Ginsenoside Metabolite Compound K Suppresses T-Cell Priming via Modulation of Dendritic Cell Trafficking and Costimulatory Signals, Resulting in Alleviation of Collagen-Induced Arthritis

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ABSTRACT
Ginsenoside metabolite compound K (CK; 20-O-α-glucopyranosyl-20(S)-protopanaxadiol), a novel ginsenoside metabolite, belongs to the dammarane-type triterpene saponins, according to its structure. The anti-inflammatory activity of CK has been identified in several studies. Our study demonstrated that CK exerted an anti-inflammatory effect in collagen-induced arthritis (CIA) and adjuvant-induced arthritis animal models, and this effect was due to inhibition of the abnormal activation and differentiation of T cells. However, the mechanism of CK in suppressing T-cell activation remains unclear. In this study, CK had a therapeutic effect in mice with CIA, decreased the percentage of activated T cells and dendritic cells (DCs), and increased the percentage of naive T cells in lymph nodes. The inhibitory effect on T-cell activation of CK was related to suppression of accumulation of DCs in lymph nodes. CK decreased CCL21 levels in lymph nodes and CCR7 expression in DCs and suppressed CCL21/CCR7-mediated migration of DCs, thus reducing accumulation of DCs in lymph nodes. In addition, signals for T-cell activation including major histocompatibility complex class II and costimulatory molecules, such as CD80 and CD86, were suppressed by CK, and the proliferation of T cells induced by DCs was inhibited by CK. In conclusion, this study demonstrated that CK downregulated DC priming of T-cell activation in CIA, and suppression of CCL21/CCR7-mediated DC migration and signaling between T cells and DCs might be the potential mechanism. These results provide an interesting, novel insight into the potential mechanism by which CK contributes to the anti-inflammatory effect in autoimmune conditions.

Introduction
Numerous studies have confirmed the role of T cells in the pathogenesis of autoimmune diseases such as rheumatoid arthritis (RA). The excessive activation of T cells causes undesirable immune responses against self-antigens, which then result in chronic inflammatory and autoimmune response (Pablos and Cañete, 2013; Rodeghero et al., 2013). In our previous study, a decreased percentage of naive T cells, a raised percentage of activated T cells, and hyperplasia of T cells were observed in autoimmune arthritis animal models (Chang et al., 2011; Liu et al., 2011; Song et al., 2013; Zhang et al., 2013). Ginsenoside metabolite compound K (CK; 20-O-α-glucopyranosyl-20(S)-protopanaxadiol; C36H42O8; mol. wt., 622.88) belongs to the dammarane-type triterpene saponins, according to its structure (Fig. 1) (Chi and Ji, 2005). It is a degradation product of ginsenoside in the intestine by bacteria and is the major form of ginsenoside absorbed into the body (Tawab et al., 2003; Lee et al., 2009) and can be transformed from ginsenoside by food micro-organisms in vitro (Chi and Ji, 2005). The anti-inflammatory activity of CK has been identified in several studies. CK suppresses production of proinflammatory cytokines, such as tumor necrosis factor (TNF)-α, interleukin (IL)-12, and IL-6, and inhibits cyclooxygenase-2 expression (Eriksson et al., 2003; Bailey et al., 2007). The role of DCs in the T-cell response in inflammatory conditions (Leung et al., 2002; Pablos and Cañete, 2013) is complex class II (MHCII)-TCR and B7 (CD80, CD86)-CD28 signals for T-cell activation including major histocompatibility complex class II and costimulatory molecules, such as CD80 and CD86, were suppressed by CK, and the proliferation of T cells induced by DCs was inhibited by CK. In conclusion, this study demonstrated that CK downregulated DC priming of T-cell activation in CIA, and suppression of CCL21/CCR7-mediated DC migration and signaling between T cells and DCs might be the potential mechanism. These results provide an interesting, novel insight into the potential mechanism by which CK contributes to the anti-inflammatory effect in autoimmune conditions.

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ABBREVIATIONS: APC, antigen-presenting cell; CIA, collagen-induced arthritis; CK, ginsenoside metabolite compound K; 20-O-α-glucopyranosyl-20(S)-protopanaxadiol; DC, dendritic cell; IL, interleukin; MHCII, major histocompatibility complex class II; MTX, methotrexate; RA, rheumatoid arthritis; TCR, T-cell receptor; TNF, tumor necrosis factor; Treg, regulatory T cell.
pathogenesis of autoimmune disease, such as RA, has been confirmed in numerous studies (Pulendran et al., 2010; Rodríguez-Fernández, 2013; Miles et al., 2014). Studies have demonstrated that the infiltrated DCs in the inflamed synovial tissue subsequently migrate to the draining lymph nodes and present arthritogenic peptide to T cells and induce naive T-cell activation (Pettit et al., 2000; Lutzky et al., 2007). The homing of DCs from peripheral to secondary lymphoid organs is a basic feature of their ability to prime effective T-cell immune responses. In inflammatory conditions, migration of DCs induces pathogenic T cells, such as Th1 and Th17 cells (Ma et al., 2013). Migration of DCs from peripheral tissues to lymph nodes is induced by the chemokine CCL21 and its receptor CCR7 (Tal et al., 2011). CCL21 expressed in high endothelial venules is required for antigen-laded mature DCs to migrate to lymphatic vessels, and this has been shown to increase massively during inflammation (Johnson and Jackson, 2010).

Therefore, in this study, we observed the effect of CK on T-cell activation in mice with collagen-induced arthritis (CIA) and investigated the mechanism, focusing on migration of DCs and signals provided by DCs. The results demonstrate that CK downregulated DC priming of T-cell activation in CIA, and suppression of CCL21/CCR7-mediated DC migration and signaling between T cells and DCs might be the potential mechanism.

Materials and Methods

Animals. Sixty male DBA/1 mice (18 ± 2 g) (Shanghai SLAC Laboratory Animal, Ltd., Shanghai, China) were used in this study. All mice were maintained in the specific pathogen-free animal laboratory of Anhui Medical University (Hefei, China). All experiments were approved by the Ethics Review Committee for Animal Experimentation of Institute of Clinical Pharmacology, Anhui Medical University.

Reagents. Complete Freund's adjuvant and chick type II collagen were obtained from Chondrex Inc. (Redmond, WA); Gibco RPMI 1640 medium was from Life Technologies (Grand Island, NY); CCL21 enzyme-linked immunosorbent assay kit was from Cusabio Biotech Co., Ltd. (Wuhan, China); and fluorescence-conjugated monoclonal antibodies to CD4, CD62L, CD25, CD11c, CD80, CD86, and MHCII were from BioLegend, Inc. (San Diego, CA). Anti-CD11c and T-cell MicroBeads were from Miltenyi Biotec (Bergisch Gladbach, Germany); Cell Counting Kit was from Dojindo Molecular Technologies, Inc. (Tokyo, Japan); and monoclonal antibody to CCR7 was from Abcam (Cambridge, UK). CK (purity, >98%) was provided by Zhejiang Haizheng Medicine Co., Ltd. (Taizhou, China), and methotrexate (MTX) was obtained from Shanghai Xinyi Pharmaceutical Co., Ltd. (Shanghai, China).

Induction and Treatment of CIA. Type II collagen was dissolved in 0.1 M acetic acid and emulsified with an equal volume of complete Freund's adjuvant at 2 mg/ml and then incubated overnight at 4°C. DBA/1 mice were injected intradermally twice with 0.1 ml of this emulsion (100 μg of type II collagen/mouse) at the back and the base of the tail. The day of the first immunization was defined as day 0, and the booster injection was administered into the back on day 21 (Zhang et al., 2013). Mice were divided into six groups (n = 10 per group): normal, CIA, CK 14 mg/kg, CK 26 mg/kg, CK 224 mg/kg, and MTX 2 mg/kg. After the onset of arthritis at day 27, mice were given CK by intragastric administration, once a day, for 21 days (during secondary arthritis phase). MTX (2 mg/kg) was administered intragastrically, once every 3 days, eight times. Meanwhile, the normal and model animals were given an equal volume of normal saline.

Evaluation of Arthritis. To evaluate the severity of arthritis, arthritis global assessment and swollen joint count of mice were evaluated every 3 days by two observers blinded to the treatment. The arthritis global assessment was based on symptoms in different parts of the CIA mice: ear (0 = no nodules or redness, 1 = nodules and redness on one ear), nose (0 = no connective tissue swelling or redness, 1 = evident connective tissue swelling and redness), tail (0 = no nodules or redness, 1 = evident nodules and tail redness), and paw (0 = no swelling or redness, 1 = one front or hind paw with swelling and redness, 2 = two paws with swelling and redness, 3 = three paws with swelling and redness, 4 = four paws with swelling and redness). The above-mentioned scores were tallied to form the arthritis global assessment, and a maximum value for each mouse was used. Each paw has five phalanges and one ankle or wrist joint, so the maximum swollen joint count for each mouse was 24 (Wang et al., 2013).

Preparation of Mononuclear Cells and T Cells. Lymph node single-cell suspension was collected by mechanical dissociation of lymph node through nylon mesh. Mononuclear cells were purified from the gradient interface. Then the cells were washed with phosphate-buffered saline three times and suspended in RPMI 1640 medium at a concentration of 1 × 10^7 cells/l for DC and T-cell subset assays. Then CD4^+ T cells were purified by MicroBeads (Miltenyi) for mixed lymphocyte reaction assay.

Culture of Bone Marrow–Derived DCs. Bone marrow–derived DCs were generated from mouse bone marrow cells in the presence of recombinant murine granulocyte-macrophage colony-stimulating factor and recombinant murine IL-4 (Song et al., 2014). On day 7, the dendritic proliferation clusters were collected and purified by anti-CD11c MicroBeads as immature DCs. Purified DCs were stimulated with lipopolysaccharide (100 ng/ml) for 24 hours as mature DCs.

Flow Cytometry. To assay the subsets of T cells and DCs and expression of surface markers (CD80, CD86, and MHCII) and CCR7, fluorescence-conjugated antibodies were added to single-cell suspension (100 μl) prepared previously. After gentle mixing, the samples were incubated for 20 minutes at 4°C and then analyzed using flow cytometry.

CCL21 Determination. Lymph nodes from each group were weighed and homogenized (100 mg of tissue per 1 ml of homogenization buffer). The samples were subjected to one round of freeze–thaw and then sonicated for 10 minutes. The homogenate was centrifuged at 3000g for 10 minutes. The supernatants were collected for CCL21 measurements using an enzyme-linked immunosorbent assay.

CCR7 Analysis by Western Blot. DCs prepared previously were lysed in cell lysis buffer, and denatured protein was separated by 10% SDS-PAGE and transferred electrophoretically to polyvinylidene fluoride membrane. After incubating with blocking buffer (0.05% Tween 20 phosphate-buffered saline with 5% nonfat milk) at 37°C for 2 hours, primary antibody to CCR7 was used at a final dilution of 1:2000 overnight at 4°C. After incubating with the appropriate secondary antibody at 37°C for 2 hours, immunodetection was carried out using enhanced chemiluminescence reagent according to the manufacturer's instructions. Equivalent protein loading and transfer efficiency were verified using staining for β-actin.

Transwell Assay. Mature DCs (5 × 10^5 cells) were placed in transwell chambers (5.0 μm) (Corning, Corning, NY) with or without
CK (10⁻⁹, 10⁻⁷, or 10⁻⁵ M); culture media 600 µl with CCL21 (50 ng/ml) was added in 24-well plates and then incubated at 37°C with 5% CO₂ for 18 hours. Cells migrating into the bottom chamber were determined using cell counter (Cellometer; Nexcelom Bioscience, Lawrence, MA). The migration index was calculated as the number of cells transmigrating with chemokine divided by the number of transmigrating cells in the absence of chemokine (Calpe et al., 2011).

**Mixed Lymphocyte Reaction.** Prepared DCs (5 × 10⁶ cells/well) and T cells (1 × 10⁶ cells/well) were placed in 96-well plates at a ratio of 1:20 and then cocultured at 37°C with 5% CO₂ for 44 hours. T-cell proliferation was assayed using the Cell Counting Kit assay. Ten microliters of Cell Counting Kit solution was added to each well; the absorbance at 450 nm was recorded using a 96-well plate reader (Zhu et al., 2013). Ten microliters of Cell Counting Kit solution was added to each well; the absorbance at 450 nm was recorded using a 96-well plate reader (Zhu et al., 2013).

**Statistical Analysis.** Data were expressed as mean ± S.D. Statistical significance was determined by one-sample t test, where P < 0.05 was considered statistically significant.

**Results**

**CK Alleviated the Severity of Arthritis in CIA Mice.** To assess the therapeutic effect of CK on CIA mice, the severity of arthritis was evaluated. After onset of arthritis on day 27, there was a significant increase in arthritis global assessment and swollen joint count in CIA mice. Administration of CK (56 mg/kg) alleviated the global assessment (day 45: t = 2.333, P = 0.031; day 48: t = 2.212, P = 0.040; day 51: t = 2.703, P = 0.015; Fig. 2A) and swollen joint count (day 45: t = 2.762, P = 0.013; day 48: t = 2.423, P = 0.026; day 51: t = 2.640, P = 0.023; Fig. 2B) significantly compared with untreated CIA mice. Administration of CK (224 mg/kg) alleviated the global assessment (day 45: t = 2.400, P = 0.027; day 48: t = 2.554, P = 0.020; day 51: t = 3.239, P = 0.005; Fig. 2A) and swollen joint count (day 45: t = 2.867, P = 0.010; day 48: t = 3.757, P = 0.001; day 51: t = 4.821, P = 0.001; Fig. 2B) significantly compared with untreated CIA mice.

**The Effect of CK on the Subsets of T Cells and DCs in Lymph Nodes from CIA Mice.** To evaluate the effect of CK on T-cell activation in CIA mice, the subsets of naive T cells and activated T cells were assayed in CIA lymph nodes. The results show that naive T cells decreased (t = -7.432, P = 0.001; Fig. 3, A and D) and activated T cells increased (t = 3.552, P = 0.005; Fig. 3, B and E) in CIA mice compared with normal mice. Treatment with CK (56 mg/kg, 224 mg/kg) resulted in an increase of naive T cells (CK 56 mg/kg: t = -4.654, P = 0.004; CK 224 mg/kg: t = -9.613, P = 0.000; Fig. 3, A and D) and a decrease of activated T cells in lymph nodes (CK 56 mg/kg: t = 2.763, P = 0.020; CK 224 mg/kg: t = 3.849, P = 0.009; Fig. 3, B and E). The percentage of DCs increased in CIA lymph nodes compared with normal mice (t = 5.442, P = 0.001; Fig. 3, C and F). Administration of CK (56 and 224 mg/kg) decreased the percentage of DCs (CK 56 mg/kg: t = 3.116, P = 0.01; CK 224 mg/kg: t = 5.737, P = 0.001; Fig. 3, C and F) in lymph nodes. Interestingly, the percentages of DCs and naive T cells revealed a negative correlation (R² = 0.8348, P = 0.03; Fig. 3G), whereas the percentages of DCs and activated T cells revealed a positive correlation (R² = 0.9830, P = 0.0009; Fig. 3H).

**CK Suppressed DC Migration Induced by CCL21.** CCL21 is the major chemokine that mediated DC migration from the periphery to the lymph nodes. To clarify whether the changed percentage of DCs in lymph nodes was associated with migration, a transwell assay was designed; CCL21 levels in the lymph nodes and CCR7 expression on DCs were detected. The results show that the migration index of DCs induced by CCL21 was inhibited by CK (100 nM and 10 µM) (CK 100 nM: t = 5.356, P = 0.006; CK 10 µM: t = 8.094, P = 0.001; Fig. 4A). CCL21 in the lymph nodes increased in CIA mice compared with normal mice (t = 8.216, P = 0.001; Fig. 4B). CK (56 and 224 mg/kg) decreased CCL21 levels in the lymph nodes (CK 56 mg/kg: t = 3.481, P = 0.025; CK 224 mg/kg: t = 6.702, P = 0.003; Fig. 4B). Interestingly, the percentage of DCs and CCL21 levels revealed a positive correlation (R² = 0.7802, P = 0.0470; Fig. 4C). In addition, CK (100 nM and 10 µM) decreased CCR7 expression on DCs (CK 100 nM: t = 10.119, P = 0.001; CK 10 µM: t = 5.931, P = 0.004; Fig. 4, D and E) (CK 100 nM: t = 7.469, P = 0.001; CK 10 µM: t = 11.540, P = 0.000; Fig. 4, F and G).

**CK Impaired T-Cell Stimulatory Capability of DCs.** In the next study, the T-cell stimulatory capacity of DCs was...
assayed by mixed lymphocyte reaction. T-cell proliferation was significantly induced by DCs ($t = 6.690, P = 0.001$; Fig. 5). The ability of DCs to prime T-cell proliferation was impaired when DCs were pretreated with CK (100 nM and 10 μM) before coculture with T cells (CK 100 nM: $t = 2.707, P = 0.035$; CK 10 μM: $t = 3.877, P = 0.008$; Fig. 5).

**CK Impaired CD80, CD86, and MHCII Expression on DCs.** CD80 and CD86 are important costimulatory molecules

**Fig. 3.** The effect of CK on subsets of T cells and DCs in lymph nodes from CIA mice. Subsets of naive T cells (A and D), activated T cells (B and E), and DCs (C and F) in CIA mice lymph nodes were evaluated by flow cytometry. The representative flow cytometry dot plot of each group is shown (A–C). A scatter plot of the correlation between DCs and naive T cells (G) and DCs and activated T cells (H). Values are expressed as mean ± S.D; $n = 6$ for each group. *$P < 0.05$ versus normal. **$P < 0.01$; ***$P < 0.001$ versus CIA. Comp, compensation; FL, fluorescence.
expressed on DCs, and MHCII is the molecule associated with the presenting antigen. They can provide signaling for T-cell activation. In the next study, we investigated whether CK affected the expression of these molecules on DCs. After treatment with CK (100 nM and 10 μM) in vitro, expressions of CD80 (CK 100 nM: t = 3.346, P = 0.015; CK 10 μM: t = 4.211, P = 0.006; Fig. 6, A and B), CD86 (CK 100 nM: t = 3.205, P = 0.018; CK 10 μM: t = 4.063, P = 0.007; Fig. 6, C and D), and MHCII (CK 100 nM: t = 3.174, P = 0.019; CK 10 μM: t = 3.797, P = 0.009; Fig. 6, E and F) on DCs were decreased.

Discussion

Numerous studies have confirmed the role of T cells in the pathogenesis of RA. RA has been regarded as a classic T cell–mediated chronic inflammatory autoimmune disease for years. Abnormal activation of T cells in RA are the main induced factors causing immune damage and synovial pathologic histology change (Alzabin and Williams, 2011; Rodeghero et al., 2013). In our previous study, we found that CK alleviated CIA and adjuvant-induced arthritis, downregulated the percentage of activated T cells, and upregulated naive T cells and regulatory T cells (Tregs) in spleen (Chen et al., 2014; Liu et al., 2014). Consistent with these findings, the results in this study show that CK alleviated CIA and adjuvant-induced arthritis, downregulated the percentage of activated T cells, and upregulated naive T cells and regulatory T cells (Tregs) in spleen (Chen et al., 2014; Liu et al., 2014). Consistent with these findings, the results in this study show that the severity of CIA was alleviated; naive T cells in the lymph nodes increased, whereas activated T cells in the lymph nodes decreased, when CIA mice were treated with CK. To further investigate the mechanism of CK responsible for suppressing T-cell activation, we focused on DCs, the most powerful APCs, in this study.

Secondary lymphoid organs, such as lymph nodes, are the main area where DCs interact with T cells and promote T-cell activation. DC accumulation in the secondary lymphoid tissues is associated with inflammation. A greater number of DCs in the lymph nodes and increased Th2 inflammation have been found in mice with hypersensitivity pneumonitis (Fanning et al., 2013). Our results show that CK downregulated the percentage of DCs in the lymph nodes in CIA mice. Interestingly, the percentages of DCs and naive T cells revealed a negative correlation, whereas the percentages of DCs and activated T cells revealed a positive correlation. These results suggest that the inhibitory effect of CK on T-cell activation may be associated with decreasing DC accumulation in the lymph nodes.

Tissue inflammation induces rapid mobilization of antigen-charged DCs, which can migrate to the draining lymph nodes via afferent lymphatics to induce pathogenic T cells, such as Th1 and Th17 (Ma et al., 2013; Platt and Randolph, 2013). Upregulation of CCR7 and CCL21 can lead to an increase of DCs in the lymph nodes, thus upregulating the development of inflammation. In a mouse model of hypersensitivity pneumonitis, the lung DCs of challenged Lilrb4−/− mice expressed more CCR7. In addition, the lungs of challenged Lilrb4−/− mice contained significantly greater numbers of Ag-bearing DCs and Th2 cells in lymph nodes and were consistent with the attendant exacerbated Th2 lung pathology (Fanning et al., 2013). Reducing the expression of CCR7 can inhibit DC migration to the lymph nodes and then limit the generation of adaptive immunity (Odobasic et al., 2013). Our results show that CK decreased CCL21 levels in the lymph nodes and CCR7 expression on DCs and suppressed CCL21-mediated migration of DCs. Interestingly, the percentage of DCs and CCL21 levels revealed a positive correlation (R^2 = 0.7802, P = 0.0470). These results suggest...
that the effect of CK on reducing DC accumulation in the lymph nodes may be due to the decreasing expression of CCL21 and CCR7.

Fig. 4. CK suppressed DC migration. (A) Migration of DCs induced by CCL21 was evaluated by transwell assay. The migration index was calculated as the number of cells transmigrating with CCL21 divided by the number of transmigrating cells in the absence of CCL21. **P < 0.01 versus control. The results are representative of at least three independent experiments. (B) CCL21 expression in CIA lymph nodes was assayed by enzyme-linked immunosorbent assay. Data are expressed as mean ± S.D.; n = 6 for each group. *P < 0.05; **P < 0.01 versus normal. #P < 0.01 versus CIA. (C) A scatter plot of the correlation between DCs and CXCL12 levels in lymph nodes. (D and E) CCR7 expression on DCs was assayed by flow cytometry. Isotype (red line), control (blue line), CK 10 μM (orange line). Mean fluorescence intensity is presented as mean ± S.D. **P < 0.01 versus control. Mean fluorescence intensity is presented as mean ± S.D. ##P < 0.01 versus control. The results are representative of at least three independent experiments. (F and G) CCR7 expression on DCs was assayed by Western blot. Data are expressed as mean ± S.D. **P < 0.01; ***P < 0.001 versus control. The results are representative of at least three independent experiments.

Fig. 5. CK suppressed T-cell stimulatory capability of DCs. T-cell stimulatory capability of DCs was assayed by mixed lymphocyte reaction. DCs and T cells (at a ratio of 1:20) were cocultured for 44 hours; then proliferation of T cells was assayed by Cell Counting Kit assay. Data are expressed as mean ± S.D. **P < 0.01 versus T cells. *P < 0.05; **P < 0.01 versus T cells with DCs. The results are representative of at least three independent experiments.
lesions, whereas wild-type mice injected with B6.CCR7−/− DCs presented delayed healing of the lesions (Kling et al., 2014). CCR7 and its ligands are associated with the severity of inflammation. In RA patients, the concentration of CCL19 and CCL21 in the plasma and synovial tissue, the cell-surface expression of CCR7 on circulating monocytes and CD4+ T cells, and the elevated levels of CCL21, CCL19, and CCR7 are associated with disease activity (Pickens et al., 2011; Ellingsen et al., 2014). Inhibition of CCR7 in lesional skin is a critical event for clinical remission induced by TNF blockade in patients with psoriasis (Bosè et al., 2013). Our results show that CK exerted therapeutic effects in CIA mice and decreased CCL21 and CCR7 expression. On the basis of our results and the studies previously mentioned, we suppose that the suppression of CCL21 and CCR7 may be one of the mechanisms for the anti-inflammatory effect of CK.

T-cell activation requires three major signals provided by APCs (Caporali et al., 2014). Signal 1 consists of processed antigen peptide, which is presented in the context of MHC complexes to TCR. Signal 2 consists of upregulation of costimulatory molecules (such as CD80 and CD86) for binding to their receptor (CD28) on T cells. Signal 3 consists of the
proper cytokines, providing the final differentiation and proliferation of T cells. We observed that the ability of DCs to promote T-cell proliferation was impaired by CK. In addition, this study showed that CK impaired the expression of CD80, CD86, and MHCII on DCs. Consistent with these results, our previous study showed that CK suppressed TCR and CD28 expression on T cells (Liu et al., 2014) and reduced IL-2 production and CD25 (α-chain of the high-affinity IL-2 receptor) expression on T cells (Chen et al., 2014). These data suggest that CK impairs T-cell activation by reducing the signals provided by DCs.

In our previous studies, CK increased the percentages of CD4^+CD25^+Foxp3^+ Tregs in CIA mice (Liu et al., 2014). It is well established that Tregs play a critical protective role in RA and other autoimmune diseases (Cooles et al., 2013). Tregs control the responses of APCs and effector T cells through direct interaction or through anti-inflammatory cytokine production, and they play an important role in limiting inflammation and regulating adaptive immunity (Bacchetta et al., 2007; Li and Coffman, 2009). Tregs colocalize with DCs in the lymphoid aggregate areas of the RA synovium, but cannot fully suppress DC activation and function (E et al., 2012). In this study, we found that CK suppressed DC priming of T-cell activation. Based on these studies, we can suppose that the effect of CK on suppressing DC function may be associated with Tregs in CIA mice, but the exact effect and mechanism need to be further studied.

Overall, this study demonstrated that CK suppressed DC priming of T-cell activation in CIA, and suppression of CCL21/CCL7-mediated DC migration and signaling between T cells and DCs might be the potential mechanism. These results provide an interesting, novel insight into the potential priming of T-cell activation in CIA, and suppression of CCL21/CCL7 cell-surface expression on monocytes in early rheumatoid arthritis normalized during treatment and CCL19 correlated with radiographic progression. Scand J Rheumatol 43:41–420.


References


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