigenin concentrations ranging from 10 to 100 μM did not show obvious effects on the expression of SR-B1, SR-A1 and CD36 at both mRNA ($P > 0.05$; Fig. 2A) and protein levels ($P > 0.05$; Fig. 2B). The expression of ABCA1 did not show any changes at both mRNA and protein levels in oxLDL-loaded THP-1 macrophages-treated with 10 μM of arctigenin ($P > 0.05$; Fig. 2), but was remarkably upregulated in 50 and 100 μM of arctigenin-stimulated cells ($P < 0.01$; Fig. 2). 50 μM of arctigenin significantly enhanced the expression of ABCG1 ($P < 0.01$ for mRNA; $P < 0.05$ for protein; Fig. 2). For apoE expression, 10 μM of arctigenin only significantly elevated the expression of apoE mRNA, but not protein. The expression of apoE mRNA and protein was profoundly enhanced by 50 and 100 μM of arctigenin ($P < 0.01$; Fig. 2). Our data showed that arctigenin markedly upregulated the expression of ABCA1, ABCG1 and apoE in a concentration-dependent manner at 12 h after arctigenin treatment in THP-1 macrophages (Fig. 2).

3.3. Arctigenin induces the expression of PPAR-γ and LXR-α in oxLDL-loaded THP-1 macrophages

To detect whether PPAR-γ and LXR-α are involved in the upregulation of ABCA1, ABCG1 and apoE by arctigenin, RT-PCR and Western blot analysis were performed to examine the expression of PPAR-γ and LXR-α in arctigenin-treated and oxLDL-stimulated THP-1 cells. As Fig. 3A shown, the expression of PPAR-γ and LXR-α was upregulated at transcriptional levels by 50 ($P < 0.01$) and 100 μM ($P < 0.01$) of arctigenin, but not 10 μM of arctigenin. Similarly, 50 and 100 μM of arctigenin significantly promoted the

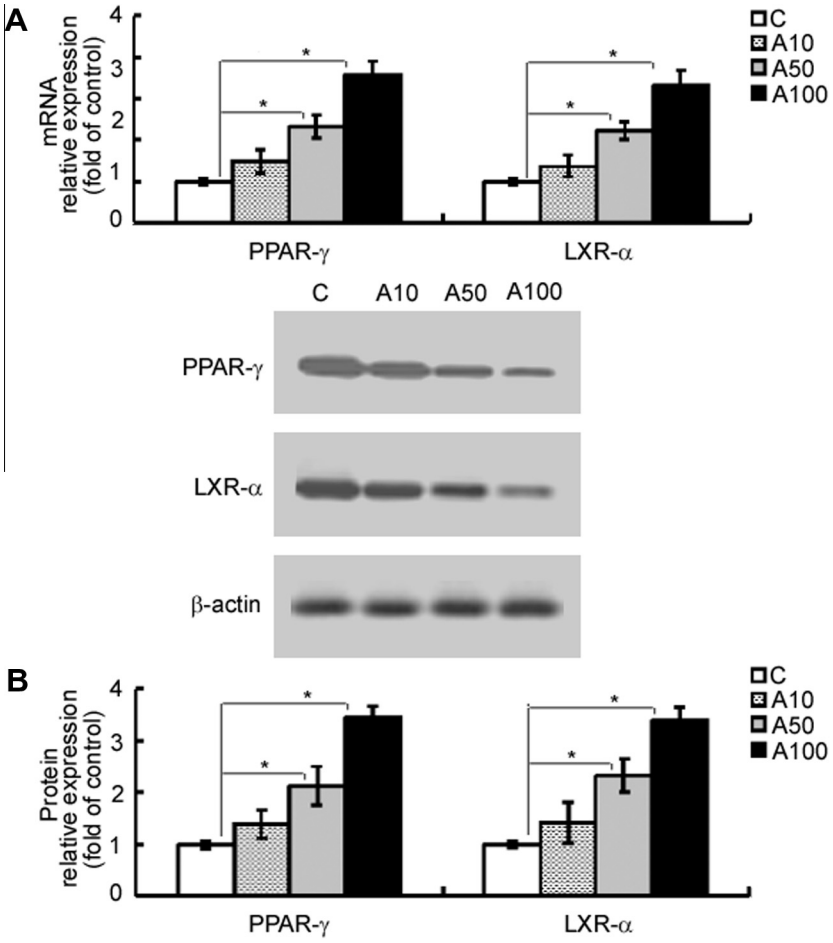

Fig. 3. The effects of arctigenin on the expression of PPARγ and LXR-α in oxLDL-loaded THP-1 macrophages. THP-1 macrophages were stimulated with 50 mg/L oxLDL for 24 h in RPMI 1640 with 0.2% BSA. Then cells were treated with 100 μM of arctigenin for another 12 h. (A) RT-PCR and (B) Western blot analysis were performed for analyzing the expression of PPARγ and LXR-α at mRNA and protein levels, respectively. Semiquantitative analysis of RT-PCR and Western blots was performed by densitometric analysis and normalized to β-actin levels. Fold changes of target gene expression were calculated by comparing their expression levels in cells with different treatments to that in control cells. Data represents mean ± SD ($n = 3$). $^{*}P < 0.05$; $^{**}P < 0.01$ versus indicated controls.

expression of PPAR-γ ($P < 0.01$; Fig. 3B) and LXR-α ($P < 0.01$; Fig. 3B) protein, but not 10 μM of arctigenin.

3.4. Specific inhibition of PPAR-γ and LXR-α by siRNA attenuates the upregulation of ABCA1, ABCG1 and apoE by arctigenin

30 nM of PPAR-γ- and LXR-α-siRNA inhibited the expression of PPAR-γ- and LXR-α proteins by 68% and 61%, respectively ($P < 0.01$; Fig. 4A and C). PPAR-γ- and LXR-α proteins were almost completely abolished by the transfection of 60 nM of PPAR-γ- and LXR-α-siRNA (by 90% for PPAR-γ and by 84% for LXRα; Fig. 4A and C).

PPAR-γ-siRNA substantially abolished arctigenin-mediated-induction of ABCA1, ABCG1, apoE and LXR-α (Fig. 4B). However, scrambled siRNA had no effects on the expression of these proteins (Fig. 4B). Arctigenin-mediated upregulation of ABCA1, ABCG1, and apoE were also significantly suppressed by LXR-α-siRNA (Fig. 4D).

4. Discussion

In the present study, we found that arctigenin profoundly enhanced apoA-I and HDL-mediated cholesterol efflux in a dose- and time-dependent manner in oxLDL-stimulated THP-1 macrophages. Western blot analysis showed that the expression of ABCA1, ABCG1 and apoE was significantly upregulated in arctigenin-treated cells. Interestingly, arctigenin also remarkably promoted the expression of PPAR-γ and LXR-α, and specific inhibition of PPAR-γ or LXR-α reversed arctigenin-mediated induction of ABCA1, ABCG1 and apoE. Our data indicated that arctigenin promotes cholesterol efflux by enhancing ABCA1, ABCG1 and apoE expression through activation of PPAR-γ/LXR-α pathway.

In this study, we found that arctigenin accelerated apoA-I and HDL-induced cholesterol efflux in oxLDL-stimulated THP-1 cells. To investigate the mechanism of how arctigenin induces cholesterol release in macrophage foam cells, we examined several key genes related to cholesterol efflux. The scavenger receptors (SR-B1, SR-A1, CD36) are responsible for uptake of modified cholesterol [11]. The expression of SR-B1, SR-A1 and CD36 were measured in arctigenin-stimulated and cholesterol-loaded THP-1 macrophages. However, our results indicated that arctigenin has no obvious effects on the expression of these scavenger receptors, in terms of SR-B1, SR-A1 and CD36. ABCA1 and ABCG1 are integral membrane proteins that utilize ATP as a source of energy for transporting lipids and other metabolites across membranes [12]. Our data showed that arctigenin dose-dependently upregulated the expression of ABCA1 and ABCG1, indicating that ABCA1 and ABCG1 play important roles in arctigenin-mediated cholesterol efflux. ApoE can induce cholesterol efflux from the macrophage that originally secreted it (autocrine effect) or from surrounding macrophages (paracrine effect), which inhibits foam cell formation and prevents atherosclerosis [13]. Western blot analysis showed that arctigenin

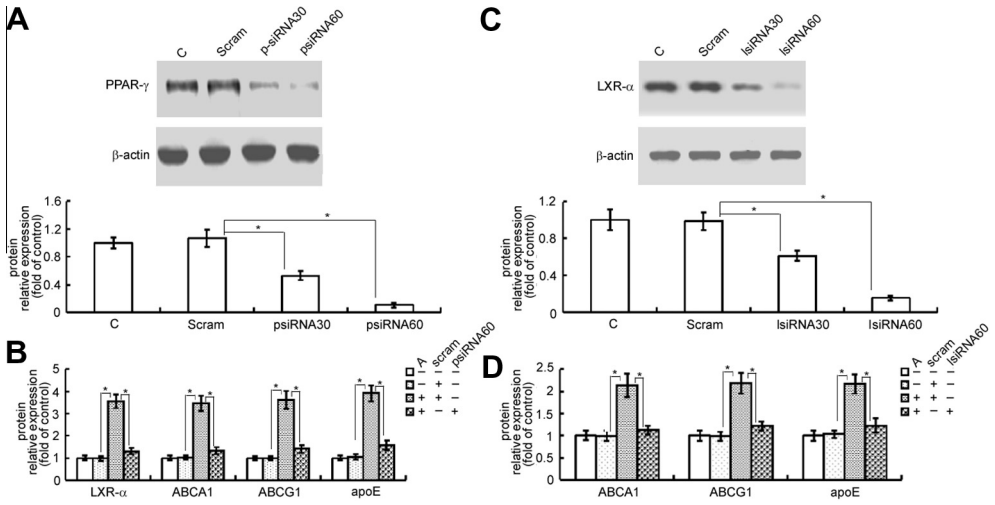

Fig. 4. Effects of PPAR-γ and LXR-α-siRNA on arctigenin-mediated gene expression in oxLDL-loaded THP-1 macrophages. After stimulation with PMA for 72 h, indicated concentrations of siRNA were added to THP-1 cells. The cells were incubated for 24 h, washed and then 50 mg/L of oxLDL were added for another 24 h in 0.2 % fatty acid-free BSA. The cells were stimulated with 100 μM of arctigenin for 12 h and collected for Western blot analysis to analyze the expression of PPAR-γ (A) and LXR-α (C). (B) The expression of LXR-α, ABCA1, ABCG1 and apoE was examined by Western blot analysis. (D) The expression of ABCA1, ABCG1 and apoE was measured by Western blot analysis. β-Actin was used as an internal control. Fold changes of target gene expression were calculated by comparing their expression levels in cells with different treatment to those in control cells. Data represents mean ± SD ($n = 3$). $^{*}P < 0.05$; $^{**}P < 0.01$ versus indicated controls.

enhanced the expression of apoE. These results suggested that arctigenin enhances cholesterol efflux through multiple signaling pathways. Actually, many extracts from Chinese traditional medicine, such as resveratrol and quercetin, have been shown promotive effects on cholesterol efflux through different signaling pathways [14–16]. Our results suggest that arctigenin might be another candidate for antiatherosclerotic therapy.

How does arctigenin enhance the expression of ABCA1, ABCG1 and apoE? Nuclear receptors offer one strategy to integrate multiple signaling pathways. PPAR-γ and LXR-α are two key nuclear receptors that play pivotal roles in macrophage cholesterol homeostasis and inflammation [17]. Both PPAR-γ and LXR-α are implicated in the transactivaiton of ABCA1, ABCG1 and apoE, integral players in cholesterol efflux [17]. We detected the effects of arctigenin on the expression of PPAR-γ and LXR-α. Interestingly, arctigenin significantly enhanced the expression of PPAR-γ and LXR-α in a dose-dependent manner. Moreover, silencing of LXR-α by LXR-α-siRNA suppressed arctigenin-mediated upregulation of ABCA1, ABCG1 and apoE, indicating that arctigenin-induced expression of ABCA1, ABCG1 and apoE is dependent on LXR-α signaling, which is consistent with the findings that ABCA1, ABCG1 and apoE are the direct targets of LXR-α in cholesterol efflux [17–19]. LXR-α is identified as one target gene of PPAR-γ and directly regulate ABCA1 expression [17]. Specific targeting of PPAR-γ by PPAR-γ-siRNA abolished the expression of LXR-α, ABCA1, ABCG1 and apoE, indicating that PPAR-γ is an upstream regulator in arctigenin-mediated promotion of cholesterol efflux. Our results suggested that arctigenin-induced upregulation of ABCA1, ABCG1 and apoE is dependent on PPAR-γ/LXR-α signaling pathway.

Arctigenin exhibits potent suppressive effects on the production of NO through down-regulation of iNOS protein expression and inhibition on the iNOS enzymatic activity in LPS-stimulated macrophages RAW 264.7 and THP-1 [8]. Excess NO decreases LXR-α-ABCA1-dependent cholesterol efflux in macrophage foam cells [9]. We therefore hypothesized that arctigenin might promote cholesterol efflux in macrophage foam cells. Interestingly, we found that arctigenin enhanced cholesterol efflux in cholesterol-loaded macrophages in a dose and time dependent manner. Whether arctigenin-induced cholesterol efflux is dependent on the suppression of NO, is still unclear. In further studies, this question will be addressed.

Taken together, our data indicate that arctigenin promotes cholesterol efflux through increasing the expression of ABCA1, ABCG1 and apoE by PPAR-γ/LXR-α signaling.

References

[1] C.K. Glass, J.L. Witztum, Atherosclerosis: the road ahead, Cell 104 (2001) 503–516.
[2] N.R. Webb, K.J. Moore, Macrophage-derived foam cells in atherosclerosis: lessons from murine models and implications for therapy, Curr. Drug Targets 8 (2007) 1249–1263.
[3] M. Cuchel, D.J. Rader, Macrophage reverse cholesterol transport: key to the regression of atherosclerosis?, Circulation 113 (2006) 2548–2555
[4] R. Ohashi, H. Mu, X. Wang, Q. Yao, C. Chen, Reverse cholesterol transport and cholesterol efflux in atherosclerosis, QJM 98 (2005) 845–856.
[5] S. Awale, J. Lu, S.K. Kalauni, Y. Kurashima, Y. Tezuka, S. Kadota, H. Esumi, Identification of arctigenin as an antitumor agent having the ability to eliminate the tolerance of cancer cells to nutrient starvation, Cancer Res. 66 (2006) 1751–1757.
[6] M.K. Cho, Y.P. Jang, Y.C. Kim, S.G. Kim, Arctigenin, a phenylpropanoid dibenzylbutyrolactone lignan, inhibits MAP kinases and AP-1 activation via potent MKK inhibition: the role in TNF-alpha inhibition, Int. Immunopharmacol. 4 (2004) 1419–1429.
[7] T. Matsumoto, K. Hosono-Nishiyama, H. Yamada, Anti-proliferative and apoptotic effects of butyrolactone lignans from *Arctium lappa* on leukemic cells, Planta. Med. 72 (2006) 276–278.
[8] F. Zhao, L. Wang, K. Liu, In vitro anti-inflammatory effects of arctigenin, a lignan from *Arctium lappa* L., through inhibition on iNOS pathway, J. Ethnopharmacol. 122 (2009) 457–462.
[9] J.F. Zhao, S.K. Shyue, S.J. Lin, J. Wei, T.S. Lee, Excess nitric oxide impairs liver X receptor α-ATP-binding cassette transporter A1-dependent cholesterol efflux in macrophage foam cells, J. Cell. Physiol. 229 (2014) 117–125.
[10] M. Ayaori, S. Sawada, A. Yonemura, N. Iwamoto, M. Ogura, N. Tanaka, K. Nakaya, M. Kusuhara, H. Nakamura, F. Ohsuzu, Glucocorticoid receptor regulates ATP-binding cassette transporter-A1 expression and apolipoprotein-mediated cholesterol efflux from macrophages, Arterioscler. Thromb. Vasc. Biol. 26 (2006) 163–168.
[11] D.L. Williams, M.A. Connelly, R.E. Temel, S. Swarnakar, M.C. Phillips, M. de la Llera-Moya, G.H. Rothblat, Scavenger receptor BI and cholesterol trafficking, Curr. Opin. Lipidol. 10 (1999) 329–339.
[12] M. Dean, Y. Hamon, G. Chimini, The human ATP-binding cassette (ABC) transporter superfamily, J. Lipid Res. 42 (2001) 1007–1017.
[13] D.E. Dove, M.F. Linton, S. Fazio, ApoE-mediated cholesterol efflux from macrophages: separation of autocrine and paracrine effects, Am. J. Physiol. Cell. Physiol. 288 (2005) C586–C592.
[14] I. Voloshyna, O. Hai, M.J. Littlefield, S. Carsons, A.B. Reiss, Resveratrol mediates anti-atherogenic effects on cholesterol flux in human macrophages and endothelium via PPARγ and adenosine, Eur. J. Pharmacol. 698 (2013) 299–309.
[15] Y.C. Chang, T.S. Lee, A.N. Chiang, Quercetin enhances ABCA1 expression and cholesterol efflux through a p38-dependent pathway in macrophages, J. Lipid Res. 53 (2012) 1840–1850.

[16] S.M. Lee, J. Moon, Y. Cho, J.H. Chung, M.J. Shin, Quercetin up-regulates expressions of peroxisome proliferator–activated receptor γ, liver X receptor α, and ATP binding cassette transporter A1 genes and increases cholesterol efflux in human macrophage cell line, Nutr. Res. 33 (2013) 136–143.
[17] A. Majdalawieh, H.S. Ro, PPARgamma1 and LXRalpha face a new regulator of macrophage cholesterol homeostasis and inflammatory responsiveness, AEBP1, Nucl. Recept. Signal. 8 (2010) e004.
[18] A. Chawla, W.A. Boisvert, C.H. Lee, B.A. Laffitte, Y. Barak, S.B. Joseph, D. Liao, L. Nagy, P.A. Edwards, L.K. Curtiss, R.M. Evans, P.A. Tontonoz, PPAR γ-LXR-ABCA1 pathway in macrophages is involved in cholesterol efflux and atherogenesis, Mol. Cell. 7 (2001) 161–171.
[19] R. Galetto, M. Albajar, J.I. Polanco, M.M. Zakin, J.C. Rodriguez-Rey, Identification of a peroxisome-proliferator–activated-receptor response element in the apolipoprotein E gene control region, Biochem. J. 357 (2001) 521–527.