Adipose-derived stem cells ameliorate colitis by suppression of inflammasome formation and regulation of M1-macrophage population through prostaglandin E2

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Adipose-derived stem cells ameliorate colitis by suppression of inflammasome formation and regulation of M1-macrophage population through prostaglandin E2

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ABSTRACT

Adipose-derived stem cells ameliorate colitis by suppression of inflammasome formation and regulation of M1-macrophage population through prostaglandin E2

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Inflammatory bowel disease (IBD) is an idiopathic disease caused by a dysregulated immune response to intestinal microbes in an individual with a genetic predisposition. Therefore, alleviation of inflammation is very important to treat IBD. Mesenchymal stem cells (MSCs) have been highlighted as new candidates for treating autoimmune disease based on their immunomodulatory properties. In this study, we investigated the anti-inflammatory mechanism and therapeutic effects of adipose tissue-derived MSCs (ASCs) using THP-1

macrophages and dextran sodium sulfate (DSS)-induced mice with chronic colitis. LPS-treated THP-1 cells expressed mRNA of CD11b, an M1 macrophage marker, at day 2. However, THP-1 co-cultured with ASCs expressed mRNA of CD206, CD68, CCL18, legumain, and IL-10, markers of M2 macrophages. In THP-1 cells co-cultured with ASCs, precursor (pro)-IL-1β, Cox-2, and NLRP3 increased dramatically compared to LPS-treated THP-1 cells. Secretion of IL-1β and IL-18 was significantly inhibited by ASCs, but PGE2 production was highly increased in co-culture conditions of THP-1 and ASCs. IL-18 secretion was inhibited by PGE2 treatment, and PGE2 inhibited inflammasome complex (ASC/Cas-1/NLRP3) formation in THP-1 cells. In the DSS-induced chronic colitis model, ASCs ameliorated colitis by decreasing the total number of macrophages and the M1 macrophage population. Our results suggest that ASCs can suppress the inflammatory response by controlling the macrophage population, and ASCs may be therapeutically useful for the treatment of IBD.

Key Words: inflammatory bowel disease, mesenchymal stem cells, macrophages, prostaglandin E2

I. INTRODUCTION

Inflammatory bowel disease (IBD) is an idiopathic disease caused by dysregulated immune response to intestinal microbes in an individual with a genetic predisposition [1, 2]. The two major types of IBD are ulcerative colitis and Crohn's disease. Innate immune cells (neutrophils, macrophages, dendritic cells, and natural killer T cells) and adaptive immune cells (B cells and T cells) strongly infiltrate the lamina propria [3], resulting in mild to severe symptoms including abdominal pain, vomiting, diarrhea, rectal bleeding, internal cramps/muscle spasms in the pelvis, and weight loss. In the United States, about 1 – 1.3 million people currently suffer from IBD [4, 5].

The goals of IBD treatment include symptom relief, long-term remission, and reduced risks of complications, which are achieved by alleviating inflammation. Anti-inflammatory drugs such as corticosteroids, azathioprine, 6 mercaptopurine (6-MP), and antibodies against tumor necrosis factor (TNF)-α (infliximab, adalimumab, certolizumab, and golimumab) or α4β7 integrin (vedolizumab) have been used to reduce symptoms [6-17]. Recently, mesenchymal stem cell (MSC) therapy has been suggested as an effective alternative therapy for immune diseases including IBD based on the immunomodulatory properties of MSCs [18-20].

MSCs have been shown to migrate to sites of tissue injury/inflammation [21, 22]. At injured sites, MSCs are be stimulated by inflammatory cytokines such as interferon (IFN)-γ, TNF-α, or interleukin (IL)-1β to express various immunosuppressive factors, including indoleamine 2,3-dioxygenase (IDO), prostaglandin E2 (PGE2), TNF-α-stimulated protein/ gene 6 (TSG-6), nitric oxide (NO), IL-6, IL-10, and HLA-G [23, 24]. PGE2 has multiple roles in proliferation, apoptosis, tissue repair, angiogenesis, inflammation, immune surveillance, and tumor growth [25-27]. PGE2 can induce acute local inflammation and activate immune reaction at the entry site of pathogens, but it has suppressive roles in the proliferation and differentiation of T cells, macrophages, and monocytes, as well as in the cytotoxic activity of NK cells and cytotoxic T lymphocytes [28-30]. Moreover, PGE2 augments the synthesis of anti-inflammatory cytokine IL-10 and decreases production of the pro-inflammatory cytokines TNF-α, IFN-γ, and IL-12 by DCs and macrophages. Therefore, PGE2 secreted by MSCs can regulate inflammation and ameliorate immune disease including IBD. In this study, we analyzed whether MSCs co-cultured with activated THP-1 cells secrete PGE2. We also evaluated the anti-inflammatory roles of PGE2 on THP-1 and the therapeutic effects of PGE2 secreted by MSCs in dextran sodium sulfate (DSS)-induced chronic colitis in mice.

II. MATERIALS AND METHODS

1. Animal studies

Female C57BL/6 mice (5- to 8-week-old) were purchased from Orient Bio, Inc. (Seongnam, Korea). Mice were maintained in a 12-h light/12-h-dark cycle under specific pathogen free (SPF) conditions at 25 ℃. All animal care and experiments were conducted in accordance with The Guide for Animal Experiments published by the Korea Academy of Medical Sciences, after approval of the Institutional Animal Care and Use Committee of Yonsei University, Wonju College of Medicine. To induce chronic colitis, mice were administered drinking water with 3% DSS (Molecular weight 36–50 kDa; MP Biomedicals, Santa Ana, CA, USA) for 5 days ad libitum, followed by 5 days of normal water without DSS. The DSS administration cycle was repeated once. Mice were divided into four groups: control group that did not drink DSS, colitis group administered with 3% DSS, colitis + ASCs group infused with ASCs (1×10^6 cells) by intraperitoneal injections twice on days 6 and 16, and colitis + ASCs + celecoxib group that was administered celecoxib (3mg/kg; Sigma, San Diego, CA, USA) once a day for the first 3 days after ASC infusion on days 6 and 16. Animal weights were monitored and recorded daily. Mice were sacrificed on day 20 to isolate the colon, and the entire colon length was measured.

2. Cell culture

Human adipose tissue (AT) from three healthy donors (age, 24–38 years) was obtained from Wonju Severance Christian Hospital (Wonju, Korea) by elective liposuction procedures under anesthesia according to procedures approved by the Institutional Review Board of Yonsei University Wonju College of Medicine. Informed consent was obtained from all donors. Mononuclear cells were isolated using a modified protocol described by Zuk et al. [31]. Briefly, lipoaspirates were extensively washed with phosphate-buffered saline (PBS, Welgene, Gyeongsan, Korea) to remove contaminating blood cells and local anesthetics. Mononuclear cells were obtained from the digested lipoaspirates with 0.075% type IA collagenase (Sigma) in PBS. A total of 5×10^6 mononuclear cells were seeded in 100-mm culture dishes with low glucose Dulbecco's minimal essential medium (LG-DMEM, Gibco BRL, Rockville, MD, USA) containing 10% FBS and penicillin/streptomycin. After 2 days, the medium was changed to remove non-adherent cells. Thereafter, the cell culture medium was changed twice weekly, and cells were passaged with 0.25% trypsin/0.1% EDTA (Gibco BRL) until reaching 90% confluency.

The human monocytic cell line THP-1 was maintained in complete RPMI-1640 (Gibco BRL: supplemented with 10% FBS, penicillin/streptomycin and 2 mM

L-glutamine). Macrophage differentiation of THP-1 was induced with 100nM of phorbol ester 12-O-tetradecanoylphorbol-13-acetate (TPA, Sigma) for 2 days and was co-cultured with or without ASCs in transwell plates (Corning, Lowell, MA, USA) while treating with 1μg/ml lipopolysaccharide (LPS, Sigma). To analyze cytokines or PGE2, conditioned medium was collected, filtered (0.45 μm), and stored at -80℃ until needed.

3. Immunohistochemistry

After measuring the entire colon length, colons were fixed in 10% formalin, embedded in paraffin, and sectioned at 5 μm slices with a microtome. For immunohistochemical staining of macrophages, deparaffinized slices were blocked with 0.3% Triton-X 100 and 5% normal horse serum in TBS (20 mM Tris pH 7.6, 137 mM NaCl) and incubated with anti-F4/80 antibodies (Abcam, Cambridge, MA, USA) overnight at 4℃. After washing three times in TBS, slices were incubated with secondary antibody (Abcam) for 30 min. Slides were counterstained with hematoxylin. To assess macrophages, digital images at 200X magnification were obtained (Zeiss Axioimager M1, Göttingen, Germany). Both positive cells and total cells were counted from three random fields, with over 200 cells counted per field.

4. IL-1β and PGE2 measurement

Human IL-1 beta/IL-1F2 Quantikine Kit (R&D Systems, Minneapolis, MN, USA) and PGE2 Assay Kit (R&D) were used to analyze the secretion levels of IL-1β and PGE2 in conditioned media obtained by co-culturing THP-1 with ASCs according to the manufacturer's instructions.

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

Total RNA was extracted from 1×10^5 cells using TRIzol Reagent according to the manufacturer's instructions (Gibco BRL). Total RNA (2 μg) was reverse-transcribed with M-MLV reverse transcriptase (Bioneer, Daejeon, Korea) for one hour at 42℃ in the presence of oligo-dT primer. PCR was performed using *Taq* DNA polymerase (Bioneer). Specific primers used for PCR assays are listed in Table 1. Amplified products were electrophoresed on a 2% agarose gel and photographed using a ChemiDoc XRS+ system (Bio-Rad, Hercules, CA, USA).

Table 1. RT-PCR primers for validation of gene expression

	Primer sequence (5′→3′)	Length (bp)	Annealing temperature (℃)
CD11b	TTACTTGGGTTATGCTGCCG GAACAGCATCACACTGCCA	320/323	62
CD36	CTGAGGACTGCAGTGTAGGAC TCTTCGAGGACAACTTGCTTTT	428	62
CD206	ACAACAAAAGCTGACACAAGGA AGGACAGACCAGTACAATTCAG	379	62
CD68	CTTTGCTGCCATCCTTCACG AGATGCCCCAGGCCTCTC	489	62
CCL18	AGCCAGGTGTCATCCTCCTA GTGGTGCTGAGCAAAACCATT	486	62
Legumain	AAAGAGACCCTGCACAAGCA AACGCATACTCGTACGTGGG	464	62
IL-10	TTCGAGATCTCCGAGATGCC AGTTCACATGCGCCTTGATG	210	58
GAPDH	GAAAGCCTGCCGGTGACTAA GATGGCATGGACTGTGGTCA	647	62

6. Immunoblotting

Cells were lysed in sodium dodecyl sulfate/polyacrylamide gel electrophoresis (SDS–PAGE) sample buffer (62.5 mM Tris-HCl (pH 6.8), 1% SDS, 10% glycerol, and 5% β-mercaptoethanol), boiled for 5 min, subjected to SDS–PAGE, and transferred to an Immobilon membrane (Millipore, Darmstadt, Germany). The membrane was blocked with 5% skim milk in TBST (Tris-HCl buffered saline containing 0.1% Tween 20) and then incubated with primary antibodies against IL-1β, NLRP3 (1:1000, purchased from Cell Signaling Technology, Danvers, MA, USA), F4/80 (1:1000, purchased from Abcam), CD206, IL-18, Cox-2, and GAPDH (1:1000, Santa Cruz Biotech, Santa Cruz, CA, USA). Bound primary antibodies were reacted with HRP-conjugated secondary antibodies (1:2000, Cell Signaling Technology), treated with EZ-Western Lumi Pico (Dogen, Seoul, Korea), and visualized using the ChemiDoc XRS+ system (Bio-Rad).

7. Statistical analysis

Data were expressed as the mean ± standard error of the mean (SEM) using one-way analysis of variance (ANOVA; Scheffe's test). Any difference was considered statistically significant at $*P<0.05$, $**P<0.005$, and $***P<0.0001$.

III. RESULTS

1. Phenotypic changes of macrophages by ASCs

A large number of macrophages are present in the small and large intestines of mammals [32], where they play a key role in the development and progression of IBD. To analyze phenotypic changes of macrophages induced by ASCs, TPA-induced macrophages (THP-1) were co-cultured with ASCs for 2 days, and then macrophage markers were evaluated. LPS-treated THP-1 cells expressed mRNA of CD11b, an M1 macrophage marker, at day 2, but THP-1 co-cultured with ASCs expressed mRNA of CD206, CD68, CCL18, legumain, and IL-10, markers of M2 macrophages (Fig. 1A). Similarly, CD206 proteins were detected in THP-1 co-cultured with ASCs (Fig. 1B). These results suggest that LPS induces M1 differentiation, while ASCs induce transition from M1 to M2 macrophages. However, as shown in Fig. 1, ASCs seem to have limited ability to change the phenotype of macrophages.

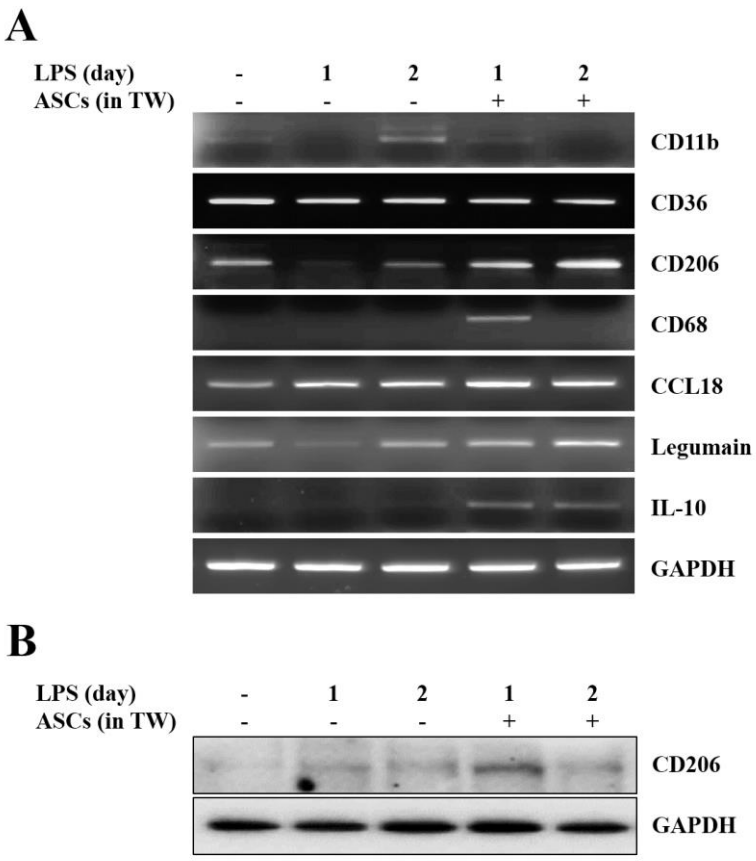


Figure 1. Phenotypic changes of macrophages by ASCs. TPA-treated THP-1 cells were in-directly co-cultured with ASCs for 2 days. A) Changes in M1- and M2-macrophage markers in THP-1 cells by ASCs. M1- and M2-macrophage markers of THP-1 cells were analyzed by RT-PCR. B) Western blot analysis of CD206.

2. Suppression of inflammatory cytokines by ASCs

Next, we analyzed expression of inflammatory cytokines IL-1β and IL-18 in M1 macrophages. Unexpectedly, precursor (pro)-IL-1β was observed to be much higher in THP-1 co-cultured with ASCs than in the LPS-treated group. Expression of NLRP3, a component of the inflammasome that plays an important role in secretion of pro-IL-1β and -IL-18 into culture media, was also dramatically increased. Moreover, expression of Cox-2, an enzyme important for PGE2 production, was greatly increased (Fig. 2A). Interestingly, active IL-18 was not observed by immunoblotting in the culture medium obtained from co-cultured THP-1 with ASCs, and expression of IL-1β was significantly decreased by ASCs (Fig. 2A&2B). PGE2 level was determined by ELISA assay, and PGE2 was dramatically increased in conditioned media from THP-1 co-cultured with ASCs, similar to Cox-2 expression (Fig. 2C). Our results suggest that, although ASCs induce increased expression of pro-IL-1β and NLRP3 in THP-1 cells, ASCs inhibit the secretion of inflammatory cytokines through PGE2.

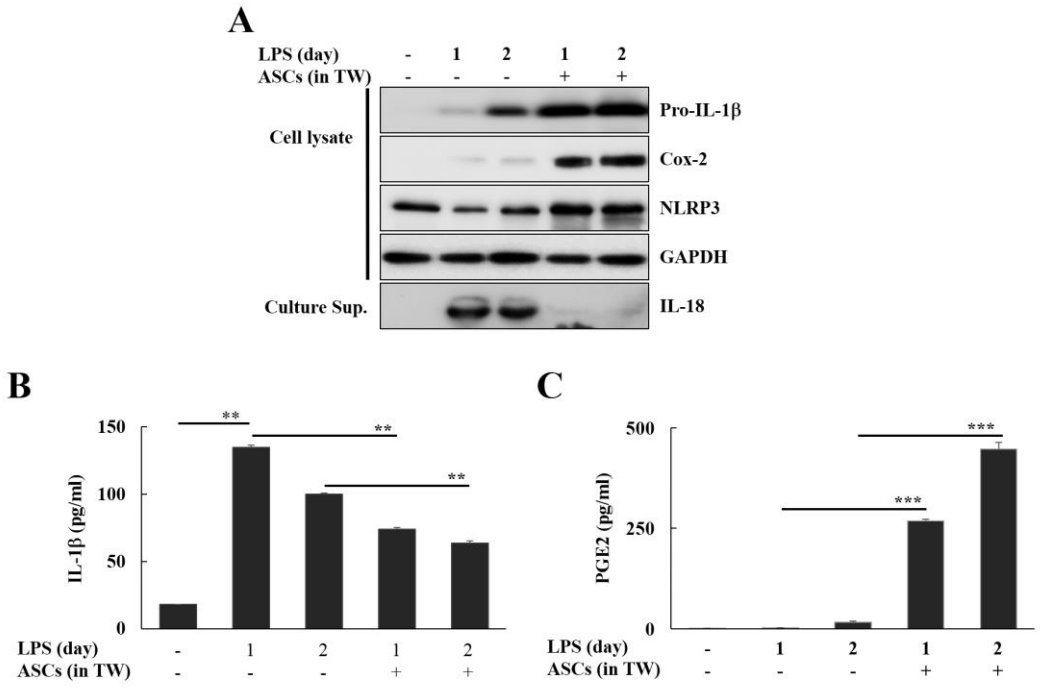


Figure 2. Suppression of inflammatory cytokines by ASCs. A) Western blot analysis of proteins involved in inflammation. Pro-IL-1β, Cox-2, and NLRP3 were detected from lysates of THP-1 cells co-cultured with or without ASCs. Secreted IL-18 was detected from conditioned medium. B) Quantitative analysis of secreted IL-1β. Secreted IL-1β was detected from conditioned medium by IL-1β-ELISA kit according to the manufacturer's instructions. C) Quantitative analysis of PGE2 production. The amount of PGE2 was evaluated from conditioned medium by PGE2-ELISA kit according to the manufacturer's instructions. Results are expressed as mean±SD; *P<0.05, **P<0.01, and

***P<0.001 versus LPS. Statistical analysis was performed using one-way ANOVA.

3. PGE2-dependent suppression of inflammatory cytokines

Therefore, we examined whether PGE2 plays an important role in IL-18 secretion. THP-1 cells were treated with 50 to 1000 nM of PGE2 and LPS, and the expression of inflammation-related proteins was analyzed. Co-treatment with PGE2 and LPS induced elevation of pro-IL-1β, Cox-2, and NLRP3 expression in THP-1 cells. However, secreted IL-18 disappeared in a PGE2-dose dependent manner (Fig. 3A). In addition, elevated expression of pro-IL-1β, Cox-2, and NLRP3 with ASCs was down-regulated by celecoxib, an inhibitor of Cox-2. Active IL-18 also decreased after celecoxib treatment, both in THP-1 cells treated with LPS and those co-cultured with ASCs (Fig. 3B). Interestingly, PGE2 suppressed formation of the inflammasome complex. In particular, ASC and caspase-1 binding to NLRP3 were inhibited by PGE2 (Fig. 3C). These results suggest that PGE2 secreted in THP-1 during co-culturing with ASCs has anti-inflammatory activity through suppression of inflammasome formation and inhibition of IL-18 secretion.

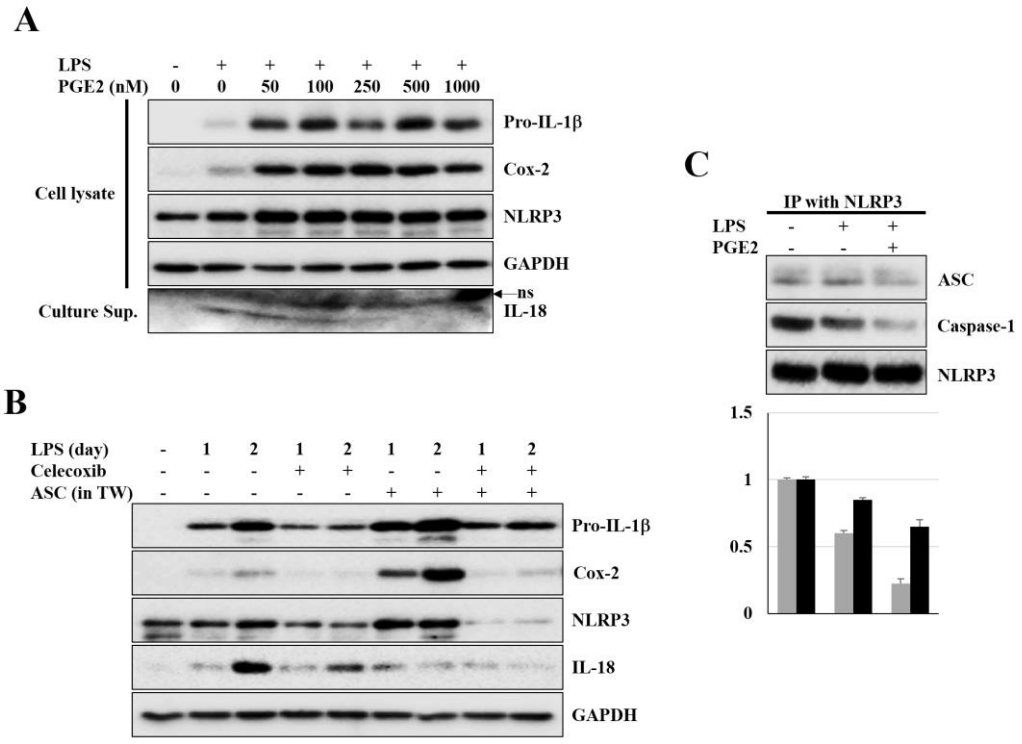


Figure 3. PGE2-dependent suppression of inflammatory cytokines. A) Western blot analysis of proteins involved in inflammation after PGE2 treatment. THP-1 cells were co-treated with LPS and PGE2. Pro-IL-1β, Cox-2, and NLRP3 were detected from lysates of THP-1 cells co-cultured with or without ASCs. Secreted IL-18 was detected from conditioned medium. B) Effects of celecoxib on inflammation-related protein expression. During celecoxib treatment, THP-1 cells were co-cultured with ASCs. Celecoxib inhibited pro-IL-1β, Cox-2, NLRP3, and active IL-18. C) Immunoprecipitation to evaluate oligomer formation of

inflammasome. THP-1 cells were treated with PGE2 and LPS for 1 day and NLRP3 antibody was used for immunoprecipitation of inflammasome components.

4. Therapeutic effects of ASCs in DSS-induced colitis mice

To confirm whether the therapeutic effects of ASCs were associated with PGE2 in mice with DSS-induced colitis, ASCs were transplanted with or without celecoxib administration in mice with colitis. Body weight gradually recovered from 14 and 17 days in ASC-injected groups without or with celecoxib administration, respectively (Fig. 4A). Colon length also significantly recovered in ASC-injected groups, and although not significant, colon length tended to decrease with celecoxib compared to ASC-injected groups (Fig. 4B). Histologic improvement was observed in the ASC-injected group and ASCs + celecoxib group (Fig. 4C). The number of macrophages in colon tissues of mice with DSS-induced colitis increased by about 2 times compared with normal mice, but ASCs significantly decreased the number of macrophages. However, in the ASCs + celecoxib group, the number of macrophages also increased. In particular, M1 macrophages were significantly increased in colitis, but they were decreased in the ASC-injected group to a similar extent as in the control group. The M1 macrophage population increased again in the ASC + celecoxib group (Fig. 4C). These results suggest that ASCs relieve inflammation by lowering the total number of macrophages and decreasing the M1 population. In addition, since the number of M1 macrophages and total number of macrophages increased in the

ASC + celecoxib group, PGE2 produced by ASCs in macrophages might suppress the inflammatory response by controlling the macrophage population.

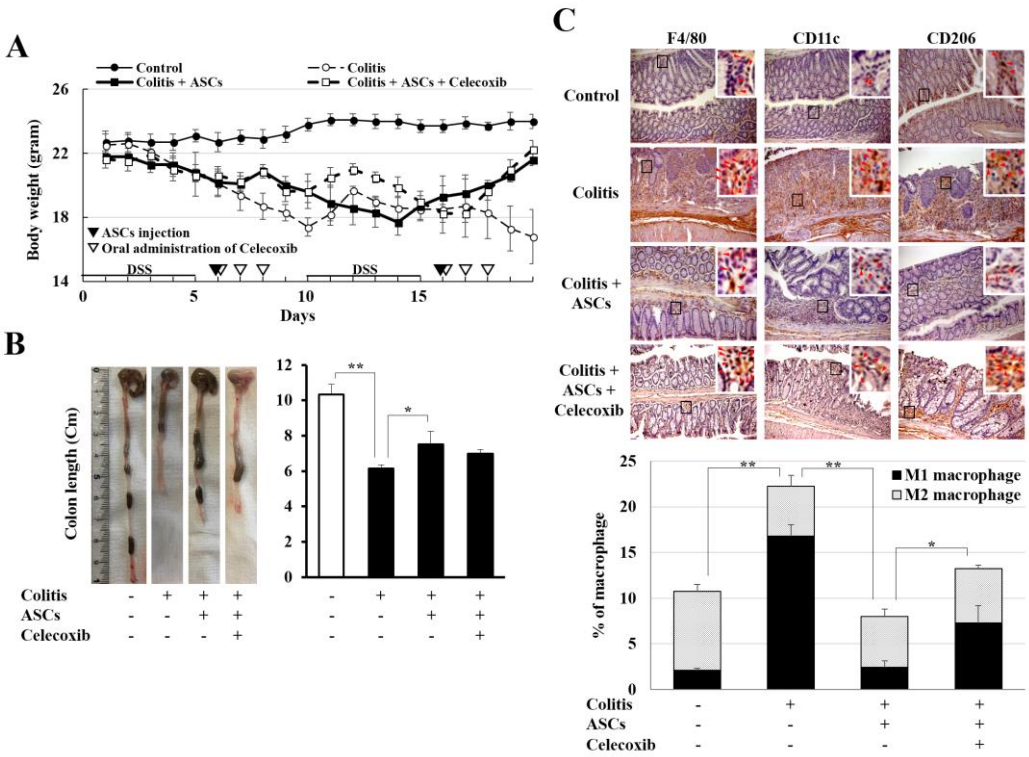


Figure 4. Therapeutic effects of ASCs in DSS-induced colitis mice. A) Study design and change of body weight. B) Colon length of each group (n=5). After immunohistochemical staining of F4/80 for microphages, CD11c for M1-macrophages, or CD206 for M2-macrophages, cells were counted from three random fields, with over 200 cells counted per field. The number of positive cells was expressed as a percentage of the total cell number. Results are expressed as mean±SD; *P<0.05 and **P<0.01. Statistical analysis was performed using a one-way ANOVA.

IV. DISCUSSION

We report that ASCs induced M1 to M2 macrophage transition and increased inflammation related genes including pro-IL-1β, Cox-2, and NLRP3 in THP-1 cells, while they inhibited secretory IL-1β and IL-18. This phenomenon is thought to be PGE2 dependent. In other words, when PGE2 was treated with THP-1 cells, expression of pro-IL-1β, Cox-2, and NLRP3 increased, but secretion of IL-18 was suppressed, similar to THP-1 cells co-cultured with ASCs. In the DSS-induced chronic colitis model, ASCs decreased the frequency of macrophages, especially M1 macrophages. However, in the ASCs + celecoxib group, the frequency of M1 macrophages increased compared to the ASCs group. These results suggest that PGE2, which is remarkably produced by co-culture of ASCs and THP-1, induces M1-M2 transition of macrophages and reduces the M1 population, thereby contributing to the relief of IBD-like inflammatory diseases.

Normally, macrophages have a central role in the epithelial barrier as well as in maintenance of mucosal homeostasis. Macrophages have an opposing 'classically activated' phenotype with proinflammatory microbicidal properties (M1) and ‘alternatively activated’ phenotype with anti-inflammatory properties (M2) that vary according to environmental cues [33, 34]. Previous studies indicate that

macrophages infiltrate the mucosa and submucosa more in IBD than in controls, with a shift toward the proinflammatory state of macrophage subpopulations (M1), indicating that macrophages have an important role in the pathogenesis of IBD [35].

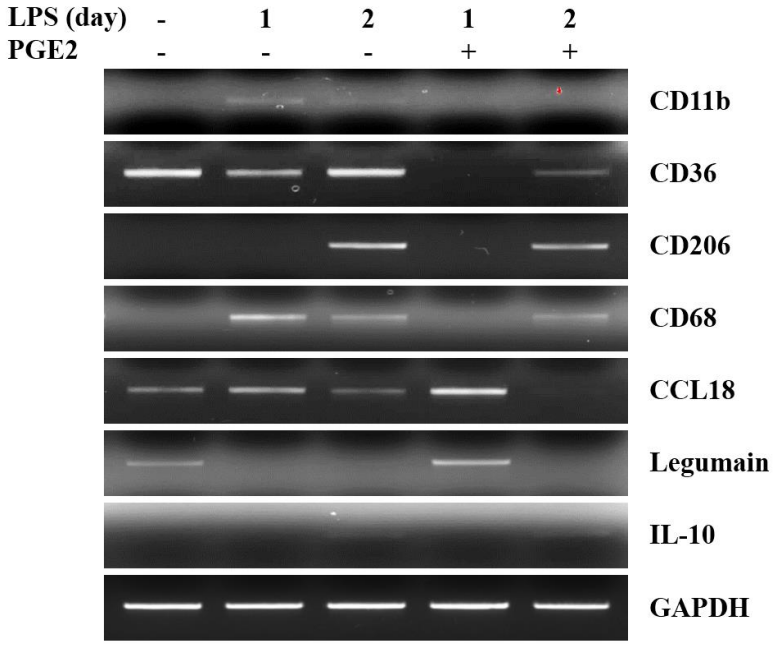
PGE2 has been reported to have dual pro-inflammatory and anti-inflammatory actions [36] and to regulate innate immunity via macrophages, neutrophils, or natural killer cells [36-38]. In macrophages, PGE2 promotes M2 polarization via the cAMP/CREB signaling pathway [39, 40]. Our results also showed that production of PGE2 and expression of M2 markers significantly increased in THP-1 cells when ASCs and THP-1 cells were co-cultured. However, THP-cells treated with exogenous PGE2 and LPS down-regulated the expression of M1 markers rather than increased expression of M2 markers (Suppl Fig. 1). Moreover, in the DSS-induced chronic colitis model, ASCs decreased the total number of macrophages, specifically the number of M1 macrophages. When the Cox-2 inhibitor celecoxib was administered, M1 macrophages again increased significantly, and colon length was increased compared to the ASC-injected group. Interestingly, changes in the M2 macrophage population in colitis, colitis + ASCs, and colitis + ASCs + celecoxib were not observed. These results suggest that ASCs or PGE2, which is induced by ASCs, regulates M1 macrophage populations in addition to the M1 to M2 transition. Regulation of M1 macrophage populations

may be more important in mitigating the inflammatory response in the in vivo system.

In addition, PGE2 has been reported to inhibit the nucleotide-binding domain and leucine-rich repeat–containing protein (NLR)P3 inflammasome activation in human primary macrophages and to decrease IL-1β secretion [41]. In macrophages, NLRP3 is predominantly expressed as an inflammasome sensor molecule. It creates the inflammasome by forming multiprotein oligomers with adaptor protein ASC and Caspase-1. Activation of the inflammasome promotes maturation of inflammatory cytokines IL-1β and IL-18 and causes their secretion from cells. Similarly, we found that PGE2 inhibits NLRP3 inflammasome formation and decreases IL-18 secretion in THP-1 cells. Nevertheless, expression of NLRP3 and pro-IL-1β was markedly increased in THP-1 cells treated with PGE2 or co-cultured with ASCs. Therefore, although PGE2 may increase expression of NLRP3 and pro-IL-1β, suppression of inflammatory cytokine secretion by inhibition of NLRP3 inflammasome formation is considered an important mechanism to control the inflammatory response.

In addition to the previously reported mechanism that PGE2 induces M1 to M2 macrophage transition and inhibits NLRP3 inflammasome activity, PGE2 modulates NLRP3 inflammasome formation or suppresses the M1 macrophage

population to mitigate the inflammatory response in animal systems.



Supplement figure 1. Phenotypic changes of macrophages by PGE2. TPA-treated THP-1 cells were cultured with LPS and exogenous PGE2 for 2 days. A) Changes in M1- and M2-macrophage markers in THP-1 cells. Markers of THP-1 cells were analyzed by RT-PCR. B) Western blot analysis of CD206.

V. CONCLUSION

In conclusion, the present study demonstrates that PGE2 or ASC therapy is expected to be useful for the treatment of inflammatory diseases such as IBD through regulation of macrophage activity.

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국문요약

만성염증성 장질환에서 지방유래 줄기세포의 M1 대식세포의 조절과 inflammasome 형성 억제에 의한 치료효과 연구

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의학과

염증성 장 질환 (IBD)은 유전 적 소인을 가진 개체의 장내 미생물에 대한 조절되지 않는 면역 반응에 의해 발생하는 특발성 질환이다. 따라서, 염증의 완화는 IBD를 치료하는 데 매우 중요하다. 최근, 중간엽줄기세포 (MSC)가 면역조절능력이 우수하다는 근거로 자가면역질환을 치료하기 위한 새로운 후보로 주목 받고있다. 본 연구에서는 THP-1 대식세포 및 덱스트란황산나트륨 (DSS) 유발 만성 대장염 마우스를 이용하여 지방조직유래 중간엽줄기세포 (ASC)의 항염증 메커니즘과 치료 효과를 조사하였다. LPS 처리를 한 THP-1 세포는 2 일째 염증유발 기능을 하는 M1 대식 세포 마커 인 CD11b의

mRNA를 발현이 증가하였다. 그러나 ASC와 공동 배양 한 THP-1 세포에서는 CD206, CD68, CCL18, legumain 및 IL-10과 같은 항염증 기능을 하는 M2 대식 세포 마커가 증가되었다. ASC와 공동 배양 한 THP-1 세포에서는 대표적인 염증성 사이토카인인 IL-1β 및 IL-18의 분비는 유의하게 억제되었으나, 염증성 사이토카인의 전구체인 pro-IL-1beta, Cox-2 및 NLRP3는 LPS 처리 THP-1 세포에 비해 크게 증가했다. 또한, ASC의 공동 배양 조건에서 프로스타글란딘 E2 발현이 증가하였다. 프로스타글란딘 E2의 역할을 알아보기 위해 프로스타글란딘 E2를 처리하였을 때, IL-18 분비가 억제 되며, THP-1 세포의 inflammasome complex (ASC / Cas-1 / NLRP3) 형성이 억제되었다. DSS 유발 만성 대장염 마우스를 이용한 동물실험에서 ASC를 주입하였을 때 총 대식세포의 감소, M1 대식세포의 감소를 보여주었다.

이와 같은 결과 종합하였을 때, ASC는 프로스타글란딘 E2를 통하여 대식세포의 분율 및 염증성 사이토카인의 분비를 조절함으로써 항염증작용을 나타낼 수 있었다. 따라서 ASC는 염증성장질환의 유용한 치료방법으로 사용될 수 있다고 생각된다.

핵심되는 말: 염증성장질환, 중간엽줄기세포, 대식세포, prostaglandin E2