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Short Communication

Effects of various flavonoids isolated from *Scutellaria baicalensis* roots on skin damage in acute UVB-irradiated hairless mice

Yoshiyuki Kimura[a] and Maho Sumiyoshi[b]

[a]Division of Biochemical Pharmacology, Department of Basic Medical Research, and [b]Division of Functional Histology, Department of Functional Biomedicine, Ehime University Graduate School of Medicine, Shitsukawa, Toon City, Ehime, Japan

Abstract

Objectives Solar ultraviolet (UV) radiation causes skin damage including increasing skin thickness, edema and flush. *Scutellaria baicalensis* roots have been traditionally used as a remedy for allergic inflammatory diseases in China and Japan. In this study, we examined the effects of four flavonoids isolated from these roots, namely 2′,5, 5′,7-tetrahydroxy-6′,8-dimethoxyflavone (**1**), skullcapflavone II (**2**), 2(*S*)-2′,5,6′,7-tetrahydroxyflavanone (**3**) and 2(*R*), 3(*R*)-2′,3,5,6′,7-pentahydroxyflavanone (**4**), on acute UVB irradiation-induced skin damage in hairless mice.

Methods The four flavonoids were orally administered twice daily, at doses of 10 and 50 mg/kg, for 14 consecutive days. The UVB irradiation was performed at a dose of 200 mJ cm^{-2} on days 7 and 8 after beginning oral administration of the four flavonoids.

Key findings Compounds **1** and **4** prevented increases in skin thickness, levels of matrix metalloproteinases (MMP)-2 and MMP-9, and vascular endothelial growth factor (VEGF) induced by UVB irradiation. The other two flavonoids **2** and **3** had no effect.

Conclusions Compounds **1** and **4** isolated from *Scutellaria baicalensis* roots may be useful for preventing skin inflammation induced by acute UVB irradiation.

Keywords flavonoids; MMP; *Scutellaria baicalensis*; ultraviolet B; VEGF

Introduction

Since ancient times, the roots of *Scutellaria baicalensis* Georgi (Labiatae) have been used to treat allergic and inflammatory diseases in China and Japan. Kimura and co-workers reported that baicalein and its related new flavanones and flavones have antibacterial,[1] anti-lipid peroxidative[2,3] and anti-arthritis[4] activities. They also inhibit 5-lipoxygenase activity in leukocytes[5–8] and inhibit adhesion molecule expression and plasminogen activator inhibitor-1 (PAI-1) production induced by inflammatory cytokines (TNF-α and IL-1β) in human umbilical vein endothelial cells (HUVECs).[9–13] It is well established that the symptoms of skin ageing, such as wrinkles and pigmentation, develop earlier in sun-exposed skin than in unexposed skin: a phenomenon referred to as photoageing. Ultraviolet (UV) B radiation is an important environmental factor because of its hazardous effect on health, which includes the generation of skin cancers,[14] suppression of the immune system[15] and premature skin ageing.[16] In a series of studies on the effects of natural products on UV-induced skin damage, we found that the oral administration of turmeric extract, and an olive leaf extract and its component oleuropein prevented increases in skin thickness induced by acute or chronic UVB irradiation in mice.[17–19] Recently, we also reported that baicalein and wogonin isolated from *S. baicalensis* roots inhibited the UVB-irradiation-induced skin damage by suppressing increases in the levels of metalloproteinase (MMP-9) and vascular endothelial factor (VEGF).[20] The effects of four flavonoids (skullcapflavone II, 2(*S*)-2′,5,6′,7-tetrahydroxyfalvanone, 2*(R)*,3(*R)*-2′,3,5,6′,7-pentahydroxyflavanone and 2′,5,5′,7-tetrahydroxy-6′,8-dimethoxyflavone) isolated from these roots on inflammation of the skin induced by acute UVB irradiation are not yet clarified. In this study we examined the effects of the oral administration of the four flavonoids on inflammation of the skin induced by acute UVB irradiation in hairless mice.

Correspondence: Yoshiyuki Kimura, Division of Biochemical Pharmacology, Department of Basic Medical Research, Ehime University Graduate School of Medicine, Shitsukawa, Toon City, Ehime 791-0295, Japan.
E-mail: yokim@m.ehime-u.ac.jp

Materials and Methods

General experimental procedures

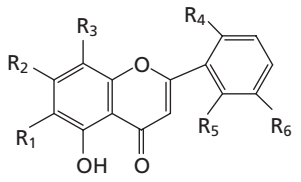
Melting points, determined using a Yamato (Tokyo, Japan) MO-21 capillary apparatus, were determined. IR, UV and ORD spectra were measured using a Shimadzu (Kyoto, Japan) IR-400 spectrometer and a JASCO (Tokyo, Japan) ORD/UV-5 spectrometer, respectively. ^{1}H NMR (499.83 Hz) spectra were recorded in DMSO-d_6, $CDCl_3$ and solvent + D_2O using a Varian (Palo Alto, CA, USA) Unity Inova 500 spectrometer. Column chromatography was performed using silica gel 60 (70–230 mesh, ASTM; Merck, Germany) as adsorbent.

Materials

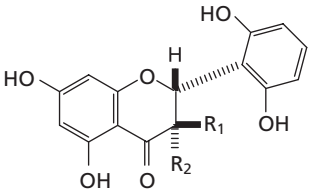
The dried roots of *Scutellaria baicalensis* Georgi (Labiatae) were purchased from Mikuni Co. (Osaka Japan). The four flavonoids, 2′,5,5′,7-tetrahydroxy-6′8-dimethoxyflavone (**1**), skullcapflavone II (**2**), 2(*S*)-2′,5,6′,7-tetrahydroxyflavanone (**3**) and 2(*R*),3(*R*)-2′,3,5,6′,7-pentahydroxyflavanone (**4**) were isolated from the ethylacetate and the methanol extracts of the dried roots of *S. baicalensis* according to a method used in previous studies.[2,3] The structures of the four flavonoids are shown in Figure 1. The four flavonoids (**1**, **2**, **3** and **4**) were analysed by HPLC (GLLIVER-HPLC System, JASCO Co., Tokyo, Japan) under the following conditions: mobile phase, solvent (A) 0.1% trifluroacetic acid and (B) acetonitrile; gradient profile, 0–25 min 20% acetonitrile; 25–30 min 70% acetonitrile. The column was a TSK-Gel ODS-120T column (150 × 4.6 mm ID) (TOSO, Co. Tokyo, Japan), the UV detector was at 280 nm, and the column temperature was 35°C. All chemicals used in this study were of reagent grade and purchased from Wako Pure Chemical Co. (Osaka, Japan). A mouse VEGF enzyme-linked immunosorbent assay (ELISA) kit and tissue protein extraction reagent were purchased from R&D Systems (Minneapolis, MN, USA) and Pierce Co. (Rockford, IL, USA), respectively.

Compound 1

Yellow needles from *n*-hexane and ethylacetate mixture, mp 250°C (decomp.). IR ν (nujol, max) cm^{-1}: 3500–3150 (OH),



2′, 5, 5′,7-Tetrahydroxy-6′,8-dimethoxyflavone (**1**): R_1=H, R_2=R_4=R_5-OH, R_3=R_6=OCH_3
Skullcapflavone II (**2**): R_1=R_2=R_3=R_4=OCH_3, R_5=H, R_6=OH



2(*S*)-2′,5,6′,7-Tetrahydroxyflavanone (**3**): R_1=R_1=H
2(*R*), 3(*R*)-2′,3,5,6′7-Pentahydroxyflavanone (**4**): R_1=OH, R_2=H

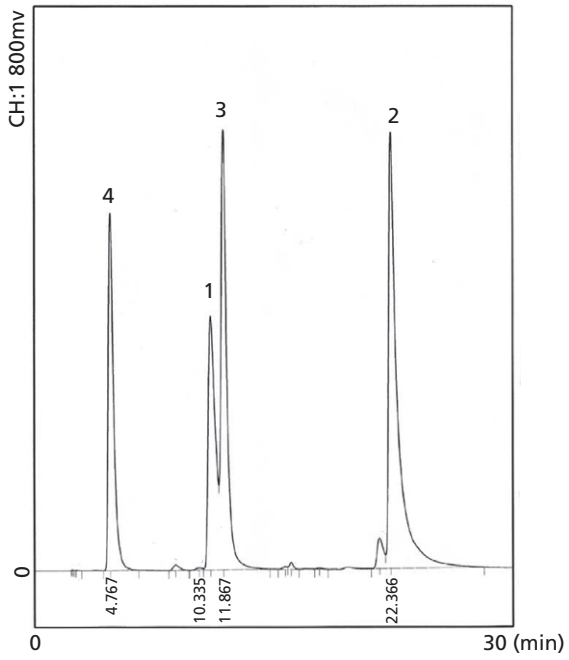


Figure 1 The structure of four flavonoids isolated from *S. baicalensis* roots and their HPLC profiles.

1650 (chelated C=O, 1610, 1580, 1570 (aromatic ring). UV λ (EtOH, max) nm (log ε): 261 (4.48), 305 sh (3.92), 337 (3.90). ^{1}H NMR (in DMSO-d_6) δppm: 3.74, 3.77 (3H each singlet, -OCH_3), 6.30, 6.35 (1H, each singlet, C3-H and C6-H), 6.61, 6.94 (1H each doublet, J = 9.0 Hz, C3′-H and C4′-H), 9.0, 9.40, 10.72 (^{1}H each broad singlet, OH), 12.56 (1H, singlet, C5-OH).

Compound 2

Yellow prisms from ethylacetate, mp 180°C. IR ν (nujol, max) cm^{-1}: 3200–3100 (OH), 1650 (C=O), 1600, 1560 (aromatic ring). UV λ (EtOH, max) nm (log ε): 270 (4.46). ^{1}H NMR (in DMSO-d_6) δppm: 3.76, 3.80, 3.82, 4.00 (3H, each singlet, -OCH_3), 6.22 (1H, singlet, C3-H), 6.56 (2H, doublet, J = 8.0 Hz, C3′-H and C5′-H), 7.24 (1H, triplet, J = 8.0 Hz, C4′-H), 10.0 (^{1}H, broad singlet, C2′-OH), 12.67 (1H, singlet, C5-OH).

Compound 3

Colorless prism from *n*-hexane and ethylacetate mixture, mp.240°C (decomp.). IR ν (nujol, max) cm^{-1}: 3450, 3200 (OH), 1640 (chelated C=O), 1610, 1517 (aromatic ring). UV λ (EtOH, max) nm (log ε): 289 (4.21). ^{1}H NMR (in DMSO-d_6) δppm:2.35 (1H, doublet, J = 4.0 Hz, C3-H), 3.40 (1H, quartet, J = 14.0, 17.0 Hz, C3-H), 5.83, 5.87 (1H each doublet, J = 2.5 Hz, aromatic H), 5.84 (1H, quartet, J = 14.0, 4.0 Hz, C2-H), 6.32 (2H, doublet, J = 9.0 Hz, aromatic H), 6.98 (^{1}H, triplet, J = 9.0 Hz, aromatic H), 9.48 (2H, singlet, OH), 12.24 (^{1}H, singlet, C5-OH). CD (c = 0.126, CH_3OH) $[\theta]^{25}$ (nm): –29000 (284), +7800 (306), +9400 (327).

Compound 4

Colorless needles from CH_3OH, mp. 221–225°C (decomp.). IR ν (nujol, max) cm^{-1}: 3460, 3250 (OH), 1640 (chelated C=O), 1615, 1510 (aromatic ring). UV λ (EtOH, max) nm (log ε): 292 (4.21), 326 sh (3.66). ^{1}H NMR (in DMSO-d_6) δppm: 5.31, 5.62 (1H, each doublet J = 12.0 Hz, C2,3-H), 5.19–5.98 (1H, broad singlet, C3-OH) 5.83, 5.90 (1H, each doublet, J = 1.5 Hz, aromatic H), 6.34 (2H, doublet, J = 8.5 Hz, aromatic H x 2), 6.96 (1H, triplet, J = 8.5 Hz, aromatic H), 9.58 (2H, broad singlet, OH × 2), 10.73 (1H, broad singlet, OH), 12.11 (1H, singlet, C5-OH).

Animals

Male albino hairless HOS:HR-1 mice (5 weeks old) were purchased from Hoshino Laboratory Animal Co. Ltd (Saitama, Japan), housed for 1 week in a temperature-controlled room at 25 ± 1°C and 60% relative humidity, and given free access to standard laboratory diet and water during this experiment. Mice were treated according to the Ethical Guidelines of the Animal Center, Graduate School of Medicine, Ehime University and the Japanese Pharmacological Society, and guide for the care and use of laboratory animals of the National Institutes of Health. The experimental protocol was approved by the Animal Studies Committee of Ehime University.

Acute UVB-induced skin damage

A UVB lamp (15 W; maximum wavelength 312 nm; intensity 100 μW/cm^2; Ieda Boueki Co., Tokyo, Japan) was used to examine the effects of the four flavonoids isolated from *S. baicalensis* roots on skin thickness and elasticity following acute irradiation. The period of irradiation was varied to control the amount of UVB energy applied to the dorsal region of each animal. The minimal erythema dose was about 36 mJ/cm^2. The four flavonoids were orally administered twice daily, for 14 consecutive days, at doses of 10 or 50 mg/kg. The UVB irradiation was performed at a dose of 200 mJ/cm^2 on days 7 and 8 after beginning oral administration of four flavonoids. Skin thickness was assessed by measuring skin-fold thickness by the described methods.[18,21] Briefly, the dorsal skin of the hairless mice was lifted up by pinching gently under anesthetization with pentobarbital. Skin-fold thickness was measured using a Quick Mini caliper (Mitsutoyo Co., Kanagawa, Japan). Ear thickness was measured by pinching gently using a Quick Mini caliper. Skin and ear thickness after UVB irradiation were measured every other day. On day 15, the mice were killed by cervical dislocation and irradiated skin was removed for analysis.

Skin vascular endothelial growth factor (VEGF) and matrix metalloproteinase (MMP) analysis

The removed skin tissue (about 100 mg) was washed in phosphate-buffered saline (PBS, pH 7.0) and cut into small pieces, and then tissue protein extraction reagent containing protease inhibitor (Pierce Co., Rockford, IL, USA) (2 ml) was added to the skin and the mixture was homogenized. The skin homogenate was centrifuged at 2000*g* for 10 min at 4°C. The VEGF content of the supernatant was determined using a VEGF ELSA kit (R&D Systems, Minneapolis, MN, USA). The MMP-2 (active and inactive forms) and MMP-9 (active and inactive form) in the supernatant were separated by electrophoresis on a 7.5% sodium dodecyl sulfate polyacrylamide gel containing 0.1% gelatin under non-reducing conditions. The gel was then washed with 50 mM Tris-HCl (pH 7.5) containing 100 mM NaCl and 2.5% Triton X-100 for 1.5 h, and incubated in 50 mM Tris-HCl containing 10 mM $CaCl_2$ and 10 μM $ZnCl_2$ at 37°C for 20 h. It was stained with 0.25% Coomassie Brilliant Blue 250 and the unstained gelatin-degraded zone was quantified using NIH Image J 1.36.

Histological examination

Dorsal skin samples (about 3 cm^2) were fixed in 10% buffered formalin for at least 24 h, progressively dehydrated in solutions containing an increasing percentage of ethanol (70, 80, 95 and 100%, v/v), cleared in Histoclear (AS-ONE, Tokyo, Japan), embedded in paraffin under vacuum, cut into sections 5-μm thick, deparaffinized, and then stained with hematoxylin-eosin (HE) and Azan stain. After three cross-sections per sample were selected, four different microscopic fields (40×, 100×, or 200×) per plate were photographed. The thickness of the epidermis and dermis were measured using a Digimatic caliper.

Statistical analysis

All values are expressed as means ± SE. Data were analysed by one-way ANOVA or repeated-measures ANOVA. When the *F*-test was significant, means were compared using

Tukey–Kramer or Dunnet's test with Stat View (SAS Institute Inc., Tokyo, Japan). Differences were considered significant at $P < 0.05$.

Results

Effects of flavonoids on skin and ear thickness

Skin and ear thickness increased significantly during days 2 to 7 after irradiation, compared to unexposed mice (normal) (Figure 2). The oral administration of compounds **1** (50 mg/kg, twice daily), **3** (50 mg/kg) and **4** (10 and 50 mg/kg) significantly inhibited the increase in skin thickness, at days 2 to 7, compared to the controls. But compounds **1** (10 mg/kg), **2** (10 and 50 mg/kg) and **3** (10 mg/kg) had no effect on the increase in skin thickness induced by UVB irradiation (Figure 2a and b). Ear thickness also increased during days 0 to 7 after UVB irradiation (Figure 2c and d). The increase in ear thickness after UVB irradiation was significantly inhibited by the oral administration of **1** (50 mg/kg), **2** (50 mg/kg) and **4** (10 and 50 mg/kg). But compounds **1** (10 mg/kg), **2** (10 mg/kg) and **3** had no effect on the increase in ear thickness induced by UVB irradiation (Figure 2c and d).

Effects of four flavonoids on the thickness of epidermis and extracellular matrix of the dermis

The thickness of epidermis and ECM in the dermis was increased by acute UVB irradiation. Compound **4** (10 and 50 mg/kg) significantly inhibited the increase in thickness of the epidermis induced by UVB irradiation compared with the control (Figure 3a and b). Compounds **1** (10 and 50 mg/kg), **2** (10 and 50/mg kg) and **3** (10 and 50 mg/kg), had no effect on the increase in the thickness of epidermis (Figure 3a). The increase in the thickness of ECM in the dermis was inhibited by the oral administration of compounds **1** (50 mg/kg) and **4** (10 and 50 mg/kg). Compounds **2** (10 and 50 mg/kg) and **3** (10 and 50/mg kg) had no effect.

Effects of four flavonoids on the expression of MMP-2, MMP–9 and VEGF

The expression of pro-MMP-2 (inactive form), MMP-2 (active form), pro-MMP-9 (inactive form) and MMP-9 (active form) in acute UVB-irradiated mice (control) was significantly greater than that of non-UVB-irradiated mice (normal) (Figures 4 and 5). Compounds **1** (10 and 50 mg/kg) and **2** (10 and 50 mg/kg) significantly inhibited the increase in MMP-9

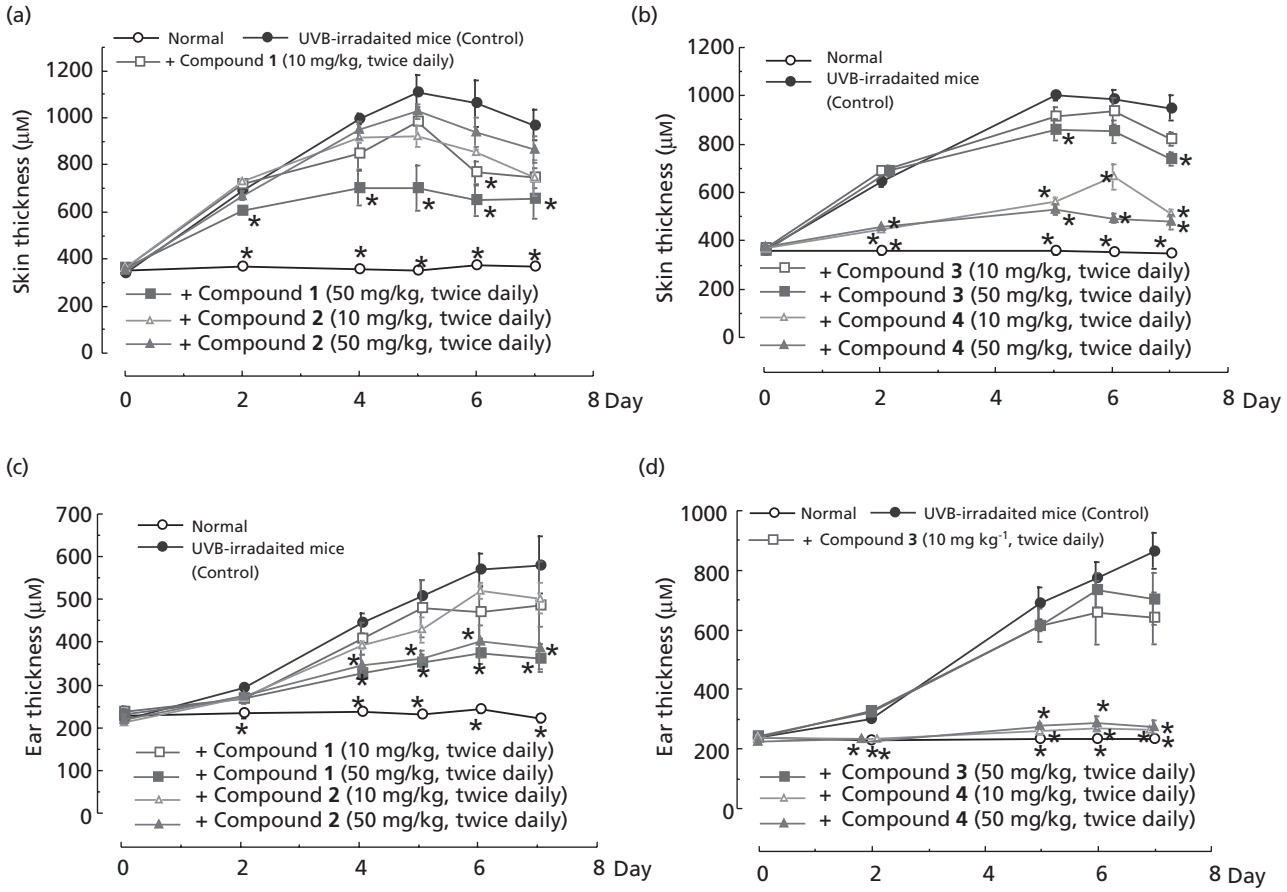


Figure 2 Effects of 2′,5,5′,7-tetrahydroxy-6′,8-dimethoxyflavone (**1**), skullcapflavone II (**2**), 2(*S*)-2′,5,6′,7-terahydroxyflavanone (**3**) and 2(*R*),3(*R*)-2′,3,5,6′,7-pentahydroxyflavanone (**4**) on skin thickness (A and B) and ear thickness (C and D) in acute UVB-irradiated mice. Values are the mean ± SE for six mice. *Significantly different from UVB-irradiated mice (control), $P < 0.05$.

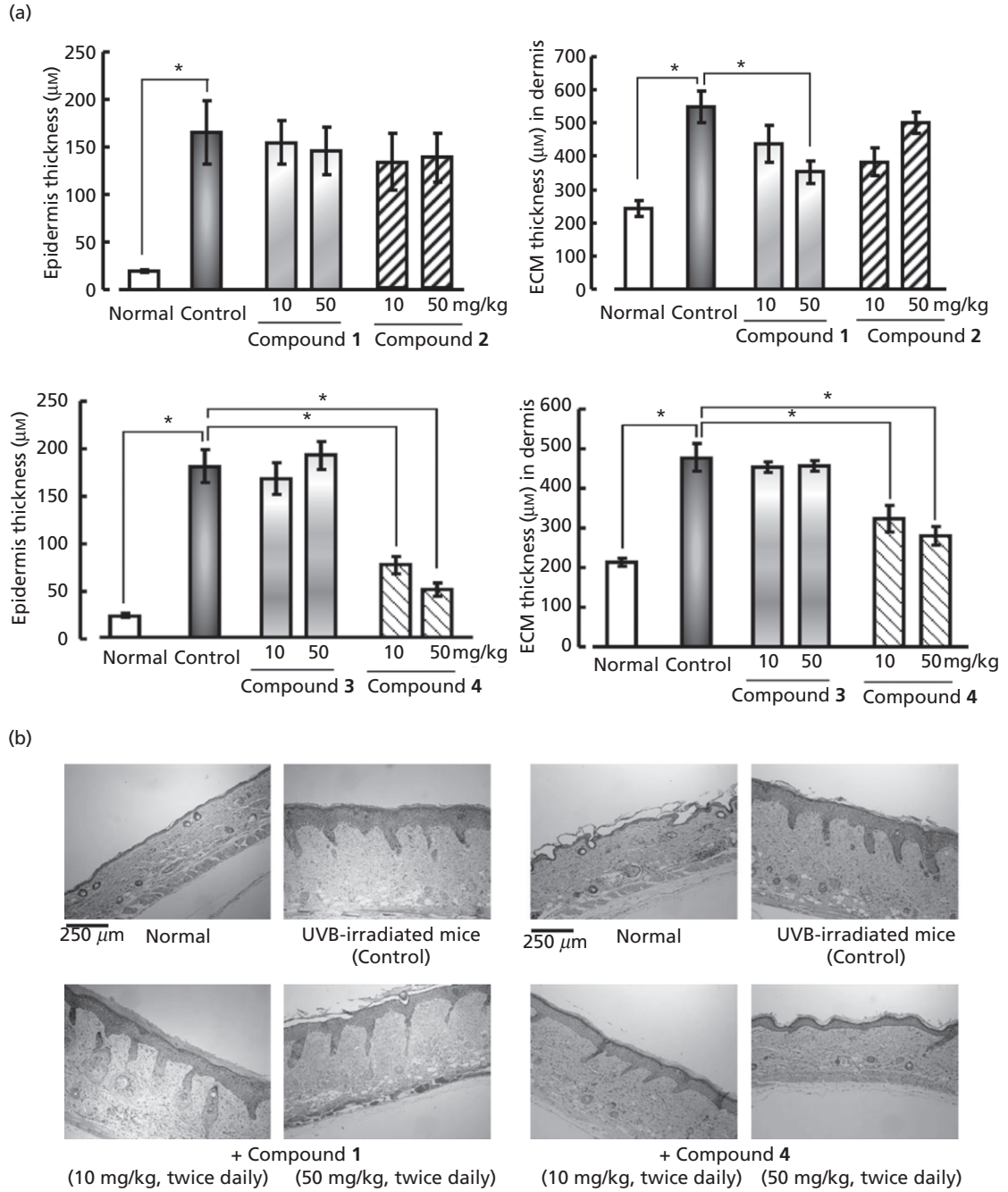


Figure 3 (A) Effects of compounds **1**, **2**, **3** and **4** on the thickness of epidermis and extracellular matrix of the dermis in acute UVB-irradiated hairless mice. Values are the mean ± SE for six mice. *Significantly different from UVB-irradiated mice (control), $P < 0.05$. (B) Light micrographs of cell stained with hematoxylin-eosin to show the thickness of epidermis and dermis in normal mice, vehicle-treated acute UVB-irradiated mice (control), compounds **1**- and **4**-treated UVB-irradiated mice.

expression compared with the control. But these flavones had no effect on the increase in pro-MMP-9 expression (Figure 4). Compounds **3** (50 mg/kg) and **4** (10 and 50 mg/kg) inhibited the increase in pro-MMP-9 and MMP-9 expression compared with the control (Figure 5). The increase in pro-MMP-2 in the control was inhibited by compound **2** (10 mg/kg), but compounds **1** (10 and 50 mg/kg) and **2** (50 mg/kg) did not affect it (Figure 5). Compounds **3** (50 mg/kg) and **4** (10 and 50 mg/kg) inhibited the increase in pro-MMP-2 expression compared with the control (Figure 5). The four flavonoids had no effect on the increases in MMP-2 induced by UVB irradiation (Figures 4 and 5).

The VEGF content of the skin was also significantly increased by acute UVB irradiation. Compounds **1** (10 and 50 mg/kg) and **4** (10 and 50 mg/kg) significantly inhibited this increase, but **2** and **3** did not affect it (Table 1).

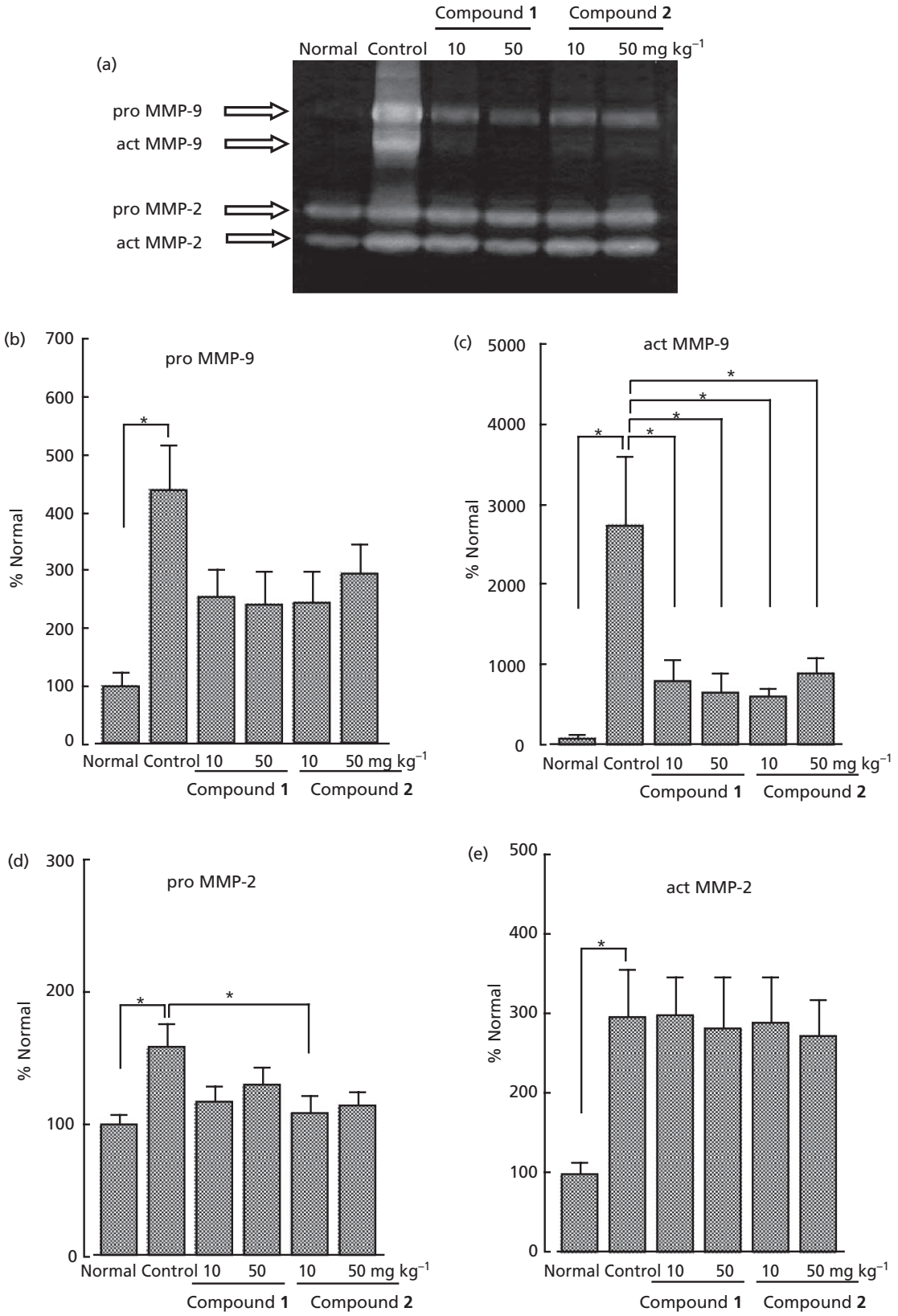


Figure 4 Effects of compounds **1** and **2** on pro-MMP-9, MMP-9, pro-MMP-2 and MMP-2 expression in the skin of acute UVB-irradiated mice. Values are the mean ± SE for six mice. *Significantly different from UVB-irradiated mice (control), $P < 0.05$.

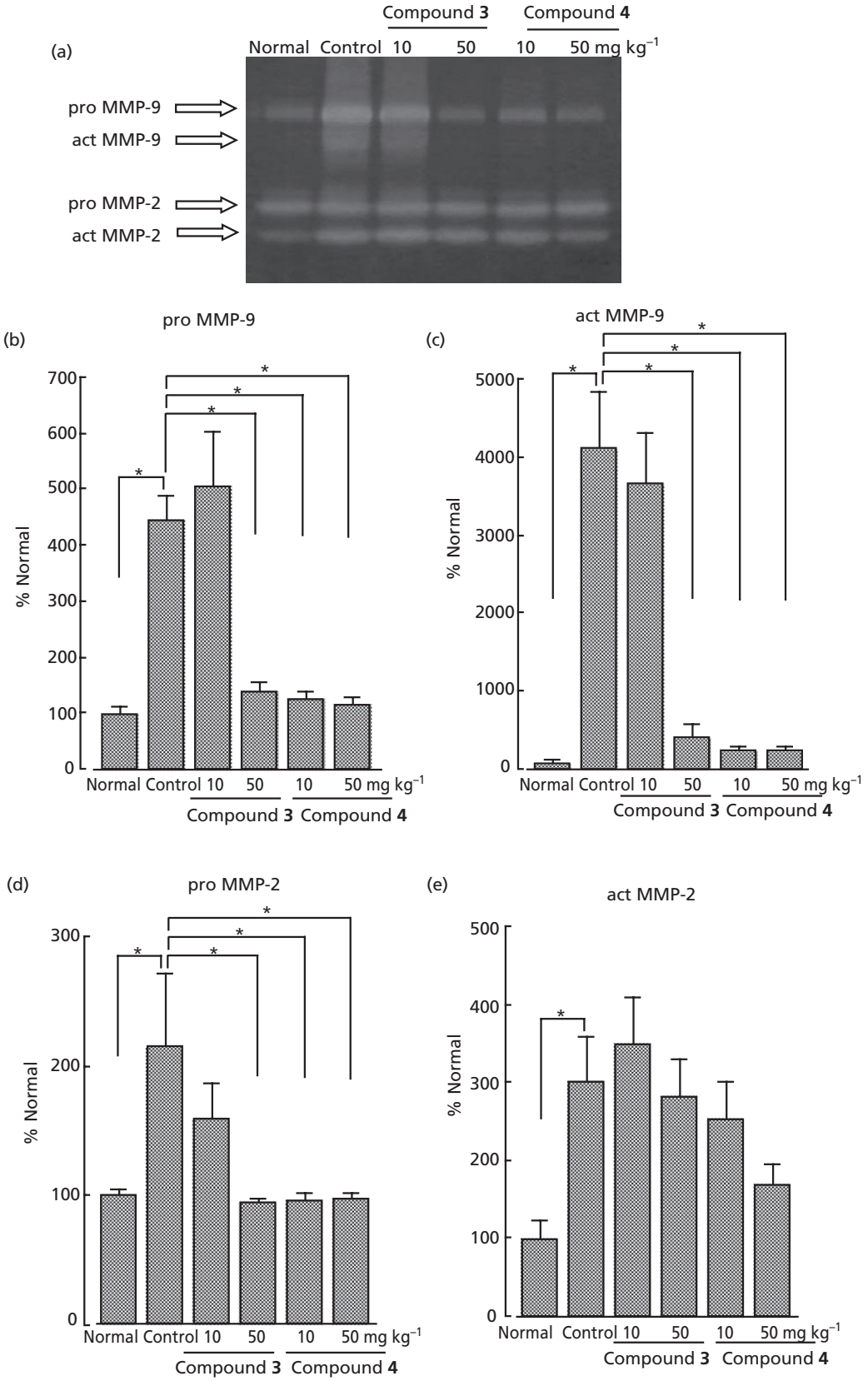


Figure 5 Effects of compounds **3** and **4** on pro-MMP-9, MMP-9, pro-MMP-2 and MMP-2 expression in the skin of acute UVB-irradiated mice. Values are the mean ± SE for six mice. *Significantly different from UVB-irradiated mice (control), $P < 0.05$.

Table 1 Effects of four flavonoids isolated from *Scutellaria baicalensis* roots on VEGF content in the skin in acute UVB-irradiated hairless mice

	VEGF (pg/mg protein)	Control (%)
Experiment 1		
Normal mice	12.72 ± 1.06*	6.31
Vehicle-treated UVB-irradiated mice (control)	201.70 ± 19.28	100
+ Compound **1**		
(10 mg/kg, twice daily)	88.47 ± 13.69*	43.9
(50 mg/kg, twice daily)	72.54 ± 19.92*	36.0
+ Compound **2**		
(10 mg/kg, twice daily)	131.18 ± 30.69	65.0
(50 mg/kg, twice daily)	176.65 ± 28.28	87.6
Experiment 2		
Normal mice	12.43 ± 0.80*	7.38
Vehicle-treated UVB-irradiated mice (control)	168.40 ± 9.53	100
+ Compound **3**		
(10 mg/kg, twice daily)	151.26 ± 21.61	89.8
(50 mg/kg, twice daily)	214.44 ± 36.30	127.3
+ Compound **4**		
(10 mg/kg, twice daily)	56.20 ± 13.15*	33.4
(50 mg/kg, twice daily)	36.87 ± 7.05*	21.9

Values are means ± SE ($n = 6$ mice). *$P < 0.05$, significantly different from UVB-irradiated mice (control).

Effects of four flavonoids on the diameter and length of blood vessels in the skin

The diameter and length of blood vessels were significantly increased by acute UVB irradiation compared to the control. Compounds **1** (50 mg/kg) and **4** (10 and 50 mg/kg) inhibited the increase in diameter of blood vessels, but **2** and **3** did not affect vessel diameter (Figure 6a and b). The increase in the vessel length was also inhibited by the oral administration of **4** (50 mg/kg), but **1, 2** and **3** had no effect on the vessel length (Figure 6a).

Discussion

Exposure to solar UV radiation has severe effects on the structure and function of skin. The number of cases of non-melanoma skin cancers is estimated at over 700 000 and is expected to rise as more UV radiation reaches the earth because of depletion of the ozone layer.[22–24] Although the relationship between non-melanoma skin cancer and UV irradiation was almost not referred in Japan, cases of skin cancer may increase with further depletion of the ozone layer. The symptoms of cutaneous aging, including wrinkles and pigmentation, develop earlier in sun-exposed skin than in unexposed skin, a phenomenon referred to as photoageing. UVB radiation is an important environmental factor because of its hazardous effects, which include genetic mutation that can lead to skin cancer,[14] suppression of the immune system,[15] skin inflammation and edema,[25] and premature skin ageing.[16] Skin alterations observed after UVB irradiation include erythema, vascular hyper-permeability, dilation of dermal blood vessels and epidermal hyperplasia.[26–29] It has been reported that levels of angiogenic factors such as VEGF, basic fibroblast growth factor and interleukin-8 are increased after acute UVB irradiation of the skin, whereas the expression of interferon-β (an anti-angiogenic cytokine) is decreased.[30–32] Furthermore, Yano *et al*.[33,34] have reported that skin vascularization is increased after acute and chronic UVB exposure, with a significant increase in both the number and the size of dermal blood vessels, associated with an upregulation of VEGF expression and downregulation of thrombospondin-1 (TSP-1) mRNA expression in the hyperplastic epidermis. Several lines of evidence suggest that TSP-1 inhibits the activation of MMP-2 and MMP-9, with potential implications for its anti-angiogenic effects and its inhibitory effect on the formation of granulation tissue.[35–37] MMPs, including MMP-2 and MMP-9, are zinc-dependent endopetidases involved in the remodelling of the extracellular matrix, and play important roles in morphogenesis, angiogenesis, arthritis, skin ulceration, tumor invasion and metastasis.[38] Moreover, the MMP-9 proteolytic system may also modulate active VEGF.[39–41]

Among the four flavonoids isolated from the roots of *Scutellaria baicalensis* we found that 2′,5,5′,7-tetrahydroxy-6′,8-dimethoxyflavone (**1**) and 2(*R*),3(*R*)-2′,3,5,6′,7-pentahydroxyflavanone (**4**) reduced the increase in skin thickness and ear thickness induced by acute UVB irradiation. The dose-response of the inhibitory effect of compounds **1** and **2** on increases in ear thickness induced by UVB irradiation is slight. Furthermore, the inhibitory effect of compound **4** on the increase in ear thickness induced by UVB irradiation is a dose-independent effect. Thus, there are no significant differences between 10 and 50 mg/kg doses of compounds **1**, **2** and **4**. The reason for this is unknown and therefore further studies are needed. Furthermore, compounds **1** and **4** also inhibited the increase in diameter and length of the skin area of blood vessels induced by acute UVB irradiation. Thus we found that these two flavonoids prevent skin inflammation induced by UVB irradiation. Furthermore, the increase in the levels of MMP-2, and MMP–9 and VEGF expression in the skin induced by irradiation is reduced by oral administration of these flavonoids. Liu *et al*.[42] have reported that a main

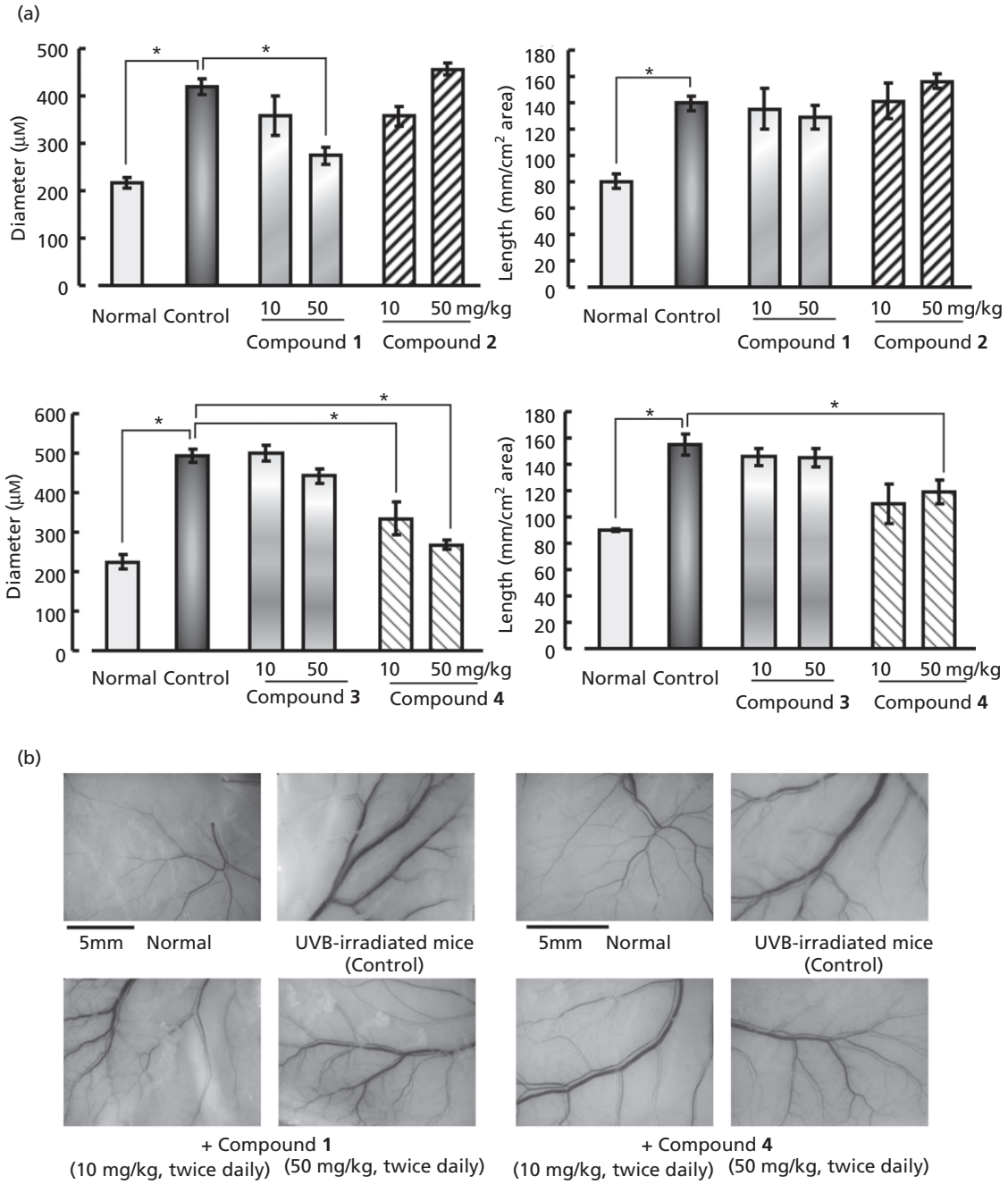


Figure 6 (A) Effects of compounds **1**, **2**, **3** and **4** on diameter and length of blood vessels of the skin in acute UVB-irradiated hairless mice. Values are the mean ± SE for six mice. *Significantly different from UVB-irradiated mice (control), $P < 0.05$. (B) Photograph showing skin blood vessels in normal mice, vehicle-treated acute UVB-irradiated mice (control), compounds **1**- and **4**-treated UVB-irradiated mice.

component – baicalein – of *Scutellaria baicalensis* roots is a potent inhibitor of angiogenesis through the inhibition of MMP-2 expression in human umbilical vein endothelial cells. Therefore, it is suggested that the inhibitory effects of compounds **1** and **4** on acute UVB irradiation-induced skin inflammation may be due to the inhibition of increases in the levels of MMP-2, MMP-9 and VEGF expression. Further studies are needed to clarify the mechanism of the inhibitory effects of compounds **1** and **4** on skin damage. Two other flavonoids, skullcapflavone II (**2**) and 2*(S)*-2′,5,6′,7-tetratahydroxyflavanone (**3**), had no effect. The structural difference between **3** and **4** is the presence (or not) of a hydroxyl group at the C-3 position in the flavanone skeleton. The structural difference between **1** and **2** is the number of methoxyl groups in the flavone skeleton. The structural differences between the four flavonoids used in this study are few and therefore studies are needed to clarify the relationship between the pharmacological actions and structures.

Experiments are now in progress to study the inhibitory effects of 2′,5,5′,7-tetrahydroxy-6′,8-dimethoxyflavone (**1**)

and 2*(R)*,3*(R)*-2′,3, 5,6′,7-pentahydroxyflavanone (**4**) on chronic UVB irradiation-induced carcinogenesis in hairless mice.

Conclusion

2′,5,5′,7-tetrahydroxy-6′,8-dimethoxyflavone (**1**) and 2*(R)*, 3*(R)*-2′,3,5,6′,7-pentahydroxyflavanone (**4**) isolated from *Scutellaria baicalensis* roots may be useful for preventing skin inflammation induced by acute UVB irradiation.

Declarations

Conflict of interest

The authors declare that they have no conflicts of interest to disclose.

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