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Clinical efficacy of facial masks containing yoghurt and *Opuntia humifusa* Raf. (F-YOP)

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Synopsis

Facial packs or masks are popular beauty treatments that are thought to improve skin quality. We formulated a yoghurt pack using natural ingredients (F-YOP), with consideration of skin affinity, safety, health, and beauty. Then, we performed an *in vitro* assessment of biological activity and *in vivo* assessments of moisture, TEWL, melanin content, and elasticity. Facial areas treated with F-YOP showed increased moisture compared to control regions: 89±6.26% (forehead), 140.72±10.19% (cheek), and 123.29±6.67% (chin). Transepidermal water loss (TEWL) values were decreased in the treated areas compared to control: 101.38±6.95% (forehead), 50.37±5.93% (cheek), and 157.81±10.88% (chin). Elasticity was decreased in the control region, whereas the treatment region did not change. The initial elasticity was maintained in the cheek. F-YOP exhibited activity on DPPH radical scavenging, SOD-like activity, and lipoxygenase activity. F-YOP treatment successfully improved the moisture, brightness, and elasticity of treated skin.

INTRODUCTION

Recently, the market for natural cosmetic products has grown to meet the demands of individuals who are concerned about health and beauty, and it continues to grow every year. A number of natural materials have been studied for their physiological effects and metabolism in order to develop them for use in skin-affinity products: black tea gel (1); *Morinda citrifolia* seeds (2); *Vitreoscilla filiformis* (3); *Scutellaria baicalensis* (4) as an antioxidant and anti-aging source; *Saxifraga stolonifera (5)* as an anti-cancer agent; *Morus alba* leaves (6); *Glycyrrhiza uralensis* (7); and *Schisandra chinesis* as a melanogenesis inhibitor.

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Skin penetration of natural materials may be improved through fermentation, which increases the nutritive content of substances and reduces the particle size. Microorganisms are the key factor for fermentation (8), and *Lactobacillus, Streptobacillus*, and *Bifodobacterium* are usually used for cosmetic materials. Yoghurt is a fermented milk product that is often used in simple diets for atopic individuals (9), but there are currently no cosmetic products based on yoghurt.

We formulated a yoghurt-based facial mask, and conducted analyses to determine its biological effects. This product also contains the Eastern prickly pear *Opuntia humifusa* Raf., which has been reported to have antioxidant and anti-inflammatory activity. A member of the Cactaceae family, the *Opuntia* genus has antioxidative components, flavonoids, quercetin, (+)-dihydroquercetin, and quercetin 3-methyl ether (10), which have radical scavenging and anti-inflammatory activities (10–14). *O. humifusa* can tolerate low temperature (15). Recently, it has been successfully cultivated in winter in Korea (11). Because of its biological effects, *Opuntia* has been used as a food and cosmetic material, but it is always processed for these uses. We presume that is because when *Opuntia* comes into contact with water, it becomes viscous.

We added intact *Opuntia* powder to yoghurt under the hypothesis that the ingredients would have a synergistic effect in terms of antioxidant and anti-inflammatory activity. In this study, we examined changes in skin elasticity, moisture, melanin content, and erythema through *in vivo* tests and compared the results of our facial mask containing yoghurt and *O. humifusa* (F-YOP) to those of a control facial mask.

MATERIALS AND METHODS

PREPARATION OF YOGHURT

Yoghurt base (commercial fermented milk) was added to sterile milk at 10 w/w% and then incubated at 42°C for 16 h, followed by overnight storage at 4°C. The yoghurt was mixed into a pack base of 4:6 (yoghurt:pack base). The pack base included de-ionized water, oil, and a viscosity-increasing agent (Table I). It is the same as the base of the control product. The resulting product (completed yoghurt and pack base, YP) contained no preservatives.

PREPARATION OF *OPUNTIA HUMIFUSA* RAF.

We purchased freeze-dried *O. humifusa* powder (Goryeo Cactus, Seoul, Korea). The powder was prepared as 3 w/w%. Immediately prior to use, the powder was mixed completely with the YP.

IN VITRO TESTS

Scavenging activity on DPPH radicals. Radical scavenging activity of the *Opuntia*/YP components was determined according to Blosi's method (1958), with some modifications (16). It was made from 0.2 mM DPPH (1,1-diphenyl-2-picryl-hydrazyl, Sigma, St. Louis, MO) solution using ethanol. The sample was dissolved in ethanol and was reacted with

Table I
Base Ingredients

Chemical name
Disodium EDTA
Glycerin
Betain
Propylene glycol
Triethanolamine
Stearic acid
Cetyl alcohol
Sorbitan stearate
Sucrose cocoate
Glyceryl stearate
PEG-100 stearate
Polysorbate 80
Macadamia ternifolia seed oil
Cetyl ethylhexanoate
Cyclomethicone
Caprylic/capric triglyceride
Tocopheryl acetate
Dimethicone
Carbomer
Xanthan gum
Water

DPPH for 10 min. Then, the absorbance was measured at 517 nm by a spectrophotometer (Spectramax, plus384, Molecular Devices, Sunnyvale, CA). The radical scavenging activity was calculated using the following equation: Radical scavenging activity (%) = $\{(OD_{control} - OD_{sample}) / OD_{control}\} \times 100$.

SOD-like activity. Activity was measured according to the method of Oyanagui (17), with minor modifications. Superoxide dismutase (SOD) activity of reactive oxygen species (ROS) was measured using the xanthine-xanthine oxidase (Sigma) system as a source of superoxide and nitroblue tetrazolium (NBT; Sigma) as a scavenger. Radical SOD activity was determined by measuring the change in absorbance by oxidation of NBT by SOD. Absorbance was measured at 560 nm by spectrophotometer. Activity (%) was calculated as compared with the control. The method of calculation was the same as that used for the DPPH method.

Inhibitory activity of lipoxygenase (LOX). Activity was measured according to the method of Galliard, with minor modifications (18). After the test sample was mixed, 1 mM linolenic acid (Sigma) and lipoxygenase (Sigma) was added. Then the sample was vortexed at regular speed and time (IMS1000; Eyela, Rikakikai Co., Ltd, Tokyo) and pre-incubated at room temperature for 10 min. After the sample was incubated, 20 w/v% trichloroacetic acid (TCA; Junsei Chemical, Koshigaya, Japan) and 0.6 w/v% thiobarbituric acid (TBA; Sigma) were added to the sample. The sample was boiled in water for 10 min and then cooled in ice water for 2–3 min. Then n-butyl alcohol (99%; Duksan Pure Chemicals Co., Ltd, Ansan, Korea) was added and the sample was centrifuged at 3,500 rpm for

5 min after vortexing for 20 sec. The absorbance was measured at 535 nm by spectrophotometer. Activity (%) was calculated as compared with the control. The calculations were performed as described above.

IN VIVO TESTS

Sixteen subjects, Korean adults of both sexes, with an average age of 29 years, participated in this study. The moisture, TEWL, melanin content, and elasticity were accessed, respectively. We measured parameters before and after treatment on the forehead, cheek, and chin of all subjects. All tests were performed at 25±0.01°C, RH 40.27±0.08%. The control product consists of the base ingredients without the active ingredients (Table I). The base of F-YOP is the same as the base of the control product. The control product is manufactured by our company and is selling in the open market.

Dermal irritation. The dermal irritation potential of each product was determined using the "primary irritant test" with ten different volunteers. The pack was applied for 24 h under occlusive patches (Haye's test chambers; Koms Company, Alphen ann den Rijn, Netherlands) on the inner forearm. The regions were then evaluated for the existence of erythema after treatment (Table II).

Water content (moisture) and TEWL value. To assess skin water content, a corneometer (Skin-O-Mat; Cosmomed-Beauty-Rent GmbH, Germany) was used to measure electrical capacitance. The amount of water loss was assessed with a TEWL (Dermalab Cortex Technology, Hadsund, Denmark) probe and expressed as $g/cm^2/hr$. The water content was measured for each of five random selected assessment regions of the forehead, cheek, and chin, and average values were calculated. For TEWL analysis, measurements were taken from two regions of the forehead, three regions of the cheek, and two regions of the chin, and average values were calculated.

Elasticity values. The elasticity of the skin was measured using an elast probe (Dermalab Cortex Technology), which applies suction to the skin surface. The suction method incorporates an elevation phase and retraction phase; each has properties that contribute to the "feel" of the skin. Elasticity was measured in six random assessment regions of the cheek, and average values were calculated.

Table II
Incidence of Erythema after F-YOP Treatment

Symptoms	Indication	Identification
No change	–	8
Weak erythema	±	2
Definite erythema or a little quantity of papules	+	0
Erythema and swelling or erythema and papule	++	0
Erythema, swelling and vesicle	+++	0
Vesicle, festering	++++	0

The percentage reporting primary irritation was 20%, and these cases were "weak erythema." The other 80% did not have primary dermal irritation.

Melanin and erythema values. Melanin and erythema values were measured by a dermaspectrometer (Dermalab Cortex Technology), which measured the erythema index of the skin as described by the Diffey and Farr melanin index (19). After calibration of the erythema/melanin indexes at zero, we performed the measurements (black: 99.9; white: 0.00). The measurement points were in four regions of the forehead, six regions of the cheek, and three regions of the chin. Average values were calculated.

Subjective assessment. We asked the participants to subjectively assess the results of using the F-YOP or control treatments. Questions included the evaluation of skin moisture, skin brightness, and skin irritation after use (highest, high, medium, low, and lowest). Final results were assessed as percentages (Table III).

Statistical analysis. The levels of significance of any differences between groups were calculated by *t*-test.

RESULTS

IN VITRO TEST

Scavenging activity on DPPH radicals. The scavenging activity was 19.71±9.24% in YP, 49.29±0.84% in yoghurt, 68.82±4.54% in *O. humifusa,* 30.66±4.30% in fermented milk, and 19.71±9.24% in the pack base, with a final concentration of 0.1% in the Y-FOP product (Table IV). Trolox (6-hydroxyl-2,5,6,8-tetramethylchloroman-2-carboxylic acid, 97%; Sigma) was used as a positive control. Trolox is a vitamin E derivative with excellent anti-oxidant effects (20). In our work, SC_{50} based on the DPPH radical scavenging rate was expressed as SC_{50}. SC_{50} is the concentration of sample needed to produce a 50% reduction in the DPPH.

Table III
Subjective Feelings Regarding Moisture, Brightness, and Irritation after F-YOP Treatment

No.	Item	Highest	High	Medium	Low	Lowest
1	Skin moisture	11.11	61.11	27.78		
2	Skin brightness		44.44	50.00		5.56
3	Skin irritation				16.67	83.33

The lowest rating indicates no effect. Values are expressed as percentages (%). Subjects who felt high moisture: 72.72%. Skin brightness was indicated at a nearly medium level. Skin irritation was not represented by 83.33% of subjects.

Table IV
Physiological Activities (%) of Raw Materials for F-YOP

	YP	Yoghurt	*O. humifusa*. Raf.	Fermented milk	positive control
DPPH	19.71 ± 9.24	49.29 ± 0.84§§	68.82 ± 4.54§§	30.66 ± 4.30§	51.13 ± 3.07§§
SOD	—	—	37.89 ± 8.75*	—	51.23 ± 11.29**
LOX	22.05 ± 0.57	31.79 ± 1.22	—	27.19 ± 0.93	53.67 ± 2.99

YP (completed yoghurt and pack base mixture), Yoghurt, *O. humifusa* Raf., and fermented milk have antioxidant and antiinflammatory effects, except that there was no LOX effect in *O. humifusa* Raf. Final concentration of samples: 1000 ug/ml (SOD; 500 ug/ml), positive control (IC_{50}): Trolox, 5ug/ml in DPPH; ascorbic acid, 33ug/ml in SOD; NDGA, 7.55ug/ml in LOX. * $p<0.05$; ** $p<0.01$; § $p<0.001$; §§ $p<0.0001$.

SOD-like activity. O. humifusa had a ROS scavenging activity of 37.89±8.75% at a final concentration of 0.1% (Table IV). The positive control was L-ascorbic acid (Sigma). The other three materials had no activities. SC_{50} values represent the concentration of sample required to scavenge 50% of the superoxide anion produced by the hypoxanthin-xanthine oxidase system.

Inhibitory activity of lipoxygenase (LOX). YP, yoghurt, and fermented milk produced 22.05±0.57%, 31.79±1.22%, and 27.19±0.93% inhibition of LOX, respectively (Table IV). The final concentration was 0.1% for all materials tested. Nordihydroguaiaretic acid (NDGA; Sigma) was used for the positive control. IC_{50} values represent the concentration of sample required to inhibit 50% of LOX. Lipoxygenase (5-LO, 5-lipoxygenase) is a human enzyme that is a member of the lipoxygenase family. It transforms essential fatty acids into leukotriences and is a current target for pharmaceutical intervention in a number of diseases. Lipid peroxidation is a free radical-related process that may be enzymatically controlled, e.g., for the generation of lipid-derived inflammatory mediators, or may be non-enzymatically regulated (21). Typically, the hydroperoxide in lipoxygenation is measured using SLO (soybean lipoxygenase) (22). This approach can be used to screen inhibitors of lipoxygenase and to confirm inflammatory effectiveness (23).

IN VIVO TEST

F-YOP and control products were applied to human subjects on both the left and right sides of the face, respectively. The F-YOP-treated region is typically more moist, with more brightness and elasticity in the skin.

Dermal irritation. The percentage of the face exhibiting primary irritation was a 20% degree of irritation, and this represented weak erythema. The remaining 80% did not have dermal irritation. The irritation level is shown in Table II.

Water content (moisture) and TEWL values. The moisture levels in the F-YOP-treated regions were superior to those in the control. For the treated areas, the moisture was 18.7% in the forehead; 24.66% in the cheek, and 17.08% in the chin, whereas the moisture in the control areas was 9.88% in forehead, 10.24% in the cheek, and 7.65% in the chin. The TEWL values were high across all regions compared to the control: 89±6.26% in the forehead, 140.72±10.19% in the cheek, and 123.29±6.67% in the chin (Figure 1).

The TEWL values for the treated regions were inferior to those of the control. For the treated regions, the TEWL values decreased to 0.13% in the forehead, and increased to 8.26% in the cheek and 5.63% in the chin, but the TEWL values in the control regions increased to 9.69% in the forehead, 16.65% in the cheek, and 9.75% in the chin. TEWL values were lower, with 101.38±6.95% in the forehead, 50.37±5.93% in the cheek, and 157.81±10.88% in the chin, compared to the control (Figure 2). The values for water content and TEWL in the skin were shown in reciprocal proportion on a graph.

Elasticity value. Elasticity decreased in the control region, and there was no change in elasticity in the F-YOP treatment region. The initial elasticity was maintained in the treatment region (Figure 3).

Melanin and erythema value. There was no significant decrease in melanin or erythema values for treatment or control. However, the erythema value decreased in F-YOP-treated

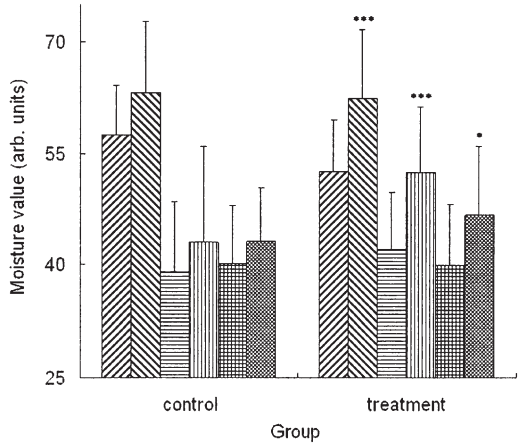

Figure 1. Moisture values for F-YOP (treatment) and control product. The values were significantly different between the two groups. ▨ forehead-before; ▧ forehead-after; ▤ cheek-before; ▥ cheek-after; ▦ chin-before; ▩ chin-after. * $p<0.05$; *** $p<0.001$.

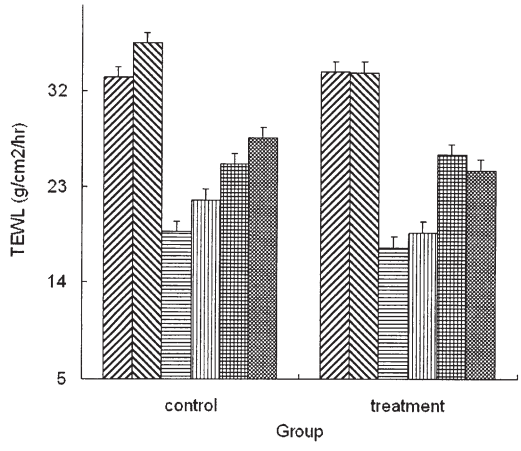

Figure 2. TEWL values for F-YOP (treatment) and control product. TEWL values were increased in the control regions and decreased or maintained in the treated regions. In the cheek, the TEWL value for treatment was lower than that for the control. ▨ forehead-before; ▧ forehead-after; ▤ cheek-before; ▥ cheek-after; ▦ chin-before; ▩ chin-after.

cheeks compared to the control, and the melanin value decreased in F-YOP-treated foreheads compared to the control (Figures 4, 5).

DISCUSSION

The present study describes the physiological effects of a face mask made of natural ingredients. We determined that the proper condition is a 10 w/w% ratio of yoghurt to sterile milk. If the yoghurt ratio varies from this, it will not be complete. The incubation must be at 42°C for 16 h because yoghurt becomes acidic over time and cannot form below 40°C. There was no change in color, flavor, viscosity, or sediment at room temperature for

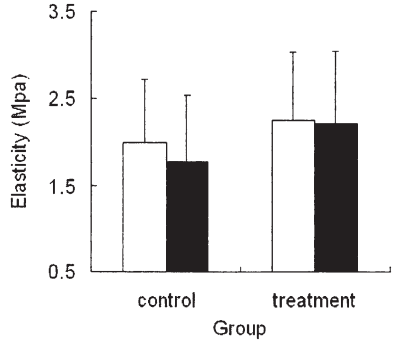

Figure 3. Elasticity values for F-YOP (treatment) and control product in the cheek. The value decreased in the control region, and was maintained at pretreatment levels in the treatment region. □ before treatment; ■ after treatment.

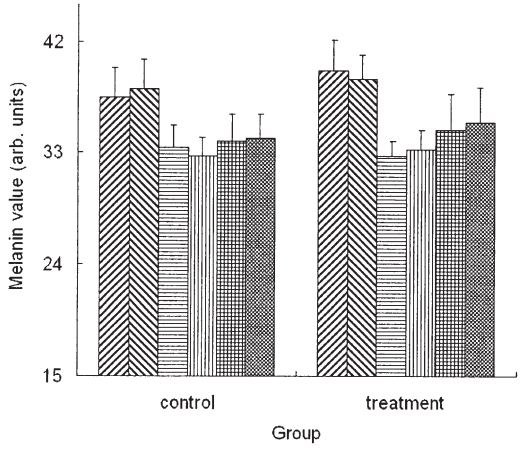

Figure 4. Melanin values for F-YOP (treatment) and control product. There was no effect on melanin in either group. The melanin value decreased in the treated forehead region, in comparison to the control. ▨ forehead-before; ▧ forehead-after; ▤ cheek-before; ▥ cheek-after; ▦ chin-before; ▩ chin-after.

six months or more. If the ratios of yoghurt to pack base are 5:5 (1:1), it has a putrid smell of yoghurt. We tested several ratios of yoghurt to pack base (1:9, 2:8, and 4:6). All were stable and did not have a putrid smell of yoghurt. In consideration of the feel upon application and unit cost, we determined that 4:6 should be the final ratio.

O. humifusa powder was mixed with YP immediately prior to use for freshness, and the optimal powder ratio was determined as 3% of the total volume. This powder tends to gel when combined with water, and so if the amount of powder is increased, it will affect the application feel and washing.

The moisture value was superior with F-YOP treatment compared to the control product that was used as the control. The TEWL decreased after treatment and the values were superior compared to the control. Elasticity decreased in the control region; however it did not change in the F-YOP-treated region. Thus, F-YOP treatment supplied efficient moisture to the skin and helped maintain elasticity.

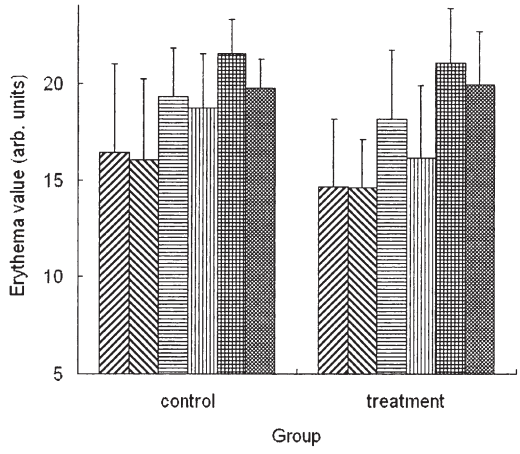

Figure 5. Erythema values for F-YOP (treatment) and control product. The erythema value decreased in the treatment region of the cheek, compared to the control. ▨ forehead-before; ▧ forehead-after; ▤ cheek-before; ▥ cheek-after; ▦ chin-before; ▩ chin-after.

For this reason, physiological materials may act as an evaluation factor. Fermented milk at 1% final concentration was comparatively more dominant than the positive control in DPPH and lipoxygenase assays. In theory, if fermented milk is added to sterile milk by 10 w/w%, this activity improves. We did not confirm this with a long-term *in vivo* practical test, but in this study, we found that the elasticity value decreased in the control region but increased in the treatment region. We believe that this product will be suitable for dry-skin patients because of the associated reduction in TEWL, increased moisture, and lack of preservatives.

There is an increasing demand for the cosmetics industry to produce products that are safe, effective, stable, and provide user convenience (19). Skin products that are made from natural materials are less likely to cause irritation in consumers with sensitive skin because there are no preservatives or additives. We studied the stability of our F-YOP product for over six months, and we did not find any changes in the deposits, color, or flavor. Our product appears to be effective at improving skin moisture and brightness, and in reducing TEWL.

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REFERENCES

(1) M. Turkoglu and N. Cigirgil, Evaluation of black tea gel and its protection potential against UV, *Int. J. Cosmet. Sci.,* **29,** 437–442 (2007).

(2) M. Masuda, K. Murata, A. Fukuhama, S. Naruto, T. Fujita, A. Uwaya, F. Isami, and H. Matsuda, Inhibitory effects of constituents of *Morinda citrifolia* seeds on elastase and tyrosinase, *Nat. Med. (Tokyo),* **63,** 267–273 (2009).

(3) Y. F. Mahe, R. Martin, L. Aubert, N. Billoni, C. Collin, F. Pruche, P. Bastien, S. S. Drost, A. T. Lane, and A. Meybeck, Induction of the skin endogenous protective mitochondrial MnSOD by *Vitreoscilla filiformis* extract, *Int. J. Cosmet. Sci.,* **28,** 277–287 (2006).
(4) R. Liang, R. M. Han, L. M. Fu, X. C. Ai, J. P. Zhang, and L. H. Skibsted, Baicalin in radical scavenging and its synergistic effect with beta-carotene in antilipoxidation, *J. Agric. Food Chem.,* **57,** 7118–7124 (2009).
(5) Z. Chen, Y. M. Liu, S. Yang, B. A. Song, G. F. Xu, P. S. Bhadury, L. H. Jin, D. Y. Hu, F. Liu, W. Xue, and X. Zhou, Studies on the chemical constituents and anticancer activity of *Saxifraga stolonifera* (L.) Meeb, *Bioorg. Med. Chem.,* **16,** 1337–1344 (2008).
(6) S. H. Lee, S. Y. Choi, H. Kim, J. S. Hwang, B. G. Lee, J. J. Gao, and S. Y. Kim, Mulberroside F isolated from the leaves of *Morus alba* inhibits melanin biosynthesis, *Biol. Pharm. Bull.,* **25,** 1045–1048 (2002).
(7) H. J. Kim, S. H. Seo, B. G. Lee, and Y. S. Lee, Identification of tyrosinase inhibitors from *Glycyrrhiza uralensis, Planta Med.,* **71,** 785–787 (2005).
(8) M. Brasca, S. Morandi, R. Lodi, and A. Tamburini, Redox potential to discriminate among species of lactic acid bacteria, *J. Appl. Microbiol.,* **103,** 1516–1524 (2007).
(9) T. Uenishi, H. Sugiura, T. Tanaka, and M. Uehara, Role of foods in irregular aggravation of skin lesions in children with atopic dermatitis, *J. Dermatol.,* **35,** 407–412 (2008).
(10) H. Dok-Go, K. H. Lee, H. J. Kim, E. H. Lee, J. Lee, Y. S. Song, Y. H. Lee, C. Jin, Y. S. Lee, and J. Cho, Neuroprotective effects of antioxidative flavonoids, quercetin, (+)-dihydroquercetin and quercetin 3-methyl ether, isolated from *Opuntia ficus-indica* var. saboten, *Brain Res.,* **965,** 130–136 (2003).
(11) J. Y. Cho, S. C. Park, T. W. Kim, K. S. Kim, J. C. Song, S. K. Kim, H. M. Lee, H. J. Sung, H. J. Park, Y. B. Song, E. S. Yoo, C. H. Lee, and M. H. Rhee, Radical scavenging and anti-inflammatory activity of extracts from *Opuntia humifusa* Raf, *J. Pharm. Pharmacol.,* **58,** 113–119 (2006).
(12) E. M. Galati, M. R. Mondello, D. Giuffrida, G. Dugo, N. Miceli, S. Pergolizzi, and M. F. Taviano, Chemical characterization and biological effects of Sicilian *Opuntia ficus indica* (L.) mill. Fruit juice: antioxidant and antiulcerogenic activity, *J. Agric. Food Chem.,* **51,** 4903–4908 (2003).
(13) J. C. Lee, H. R. Kim, J. Kim, and Y. S. Jang, Antioxidant property of an ethanol extract of the stem of *Opuntia ficus-indica* var. saboten, *J. Agric. Food Chem.,* **50,** 6490–6496 (2002).
(14) D. Butera, L. Tesoriere, F. Di Gaudio, A. Bongiorno, M. Allegra, A. M. Pintaudi, R. Kohen, and M. A. Livrea, Antioxidant activities of Sicilian prickly pear (*Opuntia ficus indica*) fruit extracts and reducing properties of its betalains: Betanin and indicaxanthin, *J. Agric. Food Chem.,* **50,** 6895–6901 (2002).
(15) G. Goldstein and P. S. Nobel, Water relations and low-temperature acclimation for cactus species varying in freezing tolerance, *Plant Physiol.,* **104,** 675–681 (1994).
(16) S. E. Lee, H. J. Hwang, J. S. Ha, H. S. Jeong, and J. H. Kim, Screening of medicinal plant extracts for antioxidant activity, *Life Sci.,* **73,** 167–179 (2003).
(17) Y. Oyanagui, Reevaluation of assay methods and establishment of kit for superoxide dismutase activity, *Anal. Biochem.,* **142,** 290–296 (1984).
(18) R. K. Hughes, S. I. West, A. R. Hornostaj, D. M. Lawson, S. A. Fairhurst, R. O. Sanchez, P. Hough, B. H. Robinson, and R. Casey, Probing a novel potato lipoxygenase with dual positional specificity reveals primary determinants of substrate binding and requirements for a surface hydrophobic loop and has implications for the role of lipoxygenases in tubers, *Biochem. J.,* **353,** 345–355 (2001).
(19) B. L. Diffey, R. J. Oliver, and P. M. Farr, A portable instrument for quantifying erythema induced by ultraviolet radiation, *Br. J. Dermatol.,* **111,** 663–672 (1984).
(20) R. Re, N. Pellegrini, A. Proteggente, A. Pannala, M. Yang, and C. Rice-Evans, Antioxidant activity applying an improved ABTS radical cation decolorization assay, *Free Rad. Biol. Med.,* **26,** 1231–1237 (1999).
(21) F. J. Romero, F. Bosch-Morell, M. J. Romero, E. J. Jareno, B. Romero, N. Marin, and J. Roma, Lipid peroxidation products and antioxidants in human disease, *Environ. Health Perspect.,* **106**(Suppl. 5), 1229–1234 (1998).
(22) T. Galliard and D. R. Phillips, Lipoxygenase from potato tubers. Partial purification and properties of an enzyme that specifically oxygenates the 9-position of linoleic acid, *Biochem. J.,* **124,** 431–438 (1971).
(23) M. Resch, A. Steigel, Z. L. Chen, and R. Bauer, 5-Lipoxygenase and cyclooxygenase-1 inhibitory active compounds from *Atractylodes lancea, J. Nat. Prod.,* **61,** 347–350 (1998).