A Comparative Study of Baby Immature and Adult Shoots of Aloe Vera on UVB-Induced Skin Photoaging in vitro

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INTRODUCTION

Ultraviolet (UV) irradiation induces photo-damage of the skin, which in turn causes depletion of the dermal extracellular matrix and chronic alterations in skin structure. Skin wrinkle formations are associated with collagen synthesis and matrix metalloproteinase (MMP) expression. The production of type I procollagen is regulated by transforming growth factor-β1 (TGF-β1) expression; the activation of MMP is also correlated with an increase of interleukin-6 (IL-6). Aloe barbadensis M. (Aloe vera) is widely used in cosmetic and pharmaceutical products. In this study, we examined whether baby aloe shoot extract (BAE, immature aloe extract), which is from the one-month-old shoots of Aloe vera, and adult aloe shoot extract (AE), which is from the four-month-old shoots of Aloe vera, have a protective effect on UVB-induced skin photoaging in normal human dermal fibroblasts (NHDFs). The effects of BAE and AE on UVB-induced photoaging were tested by measuring the levels of reactive oxygen species, MMP-1, MMP-3, IL-6, type I procollagen, and TGF-β1 after UVB irradiation. We found that NHDF cells treated with BAE after UVB-irradiation suppressed MMP-1, MMP-3, and IL-6 levels compared to the AE-treated cells. Furthermore, BAE treatment elevated type I procollagen and TGF-β1 levels. Our results suggest that BAE may potentially protect the skin from UVB-induced damage more than AE. Copyright © 2013 John Wiley & Sons, Ltd.

Keywords: baby aloe (immature aloe); aloe; photoaging; MMPs; procollagen type I; IL-6.

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Ultraviolet (UV) light is composed of UVA (320–400 nm), UVB (290–320 nm), and UVC (100–280 nm). Excessive or repeated exposure to UV irradiation, particularly UVB exposure, is thought to be responsible for most sunburn and damage to the skin. In photoaged skin, the most noticeable changes, such as the degradation of matrix components including type I collagen, elastin, proteoglycans, and fibronectin, are found in the dermis (Fisher et al., 2002; Fisher et al., 2009). The cleavage of collagen by matrix metalloproteinases (MMPs) is not completely reconstituted by de novo collagen synthesis (Quan et al., 2009), another sign of skin photoaging. The degradation of the structural extracellular matrix (ECM) increases expression of MMPs which is known as interstitial collagenase. Cherr et al. reported that induction of interleukin-6 (IL-6) by UVB irradiation has an effect on the protein levels of MMP-1 and MMP-3, which particularly target collagen type I and III. Transforming growth factor-β1 (TGF-β1) is well known as the major regulator of type I procollagen synthesis in the dermal fibroblast (Quan et al., 2002).

Aloe barbadensis M. (Aloe vera), one of the Asphodelaceae plants, has been widely used in functional food supplements, cosmetics, and herbal medicines (Wynn et al., 2005; Djuv and Nilsen, 2012; Shimpo et al., 2002). It has been known for containing polyphenolic structures—aloins, aloemodin, and aloesin—which have been shown to have biological effects, including antioxidant effects (Wamer et al., 2003; Bawankar et al., 2012). Especially, aloesin had been demonstrated to be a competitive inhibitor of tyrosinase and shown to have radical scavenging activity (Jones et al., 2002; Yagi et al., 2002). Many studies have focused on the dermatological effects of Aloe vera, including moisturizing, anti-inflammatory effect, anti-scabies, anti-psoriatic activity, and wound healing (Dal’Belo et al., 2006; Byeon et al., 1998; Oyelami et al., 2009; Dhanabal et al., 2012; John et al., 1995). However, the protective effects of Aloe vera on UV-induced photoaging of the skin have not been reported. Furthermore, there has been no research on the standardization of activity changes in aloe growth patterns. Accordingly, we wanted to investigate the anti-photoaging effects of Aloe vera and its active component, aloesin. Among more than 500 aloe species, only a few, including Aloe vera, have been cultivated. It is grown in hot regions, such as South Texas, USA; Mexico; India; and Africa (Moghaddasi and Verm, 2011). It is also cultivated in Jeju Island, South Korea. In this study, we examined Aloe vera...
from Jeju Island. To maximize the biological activity of aloe to control UVB-induced skin damage, we attempted to determine the protective effect of aloe according to time of growth. First, we measured the standard substance from Aloe vera by using ultra-performance liquid chromatography-quadrupole-time of flight mass spectrometry (UPLC-Q-TOF-MS). Based on result, we created a new type of aloe, which depends on the pattern of time of growth; named baby aloe shoot extract (BAE), which is from the one-month-old shoots of Aloe vera, and compared it with adult aloe shoot extract (AE), which is from the four-month-old shoots of Aloe vera. Second, we found that superoxide dismutase activity is higher in BAE than in AE; LC-MS data showed different types of metabolites, depending on the pattern of time of growth (data not shown).

The aim of this study was to compare the effect of BAE and AE with regard to UVB-induced skin damage in normal human dermal fibroblasts (NHDFs) in vitro. Reactive oxygen species (ROS), MMP-1, MMP-3, IL-6, type I procollagen, and TGF-β1 production in UVB-exposed and non-exposed NHDFs was examined in the presence of BAE and AE. This report is the first to demonstrate the ability of Aloe vera extract to protect the skin against photoaging. Exploration of a new biomaterial such as baby aloe can regulate skin photoaging. It may be used for biomaterials of the development of cosmetics.

MATERIALS AND METHODS

Chemicals. Dulbecco’s modified Eagle’s medium (DMEM), fetal bovine serum (FBS), and penicillin streptomycin were purchased from Gibco BRL (Grand Island, NY, USA). 3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) and dimethyl sulfoxide (DMSO) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Aloesin was provided by Dr. Seon-Gil Do (Univera Co. Seoul, Korea). An ELISA kit for procollagen type I was obtained from Takara (Procollagen Type I C-Peptide EIA Kit; Takara, Shiga, Japan). ELISA kits for MMP-1, MMP-3, IL-6, and TGF-β1 were purchased from R&D Systems (Human Total MMP-1 kit, Human Total MMP-3 kit, Human Total IL-6 kit, and Human Total TGF-β1 kit; R&D Systems, Inc., Minneapolis, MN, USA).

Sample preparation. A dried sample (5 g) was extracted with 400 mL of methanol using a Twist Shaker (BioFree, Korea) for 12 h. After extraction, the extract was centrifuged at 4 °C, 3000 rpm, for 10 min. One milliliter of the supernatant was completely dried with a speed vacuum concentrator (Biotron, Korea) for 12 h. For LC-MS analysis, the dried samples were resolved with 1 mL of methanol and filtered through a 0.2 μm polytetrafluoroethylene filter.

UPLC- Q-TOF-MS analysis. UPLC was performed on a Waters ACQUITY UPLC system (Waters Corp., Milford, MA) equipped with a binary solvent delivery system, an autosampler, and a UV detector. Chromatographic separation was performed on a Waters Acquity HPLC BEH C18 column (100 x 2.1 mm i.d., 1.7 μm particle size). The elution was performed by an acetonitrile (ACN)/water gradient containing 0.1% formic acid. The gradient was linearly increased from 0 to 90% ACN in 12 min, then decreased to 0% over 3 min. Total run time, including re-equilibration of the column to the initial conditions, was 17 min. The injection volume was 5 μL, and the flow rate was 0.3 mL/min. For MS experiments, the Waters Q-TOF Premier (Micromass MS Technologies, Manchester, UK) was operated in a wide pass quadrupole mode with the TOF data being collected between m/z 100 and 1500 in negative and positive ion modes. The desolvation gas (nitrogen) was set to 600 L/h at a temperature of 200 °C, the cone gas (nitrogen) was set to 50 L/h, and the source temperature was set to 100 °C. The capillary and cone voltages were set to 3.0 kV and 40 V, respectively. Data were collected in the centroid mode, with a scan accumulation time of 0.2 s. Compounds were positively identified using authentic compounds by comparing both mass spectra and retention time.

Cell culture. NHDFs were obtained by a skin biopsy on a healthy young male donor (MCTT Core, Inc., Seoul, Korea). The cells were plated in 100-mm tissue culture dishes and cultured in DMEM supplemented with 10% heat-inactivated FBS and 1% penicillin-streptomycin at 37 °C in a humidified atmosphere containing 5% CO2. All experiments were performed using only cells between passages 6 and 10.

UVB irradiation and sample treatment. NHDFs were seeded in 40-mm tissue culture dishes (1.2 x 105 cells). When cells reached over 80% confluence, they were rinsed twice with phosphate-buffered saline (PBS), and all irradiations were performed under a thin layer of PBS. The plate was closed during irradiation. UVB radiation was supplied by a closely spaced array of five Sankyo Denki sunlamps, which delivered uniform radiation at a distance of 7.5 cm. The irradiance (0.1 mW/cm²) was measured using a UVB photometer (IL1700 photometer, International Light). The cells were irradiated with UVB (144 mJ/cm²) for 40 s. Immediately after irradiation, the cells were washed three times with warm PBS, after which 1980 μL of fresh serum-free medium, and 20 μL of sample were added to each well for the indicated time. Control cells were kept in the same culture conditions without UVB exposure. MMP-1, MMP-3, IL-6, type I procollagen and TGF-β1 productions were assessed in the supernatants harvested 72 h after UVB irradiation. For assay of reverse transcription polymerase chain reaction (RT-PCR), cells were harvested 24 h after UVB irradiation.

Measurement of ROS production. After 24 h of UVB irradiation (144 mJ/cm²) and sample treatment, NHDFs were stained with 30 μM 2′,7′-dichlorofluorescein diacetate (DCFH-DA; Sigma-Aldrich) for 30 min at 37 °C in a CO2 incubator. The cells were then analyzed by flow cytometry (FACSCalibur®; Becton-Dickinson, San Jose, CA, USA).

MTT assay. The MTT assay, a colorimetric assay used to measure cell viability, reduces MTT to formazan dyes producing a purple color. After 72 h of incubation, the volume of the medium was reduced to 1 mL, and 100 μL of 1 mg/mL MTT was added to each well. Next, the cells were incubated in the presence of 5% CO2 and 95% O2 at 37 °C for 2 h. The substrate-containing
medium was removed, and 1 mL of DMSO was added to each well to dissolve the formazan crystals. The plates were shaken on an orbital shaker for 30 min at room temperature. The absorbance of 100 μL aliquots was quantified by measuring the absorbance at 570 nm using a microplate reader (Molecular Devices E09090; San Francisco, CA, USA).

**Measurement of MMP-1, MMP-3, and IL-6.** The concentrations of MMP-1, MMP-3, and IL-6 in the medium were determined using commercially available ELISA kits (Human Total MMP-1 kit, Human Total MMP-3 kit, Human Total IL-6 kit; R&D Systems, Inc., Minneapolis, MN, USA) according to the manufacturer’s instructions. Each sample was analyzed in triplicate.

**Measurement of type I procollagen and TGF-β1.** The type I procollagen and TGF-β1 concentrations in the medium were determined using commercially available ELISA kits (Procollagen Type I C-Peptide EIA Kit; Takara, Shiga, Japan; Human Total TGF-β1 kit, R&D Systems, Inc., Minneapolis, MN, USA) according to the manufacturer’s instructions. Each sample was analyzed in triplicate.

**RT-PCR.** Isolation of RNA from NHDF cells treated with sample following UVB irradiation was performed according to the manufacturer’s instructions using TRIzol reagent (Invitrogen Life Technologies, Carlsbad, CA). The RNA (5 μg) was reversibly transcribed with 200 units of reverse transcriptase and 0.5 μg/μL oligo-(dT)₁₅ primer (Bioneer Co., Korea). The reaction was then

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**Figure 1.** UPLC-Q-TOF total ion chromatograms (TICs) of BAE and AE in negative mode. BAE < 10 cm, and AE > 50 cm. Peak 1: aloesin, Peak 2: aloinB, and Peak 3: aloinA (A). Box and whisker plots of significantly different metabolites (p < 0.05) analyzed by UPLC-Q-TOF-MS in different sizes of Aloe vera (Line, mean; box, standard error; whisker, standard deviation) (B). Chemical structures of aloesin (C), aloinB (D), and aloinA (E).
performed at 42°C for 60 min and was terminated by heating to 94°C for 5 min. PCR amplification of the cDNA template was performed using PCR premix (Bioneer) and the following primer pairs: MMP-1, forward 5'-ATT CTA CTG AIA TCG GGG GGG TAT GGG TAT CGC ATC CTG AG-3', reverse 5'-AGG CAG GTG GAC ACC ACC ACC CT-3'; type I procollagen, forward 5'-GAC ATC AAC GAC ATT TAA GGT CT-3'; and glyceraldehyde-3-phosphate dehydrogenase (GAPDH), sense 5'-ACC ACA GTC CAT GCC AC-3', antisense 5'-CCA CCA CCC TGT TGC TGT AG-3'. PCR was performed in a Veriti Thermal Cycler (Applied Biosystems, Foster City, CA, USA) for 30 cycles, respectively. PCR products were separated by 2.0% agarose gel electrophoresis with ethidium bromide staining under UV illumination. GAPDH was used as an internal control. Each experiment was repeated at least three times.

Statistical analysis. All experiments were carried out in triplicate. The data are expressed as means ± SD values. Statistical comparison between different treatments was performed using a one-way analysis of variance followed by Duncan's test. For statistical analysis, a Student's t-test was performed to compare individual treatments to the controls. Statistical significance was set at $p < 0.05$.

RESULTS

Identification of Aloe vera composition

Three secondary metabolites were identified and confirmed using standard compounds of both BAE and AE. They were aloesin (1, m/z 393, ESI-), aloin B (2, m/z 417, ESI-), and aloin A (3, m/z 417, ESI-). As shown in Fig. 1, relatively high contents of aloesin, aloin B, and aloin A are found in BAE than AE. Previous studies reported that phenolic metabolites such as aloin and aloesin were the main antioxidant compounds in various aloe species (Choi and Chung, 2003; Yuko et al., 1990). In particular, aloin A is the main pharmacologically active compound obtained from Aloe vera (Yuko et al., 1990). Aloesin also has been reported to have radical scavenging activity (Yagi et al., 2002).

Effects of BAE and AE on intracellular ROS production in UVB-irradiated NHDFs

NHDFs have been used to investigate UVB-induced production of ROS using FACS analysis. As shown in Fig. 2, UVB-irradiated cells noticeably increased ROS generation compared with non-irradiated cells. The increase of ROS levels was quenched by BAE and AE.

![Figure 2](https://example.com/figure2.png)

Figure 2. Levels of ROS in NHDFs treated as indicated for 24 h were measured by flow cytometry with the DCFH-DA dye. The number of cells is plotted versus the dichlorofluorescin fluorescence detected by the FL-1 channel (A). The relative ROS production of cells appears each histogram (B). Values are means ± SDs. # and * indicate significant differences ($p < 0.05$) between the UV (−) control and UV (+) control, respectively. ## $p < 0.01$ versus the normal control, * $p < 0.05$ versus UVB-irradiated control. This figure is available in colour online at wileyonlinelibrary.com/journal/ptr.
Effects of BAE and AE on MMP-1, MMP-3, type I procollagen, and TGF-β1 productions in cultured human dermal fibroblasts without UVB exposure

The effects of BAE and AE on MMP-1 and MMP-3 secretion were studied in cultured human dermal fibroblasts that were not exposed to UVB. Treatment of cells with BAE and AE at concentrations ranging from 0.1 to 25 μg/mL revealed that BAE and AE were not significantly cytotoxic to human dermal fibroblasts in the same range of concentration compared to the normal group (Fig. 3A and Fig. 3B). The normal group was not irradiated with UVB and was not treated with BAE or AE. Fig. 3 shows the production levels of MMP-1 and MMP-3 in non-irradiated cells after treatment with BAE and AE. We found that cells treated with both BAE and AE decreased MMP-1 secretion compared with normal cells. However, MMP-3 production was not suppressed by BAE and AE treatment. BAE-treated cells increased type I procollagen production, whereas it was decreased in AE-treated cells. Nevertheless, BAE and AE treatment slightly influenced type I procollagen expression. Because TGF-β1 is well known as the major regulator of type I procollagen synthesis, TGF-β1 production in the BAE- or AE-treated NHDFs was determined. Treatment with BAE at a concentration of 25 μg/mL induced greater TGF-β1 production (up to 23%) than did the control group (Fig. 3E). However, AE treatment had not influenced secretion of TGF-β1.

Effects of BAE and AE on MMP-1, MMP-3, type I procollagen, and TGF-β1 productions in UVB-irradiated cultured human dermal fibroblasts

To examine the effects of BAE and AE on UV-damaged skin, we measured MMP-1 and MMP-3 secretion in UVB-irradiated cultured human dermal fibroblasts. BAE and AE treatment showed weak cytotoxicity in the UVB-damaged skin fibroblasts. UVB-irradiated fibroblasts had higher MMP-1 and MMP-3 production than did the untreated cells. As shown in Fig. 4, both BAE and AE treatment decreased secretion of MMP-1 and MMP-3 compared with UVB-irradiated control cells. Additionally, compared to the control, BAE treatment significantly inhibited MMP-1 and MMP-3 production in a dose-dependent manner. Type I procollagen synthesizes type I collagen which is the most abundant structural protein. TGF-β1 is well known as the major regulator of type I procollagen synthesis. Therefore, type I procollagen and TGF-β1 production in UVB-irradiated NHDFs treated with BAE and AE were determined. UVB-irradiated fibroblasts had lower type I procollagen expression than did unexposed cells. However, UVB-irradiated cells treated with BAE...
and AE had higher type I procollagen expression, respectively, compared to untreated cells (Fig. 4D). In particular, treatment with BAE at 25 μg/mL induced greater type I procollagen production (up to 74%) than in UVB-irradiated controls (Fig. 4D). UVB exposure decreased TGF-β1 to approximately 40% of that in the non-irradiated cells. At low concentrations, BAE treatments (0.1 μg/mL) up-regulated TGF-β1 production to levels higher than in untreated cells. However, AE treatment did not influence secretion of TGF-β1 (Fig. 4E).

**Effects of BAE and AE on IL-6 production in UVB-irradiated NHDFs**

The imbalanced induction of MMP-1 is, in part, mediated by pro-inflammatory cytokines, such as IL-6. And it may lead to the loss of dermal interstitial collagen, a hallmark of photoaging. As shown in Fig. 5A, in non-irradiated cells, only BAE treatment showed slight decreasing of IL-6 production. UVB exposure dramatically increased IL-6 production. However, treatment with BAE at 25 μg/mL significantly decreased the level of IL-6 by approximately 45% lower than that of the irradiated cells (Fig. 5B). These results show that BAE treatment decreased UVB-induced production of IL-6 production.

**Figure 4.** Cell viability (A), MMP-1 production (B), and MMP-3 production (C), Type I procollagen level (D), and TGF-β1 production (E) in UVB-irradiated and BAE- and AE-treated cultured human dermal fibroblasts. Cells were irradiated with UVB (144 mJ/cm²) and then incubated in the absence or presence of BAE or AE for 72 h. Values are means ± SDs. # and * indicate significant differences (p < 0.05) between the UV (-) control and UV (+) control, respectively. ### p < 0.01 and #### p < 0.001 versus the normal control, * p < 0.05, ** p < 0.01 and *** p < 0.001 versus UVB-treated control.

**Figure 5.** Interleukin-6 (IL-6) production by cells non-irradiated and treated with BAE or AE (A). IL-6 production by cells exposed to UVB and treated with BAE or AE (B). Cells were incubated in the absence or presence of UVB exposure (144 mJ/cm²) and treated with BAE or AE for 72 h. Values are means ± SDs. # and * indicate significant differences (p < 0.05) between the UV (-) control and UV (+) control, respectively. ### p < 0.001 versus the normal control, ** p < 0.01, and *** p < 0.001 versus UVB-treated control.
Effects of aloesin on mRNA expressions of MMP-1 and type I procollagen in UVB-irradiated NHDFs

To examine the mRNA levels of aloesin-treated cells (24 h after treatment), MMP-1 and type I procollagen were measured in UVB-irradiated cultured NHDFs. To quantify the data, the ratio of MMP-1/GAPDH and type I procollagen/GAPDH in normal cells was arbitrarily set to 1.0, based on band signal intensity (Fig. 6B and Fig. 6C). As expected, RT-PCR data revealed that UVB irradiation elevated the mRNA levels of MMP-1 and inhibited the mRNA expression of type I procollagen within 24 h. As shown in Fig. 6C, aloesin-treated cells was strong increased the mRNA expression of type I procollagen reduced by UVB. However, RT-PCR data showed that the cells treated with aloesin was relatively weak due to the UVB-induced mRNA accumulation of MMP-1. Accordingly, this aloesin may enhance type I procollagen production through its transcriptional modulation.

DISCUSSION

*Aloe vera* has been used frequently in folk medicine. Studies on *Aloe vera* have focused on sunburns, skin dryness, and wound healing (Dal’Belo *et al*., 2006; Byeon *et al*., 1998; Atiba *et al*., 2011). The anti-inflammatory and antioxidant activities of aloe on the skin are well established (Tomasin and Gomes-Marcondes, 2011). However, there have been no studies on the relationship of *Aloe vera* and skin aging. In addition, there are no studies on the effect of aloe according to time of growth on skin photoaging. In recent years, many studies have been done to search for safe and natural ways to regulate photoaging, including wrinkles, pigmentation, and roughness (Fisher *et al*., 2000). This led us to a comparative study of AE and BAE on UVB-induced photoaging in dermal fibroblasts.

Repetitive or acute exposure of skin to UV radiation is known to induce premature skin aging, which is called photoaging (Park *et al*., 2010). Exposure to UV radiation affects the phenotype of embedded cells, including keratinocytes, fibroblasts, and dendritic cells, either through the direct effects of irradiation on the cells or indirectly by remodeled and aged ECM. Generation of ROS is thought to play a major role in the process of skin photoaging, which eventually leads to increased collagen breakdown (Han *et al*., 2010). In this study, it was shown that ROS production was increased by UVB irradiation. The increase of ROS production by UVB irradiation was quenched by both BAE and AE treatment. Besides, BAE is more potent in diminishing ROS production than that of AE. Collagen, synthesized procollagen, is one of the main building blocks of human skin. Photoaged skin shows down-regulation of procollagen production as well as induced synthesis of several MMPs, including MMP-1, MMP-3, and MMP-9, in fibroblasts (Fisher *et al*., 2009). Specifically, MMP-1 and MMP-3, responsible for the breakdown of dermal collagen, were significantly increased by UVB irradiation.
interstitial collagen and proteoglycans, are induced by UV irradiation (Brenneisen et al., 1996). It has been reported that UV irradiation contributes to the synthesis of cytokines such as IL-1 and IL-6, which increases MMP-1 in dermal fibroblasts (Onodera et al., 2000). Cherng et al. reported that secretion of IL-6 increases MMP-1 and MMP-3 production in UVB-irradiated fibroblasts (Wlaschek et al., 1997; Cherng et al., 2012). Elevating of MMP-1 and MMP-3 production by UVB irradiation has caused skin photoaging. The regulation of two major MMPs, interstitial collagenase (MMP-1), and stromelysin-1 (MMP-3) depends on the UV wavelength. Namely, these are the leading factor for regulation of skin photoaging (Brenneisen et al., 1996). In this study, we found that UVB-induced activation of MMP-1 and MMP-3 was inhibited by BAE and AE treatment. After UVB exposure, treatment of BAE at a high concentration (25 μg/mL) significantly decreased both MMP-1 and MMP-3 production. However, BAE-treated cells without UVB irradiation do not influence MMP-1 and MMP-3. In other words, cells treated with BAE following UVB irradiation had lower MMP-1 and MMP-3 production than those treated cells that were not exposed to UVB irradiation. Fig. 5 shows that BAE treatment suppressed UVB-induced IL-6 production, but AE treatment had no effect on it. These results demonstrate that activation of BAE is more potent in UVB-irradiated cells than in normal cells. However, the intermediate molecular steps from IL-6 release to MMP-1 and MMP-3 induction after UVB irradiation are still unclear.

TGF-β1 and activator protein-1 (AP-1) are two important regulators of collagen production. TGF-β1 promotes procollagen formation, while AP-1 demotes procollagen by promoting MMPs (Massague, 1998). When skin is exposed to UVB radiation, production of procollagen is decreased due to down-regulation of TGF-β1 expression (Park et al., 2011). Our earlier studies showed that royal jelly (RJ) and the rind of Punica gransatum (PG) had the protective effects against UVB-induced skin damage in NHDFs through the up-regulation of collagen expression. UVB-irradiated cells treated with 1 μg/mL RJ and 1000 μg/mL PG increased type I procollagen production (up to 64.6% and up to 20%, respectively) (Park et al., 2011; Park et al., 2010). In this study, we found that UVB irradiation led to decreased production of type I procollagen and TGF-β1 in NHDFs. Specifically, BAE-treated cells (25 μg/mL) increased production of type I procollagen (up to 74%) as well as TGF-β1 (up to 30%), whereas AE-treated cells did not (Fig. 4). These results indicate that BAE may have more strong protective effects against UVB-induced skin photoaging than that of RJ and PG. However, in the present study, we did not explore the activation of AP-1, one of regulators of collagen production, so we will investigate the mechanisms of these effects of Aloe vera in future studies.

Photoaged skin generally results from the production of ROS, which leads to increase in MMP-1 by activating of AP-1 (Wlaschek et al., 2001). Previous studies reported that phenolic metabolites such as aloin and aloesin were the main antioxidant compounds in various aloe species (Choi and Chung, 2003; Yagi et al., 2002). In this study, we examined whether aloesin, which is the active component of Aloe vera has a protective effect on UVB-induced skin photoaging in NHDFs. We measured the mRNA expressions of type I procollagen and MMP-1 using RT-PCR in aloesin-treated cells after UVB irradiation. Aloesin showed significantly increasing type I procollagen at the mRNA levels. Because BAE has higher aloesin than AE, it seems that the inhibitory effect of BAE on photoaging was stronger than that of AE in NHDFs. BAE might be a stronger regulator for the control of photo-damage caused by UVB radiation than AE based on our results. This finding supports that BAE and aloesin as new material protects skin damage with UVB exposure. Also, it will be available not only in the cosmetics industry but also in the herbal medicine industry. Although we did UPLC-Q-TOF-MS analysis to determine differences in the content of BAE and AE, we did not determine the all structures of the active constituents that inhibit UVB-induced skin damage in Aloe vera. Furthermore, we did not explore all mechanisms in this study, so further research is warranted to find the mechanisms of Aloe vera responsible for regulation of UVB-induced skin damage. Also, further investigations are needed to support if BAE and aloesin can also protect against UVB-induced skin photoaging in the model of artificial skin or skin tissue ex vivo.

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Conflict of Interest

The authors declare that there are no conflicts of interest.

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