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Oxidized low-density lipoprotein induces secretion of interleukin-1β by macrophages via reactive oxygen species-dependent NLRP3 inflammasome activation

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ABSTRACT

Oxidized low-density lipoprotein (ox-LDL) is a critical mediator of atherogenesis. Macrophage uptake of ox-LDL and their subsequent development into foam cells is the principal event in atherosclerosis. Interleukin-1β (IL-1β), a prototypic multifunctional cytokine involved in inflammation, has an important effect on the pathogenesis and progression of atherosclerosis. Here we show that the phagocytosis of ox-LDL can induce human macrophages to secrete IL-1β by activating the NLRP3 inflammasome, and we further show that the activation of the NLRP3 inflammasome is dependent on the generation of reactive oxygen species and is related to the cathepsin B pathway. Furthermore, ox-LDL can upregulate the expression of the pro-IL-1β protein, thus priming IL-1β secretion. Therefore, our results suggest that the role of ox-LDL in atherosclerosis-related inflammation may involve the activation of the NLRP3 inflammasome.

1. Introduction

The accumulation of oxidized low-density lipoprotein (ox-LDL) in atherosclerotic lesions is a principal event in atherosclerosis. Macrophages take up ox-LDL and then become foam cells and release multiple pro-inflammatory mediators, thus contributing to atherogenesis [1–3]. Ox-LDL not only enhances atherogenesis but also drives arterial inflammation to promote the progression of atherosclerosis. Interleukin-1β (IL-1β) is considered to be the prototypic 'multifunctional' cytokine in inflammation [4,5]. Previous studies have shown that IL-1β has an important effect on the initiation and progression of atherosclerosis [6–8]. The secretion of active IL-1β is rigorously controlled by two signals: one is the transcription of the IL-1β gene and the expression of IL-1β precursor protein (pro-IL-1β), which is called priming; and the other is the activation of caspase-1 by inflammasomes, which ultimately induces the cleavage of the pro-IL-1β to the mature IL-1β protein [4,9,10]. The best characterized inflammasome is the NLRP3 inflammasome, which consists of NLRP3, the ASC adaptor, and caspase-1 [11,12]. Initial studies have shown that the NLRP3 inflammasome can be activated by host-derived molecules and exogenous molecules, including bacterial pore-forming toxins, ATP, urate crystals, and particulate matter such as a silica and asbestos [13,14]. However, the exact mechanisms that activate the NLRP3 inflammasome are currently unclear.

Although it is known that ox-LDL is the most important risk factor for atherosclerosis, the mechanism by which ox-LDL drives arterial inflammation requires further clarification. In this study, we report that the phagocytosis of ox-LDL induces IL-1β secretion by macrophages through the activation of the NLRP3 inflammasome. The ox-LDL-induced activation of the NLRP3 inflammasome depends on the generation of reactive oxygen species (ROS) and may involve the cathepsin B pathway. Moreover, ox-LDL is able to upregulate the expression of the pro-IL-1β protein, thus participating in the priming of IL-1β secretion, and this activity may be associated with the NF-κB pathway. Therefore, our results suggest that the role of ox-LDL in atherosclerosis may involve the activation of the NLRP3 inflammasome.

2. Materials and methods

2.1. Reagents

Phorbol 12-myristate 13-acetate (PMA), zYVAD-fmk, Bay11-7082, cytochalasin D, diphenyleneiodonium chloride (DPI), CA-074 Me, ATP, and ultrapure lipopoly-saccharide (LPS) were purchased from Merck. *N*-acetyl-L-cysteine (NAC) was from Sigma. Granulocyte macrophage colony-stimulating factor (GM-CSF) was purchased from Abcam. Ox-LDL was obtained from Yiyuan Biotechnology. Anti-human NLRP3 antibodies were purchased from Epitomics (3560-1), anti-human caspase-1 (D7F10 3866) and

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anti-human NF-κB p65 (#4764) antibodies were obtained from Cell Signaling. Anti-IL-1β (sc-7884) antibodies were purchased from Santa Cruz. Anti-NOX2 (ab80508) antibodies were from Abcam.

2.2. Cell culture

Human peripheral blood mononuclear cells were isolated from buffy coats by density gradient centrifugation. Cells were cultured in 6-well plates in DMEM media (Gibco) supplemented with 10% FBS as well as 100 U/ml penicillin and streptomycin at a density of 3×10^6 cells/well. After 2 h, the nonadherent cells were washed away and the medium was replaced with DMEM supplemented with 10 ng/ml granulocyte macrophage colony-stimulating factor (GM-CSF). The medium was replaced every 48 h for 6–8 days until good adherence was achieved. Human THP-1 cells were obtained from the American Type Culture Collection (ATCC). THP-1 cells were differentiated for 24 h with 100 nM PMA.

2.3. Western blot analysis

Proteins in total cell lysates were separated on a NuPAGE 6–12% Bis-Tris gradient gel and transferred to a PVDF membrane. The membrane was blocked with 5% non-fat milk in PBS-Tween buffer, incubated with PBS-Tween buffer containing the anti-human NLRP3 antibody (1:500 dilution), anti-human IL-1β antibody (1:1000), or anti-human caspase-1 antibody (1:1000) overnight at 4 °C. The membrane was incubated with the secondary antibody for 1 h at room temperature. Signals were developed using an ECL Western blotting analysis system (Invitrogen) and exposing the protein side of the membrane to X-ray film.

2.4. Confocal microscopy

Cells were added to chamber slides, incubated for 60 min with ox-LDL, and then washed with PBS and fixed using 4% paraformaldehyde for 20 min. For intracellular staining, the cells were blocked and permeabilized using PBS containing 1% BSA and 0.1% Triton X-100. Cells were incubated with rabbit anti-human NF-κB p65 antibody at 4 °C overnight and subsequently incubated with Alexa Fluor 488 goat anti-rabbit secondary antibody (Invitrogen) for 30 min. Nuclear staining with DAPI was carried out for 15 min. The cells were then covered with a cover slide and imaged with a laser confocal microscope.

2.5. ROS detection

Intracellular ROS levels were measured using a cell-permeable fluorescent probe, 2′,7′-dichlorofluorescein diacetate (DCFH-DA),

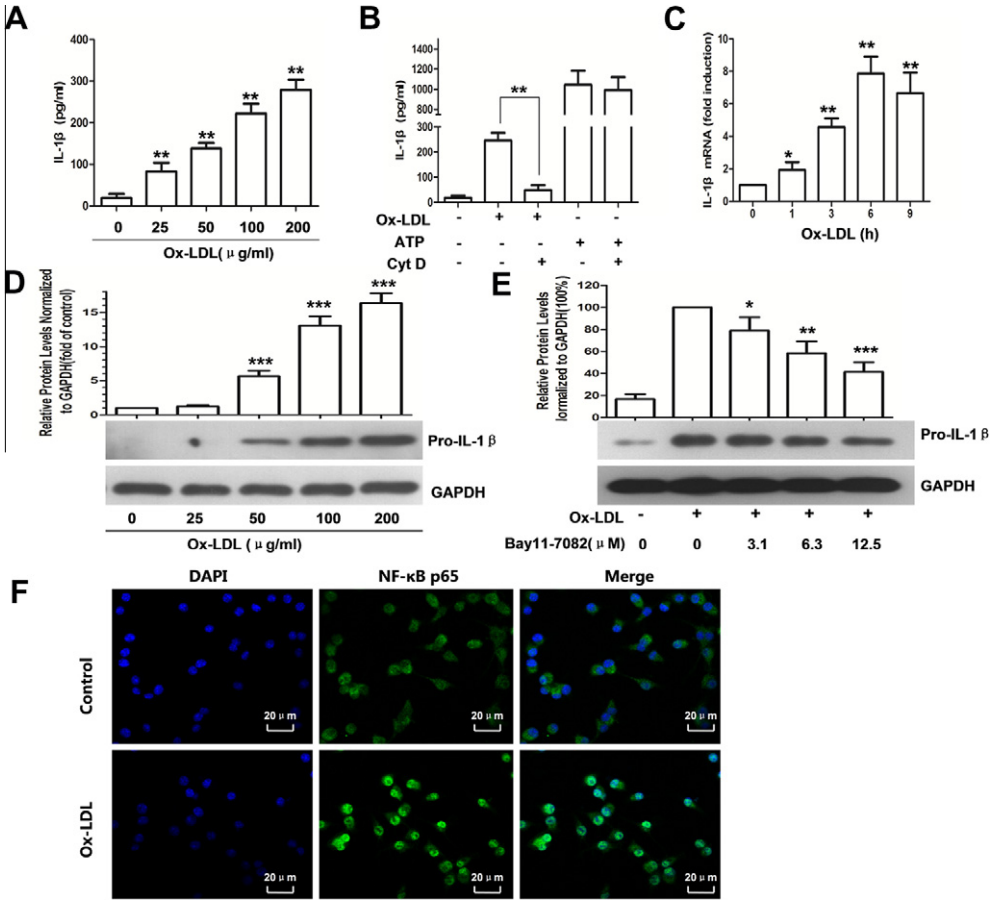

Fig. 1. Phagocytosis of ox-LDL induces IL-1β secretion. (A) ELISA measuring IL-1β release in the supernatants of human macrophages incubated with ox-LDL. (B) Human macrophages were pretreated with LPS for 2 h, followed by treatment with cytochalasin D (cyt D; 3 μM) before and during stimulation with ox-LDL (200 μg/ml) for 12 h or ATP (5 mM) for 1 h. The release of IL-1β into the supernatants was analyzed by ELISA. (C) Human macrophages were incubated with ox-LDL (200 μg/ml) for different times (0–9 h), and real-time PCR was used to analyze the expression of the IL-1β mRNA. Data are expressed relative to the control group (time 0 h). (D) Western blot analysis of pro-IL-1β expression in extracts of human macrophages incubated with ox-LDL for 12 h. Data are expressed relative to the control group (0 μg/ml of ox-LDL). (E) Human macrophages were pretreated with the NF-κB p65 inhibitor Bay11–7082 (0, 3.1, 6.3, or 12.5 μM) for 1 h, followed by incubation with ox-LDL (200 μg/ml) for 12 h. Extracts from these cells were subjected to Western blot analysis to evaluate pro-IL-1β expression. Data are expressed relative to the control group (ox-LDL (+), bay11–7082 (−)). (F) Human macrophages were incubated with ox-LDL (200 μg/ml) for 1 h, and NF-κB p65 (green) nuclear import in these cells was analyzed by confocal microscopy. The data shown are representative of three independent experiments (mean ± s.d. in A, B, C, D and E; $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.01$). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

as instructed by the manufacturer. Cells were treated with ox-LDL for 0–24 h, then incubated with fresh DCFH-DA (100 μM) in PBS for 30 min. Fluorescence was assessed using a BioTek fluorescence plate reader.

2.6. RNA interference

The siRNAs targeting human NLRP3 and control siRNA were synthesized and purified by RiboBio. The three pairs of siRNAs against NLRP3 included siRNA1 (5′-GAAAUGGAUUGAAGUGAAAdTdT-3′, 3′-dTdTCUUUACCUAACUUCACUUU-5′), siRNA2 (5′-GGAUCAAACUACUCUGUGAdTdT-3′, 3′-dTdTCCUAGUUUGAUGAGACACU-5′), and siRNA3 (5′-GGAGAGACCUUUAUGAGAAdTdT-3′, 3′-dTdTCCUCUCUGGAAAUACUCUU-5′). THP-1 cells were differentiated for 24 h with 100 nM PMA before transfection at 50–80% confluence. The siRNA transfection was performed with Lipofectamine 2000 (Invitrogen) according to the manufacturer's instructions.

2.7. Real-time quantitative PCR

Total RNA was isolated from cells using TRIzol (Invitrogen) according to the manufacturer's instructions and reverse transcribed using a PrimeScript RT reagent kit (TaKaRa) according to the instructions provided by the manufacturer. The cDNA was amplified by real-time PCR with SYBR Green (TaKaRa) and the following primers: IL-1β sense 5′-TTCAACACGCAGGACAGGTACAG-3′, IL-1β antisense 5′-CCAGGGACAGGATATGGAGCA-3′, NLRP3 sense 5′-AGCCAAGAATCCACAGTGTAACC-3′, and NLRP3 antisense 5′-AGTGTTGCCTCGCAGGTAAAG-3′. Target gene expression was assessed by the comparative cycling threshold (CT) method using the expression of the GAPDH house-keeping gene as a control.

2.8. Statistical analysis

Data are presented as the mean ± s.d. of three repeated experiments. Unpaired two-tailed Student's *t* tests were performed for all comparisons between two populations. One-way ANOVA was performed when three or more populations were compared. Values of $P < 0.05$ were considered significant.

3. Results

3.1. Phagocytosis of ox-LDL promotes IL-1β secretion

To investigate whether ox-LDL could induce the secretion of IL-1β, human monocyte-derived macrophages were incubated with ox-LDL. After incubation, the concentration of IL-1β in the supernatant was analyzed by ELISA. The results showed that ox-LDL induced the release of IL-1β in a dose-dependent manner (Fig. 1A). Foam cells, which are formed after phagocytosis of ox-LDL by macrophages, are the most important pathological sign of atherogenesis [15]. We therefore investigated whether phagocytosis was required for ox-LDL-induced release of IL-1β. Human macrophages were pretreated with LPS, followed by treatment with cytochalasin D, an inhibitor of phagocytosis, and the cells were then incubated with ox-LDL or ATP. The ox-LDL–induced release of IL-1β was reduced; however, cytochalasin D had no effect on ATP-induced secretion of IL-1β (Fig. 1B). This result suggested that phagocytosis was required for the induction of IL-1β by ox-LDL but not by ATP.

3.2. Ox-LDL participates in the priming of IL-1β secretion

The secretion of mature IL-1β requires the transcription of the IL-1β gene and the assembly of IL-1β precursor proteins; this

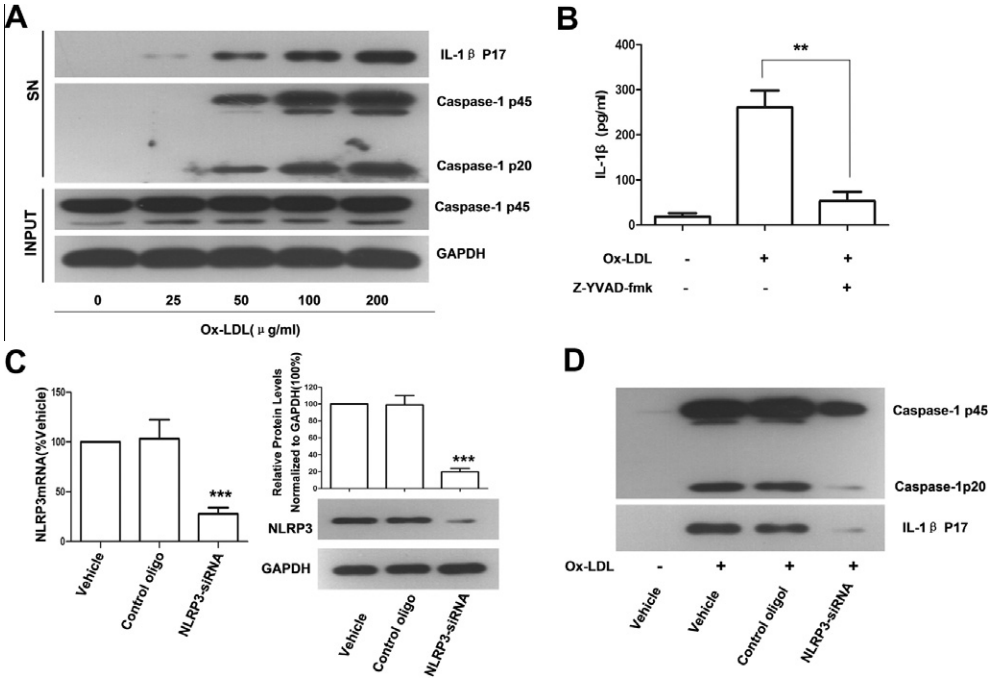

Fig. 2. Ox-LDL activates the NLRP3 inflammasome in macrophages. (A) Western blot analysis of IL-1β and caspase-1 in the supernatants (SN) and extracts (INPUT) of THP-1 macrophages incubated with ox-LDL. (B) Human macrophages were incubated with ox-LDL (200 μg/ml) in the absence or presence of the caspase-1 inhibitor zYVAD-fmk (10 mM) for 24 h, and the IL-1β in the supernatants were analyzed by ELISA. (C) THP-1 cells were differentiated for 24 h with 100 nM PMA. The cells were then transfected with NLRP3 siRNAs or a non-targeting control siRNA (control oligo) using Lipofectamine 2000 for 24 h and subsequently incubated with ox-LDL (200 μg/ml). Quantitative real-time PCR analysis of the NLRP3 mRNA levels in the cells incubated with ox-LDL for 6 h and Western blot analysis of NLRP3 protein expression in cells incubated with ox-LDL for 12 h were performed. (D) THP-1 macrophages were transfected with NLRP3 siRNAs or a non-targeting control siRNA (control oligo) and subsequently incubated with ox-LDL (200 μg/ml) for 12 h. Western blot analysis was used to measure the secretion of IL-1β and caspase-1 in the cell supernatants. Data are representative of three independent experiments (mean ± s.d. in B, C; $^{**}P < 0.01$, $^{***}P < 0.001$).

process is called priming [4]. To investigate whether ox-LDL participates in priming of IL-1β secretion, we observed the expression of the IL-1β mRNA in human macrophages. As indicated in Fig. 2A, the expression of the IL-1β mRNA was upregulated in ox-LDL–stimulated macrophages (Fig. 1C). Western blots conducted to measure the pro-IL-1β in the lysates of cells showed that ox-LDL stimulation led to pro-IL-1β protein production in a dose-dependent manner (Fig. 1D). This suggested that ox-LDL participated in the priming of IL-1β secretion by upregulating IL-1β expression.

Previous studies have shown that LPS promotes the expression of IL-1β via the NF-κB pathway [16]. To further study the mechanism by which ox-LDL promotes the expression of IL-1β, we observed the effects on NF-κB p65 nuclear import in human macrophages treated with ox-LDL using confocal microscopy. After incubating macrophages with ox-LDL, nuclear NF-κB p65 expression was clearly enhanced (Fig. 1F). We further pretreated macrophages with the NF-κB inhibitor Bay11-7082, and this was followed by treatment with ox-LDL. As shown in Fig 1E, the expression of pro-IL-1β was downregulated in a dose-dependent manner (Fig. 1E). This suggested that the priming of IL-1β secretion induced by ox-LDL might be mediated by the NF-κB pathway.

3.3. Ox-LDL–induced secretion of IL-1β is caspase-1 dependent

After caspase-1 inflammasome complex assembly is triggered, pro-caspase-1 is cleaved to active caspase-1, which induces the secretion of IL-1β. To investigate whether active caspase-1 is essential for ox-LDL–induced IL-1β release, we first measured the ox-LDL–induced cleavage of caspase-1 in THP-1 macrophages. As shown in Fig. 2A, ox-LDL dramatically induced caspase-1 p20 (cleaved caspase-1) secretion in a dose-dependent manner, while the levels of the caspase-1 precursor were not obviously altered. ELISA showed that ox-LDL–induced IL-1β secretion in THP-1 macrophages could be suppressed by a caspase-1–specific inhibitor (z-YVAD-fmk; Fig. 2B). These results suggested that ox-LDL could stimulate IL-1β secretion and that this stimulation was dependent on the cleavage and activation of caspase-1 precursor proteins.

3.4. Ox-LDL induces IL-β secretion by activating the NLRP3 inflammasome

Ox-LDL significantly increased the secretion of cleaved caspase-1 and IL-1β, suggesting that ox-LDL could activate inflammasomes. The NLRP3 inflammasome is a typical inflammasome [11,12]. To further study the role of the NLRP3 inflammasome in the ox-LDL–induced secretion of IL-1β, we depleted NLRP3 expression in THP-1 macrophages using an NRLP3-specific siRNA. Western blot analysis confirmed that NLPR3 protein expression was obviously downregulated (Fig. 2C). Importantly, the ox-LDL–induced caspase-1 activation and IL-1β secretion was significantly attenuated by silencing of NLRP3 (Fig. 2D). These data suggested that ox-LDL could induce IL-1β secretion by activating the NLRP3 inflammasome.

3.5. Activation of the NLRP3 inflammasome induced by ox-LDL is ROS dependent

Many endogenous and exogenous inflammasome activators can promote ROS generation [11,17,18]. Thus, we designed an experiment to investigate the role of ROS in ox-LDL–induced inflammasome activation. Human monocyte-derived macrophages were incubated with ox-LDL, and cellular ROS was then assessed using an ROS-specific fluorescent probe (DCFH-DA) and a BioTek plate reader. The results showed that ROS increased dramatically after stimulation with ox-LDL (Fig. 3A). When THP-1 macrophages were

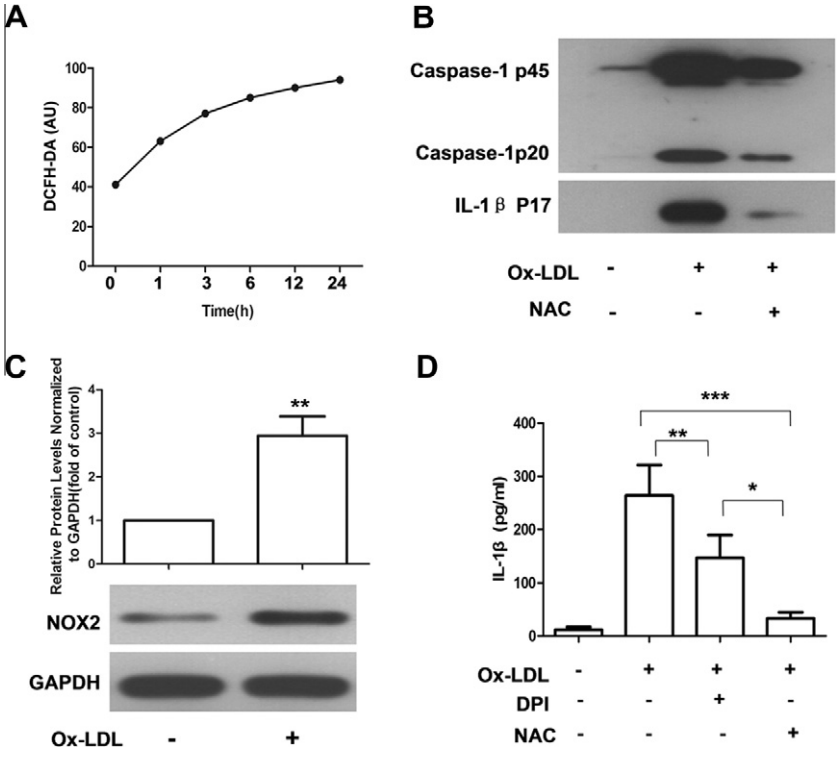

Fig. 3. The activation of the NLRP3 inflammasome induced by ox-LDL requires the presence of ROS. (A) ROS generation in human macrophages at various times after incubation with ox-LDL (200 μg/ml), as assessed with the ROS-specific fluorescent probe DCFH-DA (AU, arbitrary units). (B) Western blot analysis of IL-1β and caspase-1 secretion in THP-1 macrophages stimulated with ox-LDL (200 μg/ml) for 24 h in the presence or absence of NAC (10 mM). (C) Human macrophages were incubated with ox-LDL (200 μg/ml) for 12 h. Western blot analysis of NOX2 in ox-LDL stimulated human macrophages. (D) Human macrophages were incubated with DPI (25 μM) or NAC (10 mM) before and during stimulation with ox-LDL (200 μg/ml) for 12 h. ELISA analysis of the secretion of IL-1β in supernatant. The data shown are representative of three independent experiments (mean ± s.d. in C, D; $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$).

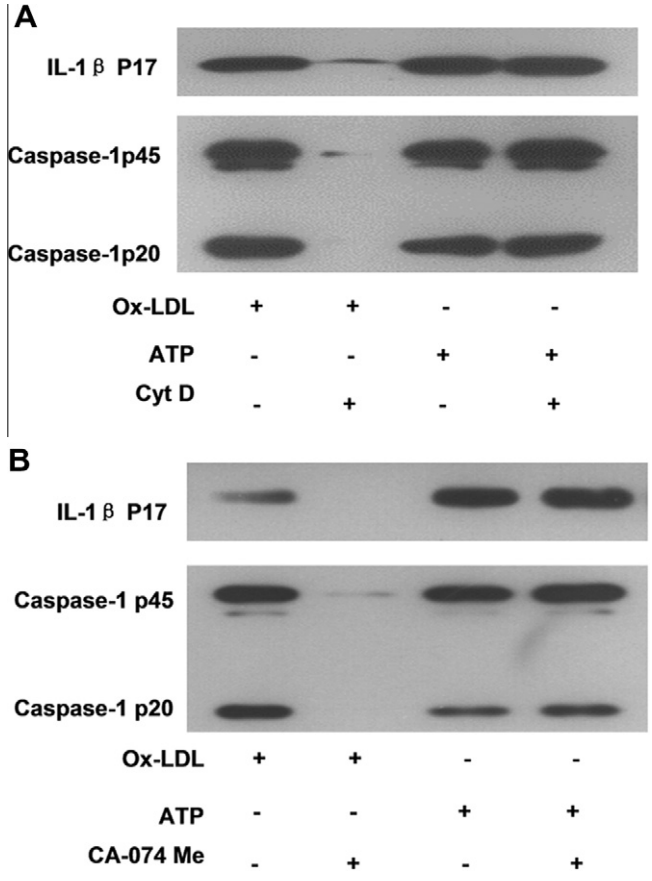

Fig. 4. IL-1β secretion induced by ox-LDL via NLRP3 activation is related to cathepsin B. (A) THP-1 macrophages were incubated with cytochalasin D (cyt D; 3 μM) before and during stimulation with ox-LDL (200 μg/ml) for 12 h or with ATP (5 mM) for 1 h, Western blot analysis of IL-1β and caspase-1 secretion in the supernatants of cells. (B) THP-1 macrophages were incubated with the cathepsin B inhibitor CA-074 Me before and during stimulation with ox-LDL (200 μg/ml) for 12 h or ATP (5 mM) for 1 h, and Western blot analysis was used to determine the secretion of IL-1β and caspase-1 in the supernatants of cells. The data shown are representative of three independent experiments.

incubated with a direct scavenger of ROS, *N*-acetyl-L-cysteine (NAC), both before and during stimulation with ox-LDL, the ox-LDL–induced secretion of IL-1β and cleaved caspase-1 was clearly reduced (Fig. 3B). This suggested that the activation of inflammasomes induced by ox-LDL required the ROS pathway. The sources contributing to ROS generation in macrophages are several, including NADPH oxidase. To clarify whether NADPH oxidase-derived ROS was involved the ox-LDL–induced activation of the NLRP3 inflammasome, human monocyte-derived macrophages were incubated with ox-LDL for 12 h. Subsequent Western blot analysis showed that the expression of NADPH oxidase 2 (NOX2) was upregulated (Fig. 3C); however, the IL-1β secretion from ox-LDL–stimulated macrophages was only partially inhibited by the NADPH oxidase inhibitor DPI. When the macrophages were pretreated with NAC, IL-1β secretion was almost completely suppressed (Fig. 3D). These results indicated that, in addition to NADPH oxidase-derived ROS, there must be other sources of ROS involve the ox-LDL–induced activation of the NLRP3 inflammasome.

3.6. IL-1β secretion is induced by ox-LDL via NLRP3 activation related to cathepsin B

Phagocytosis is essential for the activation of the NLRP3 inflammasome by crystalline [17], and our study also found that phagocytosis was required for the induction of IL-1β by ox-LDL (Fig. 1B). THP-1 macrophages were pretreated with cytochalasin D, followed by treatment with ox-LDL for 12 h or ATP for 1 h. Western blot analysis showed that release of both IL-1β and cleaved caspase-1 induced by ox-LDL were inhibited, but the ATP-induced secretion of these factors was not affected. These results suggested that phagocytosis was required for the ox-LDL–induced caspase-1–dependent release of IL-1β (Fig. 4A). Previous studies suggest that the phagocytosis of ox-LDL by macrophages can induce the release of cathepsin B by destroying lysosomes [15,19]. To observe whether the activation of the NLRP3 inflammasome by ox-LDL involved cathepsin B, THP-1 macrophages were incubated with a cathepsin B inhibitor (CA-074 Me) before and during stimulation with ox-LDL or ATP. The secretion of IL-1β induced by ox-LDL was reduced under these conditions. However, the cathepsin B inhibitor did not inhibit IL-1β release when THP-1 macrophages were stimulated with ATP (Fig. 4B). This indicated that the secretion of IL-1β induced by ox-LDL through NLRP3 activation was related to the cathepsin B pathway.

4. Discussion

IL-1β has an important effect on the pathogenesis and progression of atherosclerosis [6–8]. Our study showed that ox-LDL could promote the secretion of IL-1β through the activation of the NLRP3 inflammasome. Previous studies have demonstrated that ox-LDL can activate the expression of certain genes, which may further promote inflammation during atherosclerosis by binding to certain receptors as ligands [1,2,20]. Here, we found that the secretion of IL-1β by macrophages was dramatically increased by ox-LDL stimulation in a dose-dependent manner. Our data showed that the secretion of IL-1β was suppressed by cytochalasin D, an inhibitor of phagocytosis; however, cytochalasin D had no effect on the secretion of IL-1β induced by ATP. This observation suggested that the phagocytosis of ox-LDL was necessary for macrophages to secrete IL-1β. The secretion of active IL-1β requires the transcription of the IL-1β gene and the assembly of IL-1β precursor proteins, also known as priming [4]. Our study showed that ox-LDL could upregulate the expression of IL-1β to participate in the priming of IL-1β secretion. Previous studies have shown that LPS or DMSO promotes the expression of IL-1β through the NF-κB pathway [21–23]. In our study, we confirmed that ox-LDL could promote the nuclear import of NF-κB p65 in human macrophages. Meanwhile, the ox-LDL–induced expression of the IL-1β precursor protein could be inhibited by the NF-κB–specific inhibitor Bay11-7082. All of these data suggested that ox-LDL promoted the expression of the IL-1β precursor protein through the NF-κB pathway.

The secretion of IL-1β is controlled by the caspase-1 inflammasome. After caspase-1 inflammasome complex assembly is triggered, pro-caspase-1 is cleaved to active caspase-1 and the secretion of IL-1β is induced. Here, we found that the secretion of cleaved caspase-1 p20 by THP-1 macrophages was strongly increased after ox-LDL stimulation. Moreover, the secretion of IL-1β could be inhibited by z-YVAD-fmk, a caspase-1–specific inhibitor. This indicated that ox-LDL promoted the cleavage of the caspase-1 precursor protein to generate cleaved caspase-1 and ultimately induced the cleavage of the precursor pro-IL-1β to mature IL-1β. The NLRP3 inflammasome is a well-characterized inflammasome that consists of NLRP3, the ASC adaptor, and caspase-1 [11,12]. NLRP3 is the core constituent of this complex and it can recognize endogenous or exogenous damage signals to induce the assembly of the inflammasome complex [12]. To investigate whether ox-LDL could activate the NLRP3 inflammasome, we knocked down NLRP3 expression in THP-1 macrophages using siRNA. When NLRP3 was knocked down, the secretion of IL-1β induced by ox-LDL was eliminated. This demonstrated that ox-LDL could activate

the NLRP3 inflammasome to promote caspase-1–dependent secretion of IL-1β.

Previous studies have shown that ROS are generated by many NLRP3 agonists [11,17,18]. In our study, ox-LDL enhanced the accumulation of ROS in human macrophages. Meanwhile, the secretion of IL-1β induced by ox-LDL was obviously inhibited by the ROS inhibitor NAC. These results demonstrated that ox-LDL–induced activation of the NLRP3 inflammasome was dependent on the generation of ROS. NADPH oxidase is the main source of ROS generation in macrophages. Our data showed that the expression of NADPH oxidase 2 (NOX2) was upregulated by ox-LDL. Although IL-1β secretion was almost completely suppressed when the macrophages were pretreated with NAC, ox-LDL–induced IL-1β secretion was, however, only partially inhibited by the NADPH oxidase inhibitor. This suggested that, in addition to NADPH oxidase-derived ROS, there must be other sources of ROS involve the ox-LDL–induced activation of the NLRP3 inflammasome. As such, the underlying mechanism of ROS generation in the ox-LDL–induced activation of the NLRP3 inflammasome requires further clarification.

Phagocytosis is essential for the activation of the NLRP3 inflammasome by crystalline or amyloid-β [14,17,19,24]. In our study, we found that phagocytosis was also necessary for the ox-LDL–induced activation of the NLRP3 inflammasome. Additionally, the secretion of cleaved caspase-1 by ox-LDL–stimulated macrophages was suppressed by cytochalasin D. It has been demonstrated that the destruction of the lysosome can induce the release of lysosomal enzymes such as cathepsin B, which activates the NLRP3 inflammasome [14,24,25]. Previous studies have shown that the phagocytosis of ox-LDL by macrophages can induce the release of cathepsin B by destroying the lysosome [15,19]. We found that the secretion of IL-1β induced by ox-LDL was reduced after macrophages were pretreated with the cathepsin B inhibitor CA-074 Me. The release of cleaved caspase-1 by ox-LDL was also inhibited by CA-074 Me. These results indicated that the ox-LDL–induced activation of the NLRP3 inflammasome may involve the cathepsin B pathway.

In this article, we suggested that the phagocytosis of ox-LDL could promote the release of IL-1β by activating the NLRP3 inflammasome, which was dependent on ROS and relate to the cathepsin B pathway. Moreover, ox-LDL may participate in the priming of IL-1β secretion. Therefore, the role of ox-LDL in inflammation associated with atherosclerosis may involve the activation of the NLRP3 inflammasome.

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