Available online at www.sciencedirect.com
ScienceDirect
Biochemical and Biophysical Research Communications 352 (2007) 244–250
BBRC
www.elsevier.com/locate/ybbrc

Discoidin domain receptor 2 is involved in the activation of bone marrow-derived dendritic cells caused by type I collagen

Ji-Eun Lee [a], Chon-Sik Kang [a], Xiu-Ying Guan [a], Beom-Tae Kim [b], Sang-Hyun Kim [c], Young-Mi Lee [d], Woo-Sung Moon [e,*], Dae-Ki Kim [a,*]

[a] *Department of Immunology, Chonbuk National University Medical School, Jeonbuk 561-180, Republic of Korea*
[b] *Research Center of Bioactive Materials, College of Natural Science, Chonbuk National University, Jeonbuk 561-756, Republic of Korea*
[c] *Department of Pharmacology, Kyungpook National University Medical School, Taegu 700-422, Republic of Korea*
[d] *Department of Oriental Pharmacy, College of Pharmacy, Wonkwang University, Iksan, Jeonbuk 570-749, Republic of Korea*
[e] *Department of Pathology, Chonbuk National University Medical School, Jeonbuk 561-180, Republic of Korea*

Received 3 November 2006
Available online 13 November 2006

Abstract

Discoidin domain receptors (DDRs), DDR1 and DDR2, are non-integrin receptor tyrosine kinases for collagen in many cell types. In this study, we investigated the contributions of DDRs to the activation of mouse bone marrow-derived dendritic cells (DCs) by type I collagen (ColI). Our data showed that transcript and protein of DDR2 were expressed constitutively in immature DCs and upregulated in TNF-α-stimulated mature DCs. ColI treatment induced DDR2 phosphorylation and subsequently induced the upregulation of IL-12 production, CD86 expression, and antigen uptake activity by immature DCs. Depletion of DDR2 by specific siRNA attenuated significantly an increase in expression of IL-12 and CD86 in ColI-treated DCs. Additionally, DDR2–ColI interaction upregulated the ability of mature DCs to activate allogeneic T cells. These findings suggest that DDR2 is a critical collagen receptor for DC activation and that DDR2–collagen interaction plays an important role in the functional capacity of DCs regulating immune responses.

Keywords: Dendritic cells; Collagen; Discoidin domain receptor-2; Maturation

Dendritic cells (DCs) are the most central player in the initiation and direction of immune responses as effective antigen-presenting cells (APCs). In general, DCs have been found in the immature state in epithelia and interstitial space of most tissues. In the presence of tissue damage, inflammatory cytokines or pathogens, immature DCs start a differentiation process called maturation and than migrate to lymph nodes, to present the encountered antigen to T cells, thereby inducing a primary or secondary immune response [1–3]. Hallmarks of this maturation process to induce T cell responses are the upregulation of cell surface major histocompatibility complex class I and II and co-stimulatory molecules, such as CD80, CD86, and CD83. Several cytokines, including IL-12, TNF-α, and IL-10 are released by DCs during maturation [4–6]. In most tissues, DCs are present in immature state that lacks the requisite accessory signals for T cell activation [7]. Thus, understanding of DC maturation signals is important for immunotherapeutic protocols of a variety of immune diseases. The inflammatory stimuli, including TNF-α, IL-1β, and lipopolysaccharide (LPS), have been known to enhance antigen presenting ability of T cells in the maturation process of DCs and upregulate production of cytokines, including IL-12 [8–10]. In addition, various factors, such as CD40 ligand, CpG motifs in bacterial DNA, haptens, and apoptotic cells, also have been known to promote the expression of MHC and costimulatory molecules and are responsible for DC maturation [11–14].

* Corresponding authors. Fax: +82 63 270 3135.
E-mail addresses: daekim@chonbuk.ac.kr (D.-K. Kim), mws@chonbuk.ac.kr (W.-S. Moon).

0006-291X/$ - see front matter © 2006 Elsevier Inc. All rights reserved.
doi:10.1016/j.bbrc.2006.11.010

Adhesion molecules play an important role in DC adhesion and migration through receptors [15]. Connective tissue DCs accumulate at sites of extracellular matrix (ECM) deposition and exhibit the binding of ECMs, such as fibronectin, laminin, and collagen through members of integrins [16,17]. Several studies have shown that collagen accumulated at the inflammatory areas may play a critical role in regulating the function of DCs. Especially, the engagement of type I collagen (ColI) promotes the expression of co-stimulatory molecules and secretion of cytokines in murine and human DCs [18,19]. However, the mechanism of collagen-mediated DC maturation remains to be understood. Although, bone marrow-derived DCs express a typical collagen receptor VLA-2, their maturation by ColI is not inhibited by blocking antibody against β1-integins [20,21], suggesting that the activating effect of collagen is mediated by the different receptors. Recently, Discoidin domain receptors (DDRs), DDR1 and DDR2, have been identified as a novel family of tyrosine kinase receptors for collagen [22]. These receptors are expressed in early embryonic development and many adult tissues. Both receptors are activated by fibrillar collagens (types I, II, and III), but DDR1 can be activated by nonfibrillar collagens (type IV). DDR1 has been found in lung, kidney, breast, and brain tissue, while DDR2 is expressed at highest levels in skeletal muscle, skin, kidney, and lung tissue. DDR1 appears to be a key regulator of cell morphogenesis, differentiation, and proliferation in several organs [23], and DDR2 is involved in ECM remodeling during morphogenesis and tissue repair [24]. However, the DC activation event mediated by DDRs is not well-understood.

In the present study, we investigated whether collagen receptors DDR1 and DDR2 are involved in ColI-mediated maturation of mouse bone marrow-derived DCs.

Materials and methods

Animals and reagents. Female 8- to 10-week-old C57BL/6 mice (H-2K^b and I-A^b) were obtained from the Damool Science Animal Facility (Gyeonggi, Korea). Recombinant murine GM-CSF, TNF-α, and IL-4 were purchased from Biosource (Camarillo, CA). Purified anti-mouse β1 mAb (Ha2/5) and control IgM were from BD Pharmingen (San Diego, CA) and anti-phospho-tyrosine mAb (p-Tyr-100) from Cell Signaling (Beverly, MA). Anti-DDR1 (C-20), anti-DDR2 (C-19), DDR2-specific siRNA, and scrambled control siRNA were from Santa Cruz Biotechnology (Santa Cruz, CA). All reagents were from Sigma Co. (St. Louis, MO).

Generation of bone marrow-derived DCs. Dendritic cells (DCs) were established from the bone marrow of C57BL/6 mice as described by Inaba et al. [25]. Briefly, bone marrow cells were harvested from the femur and tibia of sacrificed mice, depleted erythrocytes with 0.14 M NH_4Cl buffer (pH 7.2), and then laid carefully on two volumes of Opti-Prep solution (Oxis-Shied, Oslo, Norway). After centrifugation (500*g*) for 5 min, the cells collected from the interface were cultured in RPMI1640 (Gibco, Carlsbad, CA) containing 10% FBS, 50 μM of 2-mercaptoethanol, 10 ng/ml GM-CSF, and 10 ng/ml IL-4. Medium exchange was carried out every 2 days by gently swirling the plates. On day 6, nonadherent and loosely adherent cells consisting of immature DCs were CD11c positive (>90%). DCs were incubated with serum-free medium containing 10 ng/ml rmGM-CSF, on ColI-coated or non-coated plates (10.5 μg/cm^2) until the indicated time points. For DDR2 depletion, DDR2-specific siRNA (2 μg) was transfected into DCs (1×10^6) using Effectene transfection reagent (Qiagen, Valencia, CA) according to the manufacturer's protocol. The scrambled siRNA was used as a negative control. Depletion of target protein in transfectants was analyzed by Western blot analysis.

ELISA of cytokines. Levels of IL-12 (p40/p70), IL-2, and IFN-γ were measured using a commercial ELISA kit (Biosource) according to the manufacturer's instructions. Briefly, the capture Ab-coated plates were blocked with ELISA buffer containing 50 mM Tris–HCl (pH 7.2), 0.5% BSA, 2 mM EDTA, 150 mM NaCl, 0.05% Tween 20 for 1 h. Samples (50 μl) were added into ColI-coated microtiter plates and incubated overnight at 4 °C. Plates were stained with biotinylated detection antibody and streptavidin–HRP for 1 h, washed with ELISA buffer and then TBA substrate, and 0.04% H_2O_2 in PBS were added to the plates. Reaction was stopped by adding 4 M H_2SO_4 and then optical density at 405 nm was measured using a microplate reader.

RT-PCR. Total RNA was extracted using TRIzol reagent (Invitrogen, Carlsbad, CA). For cDNA synthesis, RNA (2 μg) was reverse-transcribed in a reaction buffer containing oligo(dT) and MMLV reverse transcriptase (iNtRON, Korea) for 1 h at 42 °C. Specific primers were synthesized as follows: DDR1 forward 5′-ACCAACTTCAGCAGCTTGGAGCTG-3′ and reverse 5′-atcttgcgggtcctctacttca-3′ (618 bp of DDR1a, 729 bp of DDR1b); DDR2 forward 5′-CAAGGACCCAAACATCATCC-3′ and reverse 5′-GATGCATCATCACTCGGCTCC-3′ (619 bp); integrin α2 forward 5′-AGAACCCACTCCTGTATCTGAC and reverse 5′-5′-GA GTTCTGTGGTCTCATCCATC (520 bp); GAPDH forward 5′-GAAG GGCTCATGACCACAGTCCATG-3′ and reverse 5′-tgttgctgtagccgtatt cattgtc-3′ (450 bp). PCR amplification was performed as follows: denaturation (94 °C, 30 s), annealing (60 °C, 30 s), and extension (72 °C, 50 s). Final PCR products were separated on a 1.2%-agarose gel and photographed.

FACS analysis. Cells (1×10^5) were washed in PBS containing 0.1% sodium azide and incubated with anti-FcγRIIb (2.4G2) to block nonspecific binding of IgG on ice for 10 min and then with specific mAb (BioLegend, San Diego, CA) on ice for 30 min. FITC- or PE-conjugated mAbs used to detect I/A^b (AF6-120), CD80 (16-10A1), CD86 (GL-1), and CD11c (N418). Cells were also stained with an appropriate isotype-matched Ig as negative control. The cells (1×10^5) were analyzed using the FACS Calibur (Becton–Dickinson, San Diego, CA). Data were presented as histograms and mean-fluorescence intensity (MFI).

Western blot analysis. Cells were lysed in ice-cold lysis buffer (iNtRON Biotech, Korea) for 20 min and centrifugated (12,000*g*) for 20 min. The protein concentrations in cell extracts were measured using a bicinchoninic acid. Lysates (25 μg/lane) were separated by 8% SDS–PAGE and transferred to PVDF membranes. After blocking with 5% skimmed milk for 1 h, the membranes were incubated with anti-DDR2 or anti-p-Tyr Ab at 4 °C, washed in TBS containing 0.1% Tween 20, and incubated with HRP-conjugated secondary antibody for 45 min. Finally, proteins were visualized using an enhanced chemiluminescence WB detection kit (Amersham Pharmacia Biotech).

Antigen uptake activity. For measurement of antigen uptake activity of immature DCs, dextran–FITC was used as antigen. Cells (1×10^5) seeded on ColI-coated or non-coated plates were treated with dextran–FITC (100 μg/ml) for 90 min at either 37 °C or 4 °C, as negative control of endocytosis. The excess dextran–FITC was washed with ice-cold washing buffer (PBS containing 0.1% sodium azide) and the cells were stained with PE-conjugated anti-CD11c for 20 min on ice. The stained cells were washed and then analyzed using FACS Calibur. Data were presented as the mean-fluorescence intensity (MFI) ± SEM of triplicate.

Mixed lymphocyte reaction (MLR). For allogeneic MLR, CD3$^+$ responder T cells (purity >95%) were purified from splenocytes of C57BL/6 mice by MACS column (Miltenyi Biotec, Germany) according to manufacturer's instruction. Immature DCs were cultured on ColI-coated plates for 48 h. Allogeneic T cells (1×10^5) were placed in 96-well plates with ColI-treated DCs (10,000 cells) as stimulators. On day 4, 2 μCi/well [^{3}H] thymidine was added and incubated for 16 h and incorporation of radioactivity was measured using liquid scintillation counter (TRI-CARB 2300TR, Packard Co., Meriden, CT). To measure the release of IL-2 and IFN-γ by activated T cells, the levels released after co-culture for 3 days

were analyzed using ELISA kit. To examine the effect of DDR2 depletion in MLR, DCs transfected with DDR2-siRNA and incubated for 48 h, were used as stimulators. All data are presented as means of triplicate.

Statistical analysis. All values are presented as means ± SEM. Statistical significance was determined using the Student's *t*-test. All *p*-values <0.05 were considered to reflect a statistically significant difference.

Results

Collagen induces DDR2 phosphorylation in BM-derived DCs

We first examined whether BM-derived DCs express DDR1 and DDR2 by RT-PCR. As shown in Fig. 1A, during DC development from BM cells, transcripts of DDR2 and integrin α2 were detected after 30 cycles of PCR amplification, while DDR1 transcripts (DDR1a and b) were after 35 cycles. Moreover, the level of DDR2 transcript was upregulated in DCs stimulated with TNF-α, whereas transcripts of integrin α2 and DDR1 were rapidly downregulated (Fig. 1B). We also found that BM-derived DCs express DDR2 protein and the level of DDR2 protein was upregulated in immature DCs stimulated with TNF-α (Fig. 1C), whereas DDR1 proteins were not detected in both immature and activated DCs (data not shown). Next, to examine DDR2 activation by ColI, immature DCs were incubated on ColI-coated culture plates. As shown Fig. 1D, ColI induced DDR2 phosphorylation in DCs and the level of DDR2 phosphorylation reached a peak at 1 h. These results suggest that DDR2 is a collagen receptor expressing in both immature and mature DCs.

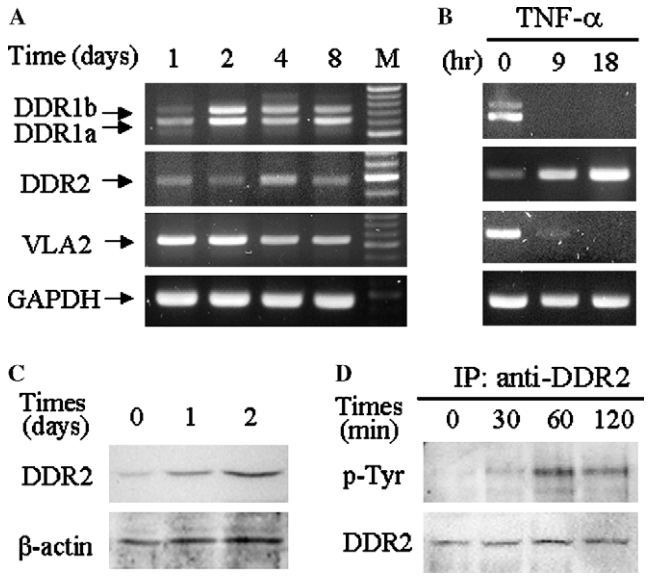

Fig. 1. Expression and phosphorylation of DDR2 collagen receptor on BM-derived DCs and TNF-α-stimulated DCs. (A) BM cells were incubated in the presence of GM-CSF and IL-4 during development of DCs. (B) Immature DCs established by culturing for 6 days were stimulated with TNF-α (20 ng/ml) for 9 and 18 h. Total RNA was extracted to analyze transcripts of DDR1, DDR2, and VLA2 (integrin α2) by RT-PCR. GAPDH was analyzed as constitutive control. (C) Immature DCs were treated with TNF-α for 1 and 2 days. Cell lysates were separated by SDS–PAGE and the level of DDR2 proteins was analyzed by Western blot analysis. (D) DCs were incubated on ColI-coated plates for indicating times (minute). Cell lysates were immunoprecipitated with anti-DDR2 Ab and immunoblotted with anti-phospho-Tyr mAb (P-Tyr-100) or anti-DDR2 Ab to analyze DDR2 phosphorylation.

Collagen enhances IL-12 production by DCs via DDR2

Since ColI has been known to promote cytokine production by BM-derived immature DCs [26], we examined the contribution of DDR2 to IL-12 production by DCs. TNF-α was used as an activating control to activate DCs [27]. To eliminate bacterial LPS contamination, ColI-coated plates were treated with polymycin B for 2 h. As shown in Fig. 2A, ColI treatment increased significantly IL-12 production by immature DCs and the level of IL-12 reached a peak at 16 h. The level of IL-12 produced by ColI was 2-fold higher than that of TNF-α and the combination of TNF-α and ColI upregulated synergistically IL-12 production by DCs. Moreover, ColI-induced upregulation of IL-12 was also observed in DCs maturated by TNF-α (Fig. 2B). To examine whether DDR2 is involved in ColI-induced IL-12 production, DDR2-specific siRNA or control siRNA was transfected into DCs to deplete DDR2. However, we found that most protocols for siRNA transfection caused the maturation of DCs. Therefore, the mature DCs induced by siRNA transfection were used for the following experiments. ColI-induced IL-12 production was attenuated significantly in DCs transfected with DDR2-siRNA, but not in control siRNA (Fig. 2C). In addition, ColI-induced IL-12 production was not affected by pretreatment of blocking antibody against integrins (Fig. 2D). These results indicate that ColI-induced DC activation is mediated through DDR2, but not through integrins.

Collagen enhances the expression of CD86 by DCs via DDR2

The costimulatory molecules, such as MHC II, CD80, and CD86, have been known to be associated with maturation of DCs. We examined the role of DDR2 on the cell surface phenotype of DCs stimulated with ColI. The expression of costimulatory molecules was analyzed in CD11c high-expressing DCs by flow cytometry. We confirmed that MHC II, CD80 and CD86 were expressed on the surface of DCs in response to ColI (data not shown). Interestingly, the expression of CD86, but not CD80, and MHC II, was upregulated significantly in response to ColI compared to that of TNF-α as activating control (Fig. 3A). To examine whether DDR2–ColI interaction regulates CD86 expression, DDR2 siRNA or control siRNA was transfected into DCs. Although siRNA transfection procedure resulted in the maturation of DCs, the level of CD86 was upregulated by ColI. Moreover, ColI-induced expression of CD86 was attenuated significantly by DDR2 depletion, but not by tansfection of control siRNA (Fig. 3B). These results indicate that DDR2 is involved in upregulation of CD86, a costimulatory molecule, in ColI-stimulated DCs.

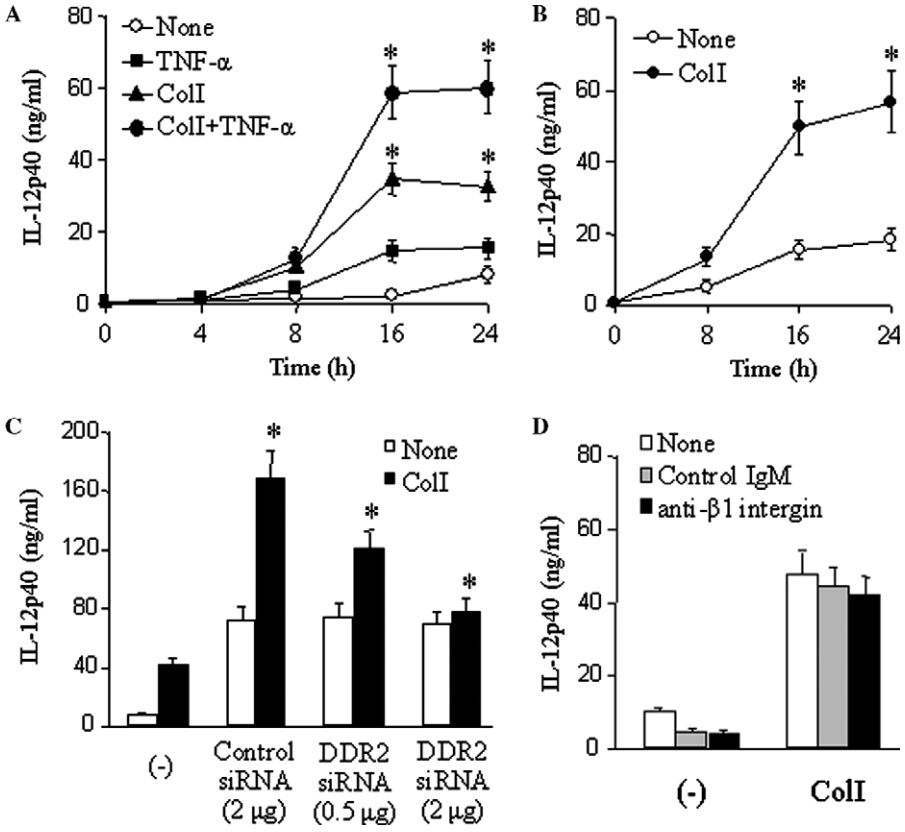

Fig. 2. Enhanced IL-12 production in ColI-stimulated DCs via DDR2. (A) BM-derived immature DCs (5×10^4 cells/well) were stimulated with ColI, TNF-α (20 ng/ml) or ColI plus TNF-α in a time-dependent manner. (B) To examine the effect of collagen on mature DCs, immature DCs were stimulated with TNF-α for 48 h. The activated cells were treated with ColI in a time-dependent manner. (C) For DDR2 depletion, DDR2-siRNA or control siRNA was transfected into immature DCs. After 48 h, cells were treated with ColI for 16 h. (D) DCs were pretreated with anti-β1 integrins mAb or control IgM for 10 min and then stimulated with ColI for 16 h. All supernatants were collected and levels of IL-12 (p40/p70) were measured by ELISA. Each data represents means ± SEM of three independent experiments ($^*p < 0.05$).

Collagen enhances the antigen uptake activity by DCs

To examine whether ColI regulates the antigen uptake activity of DCs, immature DCs were cultured on ColI-coated or non-coated plates in the presence of dextran–FITC for 90 min at 37 °C or 4 °C as a negative control of endocytosis. Dextran–FITC uptake activity of DCs was upregulated by treatment of ColI (Fig. 3C) at 37 °C, but not at 4 °C. We next analyzed ColI-induced antigen uptake activity of DDR2-depleted DCs. However, processing for siRNA transfection caused DC maturation and attenuated Dextran–FITC uptake of DCs. Thus, ColI treatment did not enhance antigen uptake activity of siRNA transfectants (data not shown). Although we could not found a direct involvement of DDR2 in regulating antigen uptake of immature DCs, our results support that DDR2–ColI interaction might enhance the antigen uptake activity of DDR2-expressing immature DCs.

Collagen enhances the allogeneic T cell reaction by DCs via DDR2

It has been known that antigen uptake activity of DCs regulates their potential ability to stimulate T cells through antigen presentation. We therefore examined the effect of DDR2–ColI interation on the capacity of DCs to stimulate naive T cells. DCs stimulated with ColI for 48 h were cocultured with allospecific CD3 + T cells purified from splenocytes for 3 days. As reported previously, ColI-stimulated DCs prominently enhanced proliferation and cytokine release of allogeneic T cells [18]. Although processing for siRNA transfection caused DC maturation and depletion of DDR2 significantly reduced ColI-induced ability of DCs upregulating allogeneic T cell proliferation (Fig. 4A). In addition, ColI-treated DCs augmented the production of IL-2 and IFN-γ by allogenic T cells, and these abilities also were significantly attenuated by DDR2 depletion (Fig. 4B and C). These results indicate that ColI might upregulate T cell activation through increasing the functional activity of DCs via DDR2.

Discussion

In the present study, we demonstrated that a novel collagen receptor DDR2 plays a critical role in the activation of DCs stimulated with ColI. Our data showed that DDR2 was expressed constitutively in BM-derived immature DCs and upregulated prominently in TNF-α-stimulated mature

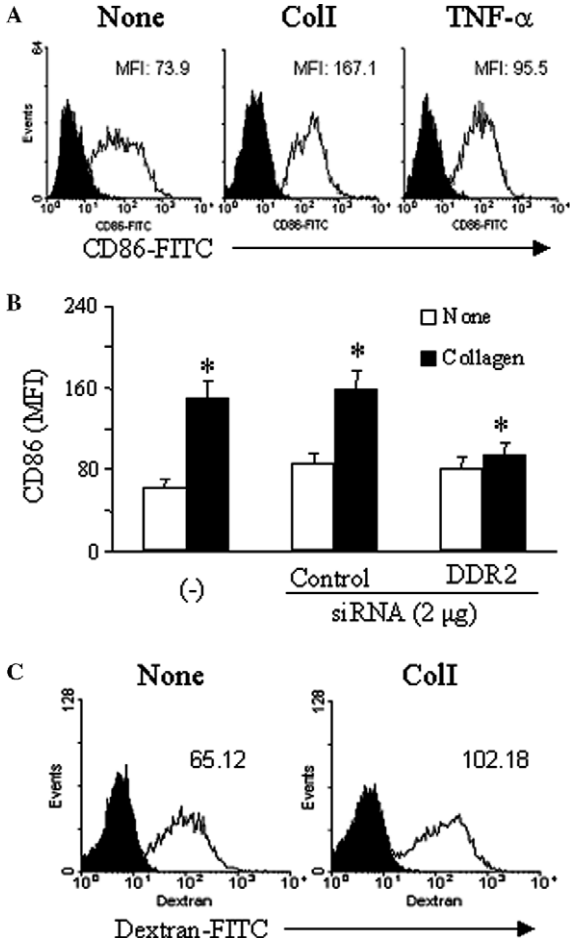

Fig. 3. Enhanced CD86 expression and antigen uptake activity in ColI-stimulated DCs via DDR2. (A) DCs (1×10^5) were stimulated with ColI or TNF-α for 48 h and immunostained with directly FITC-conjugated anti-CD86 or control IgM. Live cells were gated to separate CD11c high-expressing DCs and presented as open histogram for CD86 expression or solid for control IgM. (B) For DDR2 depletion, DCs were transfected with DDR2-specific or nonspecific siRNA (2 μg/well) and incubated for 48 h. Transfectants were incubated on ColI-coated or noncoated plates for 48 h and immunostained with FITC-conjugated anti-CD86 to be analyzed by flow cytometry. (C) To test antigen uptake activity, immature DCs were cultured with FITC–dextran for 90 min at 37 °C (open) or 4 °C (black). The cells were analyzed using FACS Calibur. Each data represents one of the mean-fluorescence intensity (MFI) of three independent experiments ($^*p < 0.05$).

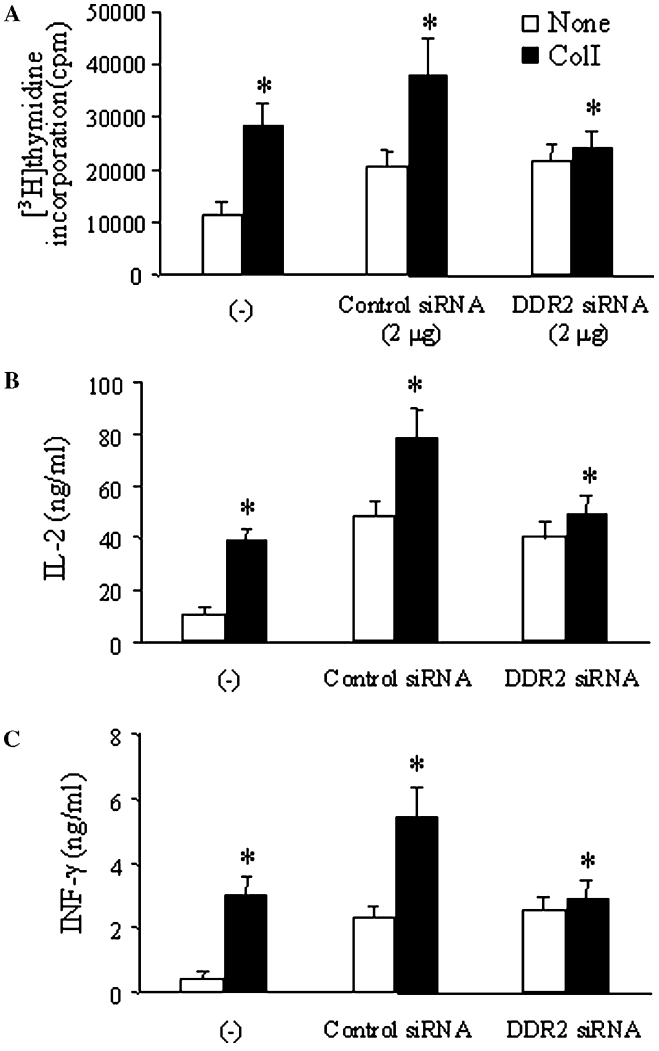

Fig. 4. Enhanced allogeneic mixed T cell reaction by ColI-stimulated DCs via DDR2. (A) For allogeneic T cell proliferation, CD3 + T cells (1×10^5) purified from splenocytes were cocultured with ColI-treated DCs (1×10^4) for 3 days and then pulsed with 2 μCi [^{3}H]thymidine for an additional 16 h. [^{3}H]thymidine incorporation in harvested cells was measured by liquid scintillation counter. To assess cytokine production of T cells by ColI-stimulated DCs, T cells were cocultured with ColI-treated DCs. After 3 days, levels of IL-2 (B) and IFN-γ (C) in the supernatant were measured by ELISA. For DDR2 deletion, DCs (1×10^6 cells) were transfected with specific or nonspecific siRNA (2 μg) and incubated for 48 h. The cells were stimulated with ColI for 24 h and used as stimulator of T cells. All data represent means ± SEM of three independent experiments ($^*p < 0.05$).

DCs, whereas other collagen receptors were downregulated rapidly by the activation. Previous work has shown that interaction of murine DCs with collagen upregulated allostimulatory capacity and release of IL-6 by allogeneic T cells [18]. It implicates that DDR2 may be a major collagen receptor for upregulating ability of DCs. Our data also showed that ColI could enhance IL-12 release, CD86 expression, and allogeneic T cell reaction of DCs through DDR2. Although we could not find the direct effect of DDR2–ColI interaction on the activation of immature DCs because of undesirable DC maturation caused by siRNA transfection procedure, our findings that ColI treatment induces DDR2 phosphorylation only in immature DCs and enhances their functional ability ultimately support the involvement of DDR2 in the activation of DCs regardless of maturation.

In addition to DDR2, we cannot exclude the involvement of DDR1 in the ColI-induced activation of immature DCs, since DDR1 transcripts were detected in immature DCs, but not mature DCs. A recent study has reported that DDR1 was expressed in human monocyte-derived immature DCs and phosphorylated by anti-DDR1 mAb to enhance the activation of DCs, but anti-DDR1 mAb alone was not sufficient to induce full maturation of immature DCs into mature DCs [28]. Therefore, it is possible that

ColI can respond at least partially to BM-derived immature DCs via DDR1.

Little is known about the regulation of DDR2 activation in cellular responses. Studies of the role of DDR2 in cellular function have primarily focused on regulation of MMP expression and collagen remodeling. Enhanced DDR2 expression has been found in activated cells, including vascular smooth muscle cells, fibroblasts, and chondrocytes [29–31]. Additionally, DDR2 phosphorylation was detected only in injury tissues, accompanying increased collagen deposition [32] and resulted in the production of matrix metalloproteinases (MMPs), which mediate collagen degradation during pathophysiological events, such as skeletal formation, cellular migration, inflammation, wound healing, arthritis, and cancer [30–34]. MMP-1 exhibits an unique ability to cleave the major fibrillar collagens (types I, II, and III), which are DDR2 ligands [35], and regulate cell migration through binding to ColI during wound healing [36]. In particular, MMP-9 enhances migration of DCs through the basement membranes [37]. Taken together, these findings indicate that DDR2 activation in injury may play an important role in DC migration to homing sites through collagen degradation by MMP produced by collagen–cell interaction.

In conclusion, we have demonstrated that DDR2 may be a major collagen receptor mediating DC activation and that DDR2–ColI interaction upregulates the functional activity of DCs. Our findings thus provide novel evidence that DDR2 plays an important role in DC activation regulating cellular immunity. However, further studies should be necessary to elucidate the role of DDR2 in the activation of human DCs for understanding the regulation of DC functions in immune diseases.

Acknowledgment

This paper was supported by Grant from the Korea Science and Engineering Foundation (R01-2005-000-10942-0).

References

[1] J. Banchereau, R.M. Steinman, Dendritic cells and the control of immunity, Nature 392 (1998) 245–252.
[2] J.L. Rodriguez-Fernandez, A.L. Corbi, Adhesion molecules in human dendritic cells, Curr. Opin. Invest. Drugs 6 (2005) 1103–1111.
[3] M. Rossi, J.W. Young, Human dendritic cells: potent antigen-presenting cells at the crossroad of innate and adaptive immunity, J. Immunol. 175 (2005) 1373–1381.
[4] M. Cella, F. Sallusto, A. Lanzavecchia, Origin maturation and antigen presenting function of dendritic cells, Curr. Opin. Immunol. 9 (1997) 10–16.
[5] D.N.J. Hart, Dendritic cells: unique leukocyte populations which control the primary immune response, Blood 90 (1997) 3245–3287.
[6] R.M. Steinman, The dendritic cell system and its role in immunogenicity, Annu. Rev. Immunol. 9 (1991) 271–296.
[7] G. Girolomoni, P. Ricciardi-Castagnoli, Dendritic cells hold promise for immunotherapy, Immunol. Today 18 (1997) 102–104.
[8] J. Banchereau, F. Briere, C. Caux, J. Davoust, S. Lebecque, Y.J. Liu, B. Pulendran, K. Palucka, Immunobiology of dendritic cells, Annu. Rev. Immunol. 18 (2000) 767–811.
[9] I. Mellman, R.M. Steinman, Dendritic cells: specialized and regulated antigen processing machines, Cell 106 (2001) 255–258.
[10] A. Lanzavecchia, F. Sallusto, Regulation of T cell immunity by dendritic cells, Cell 106 (2001) 263–266.
[11] M.A. Morse, H.K. Lyerly, E. Gilboa, E. Thomas, S.K. Nair, Optimization of the sequence of antigen loading and CD40-ligand-induced maturation of dendritic cells, Cancer Res. 58 (1998) 2965–2968.
[12] T. Jakob, P.S. Walker, A.M. Krieg, M.C. Udey, J.C. Vogel, Activation of cutaneous dendritic cells by CpG-containing oligodeoxynucleotides: a role for dendritic cells in the augmentation of Th1 responses by immunostimulatory DNA, J. Immunol. 161 (1998) 3042–3049.
[13] S. Singh, S. Aiba, H. Manome, H. Tagami, The effects of dexamethasone, cyclosporine, and vitamin D (3) on the activation of dendritic cells stimulated by haptens, Arch. Dermatol. Res. 291 (1999) 48–554.
[14] P. Rovere, C. Vallinoto, A. Bondanza, et al., Bystander apoptosis triggers dendritic cell maturation and antigen-presenting function, J. Immunol. 161 (1998) 4467–4471.
[15] C. Ammon, S.P. Meyer, L. Schwarzfischer, S.W. Krause, R. Andreesen, M. Kreutz, Comparative analysis of integrin expression on monocyte-derived macrophages and monocyte-derived dendritic cells, Immunology 100 (2000) 364–369.
[16] T.C. Tsao, W. Xia, G.M. Rodberg, C.E. Pinto, R.L. Kradin, Interferon-gamma and tumor necrosis factor-alpha promote the binding of dendritic cells to fibronectin, Pathobiology 62 (1994) 120–126.
[17] D.A. McCarthy, M.G. Macey, P.A. Bedford, S.C. Knight, D.C. Dumonde, K.A. Brown, Adhesion molecules are upregulated on dendritic cells isolated from human blood, Immunology 92 (1997) 244–251.
[18] K. Mahnke, R.S. Bhardwaj, T.A. Luger, T. Schwarz, S. Grabbe, Interaction of murine dendritic cells with collagen up-regulates allostimulatory capacity, surface expression of heat stable antigen, and release of cytokines, J. Leukoc. Biol. 60 (1996) 465–472.
[19] U. Brand, I. Bellinghausen, A.H. Enk, H. Jonuleit, D. Becker, J. Knop, J. Saloga, Influence of extracellular matrix proteins on the development of cultured human dendritic cells, Eur. J. Immunol. 28 (1998) 1673–1680.
[20] R.M. Suri, J.M. Austyn, Bacterial lipopolysaccharide contamination of commercial collagen preparations may mediate dendritic cell maturation in culture, J. Immunol. Methods 214 (1998) 149–163.
[21] W. Vogel, C. Brakebusch, R. Fassler, F. Alves, F. Ruggiero, T. Pawson, Discoidin domain receptor 1 is activated independently of β_1 integrin, J. Biol. Chem. 275 (2000) 5779–5784.
[22] W.F. Vogel, R. Abdulhussein, C.E. Ford, Sensing extracellular matrix: An update on discoidin domain receptor function, Cell. Signal. 18 (2006) 1108–1116.
[23] G. Hou, W.F. Vogel, M.P. Bendeck, Tyrosine kinase activity of discoidin domain receptor 1 is necessary for smooth muscle cell migration and matrix metalloproteinase expression, Circ. Res. 90 (2002) 1147–1149.
[24] W. Vogel, G.D. Gish, F. Alves, T. Pawson, The discoidin domain receptor tyrosine kinases are activated by collagen, Mol. Cell 1 (1997) 13–23.
[25] K. Inaba, M. Inaba, N. Romani, et al., Generation of large numbers of dendritic cells from mouse bone marrow cultures supplemented with granulocyte/macrophage colony-stimulating factors, J. Exp. Med. 176 (1992) 1693–1702.
[26] U. Brand, I. Bellinghausen, A.H. Enk, H. Jonuleit, D. Becker, J. Knop, J. Saloga, Allergen-specific immune deviation from a TH2 to a TH1 response induced by dendritic cells and collagen type I, J. Allergy Clin. Immunol. 104 (1999) 1052–1059.
[27] M.B. Lutz, C.U. Assmann, G. Girolomoni, P. Ricciardi-Castagnoli, Different cytokines regulate antigen uptake and presentation of a precursor dendritic cell line, Eur. J. Immunol. 26 (1996) 586–594.
[28] W. Matsuyama, M. Faure, T. Yoshimura, Activation of discoidin domain receptor 1 facilitates the maturation of human monocyte-derived dendritic cells through the TNF receptor associated factor

6/TGF-beta-activated protein kinase 1 binding protein 1 beta/p38 alpha mitogen-activated protein kinase signaling cascade, J. Immunol. 171 (2003) 3520–3532.

[29] K.G. Shyu, Y.M. Chao, B.W. Wang, P. Kuan, Regulation of discoidin domain receptor 2 by cyclic mechanical stretch in cultured rat vascular smooth muscle cells, Hypertension 46 (2005) 614–621.

[30] L. Xu, H. Peng, D. Wu, K. Hu, M.B. Goldring, B.R. Olsen, Y. Li, Activation of the discoidin domain receptor 2 induces expression of matrix metalloproteinase 13 associated with osteoarthritis in mice, J. Biol. Chem. 280 (2005) 548–555.

[31] E. Olaso, J.P. Labrador, L. Wang, K. Ikeda, F.J. Eng, R. Klein, D.H. Lovett, H.C. Lin, S.L. Friedman, Discoidin domain receptor 2 regulates fibroblast proliferation and migration through the extracellular matrix in association with transcriptional activation of matrix metalloproteinase-2, J. Biol. Chem. 277 (2002) 3606–3613.

[32] E. Olaso, K. Ikeda, F.J. Eng, L. Xu, L.H. Wang, H.C. Lin, S.L. Friedman, DDR2 receptor promotes MMP-2-mediated proliferation and invasion by hepatic stellate cells, J. Clin. Invest. 108 (2001) 1369–1378.

[33] A.E. Yu, A.N. Murphy, W.G. Stetler-Stevenson, Structure, activation, regulation, and substrate specificity, in: W.C. Parls, R.P. Mecham (Eds.), Matrix metalloproteinases, Academic Press, San Diego, 1998, pp. 85–113.

[34] R. Visse, H. Nagase, Matrix, metalloproteinases and tissue inhibitors of metalloproteinases: structure, function, and biochemistry, Circ. Res. 92 (2003) 827–839.

[35] J.J. Jeffrey, Interstitial collagenases, in: W.C. Parls, R.P. Mecham (Eds.), Matrix Metalloproteinases, Academic Press, San Diego, 1998, pp. 15–42.

[36] J.A. Dumin, S.K. Dickson, T.P. Stricker, M. Bhattacharyya-Pakrasi, J.D. Roby, S.A. Santoro, W.C. Parks, Pro-collagenase-1 (matrix metalloproteinase-1) binds the alpha(2)beta(1) integrin upon release from keratinocytes migrating on type I collagen, J. Biol. Chem. 276 (2000) 29368–29374.

[37] V. Chabot, P. Reverdiau, S. Iochmann, A. Rico, D. Senecal, C. Goupille, P.Y. Sizaret, L. Sensebe, CCL5-enhanced human immature dendritic cell migration through the basement membrane in vitro depends on matrix metalloproteinase-9, J. Leukoc. Biol. 79 (2006) 767–778.