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TLR2 stimulates ABCA1 expression via PKC-η and PLD2 pathway

Dae-Weon Park [a], Hyung-Kyoung Lee [a], Ji Hyo Lyu [a], Haemin Chin [a], Sang Wook Kang [b], Young-Jo Kim [b], Yoe-Sik Bae [c], Suk-Hwan Baek [a,*]

[a] Department of Biochemistry & Molecular Biology, Aging-Associated Vascular Disease Research Center, Republic of Korea
[b] Department of Internal Medicine, College of Medicine, Yeungnam University, Daegu 705-802, Republic of Korea
[c] Department of Biological Sciences, Sungkyunkwan University, Suwon 440-746, Republic of Korea

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ABSTRACT

ATP-binding cassette transporter A1 (ABCA1) is a membrane-bound protein that regulates cardiovascular disease including atherosclerosis by the efflux of excess cholesterol from cells and by suppression of inflammation. Using a mouse macrophage cell line Raw264.7, we studied the importance of toll-like receptor 2 (TLR2) on ABCA1 expression and the signaling pathway responsible for TLR2-mediated ABCA1 expression. Interestingly, our data demonstrated that treatment of macrophages with TLR2 agonist Pam_3CSK_4 significantly increased ABCA1 mRNA and protein levels. We found that ABCA1 induction is myeloid differentiation primary response gene 88 (MyD88)-dependent as well as TLR2-dependent. ABCA1 induction upon Pam_3CSK_4 is controlled by protein kinase C-η (PKC-η) and phospholipase D2 (PLD2). Furthermore, direct treatment of dioctanoyl phosphatidic acid (diC_8PA) into cells also induced ABCA1 mRNA and protein indicating that PLD2-mediated PA involve in the TLR2-stimulated ABCA1 expression. Cumulatively, these results demonstrate for the first time that activation of PKC-η and PLD2 signaling pathway is an important mechanism for regulation of TLR2-induced ABCA1 expression.

1. Introduction

Atherosclerosis is a chronic and progressive inflammatory disease of the arteries that is characterized by subendothelial accumulation of lipid-rich macrophages, called foam cells [1]. Although the mechanism has not been clearly established, toll-like receptors (TLRs) in atherosclerotic lesions contribute to the formation of foam cells [2,3]. Using a genetically defined apolipoprotein E-deficient model, pathogen-mediated inflammatory atherosclerosis occurs via TLR2-dependent manner [4,5]. Several studies also suggest the relevance of TLRs in the atherosclerosis pathology, either using human or animal models [6]. Especially, TLR2 and TLR4 participate in the induction and progression of the atheroma [7].

Reverse cholesterol transport (RCT) is a pathway by which accumulated cholesterol is transported from the vessel wall to the liver for excretion, thus preventing atherosclerosis. ATP-binding cassette transporter A1 (ABCA1) is one of the plasma membrane proteins that export excess cholesterol derived from internalized lipoproteins [8]. Specific disruption of the macrophage ABCA1 gene aggravates atherosclerosis development in low-density lipoprotein (LDL) receptor knockout mice [9], and macrophages obtained from ABCA1 knockout mice are deficient in cholesterol efflux [9,10]. Overexpression of ABCA1 inhibits atherosclerosis in mice models [11]. Macrophages obtained from transgenic mice that overexpress ABCA1 have an elevated cholesterol efflux compared with normal macrophages [11,12].

Foam cell formation is a key event for the accumulation of lipids and can be enhanced by the presence of bacteria or lipopolysaccharide (LPS), TLR4 agonist [13]. Our previous studies also demonstrated that various TLR agonists increased foam cell formation [14]. Transporters implicated in changes of foam cell formation include the ABCA1 [15]. However, early investigation of the molecular mechanism underlying LPS-mediated ABCA1 expression is very controversial. While Kaplan et al. [16] reported that bacterial LPS induced ABCA1 expression; another report suggested that LPS down regulates ABCA1 expression, leading to suppression of cholesterol efflux [17]. Therefore, we clarified such opposite results and studied by what mechanism ABCA1 expression is regulated.

In this study, we demonstrated that TLR2 agonist Pam_3CSK_4 increased ABCA1 expression, and such expression was regulated

Abbreviations: TLR, toll-like receptor; PKC-η, protein kinase C-η; PLD2, phospholipase D2; ABCA1, ATP-binding cassette transporter A1; MyD88, myeloid differentiation primary response gene 88; RCT, reverse cholesterol transport; PA, phosphatidic acid; BMDM, bone marrow-derived macrophage.

* Corresponding author. Address: Department of Biochemistry & Molecular Biology, Aging-Associated Vascular Disease Research Center, 210 Main Building, College of Medicine, Yeungnam University, 170 Hyeonchung-ro (317-1 Daemyung-5 Dong), Nam-gu, Daegu 705-802, Republic of Korea. Fax: +82 53 623 8032.
E-mail address: sbaek@med.yu.ac.kr (S.-H. Baek).

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through protein kinase C-η (PKC-η) and PLD2 signaling. This report demonstrates for the first time that TLR2-induced ABCA1 expression is associated with phosphatidic acid (PA) derived from PLD2.

2. Materials and methods

2.1. Cells lines and reagents

Raw264.7 cell lines were purchased from the American Type Culture Collection (ATCC, Manassas, VA) and cultured in Dublecco's modified Eagle's medium supplemented with 10% fetal bovine serum and 1% antibiotics (100 U/ml penicillin and 100 μg/ml streptomycin) at 37 °C in 5% CO_2. Bone marrow-derived monocytes were differentiated into bone marrow-derived macrophages (BMDM) for 5–7 days in DMEM supplemented with 10% L929 cell-conditioned medium (as a source of macrophage-colony stimulating factor; M-CSF). This study was conducted in accordance with the guidelines for the care and use of laboratory animals provided by Yeungnam University and all experimental protocols were approved by the Ethics Committee of Yeungnam University, Republic of Korea. Cell-culture reagents, including fetal bovine serum (FBS), were obtained from Life Technologies (Grand Island, NY). ABCA1 antibody was obtained from Novus Biologicals (Littleton, CO) and PLD2 antibody was from Santa Cruz Biotechnology (Santa Cruz, CA). β-Actin antibody, *Escherichia coli* LPS (0111:B4), 1-butanol (BuOH), *t*-BuOH and Polymyxin B were obtained from Sigma–Aldrich (St. Louis, MO). Dioctanoyl phosphatidic acid (diC_8PA) was obtained from Avanti polar lipids (Alabaster, AL). Pam_3CSK_4, various TLR agonists were purchased from InvivoGen (San diego, CA). Reverse transcription-polymerase chain reaction (RT-PCR) kits were from Takara Bio (Japan). siRNA and TRIzol were purchased from Invitrogen (Carlsbad, CA). PKC-η pseudosubstrate (PS) peptide inhibitor was from Peptron (Daejeon, South Korea).

2.2. siRNAs and transfection

Stealth control and gene-specific siRNAs against the following target genes were designed using the Block-IT Stealth RNAi designer (Invitrogen): (i) TLR2, 5′-UUA AAG GGC GGG UCA GAG UUC UCC A-3′; (ii) MyD88, 5′-ACC ACC AUG CGG CGA CAC CUU UUC U-3′; (iii) PLD2, 5′-AAG ACU UUG UGU CUC UGG AGG UCC C-3′; (iv) PKC-η, 5′-UUG UGU GGG AUG UUG AUG CCA AAC C-3′. For transfection experiments, Raw264.7 cells were plated in 35 mm petri-dish and transfected with siRNA at a final concentration of 150–200 pM using nucleoporation reagents from Lonza (Allendale, NJ). Cells were nucleoporated according to the manufacturer's protocol and incubated for 24 h before Pam_3CSK_4 stimulation.

2.3. Reverse transcription-polymerase chain reaction (RT-PCR)

Total RNA was extracted from cells using TRIzol. First-strand cDNA was synthesized from 1 μg total RNA with random primers, oligo-dT, and reverse transcriptase (Promega, Madison, WI). Cycling conditions for PCR were 95 °C for 5 min, followed by 26–33 cycles at 95 °C for 1 min, 63 °C for 1 min, and 72 °C for 1 min. For semi-quantitative PCR, target genes expression was normalized to β-actin transcription. The sequences of PCR primers used in the present study were as follows: (i) ABCA1 forward, 5′-GGA AGT GGC CTG GCC TCT CT-3′, reverse, 5′-GAG AAC AGGCGA GAC ACG AT-3′; (ii) TLR2 forward, 5′-CGC CCT TTA AGC TGT GTC TC-3′, reverse, 5′-TTA TCT TGC GCA GTT TGC AG-3′; (iii) MyD88 forward, 5′-CCA GTA TCC TGC GGT TCA TCA-3′, reverse, 5′-GCT CCG CAT CAG TCT CAT CTT-3′; (iv) PKC-η forward, 5′-AAG GAA ATC GAC TGG GCC-3′, reverse, 5′-CAG TTG CAA TTC CGG TGA C-3′; (v) PLD2 forward, 5′-CTG GCC ATC TAT GAC CTT CAG C-3′, reverse, 5′-GCC ACC TCT TGG ACC ATC GAT A-3′; (vi) β-actin forward, 5′-TCC TTC GTT GCC GGT CCA CA-3′, reverse, 5′-CGT CTC CGG AGT CCA TCA CA-3′.

2.4. Western-blot analysis

Macrophages were cultured in 35 mm petri-dishes and treated with Pam_3CSK_4 in the presence or absence of inhibitors. Cell pellets were resuspended in lysis buffer (50 mM Tris–HCl, pH 8.0; 5 mM EDTA; 150 mM NaCl; 0.5% Nonidet P-40; 1 mM phenylmethylsulfonyl fluoride; and protease inhibitor cocktail). Proteins were separated on an 8% reducing SDS–PAGE gels and transferred onto nitrocellulose membranes in 20% methanol, 25 mM Tris, and 192 mM glycine. Membranes were then blocked with 5% non-fat dry milk and incubated overnight with primary antibody at 4 °C before washing, followed by 1 h incubation with horseradish-peroxidase conjugated secondary antibody, further wash steps, and development with an enhanced chemiluminescence system (LAS 3000, GE Healthcare, Buckinghamshire, UK).

3. Results

3.1. Pam_3CSK_4 induces ABCA1 expression

We investigated whether the TLR2 agonist Pam_3CSK_4 can induce ABCA1 expression in Raw264.7 macrophages cell line. Pam_3CSK_4 induced both mRNA and protein of ABCA1 in a time-dependent manner (Fig. 1A and B). To prove the general effect of TLR agonist for ABCA1 expression, we tested other types of TLR agonists. TLR agonists Pam_3CSK_4 (TLR1/2), HKLM (TLR2), LPS (TLR4), Flagellin (TLR5), FSL1 ($Pam_2CGDPKHPKSF$) (TLR6/2) and

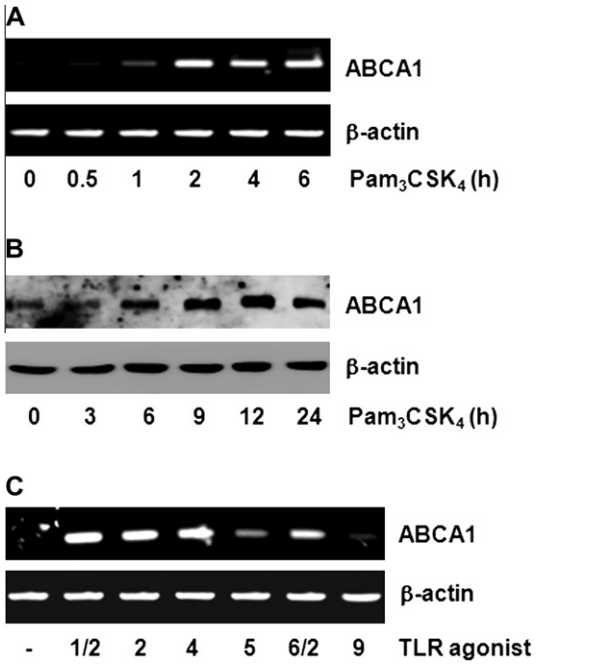

Fig. 1. Pam_3CSK_4 induces ABCA1 mRNA and protein in a time-dependent manner. (A and B) Raw264.7 cells were treated with Pam_3CSK_4 (100 ng/ml) for the times indicated. ABCA1 mRNA expression was determined by RT-PCR and normalized to a β-actin control. ABCA1 protein expression was determined by western blotting using ABCA1 antibody. (C) Raw264.7 cells were treated with the various TLR agonists for 2 h and ABCA1 mRNA expression was determined by RT-PCR and normalized to a β-actin control.

CpG ODN 1668 (TLR9, Type B) induced ABCA1 expression in mRNA level (Fig. 1C). These results suggest that ABCA1 expression by TLR agonists is a general characteristic shown in TLRs.

3.2. ABCA1 induction by Pam_3CSK_4 is TLR2-MyD88 signaling-dependent

We tested whether Pam_3CSK_4 affects ABCA1 expression in BMDM. While ABCA1 expression was induced by Pam_3CSK_4 in BMDM from WT mice, its expression was not affected in BMDM from mice with TLR2 deletions (TLR2 KO; Fig. 2A). MyD88 is the best characterized adaptor protein in TLR2 signaling. We therefore tested its involvement in ABCA1 expression by Pam_3CSK_4 using siRNA. MyD88 siRNA transfected into cells could successfully down-regulate its respective target gene. We found that in Pam_3CSK_4-treated cells, ABCA1 expression was increased in control siRNA transfected cells, but was partially reduced in MyD88 siRNA transfected cells (Fig. 2B and C).

3.3. PKC-η is involved in TLR2-mediated ABCA1 expression

Previous studies have been reported that some PKC is regulator for ABCA1 expression [18,19]. Among the PKC isoforms, PKC-η is predominantly expressed in Raw264.7 cells [13]. We therefore sought to determine the role of PKC-η in ABCA1 expression. We confirmed this involvement of PKC-η in ABCA1 expression by RNAi; PKC-η knockdown blocked increase by Pam_3CSK_4 of ABCA1 expression. (Fig. 3A and B). The PKC-η PS peptide is often used as an inhibitor of PKC-η activity; in our hands, it inhibited ABCA1 expression upon Pam_3CSK_4 stimuli (Fig. 3C and D). These results suggest that the PKC-η is important for TLR2-mediated ABCA1 expression.

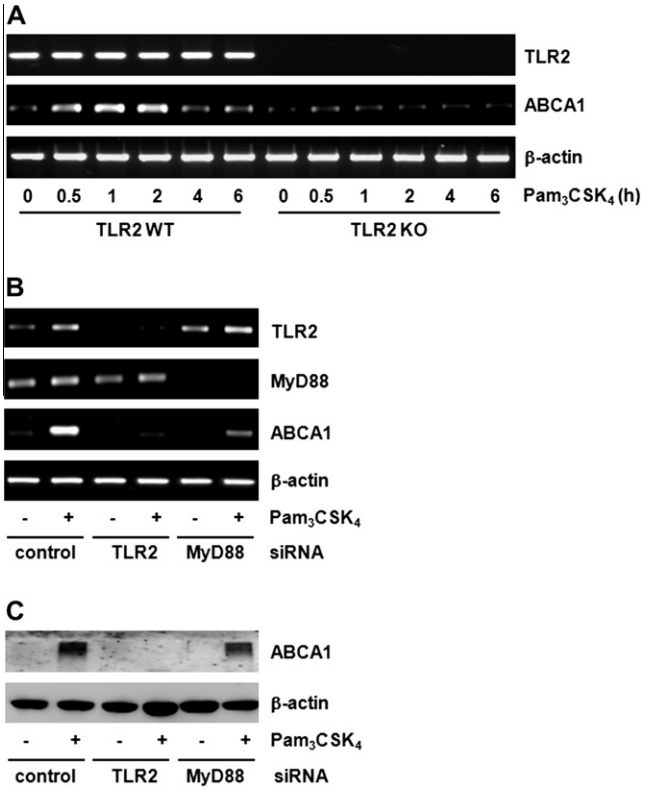

Fig. 2. Pam_3CSK_4 induces ABCA1 expression by a TLR2- and MyD88-dependent pathway. (A) BMDMs were isolated from WT or TLR2 KO mouse bone marrow and treated with Pam_3CSK_4 (100 ng/ml) for the times indicated. Indicated genes expression was determined by RT-PCR and normalized to a β-actin control. (B and C) Raw264.7 cells were transfected with control, TLR2, or MyD88 siRNA (200 pM) for 24 h. Cells were stimulated with vehicle or Pam_3CSK_4 (100 ng/ml). After 2 h, indicated genes expression was determined by RT-PCR and normalized to a β-actin control (B). After 12 h, ABCA1 protein expression was determined by western blotting and normalized to a β-actin (C).

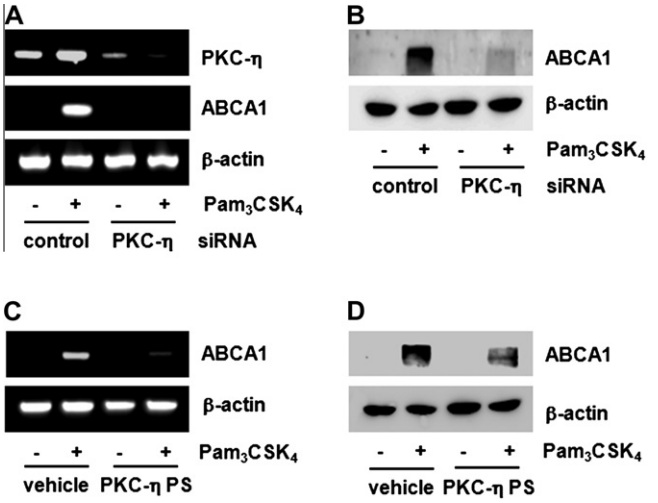

Fig. 3. PKC-η mediates ABCA1 induction by Pam_3CSK_4. (A and B) Raw264.7 cells were transfected with either control or PKC-η siRNA (200 pM) for 24 h. Cells were stimulated with vehicle or Pam_3CSK_4 (100 ng/ml). (A) After 2 h, PKC-η and ABCA1 mRNA expression was determined by RT-PCR and normalized to a β-actin control. (B) After 12 h, ABCA1 protein expression was determined by western blotting and normalized to a β-actin. (C and D) Raw264.7 cells were pretreated with PKC-η PS inhibitor (50 μM) and stimulated with Pam_3CSK_4 (100 ng/ml). ABCA1 mRNA (C) and protein (D) expression were determined by RT-PCR or western blot, respectively.

3.4. PLD2-mediated PA is an essential regulator for ABCA1 expression

Previous studies have been reported that PKC is activator for PLD activity [13]. We studied the function of PLD2 in ABCA1 regulation by TLR2. PLD2, rather than PLD1, is dominantly expressed in Raw264.7 cells [20], therefore we tested the effects of PLD2 using siRNA and inhibitor methods. Compared to control siRNA transfected cells, PLD2 siRNA transfected cells had decreased expressions of PLD2 mRNA and protein levels. Therefore ABCA1 mRNA and protein expressions were also decreased in PLD2 siRNA transfected cells (Fig. 4A and B). 1-BuOH is the most widely used PLD inhibitor. Pretreatment of 1-BuOH in Raw264.7 cells inhibited ABCA1 mRNA expression by Pam_3CSK_4, while pretreatment of *t*-BuOH, an alcohol control, showed no effect (Fig. 4C). Pretreatment of 1-BuOH also showed a similar pattern in ABCA1 protein expressions (Fig. 4D). These results suggest that PLD2 is an essential mediator in Pam_3CSK_4 induced ABCA1 expression. Effects of PLD2 were shown in Pam_3CSK_4 induced ABCA1 expression. PLD2 produces a secondary messenger, PA, and therefore we confirmed the direct effect of PA using diC_8PA. Treatment of diC_8PA induced both mRNA and protein expressions of ABCA1 as shown by Pam_3CSK_4 (Fig. 4E and F). Polymyxin was used to prove that ABCA1 expression induced by Pam_3CSK_4 and diC_8PA is not a result from toxin contamination. Pretreatment of polymyxin, a toxin scavenger, strongly inhibited ABCA1 induced by LPS, but had no effect on ABCA1 expressions induced by either diC_8PA or Pam_3CSK_4 (Fig. 4G).

4. Discussion

There are currently 11 types of TLR in mammalian cells known. TLR2 and TLR4 exist at the plasma membrane while TLR3 and TLR9 exist at the endosome [21]. TLR4 is most widely studied, and LPS is the representative ligand. Baranova et al. [17] reported that LPS reduces ABCA1 expression and thereby inhibits cholesterol efflux.

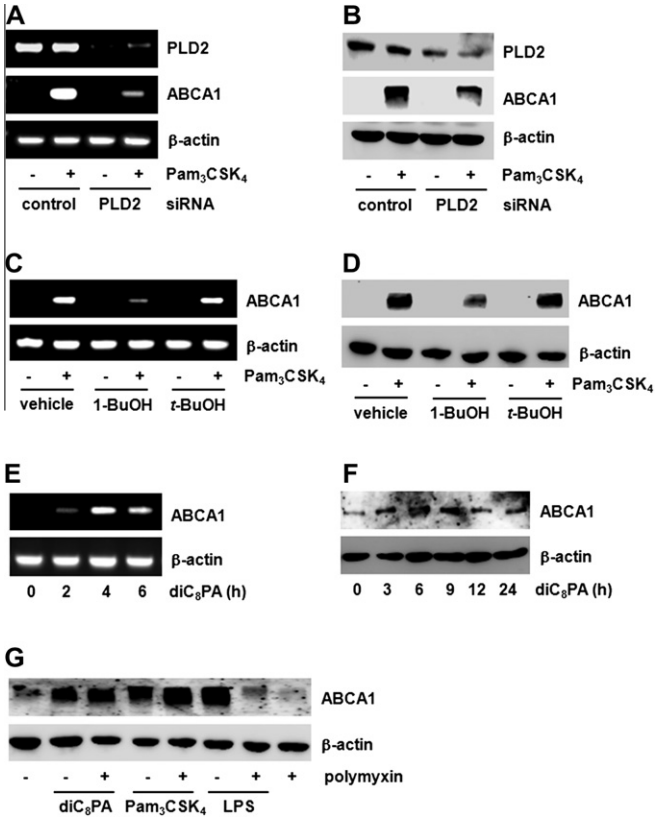

Fig. 4. PLD2 mediates ABCA1 induction by Pam_3CSK_4. (A and B) Raw264.7 cells were transfected with either control or PLD2 siRNA (200 pM) for 24 h. Cells were stimulated with vehicle or Pam_3CSK_4 (100 ng/ml). After 2 h, PLD2 and ABCA1 mRNA expression was determined by RT-PCR and normalized to a β-actin control (A). After 12 h, PLD2 and ABCA1 protein expression was determined by western blotting and normalized to a β-actin (B). (C and D) Raw264.7 cells were pretreated with PLD inhibitor (0.3% 1-butanol) or an alcohol control (0.3% *t*-butanol) and stimulated with Pam_3CSK_4 (100 ng/ml). ABCA1 mRNA expression were determined by RT-PCR and normalized to a β-actin control (C). ABCA1 protein expression was determined by western blotting and normalized to a β-actin (D). (E and F) Raw264.7 cells were treated with diC_8PA (20 μM) for the times indicated. ABCA1 mRNA expression was determined by RT-PCR and normalized to a β-actin control (E). ABCA1 protein expression was determined by western blotting and normalized to a β-actin (F). (G) Raw264.7 cells were treated with the diC_8PA, Pam_3CSK_4 or LPS in the absence or presence of polymyxin for 12 h and ABCA1 protein expression was determined by western blotting and normalized to a β-actin.

However, Kaplan et al. [16] reported that LPS increases ABCA1 expression. Our results showed similar trends with reports of Kaplan et al. Currently there are no reports shown on ABCA1 expression by other TLRs except TLR4, but our results showed that TLR2 and other types of TLR agonists (TLR1/2, TLR2, TLR4, TLR5, TLR6/2, and TLR9) increased ABCA1 expression as well.

Elevation of ABCA1 expression is assumed to inhibit foam cell formation and atherosclerosis by effluxing intracellular cholesterol [22]. However, many other studies reported that TLR stimulation promotes atherosclerosis [2,3]. This is very opposite phenomena. Our previous results also showed that LPS or Pam_3CSK_4 promoted foam cell formation in macrophages [13,14]. It is not easy to interpret foam cell formation increase amongst the elevation of ABCA1 expression. Our hypothesis is that although TLR2 stimulation promotes foam cell formation and atherosclerosis as a final output, cells will have a defense mechanism against the process of atherosclerosis. Elevation of ABCA1 expression is also one of the defense factors in TLR2-induced foam cell formation.

Previous studies reported about defense factors of atherosclerosis such as LRP1, heme oxygenase-1 (HO-1), or regulator of G-protein signaling 2 (RGS2) [23–26]. Especially, HO-1 is a well known protein for inhibiting atherosclerosis progression [23]. Recently, RGS2 is also studied as a protein inhibiting foam cell formation [24]. RGS2 is capable of deactivating G protein subunit in G-protein coupled receptor, and acts as a turn-off switch on the process of hypertension [27]. RGS2 rapidly decreases with TLR stimulation in Raw264.7 cells [24]. This phenomenon is probably due to its mechanism in interrupting the turn-off function of RGS2 in macrophages. Therefore, while various positive regulators participate in TLR2-induced foam cell formation, there must be defense mechanisms such as ABCA1 participating, and the balance of this will decide whether atherosclerosis will be promoted or inhibited.

Pam_3CSK_4 is an agonist that usually combines with TLR1/TLR2 heterodimer receptor and transmits signals. However, in TLR2 KO macrophages, ABCA1 increase by Pam_3CSK_4 did not occur. These results show that Pam_3CSK_4 induced ABCA1 expression is TLR2-dependent. The importance of TLR2 in ABCA1 expression was confirmed by siRNA method. TLR2 siRNA transfected cells had decreased TLR2 expression. TLR2 knock-down cells had decreased ABCA1 mRNA and protein expression induction, which are Pam_3CSK_4 effects (Fig. 2B). These results explain with Fig. 2A that Pam_3CSK_4 induced ABCA1 expression is TLR2 dependent.

The effect of MyD88 acting under TLR2 was also essential. MyD88 siRNA cells showed significant decrease in not only MyD88, but also ABCA1 expression. These results suggest that ABCA1 expression by Pam_3CSK_4 is dependent upon MyD88 as well. ABCA1 expression in MyD88 siRNA cells significantly decreased, but the some expression is still remained in both mRNA and protein level. These phenomena suggest the possibility that other types of adaptor proteins or pathways can exist in ABCA1 expression other than MyD88. In fact, when TLR/IL-1R-domain-containing adapter-inducing interferon-β (TRIF) siRNA was used, which has an important part in TLR4 signaling, ABCA1 expression partially decreased, suggesting TRIF participation (personal observation), but yet there have been no reports of TRIF-dependent pathway in TLR2 signaling, and thereby requires further research.

We tried to find the importance of the downstream molecule of TLR2-Myeloid differentiation primary response gene 88 (MyD88) in ABCA1 expression. We proved that PKC-η/PLD2 pathway is important in LPS-induced foam cell formation from previous studies [13]. PKC-η/PLD2 pathway stimulated by TLR4 agonist elevated Nox-1-induced ROS production, and thereby promoted foam cell formation. PKC-η/PLD2 pathway was also confirmed to participate in TLR2 stimulated tissue factor (TF) expression and monocyte chemotactic protein-1 (MCP-1) production (personal observation). TF and MCP-1 both are known as proteins that start and promote atherosclerosis. These results suggest that among various gene expression changes in TLR-mediated foam cell formations, some genes will be regulated through the PKC-η/PLD2 pathway.

Phospholipase is an enzyme that produces secondary messengers such as arachidonic acid, phosphatidylinositol 4, 5-bisphosphate (PIP_2), and PA, and is correlated accordingly to phospholipase A_2 (PLA_2), phospholipase C (PLC), and PLD. The discovery of PLD2 function in TLR2-mediated ABCA1 expression is very significant, and although the importance of other types of phospholipases was referred earlier, the report on the function of PLD2 function is the first to be made. In ABCA1 expression, group X secretory PLA_2 mechanism was reported by Shridas et al. [28], and cytosolic PLA_2 mechanism was also reported by Zhou et al. [26]. The similarity between the two reports is that arachidonic acid by PLA_2 suppresses liver X receptor-α activation, and thereby decreases ABCA1 expression. However, PA produced by PLD2, unlike arachidonic acid, promotes ABCA1 expression. These results may suggest that ABCA1 expression regulation may differ according to the secondary messenger produced by each phospholipase mechanisms. There were no direct evidence of PLD2 function in cholesterol efflux, but Haidar et al. [29] suggested the possibility of PLD using PA and alcoholic inhibitor. Our research provides

proof to the importance of PLD by showing the increase in ABCA1 when PA is directly treated to the cell. Raw274.7 cells have dominant expression of PLD2 over PLD1, and have significantly higher PKC-η expression among the PKC isoforms. Therefore, the PKC-η/PLD2 pathway regulating ABCA1 expression in Raw264.7 cells is a very meaningful discovery, and will be one of the new targets to regulate atherosclerosis in foam cell formation.

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References

[1] G.K. Hansson, A.K. Robertson, C. Soderberg-Naucler, Inflammation and atherosclerosis, Annu. Rev. Pathol. 1 (2006) 297–329.
[2] J.E. Cole, E. Georgiou, C. Monaco, The expression and functions of toll-like receptors in atherosclerosis, Mediators Inflamm. 2010 (2010) 393946.
[3] G. Nicolaou, C. Erridge, Toll-like receptor-dependent lipid body formation in macrophage foam cell formation, Curr. Opin. Lipidol. 21 (2010) 427–433.
[4] L.K. Curtiss, P.S. Tobias, The toll of Toll-like receptors, especially toll-like receptor 2, on murine atherosclerosis, Curr. Drug Targets 8 (2007) 1230–1238.
[5] C. Hayashi, A.G. Madrigal, X. Liu, T. Ukai, S. Goswami, C.V. Gudino, F.C. Gibson 3rd, C.A. Genco, Pathogen-mediated inflammatory atherosclerosis is mediated in part via Toll-like receptor 2-induced inflammatory responses, J. Innate. Immun. 2 (2010) 334–343.
[6] P.S. Tobias, L.K. Curtiss, Toll-like receptors in atherosclerosis, Biochem. Soc. Trans. 35 (2007) 1453–1455.
[7] A.H. Schoneveld, I. Hoefer, J.P. Sluijter, J.D. Laman, D.P. de Kleijn, G. Pasterkamp, Atherosclerotic lesion development and Toll like receptor 2 and 4 responsiveness, Atherosclerosis 197 (2008) 95–104.
[8] F. Quazi, R.S. Molday, Lipid transport by mammalian ABC proteins, Essays Biochem. 50 (2011) 265–290.
[9] B. Lammers, Y. Zhao, M. Hoekstra, R.B. Hildebrand, D. Ye, I. Meurs, T.J. Van Berkel, M. Van Eck, Augmented atherogenesis in LDL receptor deficient mice lacking both macrophage ABCA1 and ApoE, PLoS ONE 6 (2011) e26095.
[10] A.R. Tall, P. Costet, N. Wang, Regulation and mechanisms of macrophage cholesterol efflux, J. Clin. Invest. 110 (2002) 899–904.
[11] M. Van Eck, R.R. Singaraja, D. Ye, R.B. Hildebrand, E.R. James, M.R. Hayden, T.J. Van Berkel, Macrophage ATP-binding cassette transporter A1 overexpression inhibits atherosclerotic lesion progression in low-density lipoprotein receptor knockout mice, Arterioscler. Thromb. Vasc. Biol. 26 (2006) 929–934.
[12] C.W. Joyce, M.J. Amar, G. Lambert, B.L. Vaisman, B. Paigen, J. Najib-Fruchart, R.F. Hoyt Jr., E.D. Neufeld, A.T. Remaley, D.S. Fredrickson, H.B. Brewer Jr., S. Santamarina-Fojo, The ATP binding cassette transporter A1 (ABCA1) modulates the development of aortic atherosclerosis in C57BL/6 and apoE-knockout mice, Proc. Natl. Acad. Sci. USA 99 (2002) 407–412.
[13] H.K. Lee, S. Yeo, J.S. Kim, J.G. Lee, Y.S. Bae, C. Lee, S.H. Baek, Protein kinase C-eta and phospholipase D2 pathway regulates foam cell formation via regulator of G protein signaling 2, Mol. Pharmacol. 78 (2010) 478–485.
[14] J.G. Lee, E.J. Lim, D.W. Park, S.H. Lee, J.R. Kim, S.H. Baek, A combination of Lox-1 and Nox1 regulates TLR9-mediated foam cell formation, Cell. Signal. 20 (2008) 2266–2275.
[15] Y. Zhao, M. Pennings, R.B. Hildebrand, D. Ye, L. Calpe-Berdiel, R. Out, M. Kjerrulf, E. Hurt-Camejo, A.K. Groen, M. Hoekstra, W. Jessup, G. Chimini, T.J. Van Berkel, M. Van Eck, Enhanced foam cell formation, atherosclerotic lesion development, and inflammation by combined deletion of ABCA1 and SR-BI in Bone marrow-derived cells in LDL receptor knockout mice on western-type diet, Circ. Res. 107 (2010) e20–31.
[16] R. Kaplan, X. Gan, J.G. Menke, S.D. Wright, T.Q. Cai, Bacterial lipopolysaccharide induces expression of ABCA1 but not ABCG1 via an LXR-independent pathway, J. Lipid Res. 43 (2002) 952–959.
[17] I. Baranova, T. Vishnyakova, A. Bocharov, Z. Chen, A.T. Remaley, J. Stonik, T.L. Eggerman, A.P. Patterson, Lipopolysaccharide down regulates both scavenger receptor B1 and ATP binding cassette transporter A1 in RAW cells, Infect. Immun. 70 (2002) 2995–3003.
[18] X. Chen, Y. Zhao, Z. Guo, L. Zhou, E.U. Okoro, H. Yang, Transcriptional regulation of ATP-binding cassette transporter A1 expression by a novel signaling pathway, J. Biol. Chem. 286 (2011) 8917–8923.
[19] Y. Yamauchi, M. Hayashi, S. Abe-Dohmae, S. Yokoyama, Apolipoprotein A-I activates protein kinase C alpha signaling to phosphorylate and stabilize ATP binding cassette transporter A1 for the high density lipoprotein assembly, J. Biol. Chem. 278 (2003) 47890–47897.
[20] H.K. Lim, Y.A. Choi, W. Park, T. Lee, S.H. Ryu, S.Y. Kim, J.R. Kim, J.H. Kim, S.H. Baek, Phosphatidic acid regulates systemic inflammatory responses by modulating the Akt-mammalian target of rapamycin-p70 S6 kinase 1 pathway, J. Biol. Chem. 278 (2003) 45117–45127.
[21] L.B. Barreiro, M. Ben-Ali, H. Quach, G. Laval, E. Patin, J.K. Pickrell, C. Bouchier, M. Tichit, O. Neyrolles, B. Gicquel, J.R. Kidd, K.K. Kidd, A. Alcais, J. Ragimbeau, S. Pellegrini, L. Abel, J.L. Casanova, L. Quintana-Murci, Evolutionary dynamics of human Toll-like receptors and their different contributions to host defense, PLoS Genet. 5 (2009) e1000562.
[22] O. Saeed, F. Otsuka, R. Polavarapu, V. Karmali, D. Weiss, T. Davis, B. Rostad, K. Pachura, L. Adams, J. Elliott, W.R. Taylor, J. Narula, F. Kolodgie, R. Virmani, C.C. Hong, A.V. Finn, Pharmacological suppression of hepcidin increases macrophage cholesterol efflux and reduces foam cell formation and atherosclerosis, Arterioscler. Thromb. Vasc. Biol. 32 (2012) 299–307.
[23] S. Immenschuh, H. Schroder, Heme oxygenase-1 and cardiovascular disease, Histol. Histopathol. 21 (2006) 679–685.
[24] H.K. Lee, D.W. Park, J.H. Bae, H.J. Kim, D.G. Shin, J.S. Park, J.G. Lee, S.J. Lee, Y.S. Bae, S.H. Baek, RGS2 is a negative regulator of STAT3-mediated Nox1 expression, Cell. Signal. 24 (2012) 803–809.
[25] A.P. Lillis, L.B. Van Duyn, J.E. Murphy-Ullrich, D.K. Strickland, LDL receptor-related protein 1: unique tissue-specific functions revealed by selective gene knockout studies, Physiol. Rev. 88 (2008) 887–918.
[26] L. Zhou, H.Y. Choi, W.P. Li, F. Xu, J. Herz, LRP1 controls cPLA2 phosphorylation, ABCA1 expression and cellular cholesterol export, PLoS ONE 4 (2009) e6853.
[27] H.L. Brinks, A.D. Eckhart, Regulation of GPCR signaling in hypertension, Biochim. Biophys. Acta 1802 (2010) 1268–1275.
[28] P. Shridas, W.M. Bailey, K.R. Talbott, R.C. Oslund, M.H. Gelb, N.R. Webb, Group X secretory phospholipase A2 enhances TLR4 signaling in macrophages, J. Immunol. 187 (2011) 482–489.
[29] B. Haidar, S. Mott, B. Boucher, C.Y. Lee, M. Marcil, J. Genest Jr., Cellular cholesterol efflux is modulated by phospholipid-derived signaling molecules in familial HDL deficiency/Tangier disease fibroblasts, J. Lipid Res. 42 (2001) 249–257.