



## Clinical trial

## Wrinkle reduction using a topical herbal cream in subjects with greater yin (Tae-eumin) type: A randomized double-blind placebo-controlled study

A-Rang Im<sup>a,1</sup>, Jiho Nam<sup>b,1</sup>, Seongwon Cha<sup>c</sup>, Young Kyoung Seo<sup>d</sup>, Sungwook Chae<sup>a,e,2,\*</sup>,  
Jong Yeol Kim<sup>b,e,2,\*\*</sup>

<sup>a</sup> KM Convergence Research Division, Korea Institute of Oriental Medicine, Daejeon, 34054, Republic of Korea

<sup>b</sup> Constitutional Medicine and Diagnosis Research Group, Korea Institute of Oriental Medicine, Daejeon, 34054, Republic of Korea

<sup>c</sup> Mibyeong Research Center, Korea Institute of Oriental Medicine, Daejeon, 34054, Republic of Korea

<sup>d</sup> Dermapro, Safety and Efficacy Evaluation of Cosmetics & Cosmeceuticals, Seocho-gu, Seoul, 06684, Republic of Korea

<sup>e</sup> University of Science and Technology, 217 Gajeong-ro, Yuseong-gu, Daejeon, 305-333, Republic of Korea

## ARTICLE INFO

## Keywords:

*Scutellaria baicalensis*

*Raphanus sativus*

Randomised controlled trial

Matrix metalloproteinases

Sasang constitutional type

Tae-eumin

Skin wrinkles

## ABSTRACT

**Introduction:** Skin aging can be caused by both intrinsic and extrinsic factors. Ultraviolet (UV) radiation is the primary environmental factor which causes skin aging. Sasang constitutional medicine (SCM) is a unique type of traditional Korean medicine. In SCM, patients are treated with herbal drugs categorized according to their Sasang constitutional type based on the Yin-Yang theory. *Scutellaria baicalensis* and *Raphanus sativus* are herbal drugs exclusively used for Tae-eumin (TE)-type subjects. These drugs are constituents of the TE formula, which is effective for managing skin diseases such as urticaria and acne. This study investigated the potential of *S. baicalensis* and *R. sativus* (SR) herbal mixtures as active ingredients in wrinkle-care cosmetics.

**Methods:** The expression of matrix metalloproteinase-1 (MMP-1) was measured *in vitro* with significant difference ( $p < 0.05$ ). A 12-week placebo randomised double blind controlled trial was conducted to evaluate the safety and clinical efficacy of a cream containing SR mixtures for application to human skin. Women between the ages of 43 and 58 years who were TE-type subjects according to SCM were selected and cream containing SR mixtures was applied for 12 weeks.

**Results:** The application of SR mixtures showed there was a significant improvement in both visual assessment and evaluation of wrinkle parameters (Rt; skin roughness, Rm; maximum roughness, Rz; average roughness, Rp; smoothness depth, and Ra; arithmetic average roughness) comparing the treatment group with the control group ( $p < 0.05$ ).

**Conclusions:** The results of this study indicate that the cream containing SR mixtures may improve skin wrinkles in TE-type subjects.

## 1. Introduction

Skin aging can be caused by both intrinsic (natural) genetically determined factors and extrinsic lifestyle-driven and environmental factors [1]. Intrinsic aging is a biological process common to all living organisms and is characterized by an age-dependent deterioration of skin function and structure [2]. Naturally aged skin is pale and finely wrinkled [3]. Extrinsic aging is mainly caused by photodamage resulting from UV radiation and can be characterized by the deterioration of the dermal connective tissue [4]. As people age, they become

increasingly concerned about the appearance of their skin. In fact, extrinsic aging caused by photodamage reflects the cutaneous histological and functional changes induced by chronic and repeated exposure to UV radiation [5]. Skin aging is characterized by a progressive reduction in skin thickness. Furthermore, changes occur in both the epidermis and dermis that are expressed as wrinkles, dry and rough skin, cell proliferation and structural abnormalities, irregular pigmentation, and the degeneration of dermal elastic fibers [6].

UVB-induced skin damage has been extensively studied from the perspective of wrinkle formation and skin inflammation [7].

\* Corresponding author at: KM Convergence Research Division, Korea Institute of Oriental Medicine, 1672 Yuseongdae-ro, Yuseong-gu, Daejeon, 34054, Republic of Korea.

\*\* Corresponding author at: Constitutional Medicine and Diagnosis Research Group, Korea Institute of Oriental Medicine, 1672 Yuseongdae-ro, Yuseong-gu, Daejeon, 34054, Republic of Korea.

E-mail addresses: [kendall@kiom.re.kr](mailto:kendall@kiom.re.kr) (S. Chae), [ssmed@kiom.re.kr](mailto:ssmed@kiom.re.kr) (J.Y. Kim).

<sup>1</sup> These authors contributed equally to this work.

<sup>2</sup> These corresponding authors contributed equally to this work.

Photodamaged skin is characterized by various undesirable esthetic manifestations, including dyspigmentation, wrinkling, roughness, laxity, dullness, lentiginosities, and the atrophy of body areas chronically exposed to the sun [8]. Among a range of external factors that induce skin aging, UV radiation is a main cause of skin damage, which can be characterized by deep wrinkles, roughness, laxity, and pigmentation [9].

Chronological aging is a natural process caused by internal physiological factors and leads to relatively small wrinkles [10]. Wrinkles on facial skin are one of the most characteristic morphological changes of aging. Wrinkles are also a prominent feature of photoaged skin and are caused by the degradation of collagen and gelatin fibers [11]. Photoaging is a hallmark of prolonged UV exposure, which causes collagen breakdown by increasing the expression of matrix metalloproteinase (MMP) enzymes, such as MMP-1 [12]. Alteration and deficiency of collagen, the major structural component of the skin, have been suggested as causes of skin wrinkling in photoaged and naturally aged skin [13].

Sasang constitutional medicine (SCM) is a unique type of traditional Korean medicine. SCM is used to treat patients with herbal drugs categorized according to Sasang constitutional type, which include the Tae-eumin (TE), Soeumin (SE), Taeyang (TY), and Soyang types (SY) [14]. Each constitution is characterized by a unique set of phenotypes, such as face shape, body shape, voice type, skin characteristics, and temperament, which are also used for the diagnosis of constitution and also according to SCM, each constitutional type corresponds to a certain group of medicinal herbs and remedies [15]. Human skin properties have been used as an important diagnostic component in traditional medicine as they change with health condition [15]. In previous study, in search of skin characteristics effectively reflecting SCM features, it appeared several skin properties such as perspiration, visco-elasticity, elasticity, and elasticity hysteresis, in several candidate body parts [16]. Accordingly, skin characteristics also differ depending on constitution. For example, the skin of SY is slippery and thin (although some SY-type subjects have thick skin). Sasang constitution have been reported to be closely related to the equilibrium of internal organ functions so TE group has strong anabolic functionality and weak catabolic functionality [17]. The skin of TE is thick and stiff (occasionally TE-type subjects have soft skin), and the skin of SE is buoyant and soft [16,18]. Therefore, different skin characteristics according to the type of constitution should be treated with different skin care formulas.

*Scutellaria baicalensis* and *Raphanus sativus* (SR) are herbal drugs used exclusively for TE. They are part of the TE formula “Cheongpyesagan-tang”, which is effective in treating skin diseases such as urticaria and acne [19,20]. *S. baicalensis* is widely used in traditional Chinese herbal medicines in Asian countries, including Korea, China, and Japan, for the treatment of various diseases, such as inflammation, hypertension, and cancer, and for its beneficial biological effects [21]. *S. baicalensis* contains a variety of flavones, phenylethanoids, amino acids, sterols, and essential oils; its four major flavonoids are baicalin, baicalensin, wogonin, and wogonoside [22]. The major active components of *S. baicalensis* are baicalin and baicalensin, which show UV-protective effects [23,24]. *R. sativus* L. belongs to the family Brassicaceae and has long been grown as a food crop worldwide. The leaves, seeds, and roots of *R. sativus* L. are used in traditional Chinese medicine [25]. Furthermore, its seeds are commonly used as stomachache, anti-cancer, and anti-inflammatory agents in traditional Korean medicine [26–28]. The present study evaluated the anti-wrinkle effects and safety of a cream containing *S. baicalensis* and *R. sativus* mixtures for use as skin applications.

## 2. Materials and methods

### 2.1. Preparation of herbal extracts

Dried *Scutellaria baicalensis* and *Raphanus sativus* were purchased

from Kwangmyung-Dang (Ulsan, Korea). All voucher specimens (2015SC and 2015 SR) were deposited in the herbal bank of the Korea Institute of Oriental Medicine. To prepare the herbal extract, *Scutellaria baicalensis* and *Raphanus sativus* were mixed together, extracted with water under reflux and then filtered. The filtrate was evaporated to yield a herbal product, which was then used as an ingredient in the topical formulation.

### 2.2. UVB irradiation and cell viability assay

HaCaT cells, an immortalized non-tumorigenic human keratinocyte cell line, were maintained in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% fetal bovine serum and 1% antibiotics (10,000 units/mL of penicillin and 10,000 µg/mL of streptomycin) at 37 °C in a humidified 5% CO<sub>2</sub> incubator. Cells were seeded and allowed to adhere for 24 h. Subsequently, the cells were treated with various concentrations of SR mixtures and exposed to UVB radiation at 20 mJ/cm<sup>2</sup>. Cells that received no pretreatment and were not exposed to UVB radiation served as the control.

HaCaT cells ( $1 \times 10^4$ ) were seeded into 96-well culture plates and treated with various concentrations of SR mixtures for 24 h. After treatment, the cell culture medium was replaced with phosphate-buffered saline (PBS). Subsequently, the cells were exposed to 20 mJ/cm<sup>2</sup> UVB radiation (UV Crosslinker, Ultra Lum, Upland, CA), and the cell viability was assessed by the reduction of [3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium (MTS)] to formazan after 1 h of irradiation, according to the manufacturer's instructions. Samples were assayed at 490 nm using a microplate fluorimeter (Molecular Devices, Sunnyvale, CA).

### 2.3. Determination of MMP-1 secretion by ELISA

MMP-1 levels in the culture medium of HaCaT cells ( $5 \times 10^4$ ) after UVB irradiation were determined using human total MMP-1 ELISA kits according to the manufacturer's instructions. Briefly, HaCaT cells were seeded into 96-well plates and treated with SR mixtures. After exposure to UVB radiation, the cell culture supernatant was collected and centrifuged at 18,200 rcf for 5 min. The levels of MMP-1 were quantified by colorimetric analysis.

### 2.4. Study subjects and methods

This study was conducted in accordance with the intent and purpose of good clinical practice regulations as described in Korean Good Clinical Practice (KGCP) and the Declaration of Helsinki, as appropriate. All participants provided written informed consent before their study participation commenced. All procedures for the recruitment, selection, and inclusion of subjects in this study were established to provide the participants with clear and precise information, allowing them to appreciate the aims of the project and the consequences of their consent. The study protocol was approved by the review committee of Dermapro (IRB No. 1-220777-A-N-02-DICN15031). Furthermore, this study was performed in accordance with the standard operating procedures (SOPs) of the Dermapro Skin Science Institute (IRB No. 1-220777-A-N-02-DICN15031). This study has been uploaded to <http://cris.nih.go.kr/cris/index.jsp> (identifier KCT0002044) from the National Research Institute of Health.

The subjects of this study were 22 TE women aged 43–58 years who met the inclusion criteria, i.e., TE subjects over 30 years who had started to develop wrinkles or already had wrinkles, as defined by the SOPs of the Dermapro Skin Science Institute (Fig. 1). The subjects were informed of the purpose and procedure of this study, the expected efficacy, and potential adverse events. A written signed consent form was obtained in advance from subjects who were interested in participating in the study. Each subject's SC type was determined before the test was begun. For determination of TE type, body mass index (BMI) was

Download English Version:

<https://daneshyari.com/en/article/8510250>

Download Persian Version:

<https://daneshyari.com/article/8510250>

[Daneshyari.com](https://daneshyari.com)