Original Article

Dietary intervention of prebiotic partially hydrolyzed guar gum improves skin viscoelasticity, stratum corneum hydration, and reduction of trans-epidermal water loss: a randomized, double-blind, and placebo-controlled clinical study in healthy humans

Mahendra P. Kapoor,[1,*] Aya Abe,[1] So Morishima,[1] Atsushi Nakajima,[2] Makoto Ozeki,[1] and Norio Sato[3]

[1]Nutrition Division, Taiyo Kagaku Co. Ltd., 1-3 Takara-machi, Yokkaichi, Mie 510-0844, Japan
[2]Ueno Asagao Clinic, 6F Kairaku Building, 2-7-5 Higashiueno, Taito-ku, Tokyo 110-0015, Japan
[3]Taiyo Kagaku Co. Ltd., 800 Yamada-cho, Yokkaichi, Mie 510-1111, Japan

(Received 25 April, 2024; Accepted 3 June, 2024; Released online in J-STAGE as advance publication 7 June, 2024)

Dietary fiber-rich diets are gaining popularity as an alternative therapy for skin health. Plant-based prebiotic partially hydrolyzed guar gum (PHGG) dietary fiber promotes gastrointestinal health, which is imperative for skin health through the gut microbiome. In this randomized, double-blind, and placebo-controlled study, the purpose was to assess the therapeutic effects of PHGG on skin hydration, trans-epidermal water loss (TEWL), and skin viscoelastic properties during the winter season. Healthy male and female subjects (*n* = 70; 9 male and 61 female; mean age: 45.5 ± 8.1 years) were recruited. They received either the 5 g PHGG dietary fiber (*n* = 35) or a 5 g placebo (*n* = 35) for twelve weeks. Skin moisture, TEWL, skin elasticity and skin color parameters, and related features were assessed at baseline, after 6 and 12 weeks, and questionnaires to evaluate the study outcomes. The results confirmed the improvement in skin conditions throughout the winter season by restoring skin hydration, reducing TEWL, and improving skin elasticity parameters. After 6 weeks of PHGG intake, there was a substantial decrease in TEWL and improvement in viscoelasticity metrics when compared to placebo. Subject satisfaction with efficacy reflected these encouraging findings, and PHGG was well tolerated, with no adverse events occurring during the study period.

***Key Words*: stratum corneum, epidermis, viscoelasticity, trans-epidermal water loss, hydration**

The quest for beautiful, flawless, and healthy skin never ends. The skin impacts physical, social, and psychological aspects of health. Therefore, for numerous reasons, it is crucial to continuously develop new strategies for maintaining the health and appearance of the skin.[1–6] In addition to the inherent and intrinsic genetic factors, extrinsic factors such as pollution, ultraviolet radiation, humidity, and poor dietary patterns are also responsible for skin damage. Prolonged exposure to dry winter environments might result in skin disorder due to the comparatively low relative humidity.[7–10] Skin stiffness after washing, number of pores, skincare product maintenance, and lifestyle are other related factors that characterize skin conditions. The skin's outermost layer, the epidermis, acts as the body's first physical defense against environmental contaminants like pollution, allergies, and pathogens. Wherein Ceramide, an intercellular lipid found in the stratum corneum of the epidermis, is crucial for water retention and maintaining the skin's physiological functions.[11,12] The overall conditions of the skin—its surface texture, color, pathological characteristics, and physiological properties such as dryness, roughness, flexibility, wrinkles, laxity, and viscoelasticity—are influenced by factors such as hydration state, i.e., the presence of an adequate amount of water in the stratum corneum, which is determined by a dynamic equilibrium between water retention and water loss from the skin surface.[13–15] A skin stratum corneum with a water level of roughly 10% is optimally hydrated to provide appropriate hydration, resulting in skin that is resilient, supple, luminous, soft, and smooth. Consequently, the moisture level of the skin is directly linked to the stratum corneum water content and is a vital component in facilitating regulated skin tissue functioning.[16–18] In addition to abnormally low humidity, the variations in external environmental temperature also affect the skin hydration level and trans-epidermal water loss (TEWL), which is a commonly used indicator to assess skin barrier function. In the absence of sweat gland activity, TEWL is the typical, constitutive loss of water vapor from the skin.[19–22]

The modification in dietary patterns has gained popularity as an adjunctive remedy for skin problems and damage. Dietary nutrients are widely recognized to alter skin disorders, and a range of populations are specifically seeking diet-based therapeutic alternatives to enhance skin health.[18,23–29] Oral administration of probiotics/prebiotics, vitamins, proteins, minerals, and a variety of other important functional phytochemicals have been reported to improve skin physiological functions and preventive and therapeutic potential in maintaining skin moisture and the extracellular matrix of skin barrier function.[18,30–33] Using biologically active dietary supplements, often classified as nutricosmeticals, to deliver better health benefits for enhancing the youthful appearance of the skin is a recent trend in skin care and rejuvenation.[34] As a result, nutritional supplements that improve human skin have been developed.[35–37]

The integrity of the intestinal barrier has been associated with

*To whom correspondence should be addressed.
E-mail: mkapoor@taiyokagaku.co.jp

doi: 10.3164/jcbn.24-69

skin health.[38] Prebiotics are believed to address skin conditions like dryness, odor, roughness, flexibility, and inflammation because variations in temperature, relative humidity, and other physiological and environmental factors can affect the skin microbiome.[8,9] The moisture level of skin stratum corneum and TEWL may also have a significant impact on the distribution and composition of the microbiome because it contributes to the cutaneous homeostasis and influences host-inflammatory responses.[39–42]

Partially hydrolyzed guar gum (PHGG) is a water-soluble prebiotic dietary fiber with comparatively low viscosity (<12 cps @5% of PHGG) and a reduced average molecular weight (~20,000 Da). PHGG is enzymatically produced from guar gum seeds utilizing β-endogalacto-mannose from an *Aspergillus niger* strain and is commercially available under the brand name Sunfiber® (or Guar fiber).[43] Prebiotic PHGG dietary fiber passes through the upper gastrointestinal tract undigested but is rapidly fermented by gut colonic bacteria into short-chain fatty acids (SCFAs) in the colon, particularly increasing butyrate-producing bacteria, which serve as a significant energy source for the gut epithelia.[44,45] PHGG can also absorb water and promote satiety by delaying digestion.[46,47] Additionally, it has been demonstrated that PHGG alleviates symptoms of diarrhea constipation, and irritable bowel syndrome (IBS).[48–50] There has recently been a noticeable increase in the clinical investigation of water-absorbing substances that modify the gastrointestinal microbiome and bacterially generated metabolites (especially butyrate) for application in dermatology and cosmetology.[51–55]

In a recent 12-week open-label, randomized, controlled trial, we reported the dose response of prebiotic PHGG on the regulation of skin moisture. The study indicated the effect of PHGG dietary fiber on retaining moisture in the skin stratum corneum, and improvement in the characteristic textural qualities of the skin could alter skin conditions among healthy human volunteers.[18] The purpose of the current study was to further evaluate and confirm the impact of prebiotic PHGG dietary fiber intake on the profiles of skin hydration, skin viscoelastic properties, and TEWL as the primary endpoints of this randomized, double-blind, placebo-controlled clinical study in healthy individuals exposed to winter conditions. The present investigation is designed to support the hypothesis and preliminary results of the previous findings. Furthermore, the new information and outcomes gleaned through self-assessment questionnaire-based research associated with skin conditions were deemed as secondary endpoints of the current study.

Materials and Methods

Study design and ethical aspects. To evaluate the impact of PHGG on the effects of environmental factors on the typical profiles of skin conditions in the winter, the current study was an extension of the prior preliminary investigation. The clinical research organization TES Holding Co., Ltd., Tokyo, Japan conducted the present randomized, double-blind, parallel-group, and placebo-controlled investigation at their research site at Ueno-Asagao Clinic, Tokyo, Japan. The study protocol was initially reviewed and approved by the ethical committee expert group of Taiyo Kagaku Co., Ltd., Japan, and subsequently approved by the institutional review board of the Ueno Asagao Clinic Ethics Review Committee and TES Holding Co., Ltd., Japan [Approval No. HR-2023-TYC03 (2022-43); date: 14, December 2022]. Finally, the protocol was registered at the University Hospital Medical Information Network-Clinical Trial Registry (UMIN-CTR; Trial tracking identification: UMIN000049980; date: 6, January 2023).

The study was performed using methodologies that fully comply with the requirements of Good Clinical Practice (GCP), and International Council for Harmonization (ICH) Guidelines for clinical trials. The study procedures were carried out under the Helsinki Declaration and its amendments and the Ethical Guidelines for Medical Research Involving Human Subjects (Ministry of Education, Culture, Sport, Science, and Technology, and Ministry of Health, Labor, and Welfare, 2021).[56] The participants were given a verbal explanation of the study protocols, procedures, and comprehensive information on the study's objectives, including data management, privacy protection, risk, health hazards, compensation, and so on. Participants were told to feel free to withdraw from the study at any time without explaining any reason. Before being screened, the recruited healthy study participants submitted written informed consent to participate in the study, and a copy of their officially signed confirmation was provided for their records.

Study schedule, test materials, supplementation, and dosage. The study was conducted in Tokyo, Japan (latitude: 35° 39′ 10.1952″ N, and longitude: 35° 50′ 22.1208″ E) during the first week of January until mid-April 2023. Variations in outdoor environmental conditions in the greater Tokyo area during the study period of 12 weeks in the winter season are presented in Supplemental Table 1* [Source: Japan Meteorological Agency (JMA)]. PHGG dietary fiber as a test product (manufactured by Taiyo Kagaku Co. Ltd., Mie, Japan; Sunfiber®) and maltodextrin (dextrose equivalence 10.0–12.0; Sanwa Starch Co. Ltd., Nara, Japan) as a placebo were considered in the present study and were ingested orally. PHGG is a unique, water-soluble, virtually tasteless, colorless, and odorless dietary fiber. In dry form, it absorbs less than 7.0% moisture and contains less than 2.0% ash. The subunits of PHGG are D-mannose and D-galactose. The purified PHGG comprises at least 92% galactomannan (a straight backbone chain of D-galacto-D-mannan) with an average molecular weight of nearly 20 kDa. Study participants were instructed to consume either a supplement of 5 g per day of PHGG, which contained nearly 4 g of soluble dietary fiber [≥80%; estimated by the Association of Official Agricultural Chemists Method (AOAC) 2009.01], or placebo once a day, either before or after meals, for 12 consecutive weeks of the study. Both treatments were identically packaged in a sachet made of aluminum foil with no cover printing (5 g by weight; size: 25 × 130 mm) supplied by Taiyo Kagaku. The dietary composition and caloric values of dosage are summarized in Supplemental Table 2*.

Study subjects, sample size, and preliminary selection (screening). The study screened 146 Japanese adult volunteers (n = 146; 24 male and 122 female subjects) to participate in a 12-week skin care remedy clinical trial in the winter season. Subjects were chosen from a pool of volunteers to ensure that the participant group represented a wide range of ages (20–60 years). The sample size estimate for the 5 g/day PHGG dose was calculated based on the preliminary open-label study on the effect of PHGG dietary fiber on skin stratum corneum hydration (SCH).[18] A rough estimate anticipated that about 60–70 volunteers would be required for the current study. As a result, we considered recruiting 70 healthy subjects (35 in each PHGG and placebo intervention group). Also, this was an adequate number of subjects that could be conveniently managed at the assigned clinical research facility. The study's inclusion criteria included being between the ages of 30 and 60, being of Japanese ethnicity, and not having any current or former skin disorders or chronic illnesses. Healthy subjects are required to be willing to participate in a 12-week clinical study and be able to provide signed informed consent.

Subjects who are aware of dry, flaky, or sagging skin. The following were the exclusion criteria: (i) body mass index (BMI) of more than 30 kg/m^2; (ii) history of serious illness, recent surgical procedure, or long-term medical conditions (severe anemia, mental disorders, sleep disorders, etc.); (iii) subjects consuming health-promoting foods and supplements, particularly dietary fiber-enriched foods, or exercise therapy or history of cosmetic treatment in the previous 6 months; (iv) subjects undergoing

*See online. https://doi.org/10.3164/jcbn.24-69

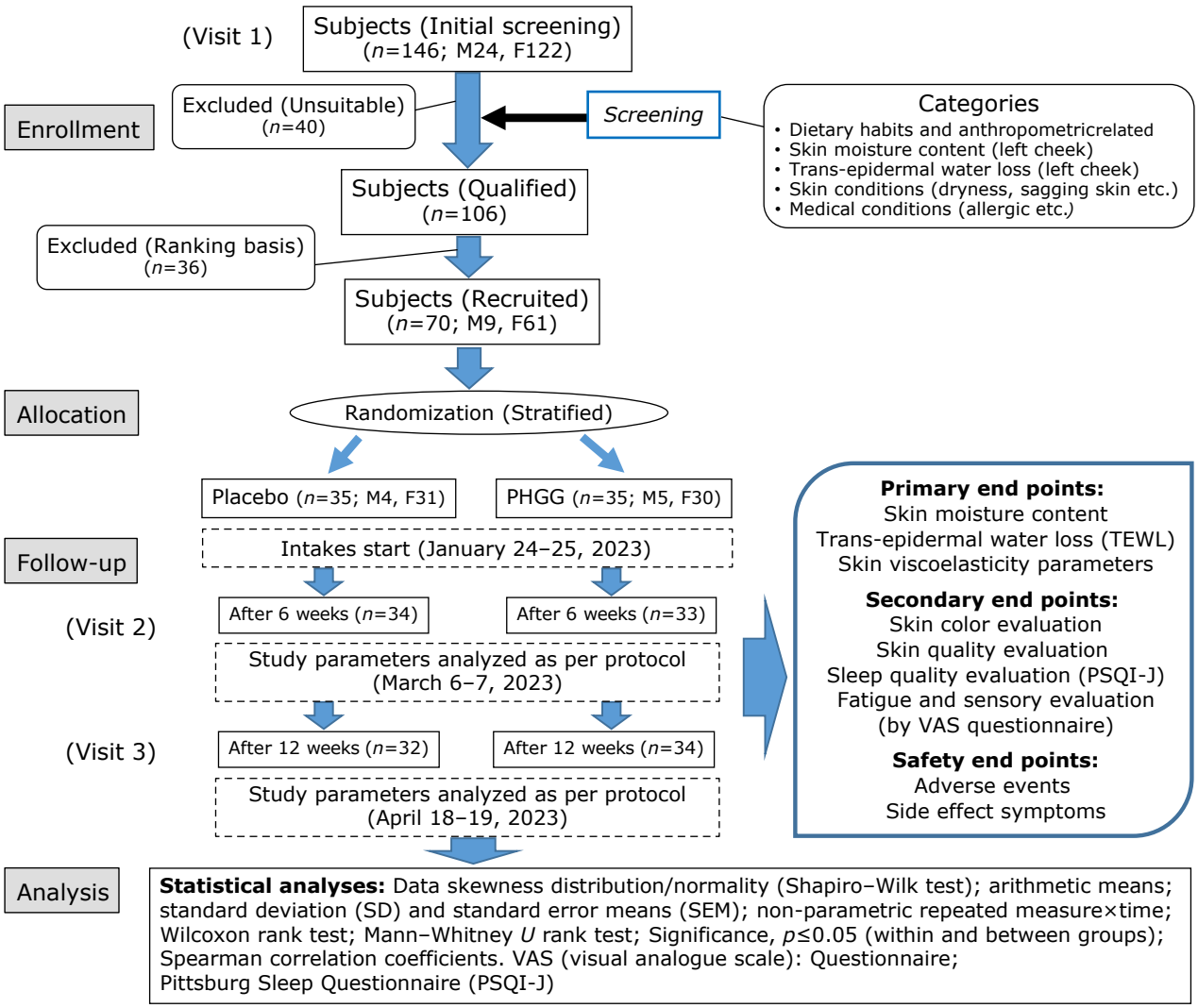

Fig. 1. A schematic illustration display the CONSORT flow chart of the study protocol design and adopted procedures in the present study.

incompatible medication regimens or with current or past drug or food allergies; (v) breastfeeding, nursing and pregnant; (vi) subjects who regularly consume alcohol (>60 g/day) and caffeine (>100 mg/day) (vii) have utilized nutraceutical items or pharmaceutical products or quasi-drugs that promise to improve skin conditions. (viii) subjects who work in shifts (i.e., shift and night shift workers), plan to travel abroad during the research period, or are engaged in work such as sales or delivery, and have to spend a lot of time outdoors activities and have exposure to diverse environmental conditions in daily life; (ix) participants in other clinical studies in the 3 months prior to start of this study; (x) habitual smokers; and (xi) declared ineligible by the medical practitioner or principal investigator of this study. Following an initial screening process, 106 volunteers ($n = 106$) were found to be qualified for the study provided they satisfied the predefined inclusion and exclusion criteria and the principal investigator declared them suitable for participation. The subjects were then screened for additional skin condition measurement indicators, including their background information. A comprehensive ranking was assigned to subjects based on their descending order of skin moisture content and TEWL measured on the left cheek as a test site. Finally, 70 healthy subjects (9 male and 61 female) with the highest overall ranking were enrolled for participation in the study and randomly assigned to receive either a PHGG dietary fiber supplement or a non-active placebo (maltodextrin).

Study protocol, procedures, parameters, and compliance. A schematic illustration of the CONSORT flow chart of the study protocol and the adopted procedural experiment design of the study are detailed in Fig. 1. Following a predetermined enlistment strategy in accordance with protocol, the enrolled 70 healthy male and female subjects ($n = 70$; 9 male and 61 female) were randomly assigned to two separate groups using a block randomization design stratified by gender, age, skin moisture content (cheek), and TEWL (cheek) factors. Subjects were divided into equal block-size groups with a 1:1 allocation ratio to receive either PHGG or placebo ($n = 35$ subjects in each group) for 12 consecutive weeks. The allocation was handled independently by an autonomous researcher at the Ueno-Asagao clinic, in Tokyo, Japan, who was essentially not engaged in the clinical test planning, conducting, protocols, or analyses of the clinical tests. According to protocol, the allocation of corresponding PHGG and placebo supplementations was entirely blinded and remained concealed from the study subjects, general instructors, data management personnel, statistical analysts, and principal investigators until the end of the clinical investigation.

The data collected during the screening was regarded as baseline information (i.e., Visit 1; T0). Several measurements were performed to evaluate the skin's biophysical characteristics

doi: 10.3164/jcbn.24-69

including the state of SCH, TEWL, skin viscoelasticity (SVE) properties, and skin color evaluation. In addition to the observational and sensory evaluation of skin conditions, fatigue, and sleep parameters were monitored using a visual analog scale (VAS) and Pittsburg sleep quality index (PSQI-J) questionnaires.[57] Groups were assigned after a baseline monitoring period of two weeks. The intake of PHGG dietary fiber and placebo supplementation started at the beginning of the final week of January 2023. Considering the possible influence of the seasonal variations, the study was performed in two phases. The subjects were instructed to visit the clinical research facility (Visit 2; T6) after completion of the initial phase of 6 weeks (T6) of respective intakes, and the assessments of the aforementioned enumerated research parameters were then repeated. Finally, after 12 weeks of the completion of the second phase of respective intakes of PHGG and placebo supplementation (Visit 3; T12), the subjects were further examined for all enlisted study parameters at the end of the clinical investigation. The key coding of the supplementation groups that received either PHGG dietary fiber or placebo was disclosed to study coordinators only after all procedural measurements were completed and reported for further analyses to estimate the parameters of the enlisted study outcomes.

The study's compliance was ensured, and the informed intervention's fidelity was assessed. Compliance was evaluated at each visit by counting the number of remaining study product sachets returned by the subject to ensure that investigational products were consumed in accordance with protocol. Adverse event data were collected by the subject reporting any adverse event using a diary log system. In general, the subjects were instructed not to drastically change their lifestyles throughout the study, including food intake patterns, lack of sleep, drinking, and so on. Subjects were prohibited from being exposed to direct sunlight, engaging in outdoor sports or farming activities, or using incompatible commercial skin care products. Additionally, to ensure that subjects are roughly in energy balance, they were advised to maintain the extent and caliber of their usual exercise routines. Male subjects were urged to use an electric shaver. Excessive exercise, high-calorie meals, sleep late at night, use of skin care products, and consumption of alcohol were prohibited 24 h prior to the study parameter measurements. Subjects were instructed to complete the subject diary every day and return it to the clinical trial facility on the scheduled visit. Subjects were given a helpline number to call if they had any problems or issues to report. The follow-up email notification or text message was sent to subjects after 3 weeks and 9 weeks of study supplementation. If necessary, the subjects were contacted independently by phone call.

Measurement of noninvasive skin biophysical functions. The study subjects followed protocol and arrived at the assigned clinical study facility after fasting (having abstained from any food or beverage) for at least 2 h before the start of experimental measurements. Subjects were provided light and comfortable clothes to wear for the measurements to rule out the possibility of the clothing influencing body temperature and to ensure that all subjects were roughly in uniformity of physical conditions. Subjects were instructed to properly wash their faces with the recommended cleanser and facial wash, rinse well with lukewarm water, and then gently pat dry with a paper towel to eliminate any remaining moisture. Before washing their faces, female subjects were instructed to remove all makeup, including mascara, using a specific makeup removal lotion. The other prescribed measurement sites, such as the inner side of the upper arm (left) and upper back (left)] were carefully cleansed with clean cotton before measuring the study parameters. Subjects were then permitted to enter the measurement chamber (temperature: 20.0 ± 1.0°C and humidity: 40 ± 5%) and sit quietly for 20 min to acclimate to the set constant ambient environmental conditions.

A room humidity of 40 and 60% is typically recommended for skin hydration measurements.[29] After becoming accustomed to the experimental chamber conditions, the prescribed physiological skin function measurements (SCH, TEWL, SVE properties, skin color evaluation, and skin quality evaluation) were performed as specified in the study protocol. Comprehensive methodologic and validation investigations have been previously published elsewhere for skin property assessment instruments utilized in the present study.[18] The measurements of skin SCH and TEWL levels, as well as other skin evaluation parameters (e.g., SVE properties, skin color assessment, and skin quality evaluation), were performed at baseline (Visit 1; T0) and after the corresponding intakes of PHGG dietary fiber supplementation and placebo for 6 weeks (Visit 2; T6) and 12 weeks (Visit 3; T12).

Skin stratum corneum hydration measurements. The skin moisturizing function was assessed using the state of the SCH. Three different sites were used to examine the measurements: the cheek (left; parietal bone), the inner side of the upper arm (left), and the upper back (left). The Corneometer CM825® (Courage-Khazaka Electronic GmbH, Köln, Germany) with a digital low-weight probe and a small measurement area (49 mm^2) was used to determine skin SCH levels, which are suitable for examining the hydration of the upper layers of the epidermis via skin surface impedance.[58] The probe head has a spring that ensures that a constant pressure of 1.0 N/cm^2 is applied to the measurement areas for 1 s, minimizing the effects of occlusions. This allowed for the detection of the frequency shift (with ±3% accuracy, between 0.9 and 1.2 MHz) of the oscillating system related to the skin capacitance. Three measurements were recorded after subtracting the highest and lowest values from the five sets of measurements that were taken at the same location. To estimate the skin SCH result, either the mean of the three measured values or the set of the three measures with the lowest standard deviation was used. These SCH are either in system-specified units, or an arbitrary unit (a.u.), and are expressed as the degree of epidermal humidity.

Trans-epidermal water loss measurements. The TEWL levels were also assessed at three sites: the cheek (left; parietal bone), the inner side of the forearm (left), and the upper back (left) after the skin SCH measurements. A calibrated Tewameter TM300® was utilized for this purpose (Courage-Khazaka Electronic GmbH) following the manufacturer's instructions. The instrument measures the TEWL of the skin stratum corneum, expressed in $g/m^2/h$. While all measurements were performed for 60 s; however, the data used for the TEWL analysis was an average value from the first 30 s recorded with a minimum SD value of 5 s.

Skin viscoelasticity functions. The viscoelastic characteristics of the skin serve as a marker of skin aging or damage and they can be measured by applying a certain pressure to the skin and observing the degree of restoration. These properties are mostly connected to the thickness and quality of the epidermis. The Cutometer MPA580® (Courage-Khazaka Electronic GmbH) was used to measure the elasticity of the face (left cheek), the region in the middle linking the lower earlobe, and the edge of the lips. The device was outfitted with a 2 mm diameter probe that suctioned onto the skin's surface, creating a perpendicular depression in its cylindrical chamber affixed to the skin measuring area of several hundred millibars. The length of the skin drawn inside the probe was then measured by a glass prism with a millimeter scale. The measurements were performed with a negative pressure of 300 mbar, a suction time of 2 s, and a relaxation phase of 2 s. Skin viscoelastic parameters were measured five times at the same spot to reduce the error; however, data was recorded for three measures and the greatest and lowest values were eliminated before averaging the values. Similarly, for the estimation of the SVE function outcome, either the measured average value or the entire set from the three measurements with

the lowest SD was selected. The parameters that are typically employed to describe SVE were evaluated. R0 (*Uf*) represents the final distension of skin sucked into the aperture of the probe when elongated. R2 (*Ua/Uf*) reflects gross elasticity (i.e., represents the recovery ratio of the skin length following elongation and constriction). R5 (*Ur/Ue*) represents pure or net elasticity (i.e., expresses the ratio of an elastic portion of relaxation to an elastic portion of suction). R6 (*Uv/Ue*) is the ratio of the viscoelasticity and elasticity portions when elongated. The R7 (*Ur/Uf*) coefficient is a biological elasticity that measures the skin's ability to return to its original position following deformation (i.e., the ratio between elastic return and total deformation).

Skin color evaluation. Skin color is the combination of melanin (yellowish-brown color) and hemoglobin (red color) and is determined by the melanin distribution on the skin epidermis level. In general, the skin color variations are caused by hormonal factors; however, the change in skin color may also be the result of environmental factors like sun exposure (i.e., tanning ability and the frequency of erythema) and seasonal winter conditions with extremely low humidity can also affect skin color by altering in the amount of melanin on the epidermis. A Chroma meter (CM-2600d; Konica Minolta Inc., Osaka, Japan) was used to measure L*, a*, and b* parameters of the standard CIE Lab (Commission Internationale de l'Eclairage) color space at the specified measurement sites. For instance, the L*a*b* color system is a three-dimensional coordinate system with an L* axis (brightness) and two orthogonal axes indicating chromaticity, notably the a* axis coordinate (red-green) and the b* axis coordinate (blue-yellow).[59] The measurement sites were under the left eye (lower inner canthus), which is adjacent to the test site region as a reference site, and the left cheek, which served as the test site. In addition to absolute color values based on the CIE color standard, the color difference can be directly measured using this device in the wavelength range of 360–740 nm, which is essentially the electromagnetic radiation of visible light sensed by the human eye and brain. Also, the difference in values between the test site and the corresponding neighboring control site of two color stimulus parameters L* and a* were estimated as ΔL* and Δa*. The software applies an algorithm to individually calculate the melanin index (MI), hemoglobin index (Hb-index), and hemoglobin with saturated oxygen index ($HbSO_2$-index, %) based on the spectral reflectance data collected by a spectrophotometer. The spectrophotometer's head was merely pressed against the skin to collect five measurements at the same location. The highest and lowest values from the measurements were then eliminated before the data was automatically averaging the selected three measurements. The measurement period was roughly 1.5 s, and the measuring area's mean average value was 8–11 mm.

Visual evaluation by a dermatologist (a professional skin specialist). A visual inspection of the subject's skin was performed by a qualified dermatologist, noticing cutaneous grooves, and cutaneous carpus, and conducting a thorough comprehensive evaluation. A digital microscope KH-1300/HXG-2016Z (Hirox Co. Ltd., Tokyo, Japan) equipped with an automatic calibration select (ACS) function and a 20–160× zoom lens was used for visual judgment. The pocket dermatoscopy DermLiteDL100 (DermLite LLC, San Juan Capistrano, CA; Jay Hewitt Co. Ltd., Tokyo, Japan) designed for skin lesions examination with magnification and cross-polarized light (3 mm white LEDs and 10× lens) was used to obtain the desired image focus. Five-point rating scale criteria (−2: extremely poor, −1: poor, 0: fair, 1: little good, and 2: very good) were used to judge the evaluation to minimize bias and ambiguity and to accurately record the severity and direction of subjects' skin conditions.

Skin quality evaluation. A digital microscope KH-1300/HXG-2016Z (Hirox Co. Ltd., Tokyo, Japan) and a pocket dermatoscopy DermLiteDL100 (DermLite LLC; Jay Hewitt Co. Ltd.) were also used to further evaluate the subject's skin quality. The quality of the skin was evaluated on the entire face based on the following skin conditions: dryness, erythema, scales, irritation, and pruritus, which is defined as a severe itching sensation. The assessment was performed utilizing a five-point Likert rating scale criteria for the examined skin symptoms (0: none, 1: minor, 2: mild, 3: moderate, and 4: severe) to reveal key insights into the subject's skin quality.

Observation evaluations and questionnaires..

Sensory and Fatigue visual analog scal questionnaire. A self-assessment questionnaire with a visual analog scale (VAS) is comprised of two sections. The first section consists of questions related to comprehensive face skin conditions like brightness, smoothness, moist feeling, moisturized level, glossiness, sagging, firmness, wrinkles, elasticity, dullness, clarity, acne and pimples, roughness, pore visibility, pimples, itchy, dryness, dark circles, etc. The other section asks questions about stress, makeup, and daily bowel movements including the subject's appreciation of their intake of study treatments. Each participant in each group completed this questionnaire on the sensory-visual analog scale, with the leftmost and rightmost sides denoting no symptomatic conditions and the worst symptoms, respectively, on the VAS horizontal line measuring 100 millimeters (in length). The subjects were required to respond to questions related to subjective sensation, and the distance marked from the left side was scored. The subjects filled out the first section of the questionnaire at baseline (T0), while both sections were completed twice, once after completion of the initial 6 weeks phase (T6) as well as after 12 weeks at the end of the clinical study (T12). The fatigue-related VAS questionnaire was assessed according to the recommended method by the Japan Society for Fatigue Science, with an emphasis on the assessments of both physical and mental fatigue.

Subjective sleep quality survey. The Pittsburg Sleep Quality Index (PSQI) is a well-validated self-reported questionnaire used to assess sleep quality and efficiency. The Japanese version of the Pittsburg Sleep Quality Index (PSQI-J) questionnaire was used to assess the participant's subjective sleep efficiency, quality, and disturbance in sleep at baseline (Visit 1; T0), after completion of 6 weeks into the initial phase (Visit 2; T6), and after 12 weeks at the end of the clinical study (Visit 3; T12). At baseline, a survey on sleep status during the previous month was evaluated. Nineteen individual items generate seven component scores, including subjective sleep quality, sleep latency, sleep duration, habitual sleep efficiency, disturbances in sleep, use of sleeping medication, and daytime dysfunction. One global score is produced by adding the scores for these seven components. The quality and efficiency of sleep decrease with increasing global score.[57]

Physical evaluation and body functions. A calibrated portable stadiometer with an accuracy of 0.1 cm was used to measure each subject's height during Visit 1 (T0) at the clinical trial facility. A digital weighing scale with a sensitivity of 0.1 kg was used to assess each subject's body mass (weight). Body mass index (BMI) was computed by dividing body weight in kilograms by height in meters squared. A palm-style semi-automatic ES-H55 Elemano (Terumo Medical Corporation, Somerset, NJ) electronic blood pressure monitor (sphygmomanometer) equipped with a double-cuff air bladder and an irregular pulse detection function was used to measure the systolic and diastolic blood pressures as well as the pulse rate. The measurements are taken from the upper arm, two to three centimeters proximal to the elbow, keeping a two-finger gap between the cuff and the arm while maintaining the cuff position at the level of the heart. All measurements were recorded digitally.

Safety aspects and related evaluations. During the investigation period, the study principal investigator or study subcontractor determined the subjective symptoms and adverse events, which were then tabulated and assessed as objective data. No

doi: 10.3164/jcbn.24-69

Table 1. The baseline clinical profiles of the healthy study subjects indicate a homogeneous representation of participants in total and age-stratified groups

Characteristics	Gender (M/F)	Age (years)	Height (cm)	Weight (kg)	BMI (kg/m^2)	SBP (mmHg)	DBP (mmHg)	Pulse rate (bpm)
Total subjects (n = 70)	9/61	45.5 ± 8.1	160.4 ± 6.4	55.0 ± 8.0	21.4 ± 2.7	115.5 ± 14.5	73.0 ± 11.7	73.8 ± 10.1
Placebo (n = 35)	4/31	45.5 ± 8.7	159.0 ± 5.9	53.4 ± 7.0	21.1 ± 2.4	116.2 ± 17.2	73.1 ± 13.2	74.0 ± 9.2
PHGG (n = 35)	5/30	45.6 ± 7.6	161.7 ± 6.7	56.5 ± 8.6	21.6 ± 3.0	114.9 ± 11.5	72.9 ± 10.1	73.7 ± 11.1
p value	1.00	0.953	0.074	0.106	0.461	0.708	0.951	0.879
≤45 years (n = 30)	5/25	37.6 ± 4.4	162.0 ± 6.5	54.2 ± 7.8	20.6 ± 2.1	108.2 ± 13.1	67.5 ± 10.6	73.5 ± 12.1
Placebo (n = 16)	2/14	37.3 ± 4.3	159.9 ± 5.7	52.8 ± 7.9	20.6 ± 2.1	109.1 ± 15.1	68.9 ± 13.3	72.8 ± 9.1
PHGG (n = 14)	3/11	37.9 ± 4.6	164.4 ± 6.8	55.8 ± 7.6	20.6 ± 2.2	107.1 ± 10.8	65.9 ± 6.4	74.3 ± 15.1
p value	0.642	0.742	0.063	0.291	0.914	0.676	0.446	0.734
≥46 years (n = 40)	4/36	51.5 ± 3.9	159.1 ± 6.1	55.6 ± 8.2	21.9 ± 2.9	121.0 ± 13.2	77.1 ± 10.8	74.1 ± 12.1
Placebo (n = 19)	2/17	52.4 ± 4.0	158.2 ± 6.1	54.0 ± 6.4	21.6 ± 2.6	122.1 ± 16.9	76.6 ± 12.4	75.1 ± 9.4
PHGG (n = 21)	2/19	50.8 ± 3.7	160.0 ± 6.2	57.0 ± 9.4	22.2 ± 3.3	120.0 ± 8.9	77.6 ± 9.4	73.2 ± 7.9
p value	1.00	0.199	0.368	0.254	0.499	0.629	0.776	0.499

p value: Placebo vs PHGG; Significance $p \leq 0.05$; (no significant differences between placebo and PHGG groups at baseline); Mean ± SD; SBP, systolic blood pressure; DBP, diastolic blood pressure; bpm, beats per min.

outliers were set. However, in any situation where there was a significant issue with the data's dependability, it was classified as a missing value. Adverse events were investigated using a subject background questionnaire, a health practitioner interview, the subject's daily diary, and a side effect assessment.

Data analysis and statistical evaluation. Statistical analyses were performed using JMP (John's Macintosh Project) statistical data analysis software package (SAS Institute Inc., Cary, NC; ver. 14.0 for Windows) in accordance with the ICH guideline E9 "Statistical Principles for Clinical Trials".[23] The mean values of the measurements of study outcomes were calculated and analyzed at each point. Unless otherwise indicated, all findings are expressed as mean ± SD. In terms of clinical relevance, the p values of ≤0.05 were deemed statistically significant, while ≤0.10 was viewed as trending toward significance.[60] The normal quantile (Q-Q) plots and histogram were used to examine the distribution of all the data, and the Shapiro-Wilk W test was used to determine the normality of the data. For a normal distribution, modest p values (p<0.05) are used to reject the null hypothesis (H0). Where necessary, data for the skewness distribution was examined, and Levene's test was used to verify the homogeneity of variance. Non-parametric statistical analysis was applied to variables that did not follow a normal distribution. When the normality test failed for non-parametric analysis in Shapiro–Wilk, the data were subjected to the Wilcoxon rank test for paired t test (within the group; comparing T6 vs T0, and T12 vs T0), and Mann–Whitney U test for independent samples t test [between groups; comparing the PHGG dietary fiber and placebo groups concerning the relative changes (Δ) in the means of study outcomes: T6–T0 and T12–T0]. The within-group effect of treatments x time as the interaction was also evaluated using a non-parametric repeated measure one-way ANOVA on ranks, with an a priori significance criterion of $p \leq 0.05$. Before analysis, any anomalies or absent data points were examined. Before analysis, any outliers or missing data points were examined. The percentage changes in each outcome within the intervention groups between two-time points (i.e., from baseline to 6 weeks and 12 weeks) were calculated as % change = [(mean value of post-supplement-mean value at baseline)/mean value at baseline] × 100.

Results

Study characteristics and subject demographics. The subject's demographic and primary baseline characteristics are listed in Table 1. The enrolled subjects (n = 70; 9 male and 61 female) aged between 30 and 60 years with an average age of 45.5 ± 8.1 years were equally distributed by stratified randomization between the PHGG dietary fiber (n = 35; 5 male and 30 female) and placebo (n = 35; 4 mele and 31 female) groups based on the following characteristics: age, gender, and their ranking of skin moisture and TEWL. The present study included all Japanese subjects with the Fitzpatrick skin classification of Type III. The majority of the subjects in this study were female (87.1%). Furthermore, according to the use of a two-sample comparison of means at the alpha (α) = 0.05 level of significance, the total sample size of each group provided more than 90% power to identify an appropriate effect size.

The average age and body mass index (BMI) of the PHGG dietary fiber and placebo groups were 45.6 ± 7.6 and 45.5 ± 8.7 years, respectively, and 21.6 ± 3.0 and 21.4 ± 2.7 kg/m^2, respectively. At baseline, there were no discernible changes in the anthropometric parameters of age, height, body weight, and BMI between the PHGG dietary fiber and placebo groups. When the groups' SCH and TEWL were assessed at baseline, there was no discernible difference between them, indicating that the groups were homogeneous. The study's subjects complied with all protocol requirements, and no significant protocol violations were noted. However, one female subject from each of the placebo and PHGG dietary fiber groups withdrew from the study after the study intakes began. Also, two female subjects withdrew from the placebo group after 6 weeks of intake for personal reasons, and one female subject who received PHGG dietary fiber was unable to complete the measurements of the study parameters after 6 weeks (Visit 2; T6), but she was available to complete the study measurements after 12 weeks (Visit 3; T12) (see schematic CONSORT flow diagram in Fig. 1). There was no discernible difference in compliance between the PHGG dietary fiber and placebo groups, as evidenced by compliance rates of more than 98.5% in both the PHGG dietary fiber and placebo groups, indicating that there was no statistically significant difference in compliance across groups.

Skin moisturizing functions. Based on the results of TEWL and SCH tests performed on every individual in each group, the skin moisturizing functions were assessed. Figure 2 depicts a

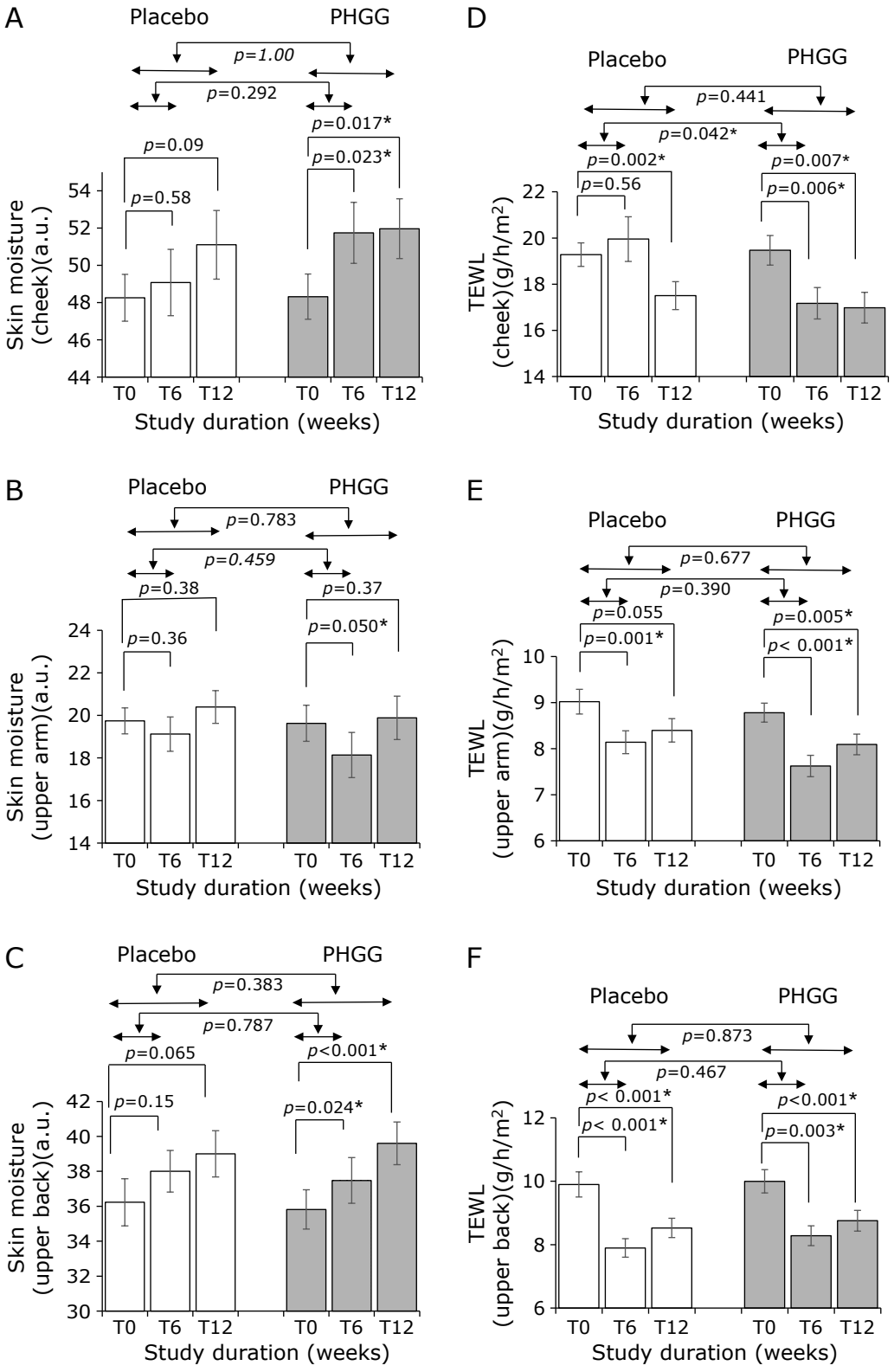

Fig. 2. Statistical comparison of the (A–C) skin moisture and (E, F) trans-epidermal water loss (TEWL) at the cheek, upper arm, and upper back of study subjects after 6 weeks (T6) and 12 weeks (T12) of consuming either placebo or PHGG dietary fiber compared to baseline (T0), as well as between the placebo and PHGG dietary fiber groups (* denote significance of *p* value; $p \leq 0.05$; Mean ± SEM).

doi: 10.3164/jcbn.24-69

Table 2. The variation in skin moisture (i.e., stratum corneum hydration level) and trans-epidermal water loss at different sites in healthy study subjects during 12 consecutive weeks of placebo or PHGG dietary fiber

Analysis parameters	Skin moisture (arbitrary unit)					
	Baseline (T0; n = 35)	6 weeks (T6; n = 34)	12 weeks (T12; n = 34)	T0 vs T6	T0 vs T12	RM × Time
	Mean ± SD	Mean ± SD	Mean ± SD	p value (within-group)		χ^2; p value
Cheek-left (parietal bone)						
Placebo	48.26 ± 7.42	49.07 ± 10.37	§51.10 ±10.42	0.584	0.087	χ^2 = 3.94; p = 0.140
PHGG	48.32 ± 7.20	#51.74 ± 9.40	51.96 ± 9.34	0.023*	0.017*	χ^2 = 2.36; p = 0.307
†p value (between groups)		p = 0.292	p = 1.00			
Arm-left (upper inner)						
Placebo	19.74 ± 3.60	19.12 ± 4.69	§20.40 ± 4.36	0.357	0.379	χ^2 = 3.32; p = 0.190
PHGG	19.63 ± 5.04	#18.14 ± 6.10	19.89 ± 5.94	0.050*	0.372	χ^2 = 3.68; p = 0.159
†p value (between groups)		p = 0.459	p = 0.783			
Back-left (upper)						
Placebo	36.23 ± 7.98	38.01 ± 6.97	§39.00 ± 7.49	0.153	0.065	χ^2 = 3.06; p = 0.216
PHGG	35.82 ± 6.66	#37.48 ± 7.53	39.60 ± 7.12	0.024*	<0.001*	χ^2 = 12.8; p = 0.002*
†p value (between groups)		p = 0.787	p = 0.383			
	Transepidermal water loss (g/h/m²)					
Cheek-left (parietal bone)						
Placebo	19.28 ± 3.01	19.95 ± 5.65	§17.51 ± 3.43	0.555	0.002*	χ^2 = 8.24; p = 0.016*
PHGG	19.469 ± 3.79	#17.17 ± 3.92	16.98 ± 3.88	0.006*	0.007*	χ^2 = 13.5; p = 0.001*
†p value (between groups)		p = 0.042*	p = 0.441			
Arm-left (upper inner)						
Placebo	9.02 ± 1.59	8.14 ± 1.45	§8.40 ± 1.44	0.001*	0.055	χ^2 = 10.2; p = 0.006*
PHGG	8.781 ± 1.22	#7.62 ± 1.33	8.09 ± 1.30	<0.001*	0.005*	χ^2 = 14.7; p<0.001*
†p value (between groups)		p = 0.390	p = 0.677			
Back-left (upper)						
Placebo	9.90 ± 2.36	7.89 ± 1.71	§8.53 ± 1.72	<0.001*	<0.001*	χ^2 = 19.2; p<0.001*
PHGG	9.99 ± 2.18	#8.28 ± 1.80	8.76 ± 1.91	0.003*	<0.001*	χ^2 = 31.1; p<0.001*
†p value (between groups)		p = 0.467	p = 0.873			

*Significance $p \leq 0.05$; RM, repeated measure (non-parametric); †Between group (change from baseline): Mann–Whitney rank test, Within group: Wilcoxon rank test; #n = 33; §n = 32.

descriptive analysis of skin SCH and TEWL at baseline (T0; before intake of study products), after 6 weeks of intake (T6) during the first phase of study, and at the end of the second phase of the clinical trial after 12 weeks of intake (T12) period of study at the cheek (left; parietal bone), the inner side of the upper arm (left), and the upper back (left) measurement sites for both groups, with corresponding results summarized in Table 2.

Skin stratum corneum hydration. The mean initial skin SCH state at T0 for the PHGG dietary fiber group and the placebo group did not differ significantly. In comparison to the inner side of the upper arm and the upper back in both groups, the measured mean skin SCH level was higher at the cheek (left; parietal bone). The mean values for subjects in the PHGG dietary fiber group were 48.32 ± 1.0 a.u. at the cheek, 35.82 ± 1.0 a.u. at the upper back, and 19.63 ± 1.0 a.u. at the inner side of the upper arm, as indicated in Table 2. In contrast, the subjects in the placebo group had mean values at the cheek, upper back, and inner side of the upper arm of 48.26 ± 1.0 a.u., 36.23 ± 1.0 a.u., and 19.74 ± 1.0 a.u., respectively. After completing the first phase of 6 weeks of consumption (T6), the PHGG dietary fiber group had significantly higher mean skin SCH levels on the cheek, upper arm, and upper back compared to the baseline (T0). Conversely, the placebo group experienced a small and non-significant rise in the SCH mean value at all measurement sites. At the end of the trial, 12 weeks (T12) after starting the treatments, the measured skin mean SCH level at the cheek and upper back compared to baseline (T0) continues to follow a similar trend. There were no statistically significant differences seen between the groups, or in the relative changes in skin SCH levels of both groups at the cheek, upper back, or upper arm sites during the T6 and T12 phases of the study.

Skin trans-epidermal water loss. The levels of TEWL measured with Tewameter for subjects in the PHGG dietary fiber and placebo groups are also presented in Table 2, along with skin SCH. For every subject in each group, the measured mean value of skin TEWL level at baseline (T0) was approximately twice as high at the cheek compared to it at the inner side of the upper arm and the upper back measurement sites. The placebo group showed a non-significant increase in the mean values of TEWL level after first phase intakes at 6 weeks (T6), whereas the PHGG dietary fiber group indicated a significant decrease compared to baseline (Fig. 2D). Additionally, there was a statistically significant difference between the groups that received PHGG dietary fiber and the placebo. Furthermore, the seasonal effect can be attributed solely in the placebo and partially in the PHGG dietary fiber intake groups to a significant decrease in mean values of both the dietary fiber and placebo after 12 weeks of intakes (T12) compared with that at baseline (T0). As a result, there was no discernible significant difference between the PHHG dietary fiber and placebo groups.

Skin elasticity parameters. The changes in the biomechanical skin parameters pertaining to skin viscoelastic properties

Table 3. The variation in primary skin viscoelasticity indices in healthy study subjects during 12 consecutive weeks of placebo or PHGG dietary fiber administration during the winter season

Description (face-left side)	Baseline (T0) Mean ± SD (n = 35)	6 weeks (T6) Mean ± SD (n = 35)	12 weeks (T12) Mean ± SD (n = 35)	‡Within group (p value)				†Between groups (p value)	
				T6 vs T0	T12 vs T0	T6 vs T12	RM × Time	T6–T0 (ΔT6)	T12–T0 (ΔT12)
Uf (R0)									
Placebo	0.310 ± 0.064	0.295 ± 0.060	§0.291 ± 0.049	0.014*	0.054	0.550	χ^2 = 5.06; p = 0.080		
PHGG	0.303 ± 0.058	#0.289 ± 0.052	0.283 ± 0.049	<0.001*	<0.001*	0.340	χ^2 = 22.5; p<0.001*	p = 0.711	p = 0.115
Ua/*Uf* (R2)									
Placebo	0.777 ± 0.060	0.799 ± 0.066	§0.803 ± 0.060	0.003*	0.001*	0.543	χ^2 = 12.0; p = 0.003*		
PHGG	0.762 ± 0.072	#0.791 ± 0.076	0.808 ± 0.067	<0.001*	<0.001*	0.002*	χ^2 = 26.8; p<0.001*	p = 0.410	p = 0.036*
Ur/*Ue* (R5)									
Placebo	0.596 ± 0.102	0.614 ± 0.115	§0.607 ± 0.102	0.058	0.19	0.299	χ^2 = 1.72; p = 0.424		
PHGG	0.585 ± 0.110	#0.627 ± 0.110	0.631 ± 0.115	<0.001*	<0.001*	0.36	χ^2 = 14.4; p<0.001*	p = 0.192	p = 0.038*
Uv/*Ue* (R6)									
Placebo	0.366 ± 0.073	0.367 ± 0.067	§0.373 ± 0.083	0.352	0.531	0.736	χ^2 = 2.37; p = 0.306		
PHGG	0.363 ± 0.071	#0.395 ± 0.095	0.404 ± 0.086	0.004*	0.005*	0.264	χ^2 = 9.13; p = 0.010*	p = 0.081	p = 0.039*
Ur/*Uf* (R7)									
Placebo	0.440 ± 0.080	0.452 ± 0.093	§0.443 ± 0.078	0.152	0.290	0.237	χ^2 = 1.31; p = 0.520		
PHGG	0.432 ± 0.093	#0.454 ± 0.097	0.458 ± 0.094	0.003*	0.001*	0.228	χ^2 = 10.2; p = 0.006*	p = 0.048*	p = 0.031*

*Significance $p \leq 0.05$; RM, repeated measure (non-parametric); †Between group: Mann–Whitney U rank test, ‡Within group: Wilcoxon rank test; R0 = Distensibility; R2 = Gross elasiticity; R5 = Net elasticity (SE); R6 = Elastic ratio (viscoelasticity/elasticity); R7 = Skin ViscoElasticity (SVE); #n = 33; §n = 32.

revealed a significant decrease in final deformation, i.e., distensibility (*Uf*; R0), from baseline at 6 weeks (T6) and 12 weeks (T12) of the study periods, regardless of PHGG dietary fiber and placebo intakes (Table 3). Whereas, there was no discernible difference in the mean values of distensibility (R0) at T0 and T6, as well as T0 and T12 between the PHGG and placebo groups. Nonetheless, over the course of the trial, there was a significant difference in treatment × time interaction between the PHGG dietary fiber and placebo groups (Table 3). However, the gross skin elasticity (SE; R2), i.e., viscous deformation (*Ua/Uf*, wherein *Ua* is a total deformation following suction) was found to be significantly increased from baseline at 6 weeks (T6) and 12 weeks (T12) of study periods in both PHGG dietary fiber and placebo groups.

During the study period, a significant within-group treatments × time interaction was noted in both intake groups. Nevertheless, the changes in gross elasticity (R2) from T0 to T6 as well as T0 to T12 between the PHGG and placebo groups could reach a significant level only after 12 weeks (T12) of intakes, which is correspond to a 6.04% increase in mean value (Table 3). The net SE without viscous deformation (*Ur/Ue*; R5), wherein *Ur* and *Ue* are immediate retraction and deformation (extensibility), respectively, showed a significant increase in skin net elasticity at T6 and T12 in the PHGG dietary fiber group, but a non-significant increase at T6 and T12 in the placebo group.

Further, the changes in net elasticity (R5) from T0 and T6 as well as T0 and T12 between PHGG and placebo groups were found to be significant only after 12 weeks (T12) of respective intakes. There was also a significant difference in treatment × time interaction between the PHGG dietary fiber and placebo groups during the study duration. Additionally, the results indicated that there was no significant variation in the viscoelastic ratio (Uv/Ue), wherein Uv denotes a delayed deformation (i.e., plasticity; R6) at T6 and T12 of the study periods in the placebo group, while the PHGG dietary fiber intake group showed a significant increase from baseline at T6 and T12. Additionally, during the course of the trial, there was a significant difference in treatment × time interaction between the PHGG dietary fiber and placebo groups. Furthermore, Table 3 indicates that a change in the viscoelastic ratio (R6) between PHGG dietary fiber and placebo groups at T0 and T6 demonstrated a trending significance, and a change in the viscoelastic ratio between T0 and T12 could reach a significant level. Likewise, the skin biological elasticity (Ur/Uf; R7) for the placebo group increased non-significantly at T6 and T12 of study periods, whereas for the PHGG dietary fiber intake group, a significant increase was noticed at T6 and T12 of study periods. Furthermore, there was a significant difference in treatment × time interaction observed between the PHGG dietary fiber and placebo groups for the study duration. Table 4 shows that there was a significant difference in the changes (from T0 to T6 as well as from T0 to T12) in skin biological elasticity (R7) between the PHGG dietary fiber and placebo groups.

Skin color variation. The variations in L*, a*, and b* values during the PHGG dietary fiber and placebo intake are presented in Table 4. At the cheek, the PHGG dietary fiber group showed a significant increase in the L* value from baseline at 6 weeks (T6) and 12 weeks (T12) of study periods. While the placebo group indicated a non-significant decrease in the L* value from baseline at 6 weeks (T6) and a slight increase at 12 weeks (T12) of study periods. Moreover, there was a significant difference in the treatment × time between the PHGG dietary fiber and placebo groups for the study duration. Whereas, the changes observed in L* values (from T0 to T6, as well as T0 to T12) between PHGG and placebo groups reach a significant level after 6 weeks (T6) of intakes (Table 4). Although both placebo and PHGG dietary fiber groups showed a reduction in redness (a*) from baseline, the decrease was statistically significant with PHGG intake at week 6 (T6) and 12 weeks (T12) compared to placebo. Similarly, there was a significant difference in treatment × time interaction between the PHGG dietary fiber and placebo groups for the study duration. However, no significant difference was observed between the PHGG dietary fiber and placebo groups. Conversely, both the PHGG dietary fiber and placebo groups showed an increase in b* values, however, no significant could emerge (Table 4).

A decrease in Hb-index value was noted for both the PHGG dietary fiber and placebo intake groups. However, the PHGG dietary fiber intake group showed a significant difference only after 6 weeks (T6). There was no between-group significance or

doi: 10.3164/jcbn.24-69

Table 4. The variation in estimated CIE-L*a*b* coordinates of the skin and skin color change markers at different sites in healthy study subjects during twelve consecutive weeks of placebo or PHGG dietary fiber administration during the winter season

Description	Parameters (analyzed)	Baseline (T0) Mean ± SD	6 weeks (T6) Mean ± SD	12 weeks (T12) Mean ± SD	‡Within group (*p* value)			†Between groups (*p* value)	
					T6 vs T0	T12 vs T0	RM × Time	T6–T0 (ΔT6)	T12–T0 (ΔT12)
Cheek (left)									
Placebo		(*n* = 35)	(*n* = 34)	(*n* = 32)					
	Brightness (L*)	61.65 ± 3.73	61.54 ± 3.69	61.78 ± 3.02	0.778	0.410	χ^2 = 1.00; *p* = 0.607		
	Redness (a*)	12.70 ± 2.42	12.68 ± 1.91	12.62 ± 2.05	0.437	0.374	χ^2 = 1.56; *p* = 0.458		
	Yellowness (b*)	16.86 ± 2.18	17.11 ± 2.13	17.15 ± 2.30	0.059	0.091	χ^2 = 3.81; *p* = 0.149		
	Melanin index	1.26 ± 0.19	1.26 ± 0.21	1.25 ± 0.20	0.745	0.410	χ^2 = 0.81; *p* = 0.67		
	Hb-index (%)	1.75 ± 0.56	1.72 ± 0.43	1.73 ± 0.46	0.256	0.287	χ^2 = 2.44; *p* = 0.296		
	$HbSO_2$-index (%)	56.63 ± 6.09	57.44 ± 5.01	56.94 ± 4.86	0.059	0.701	χ^2 = 5.69; *p* = 0.058		
PHGG		(*n* = 35)	(*n* = 33)	(*n* = 34)					
	Brightness (L*)	61.57 ± 2.637	62.85 ± 2.14	62.12 ± 2.29	<0.001*	0.002*	χ^2 = 22.7; *p*<0.001	*p* = 0.010*	*p* = 0.663
	Redness (a*)	12.87 ± 1.329	12.17 ± 1.42	12.48 ± 1.59	<0.001*	0.033*	χ^2 = 11.9; *p* = 0.003*	*p* = 0.194	*p* = 0.842
	Yellowness (b*)	16.29 ± 2.280	16.30 ± 2.27	16.35 ± 2.10	0.604	0.543	χ^2 = 0.97; *p* = 0.616	*p* = 0.284	*p* = 0.323
	Melanin index	1.28 ± 0.188	1.17 ± 0.16	1.20 ± 0.150	<0.001*	0.049*	χ^2 = 9.88; *p* = 0.007*	*p* = 0.034*	*p* = 0.719
	Hb-index (%)	1.87 ± 0.328	1.74 ± 0.35	1.80 ± 0.35	0.039*	0.274	χ^2 = 4.44; *p* = 0.108	*p* = 0.638	*p* = 0.974
	$HbSO_2$-index (%)	55.36 ± 4.926	57.36 ± 4.54	55.40 ± 4.52	0.002*	0.886	χ^2 = 11.1; *p* = 0.004*	*p* = 0.313	*p* = 0.878
Under eye (left) - reference site									
Placebo		(*n* = 35)	(*n* = 34)	(*n* = 32)					
	Brightness (L*)	61.47 ± 2.88	60.81 ± 2.96	61.70 ± 2.46	0.006*	0.355	χ^2 = 12.6; *p* = 0.002*		
	Redness (a*)	11.31 ± 1.67	11.47 ± 1.53	11.33 ± 1.62	0.180	0.385	χ^2 = 3.62; *p* = 0.164		
	Yellowness (b*)	17.42 ± 2.07	17.38 ± 1.87	17.49 ± 2.12	0.898	0.665	χ^2 = 0.44; *p* = 0.804		
	Melanin index (MI)	1.23 ± 0.19	1.23 ± 0.19	1.21 ± 0.19	0.538	0.221	χ^2 = 2.44; *p* = 0.296		
	Hb-index (%)	1.60 ± 0.39	1.66 ± 0.35	1.61 ± 0.33	0.086	0.993	χ^2 = 2.31; *p* = 0.315		
	$HbSO_2$-index (%)	52.09 ± 5.18	51.37 ± 4.99	51.41 ± 4.73	0.351	0.235	χ^2 = 2.69; *p* = 0.260		
PHGG		(*n* = 35)	(*n* = 33)	(*n* = 34)					
	Brightness (L*)	61.41 ± 2.82	61.19 ± 2.67	61.227 ± 2.84	0.041*	0.313	χ^2 = 4.55; *p* = 0.103	*p* = 0.841	*p* = 0.151
	Redness (a*)	11.40 ± 1.41	11.22 ± 1.39	11.532 ± 1.49	0.561	0.662	χ^2 = 2.03; *p* = 0.362	*p* = 0.166	*p* = 0.480
	Yellowness (b*)	17.31 ± 1.81	16.91 ± 1.67	17.067 ± 1.80	0.250	0.203	χ^2 = 0.20; *p* = 0.905	*p* = 0.235	*p* = 0.214
	Melanin index	1.23 ± 0.17	1.19 ± 0.16	1.217 ± 0.17	0.104	0.427	χ^2 = 4.70; *p* = 0.91#	*p* = 0.665	*p* = 0.476
	Hb-index (%)	1.60 ± 0.33	1.69 ± 0.31	1.681 ± 0.35	0.006*	0.035*	χ^2 = 8.06; *p* = 0.018*	*p* = 0.588	*p* = 0.223
	$HbSO_2$-index (%)	52.86 ± 5.32	51.41 ± 5.34	51.785 ± 5.94	0.023*	0.007*	χ^2 = 7.70; *p* = 0.021*	*p* = 0.219	*p* = 0.449

*Significance $p \leq 0.05$; RM, repeated measure (non-parametric); †Between group: Mann–-Whitney *U* rank test, ‡Within group: Wilcoxon rank test; a*: Red-Green chromaticity coordinate; b*: Yellow-Blue chromaticity coordinate; Hb, Hemoglobin; $HbSO_2$, Hemoglobin oxygen saturation.

treatment × time interaction observed. Hemoglobin with saturated oxygen ($HbSO_2$-index) values showed a non-significant increasing trend in the placebo intake group. However, the PHGG dietary fiber intake group indicated a significant increase after 6 weeks (T6) Additionally, there was a significant difference in the treatment × time interaction between the PHGG dietary fiber and placebo groups over the course of the study. These findings corroborate the variations in melanin levels. Both the PHGG dietary fiber intake groups and the placebo showed a decrease in the MI. There was a significant decrease in the PHGG intake group at T6 and T12, while the placebo group showed no significant decrease in the MI. There was a significant difference in the treatment × time interaction between the PHGG dietary fiber and placebo groups during the course of the trial. The changes in MI values (from T0 to T6 as well as T0 to T12) between the PHGG and placebo groups were at a significant level only after 6 weeks (T6) of respective intakes (Table 4). In contrast to the measurements at the cheek (left) described above, the changes in the L* values measured under the left eye (a reference site) indicated a significant reduction at T6 and a significant increase at T12 from baseline in the placebo consumption group. Additionally, in the PHGG dietary fiber group, there was a significant decrease in L* values at T6 from baseline in contrast to a non-significant reduction in L* values at T12. Likewise, a significant treatment × time interaction was noticed for the placebo group when compared with the PHGG dietary fiber group for the study duration.

Furthermore, for the PHGG dietary fiber and placebo intake groups, non-significant changes in the a*, b*, and MI values evaluated under the left eye could be observed within the group as well as between groups (Table 4). The reverse trends could also be noticed in the change of Hb-index and $HbSO_2$-index values, showing an increase in Hb-index and a decrease in $HbSO_2$-index values in both the placebo and PHGG dietary fiber intake groups. The PHGG dietary fiber intake group indicated a significant difference in Hb-index and $HbSO_2$-index at T6 and T12, compared to placebo, where no significant difference was observed from baseline. In addition, only the PHGG intake group showed a significant difference in treatment × time interaction for Hb-index. Also, there was a significant difference in treatment × time interaction for the $HbSO_2$-index in the PHGG dietary fiber group for the study duration.

Additionally, Fig. 3A plots the differences in two color stimulus parameters, L* and a*, between the test site (Cheek) and the

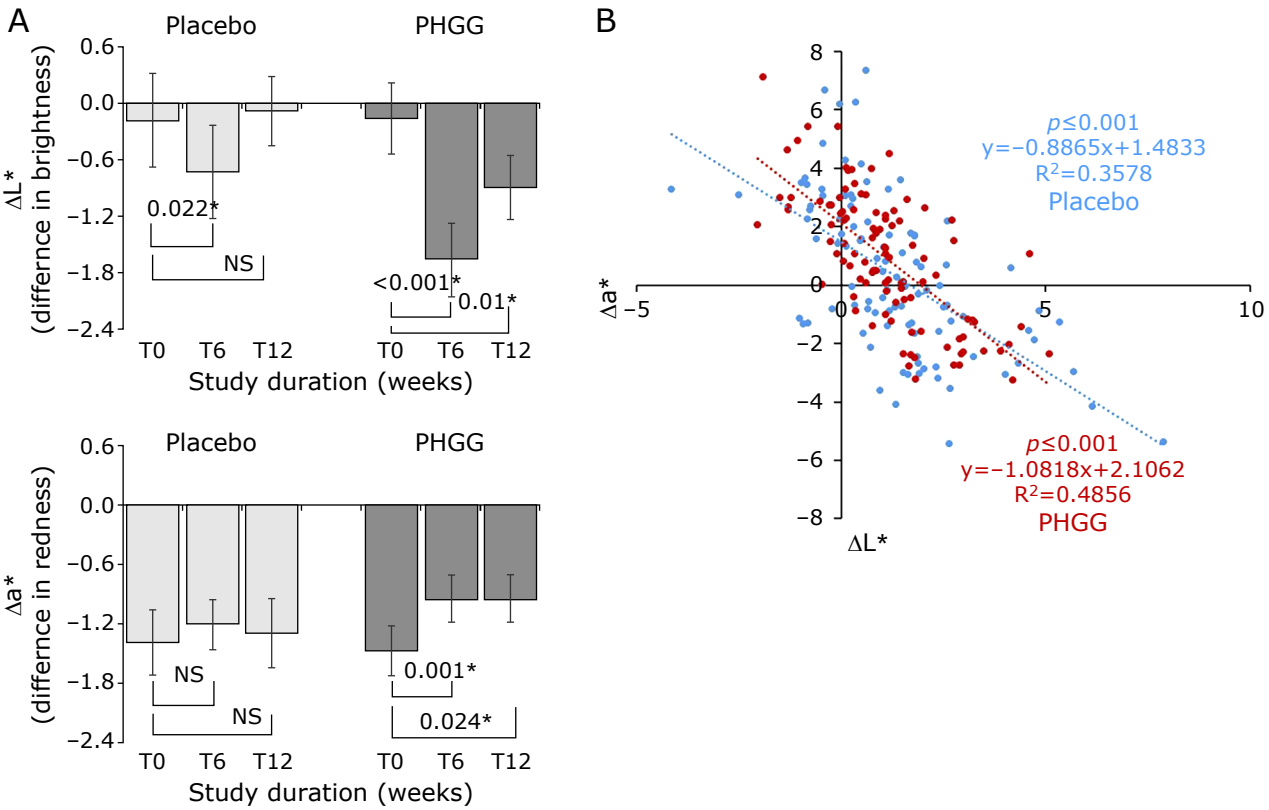

Fig. 3. (A) Plots showing the changes approximated as ΔL* and Δa* between the test site (Cheek) and the corresponding neighboring control site (Under Eye) in two color stimulus parameters, L* and a*. (B) In the correlation plot, both the placebo and PHGG dietary fiber groups indicated a significant association between ΔL* and Δa* coordinates.

corresponding neighboring control site (Under Eyes). These differences are approximated as ΔL* and Δa*. In the PHGG dietary intake group, the results revealed a significant decrease from baseline after 6 weeks (T6) and after 12 weeks (T12) in both ΔL* and Δa* values, respectively. Whereas a non-significant decrease in Δa* values could be noticed in the placebo group throughout the study duration, the decrease in ΔL* values indicated a significant difference after 6 weeks (T6) for the placebo intake group. In addition, a significant treatment × time interaction could be noted only for PHGG dietary fiber for decreasing ΔL* values, while there was no significant difference between PHGG dietary fiber and placebo intake groups for both ΔL* and Δa* values. Both the placebo and PHGG dietary fiber groups indicated a significant correlation between ΔL* and Δa* values (Fig. 3B).

Correlation analysis between functional skin parameters. Figure 4 illustrates the Spearman correlation between SCH and TEWL, featuring the skin SCH at cheeks of the subjects in the placebo and PHGG dietary fiber intake groups plotted against skin TEWL at cheeks at each time point. At baseline (T0), there was a positive, non-significant relationship between these two parameters in both the placebo and PHGG dietary fiber groups. In contrast, a negative correlation was observed between the skin TEWL and SCH at the cheek; thereby, the skin TEWL tends to decrease as the skin SCH increases. After 12 weeks, the correlation appeared significantly higher in the PHGG group in comparison to the placebo group during the course of the study.

The Spearman inter-correlation coefficients of the functional SE metrics with skin SCH and TEWL at the cheek are presented in Table 5 (the figures have been included in supplementary information; Supplemental Fig. 1*) over the study duration. In the placebo group, the SE correlation coefficients (i.e., R0, R2, R5, R6, and R7) were inversely correlated with skin SCH and did not reach statistical significance. While R2, R5, R6, and R7 remained non-significant and exhibited positive correlations with skin SCH in the PHGG dietary fiber group. Conversely, only R0 demonstrated a significant negative association with skin SCH in the PHGG group. On the contrary, all SE measures were positively correlated with skin TEWL in the PHGG dietary fiber group, although the R0, R2, and R7 SE parameters showed a negative correlation with skin TEWL in the placebo group. Except for the R6 parameter, there were no significant inter-correlations found between SE measures and skin TEWL in the placebo and PHGG dietary fiber groups (Supplemental Fig. 1B*).

In the placebo group, there was no significant correlation found between the brightness (L*), redness (a*), and yellowness (b*) components of the CIE-L*a*b* skin color system and skin SCH or skin TEWL measured at the cheek (Table 5). In the PHGG dietary fiber group the L* demonstrated a significant positive correlation with skin SCH, but no significant positive correlation with skin TEWL. Redness (a*) showed a significant positive correlation with TEWL and a significant negative correlation with skin SCH. On the other hand, the yellowness (b*) exhibited no significant positive correlation with skin SCH but a significant negative correlation with skin TEWL. Additionally, for both the PHGG dietary fiber group and the placebo group, there was a non-significant negative correlation found between skin MI and skin TEWL or the skin SCH measured at the cheek. The Hb-index was significantly positively associated with skin TEWL in both the placebo and PHGG groups, but negatively associated with skin SCH exhibited a considerable correlation only in the PHGG group. In contrast, $HbSO_2$ exhibited a negative correlation with skin TEWL and a positive correlation with skin SCH. There was a significant correlation between $HbSO_2$ and skin TEWL and skin SCH in the PHGG dietary fiber intake group compared to

*See online. https://doi.org/10.3164/jcbn.24-69

doi: 10.3164/jcbn.24-69

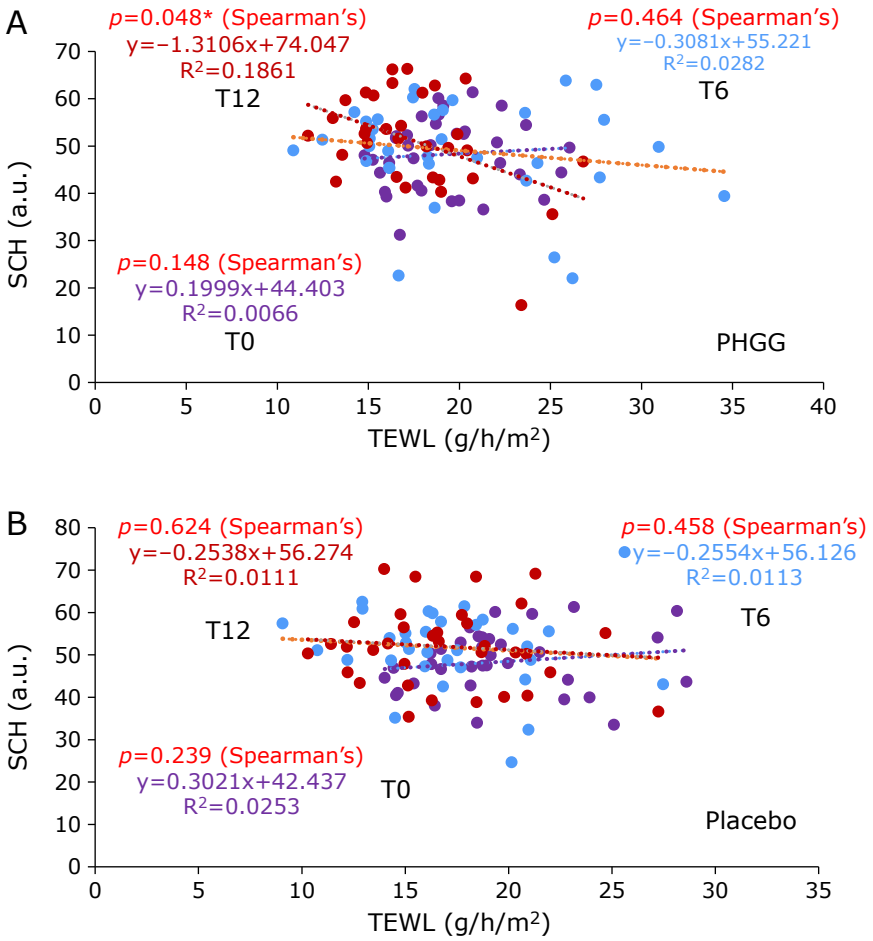

Fig. 4. The Spearman correlation between stratum corneum hydration (SCH) and trans-epidermal water loss (TEWL), featuring skin SCH at cheeks of the subjects in the (A) PHGG dietary fiber and (B) placebo intake groups plotted against skin TEWL at cheeks at each time point. See color figure in the on-line version.

Table 5. Spearman's correlation results indicate that the primary skin viscoelasticity indices, CIE-L*a*b* coordinates of the skin, and skin color change markers are associated with variation in skin moisture (i.e., stratum corneum hydration level) and trans-epidermal water loss measured at the left cheek in healthy study subjects during twelve consecutive weeks of placebo or PHGG dietary fiber administration during the winter season

Descriptions	Stratum corneum hydration (SCH)				Transepidermal water loss (TWEL)			
	Placebo		PHGG		Placebo		PHGG	
	Rho value	*p* value	Rho value	*p* value	Rho value	*p* value	Rho value	*p* value
Skin viscoelasticity parameters								
R0	−0.110	0.274	−0.277	0.022*	−0.109	0.279	0.153	0.125
R2	−0.047	0.640	0.021	0.832	−0.070	0.489	0.035	0.728
R5	−0.069	0.492	0.020	0.838	0.068	0.501	0.143	0.151
R6	−0.068	0.449	0.040	0.638	0.246	0.013*	0.035	0.729
R7	−0.003	0.980	0.009	0.926	−0.024	0.810	0.107	0.283
Skin color parameters								
L*	0.159	0.113	0.376	<0.001*	0.010	0.920	0.019	0.849
a*	−0.173	0.083	−0.283	0.004*	0.162	0.106	0.213	0.031*
b*	0.053	0.601	0.034	0.735	−0.109	0.278	−0.396	<0.001*
MI	−0.068	0.502	−0.172	0.083	−0.115	0.254	−0.173	0.083
Hb-index	−0.147	0.143	−0.310	0.002*	0.238	0.016*	0.357	<0.001*
$HbSO_2$-index	0.157	0.116	0.425	<0.001*	−0.048	0.631	−0.286	0.004*
Difference from reference site								
ΔL*	0.022	0.827	0.059	0.559	−0.022	0.827	0.078	0.433
Δa*	−0.067	0.506	−0.176	0.012*	0.120	0.232	−0.201	0.044*

*Significance $p \leq 0.05$; Spearman's corrlations.

the placebo group (Table 5, Supplemental Fig. 2*).

A strong and statistically significant linear inter-correlation between MI and CIE-L*a*b* components was observed from baseline to throughout the 12 weeks of study duration in both the placebo and PHGG dietary fiber groups. A negative correlation can be shown in Spearman's correlation coefficients between L* and MI in both placebo and PHGG groups. The results indicated that there was a mild positive association for the a* parameter and a strong positive correlation for the b* parameter with MI in both placebo and PHGG groups. However, non-significant weak inter-correlations were observed in the changes in the L*, a*, b*, and MI as a function of the Hb-Index (Supplemental Fig. 3*). Furthermore, the absence of variation in the Spearman's correlation coefficients between Δa* and ΔL* values in the dietary fiber group and placebo supports the contention that MI and Hb-Index showed no influence during the course of the study (Fig. 3A).

Visual evaluation and skin quality evaluation results. A professional dermatologist's visual assessment of the subjects' skin revealed that, while lifted cutaneous (skin) bumps, skin grooves, and an overall comprehensive evaluation of skin condition improved in both the PHGG dietary fiber and placebo groups. However, there was a noticeable and significant difference in all inspected skin conditions following the 12-week PHGG dietary fiber intake (Supplemental Table 3*). Similarly, the evaluation of skin quality across the entire face revealed improvements in conditions related to skin dryness, scales (a tiny piece of dead epidermis shed from the skin's surface), skin irritation, and pruritus (a severe itching sensation) in both the PHGG dietary fiber and the placebo groups, except erythema, which is typically determined by several skin conditioning factors (Supplemental Table 3*).

Self-assessment questionnaire results. The analysis of the sensory VAS questionnaire results of the most significant feedback from the PHGG dietary fiber group versus the placebo group at baseline (T0), after 6 weeks of study duration (T6), and at the end of the study (T12) for skin textural conditions and skin quality revealed an improvement in the majority of the sensory features (Supplemental Table 4*). The summary of the analysis revealed that both the PHGG dietary fiber and the placebo groups had similar baseline perceptions of the characteristics assessed by the self-assessment sensory VAS questionnaire. The percentage change in the parameters is displayed in Fig. 5. During the research periods, both the PHGG dietary fiber and the placebo groups showed significant improvements; however, the PHGG dietary fiber consumption was determined to be more beneficial compared to the placebo. Improvements in brightness, clarity, moisturizing feeling, glossiness, smoothness, firmness, and overall skin improvement conditions were noted, especially on the face skin. After ingesting PHGG dietary fiber, the majority of the study subjects provided positive feedback and appreciable responses. Furthermore, when comparing the PHGG dietary fiber group to the placebo, there was a substantial decrease in the appearance of skin stains, roughness, and dullness on the face. However, both the PHGG dietary fiber group and the placebo group showed similarly significant reductions in fine creases around the lips and eyes, as well as dark circles beneath the eyes. During the course of the study, the subjects in both the PHGG dietary fiber and placebo groups reported a considerable improvement in the dry and itchy conditions of the face and scalp; however, the placebo group received less appreciation. Furthermore, the physiological indicators studied (namely stress), were significantly improved in the PHGG dietary fiber group as compared to the placebo group. The complete findings of the self-assessment sensory VAS questionnaire completed by subjects in both the placebo and PHGG dietary fiber groups are presented in online supplementary Supplemental Table 4*.

Furthermore, the results of the analysis of fatigue VAS questionnaire feedback from the placebo and PHGG dietary fiber groups revealed that both groups perceived a reduction in fatigue. However, respondents in the PHGG dietary fiber intake group expressed a higher perception of fatigue reduction compared to the placebo. After 6 weeks (T6), the subjects in the PHGG dietary fiber group had a significant reduction in fatigue compared to baseline (T0), but the placebo group experienced no significant reduction ($p = 0.21$). Furthermore, there was no significant difference in fatigue between the two groups at 12 weeks (T12) of their respective intakes from baseline (Fig. 6A). While there was a significant difference in the global PSQI-J score rating between the PHGG dietary fiber intake and the placebo groups at baseline (T0), a decrease in the overall sleep rating score during the 12-week (T12) of PHGG dietary fiber consumption revealed an improvement (i.e., a lower global PSQI-J score and a better sleep regimen) in the sleep-related parameters among the subjects of the PHGG dietary fiber group (Fig. 6B). Furthermore, fatigue-related cues from restricted, deprived, or disrupted sleep may have a negative impact on facial appearance and social appeal.

The association between the PSQI-J score and fatigue score in the placebo and PHGG dietary fiber intake groups demonstrated that enhanced sleep quality significantly reduced fatigue among subjects in the PHGG intake group when compared to the placebo. The observation provided additional support for the relationship between rated fatigue and facial cues as measured by the VAS questionnaire. The correlations demonstrated how lower fatigue ratings were associated with improved ratings of skin brightness, clarity, smoothness, firmness, and overall skin condition among subjects in the PHGG dietary fiber intake group compared to placebo. Furthermore, a reduced fatigue rating may be associated with a decrease in skin dullness, roughness, face itching, and overall skin-related stress (Supplemental Table 4*).

Age-regulated functional skin parameters and correlation analysis. Based on baseline background data from the study subjects, as shown in Table 2, there was no statistically significant difference between the dietary fiber groups (PHGG and placebo) and between the stratified age groups (≤45 years and ≥46 years) among the total subjects. In addition, compared to individuals 46 years of age, those under 45 years of age had significantly lower BMI, systolic and diastolic blood pressures, and pulse rates. Aging causes the skin to undergo a variety of structural and textural alterations. The baseline background data of the study subjects presented in Table 1 revealed that there was no statistically significant difference between the placebo and PHGG dietary fiber groups in terms of total subjects or the stratified age groups of ≤45 years and the subjects aged ≥46 years. Subjects aged ≤45 years showed reduced BMI, systolic and diastolic blood pressures, and pulse rate compared to those aged ≥46 years. Aging causes the skin to undergo a variety of structural and textural alterations. As a result of intrinsic and/or extrinsic aging, SCH decreases while TEWL generally increases.[20] As tissues become thinner in aged skin, the dryness develops with age, accompanied by subjective symptoms including stiffness, itching, roughness, and burning feeling. Table 6 indicates a significant increase in SCH and SE after 6 weeks (T6) and 12 weeks (T12) from baseline, as well as a significant increase in SVE after 12 weeks in the PHGG dietary fiber intake group compared to placebo among subjects aged ≤45 years. Subjects aged ≤45 years who ingested PHGG dietary fiber experienced a non-significant decrease in TEWL. Whereas a significant decrease could be observed in TEWL among the subjects aged ≥46 years from baseline, as well as a significant increase in SCH, SE, and SVE after 6 weeks (T6) and 12 weeks (T12) of PHGG dietary fiber ingestion compared to placebo (Table 6).

In addition, there was no significant difference in basal L*, a*, and b* at the cheek (left) of the subjects aged ≥46 years compared to those aged ≤45 years after receiving placebo or PHGG

*See online. https://doi.org/10.3164/jcbn.24-69

doi: 10.3164/jcbn.24-69

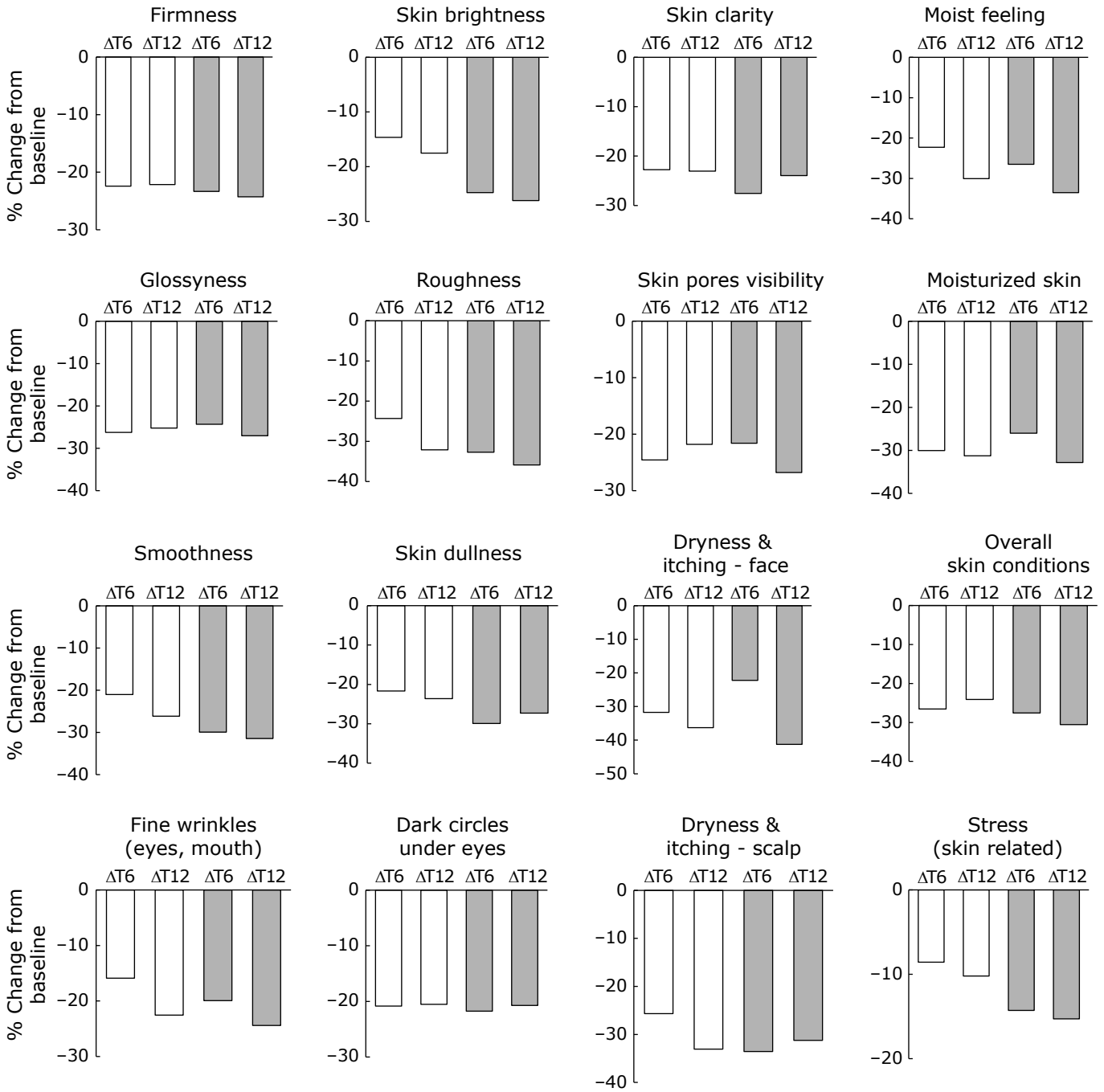

Fig. 5. The percentage change in skin texture and skin quality parameters of the self-assessed sensory VAS questionnaire feedback results from the PHGG dietary fiber group compared to the placebo group after 6 weeks (T6), and at study completion (T12) from baseline (T0).

dietary fiber from baseline (data not shown). Thus, the skin of both age groups appears to be an approximation of the same color.

Adverse events. There were no adverse events reported throughout the study. Subjects were also urged to report any adverse events involving gastrointestinal disorders, skin and subcutaneous tissues, respiratory troubles, parasite infections, and so on. There were no reports of harmful adverse effects in either group. The modest complaints mentioned were not relevant to the clinical study. The details are listed in Supplemental Table 5*.

Discussion

Various environmental conditions, particularly humidity and temperature, can affect the skin. Dermatology studies frequently probe into the influence of environmental factors on skin physiology. Skin barrier recovery in a dry environment is hindered by high relative humidity, decreased SE, and increased skin roughness.[61–63] Skin moisture, TEWL, and SVE are all interconnected factors that play an essential role in preserving skin health. Skin biomechanics, collagen generation and maintenance, and the effects of temperature and humidity fluctuations can all shed light on how SE relates to seasonal variations. The decrease in

*See online. https://doi.org/10.3164/jcbn.24-69

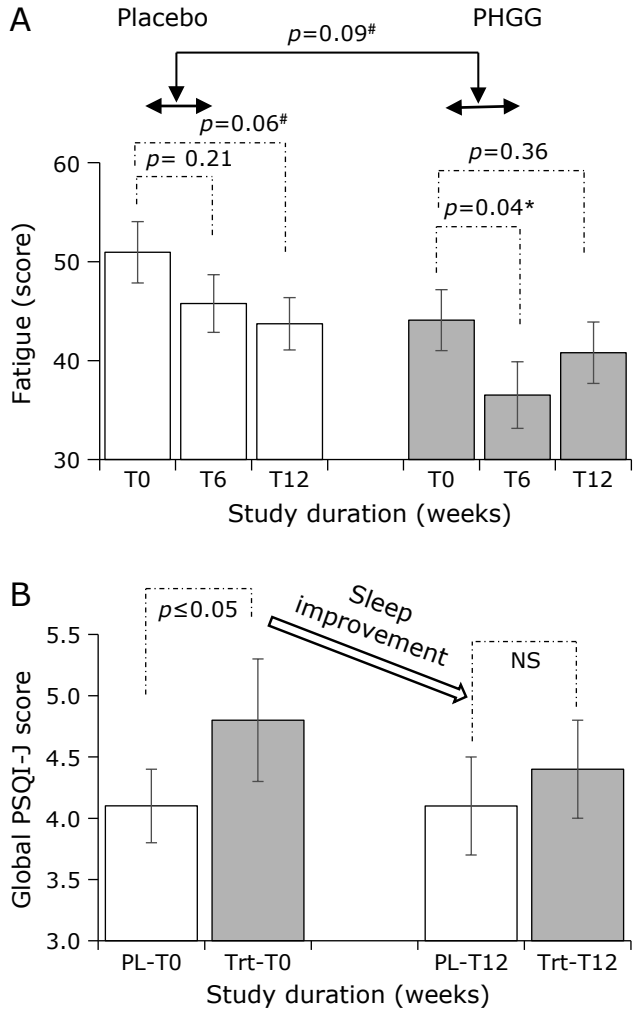

Fig. 6. (A) The results of the fatigue VAS questionnaire feedback from the placebo and PHGG dietary fiber group revealed significant reduction in fatigue after 6 weeks (T6) compared to baseline (T0) in the PHGG dietary fiber group. (B) A decrease in global PSQI-J sleep score during the 12-week (T12) of PHGG dietary fiber consumption revealed an improvement (i.e., a lower global PSQI-J score and a better sleep regimen) in the sleep-related parameters among the subjects of the PHGG dietary fiber group.

humidity and exposure to cold temperatures throughout the winter season can cause skin moisture levels to drop and induce changes in skin color and texture, resulting in itchy, rough, dry, cracked, sore, irritated, and red skin.[18] Therefore, adequate skin hydration is essential for preserving the stratum corneum integrity, which leads to improved SE and suppleness. Dietary fibers may play a favorable impact on skin health, including hydration and viscoelasticity, particularly in winter environments.

In this 12-week, randomized, double-blind, crossover, placebo-controlled study, supplementing once daily with 5 g of a PHGG prebiotic fiber (Sunfiber®) containing nearly 4 g soluble dietary fiber showed an increasing trend in skin viscoelastic properties and SCH, and a significant decrease in TEWL from baseline when compared to placebo. It has also been reported that decreases in TEWL from the stratum corneum of the epidermis are associated with improved skin barrier function.[64–66] Typically, intrinsic and extrinsic factors lead to the deterioration of skin barrier function.[41] The restoration of the skin barrier with PHGG dietary fiber ingestion may result in a reduction in TEWL and an improvement in overall skin conditions. Water can be effectively absorbed by soluble PHGG dietary fiber.[46,53] As a result, proper hydration from PHGG dietary fibers may help regulate the water balance (i.e., water retention) in the body, prevent excessive water loss through the skin in dry winter conditions, and thus help maintain hydration levels in the body, and indirectly support skin hydration, where skin ceramides play a dominant role in moisture retention and skin barrier function.[67] Furthermore, PHGG dietary fiber promotes appropriate digestion and absorption of nutrients, and functions as prebiotics to nourish beneficial gut bacteria for improved gut health.[18,41] A healthy gut microbiome has also been related to enhanced nutrient absorption, particularly those required for collagen formation and/or maintenance, as well as the efficient utilization of key vitamins and minerals for skin health.

The present study was performed in Tokyo, Japan between January and April 2023. The observed outside average seasonal temperature and humidity presented in Supplemental Table 1A* were relatively in the same range at baseline (T0; Visit 1) and after 6 weeks (T6; Visit 2) during the respective intakes of either placebo or PHGG dietary fiber.[68] Therefore, when comparing placebo and PHGG dietary fiber intakes during this study period, the seasonal environment had little impact on the study outcomes. However, the seasonal temperature and humidity were significantly different after the 12-week (T12; Visit 3) duration of the study; thus, the attributable effect of seasonal changes on the influenced outcomes of the study was included in the discussion when the placebo and PHGG intake groups were compared. The study results revealed that some efficacy for TEWL in the placebo group after 12 weeks of the study is attributable to seasonal changes in temperature and relative humidity. After 6 weeks (T6) of PHGG dietary fiber intake, the skin was considerably more moisturized (cheek: +7.1%; upper arm: +7.6%; upper back: +4.6%), and TEWL was significantly reduced (cheek: −11.8%; upper arm: −13.2%; upper back: −17.2%) compared to baseline. These findings indicated that the improved moisture content of the stratum corneum and decreased TEWL levels observed in the PHGG dietary fiber intake group, possibly due to the normal keratinization process of the epidermis were significantly improved from the baseline compared to the placebo group.[69] It has also been reported that the rate of TEWL doubles when the skin temperature rises by 7–8°C while sebum secretion increases by 10% with every 1°C rise in local temperature.[70,71] Further, the correlation ship indicated by a regression between skin SCH and TEWL confirmed TEWL threshold-like dependence on SCH, which may explain the presence of a TEWL–SCH correlation at the cheek and upper-back sites due to relatively higher threshold values. However, possibly the presence of a somewhat lower threshold may not explain the variation in TEWL reliance on SCH at the upper arm site (Table 2).

Environmental factors have an impact on SE and skin color changes over the winter. Improved SE is often associated with increased skin moisture. Skin elasticity (R5) and SVE (R7) variations are commonly associated with skin health from a clinical standpoint. In this study, SE was assessed using U and R parameters. A significant increase in R-parameters (R2, R5, R6, and R7) after 12 week consumption of PHGG dietary fiber compared to placebo (although no between-group significance was found for TEWL due to the seasonal effect) may be related to an increase in collagen content and elastic fibers such as brillin-1, fibrillin-2, and fibrillin-5 in the skin, leading to improved fiber elasticity and restoration of the skin's elastic properties, which is congruent with the study by Rue *et al.*[72] as described elsewhere. An increase in R2 and R6 values, in particular, implies a higher level of SE and moisture in the deeper layers of the skin. Furthermore, winter temperatures can diminish SE due to decreased blood flow on the skin's surface and vasoconstriction to conserve heat in response to the cold. This can cause to decrease in oxygen

*See online. https://doi.org/10.3164/jcbn.24-69

doi: 10.3164/jcbn.24-69

Table 6. The variation in skin moisture (i.e., stratum corneum hydration level), trans-epidermal water loss, and main skin viscoelasticity indices measured at the left cheek in age-stratified group of the healthy study subjects during 12 consecutive weeks of placebo or PHGG dietary fiber administration during the winter season

Parameters	≤45 years, *n* = 30 (M5; F25)							
	Placebo, *n* = 16 (M2; F14)				PHGG, *n* = 14 (M3; F11)			
	T0	T6	T12	RM × Time	T0	T6	T12	RM × Time
TWEL	19.8 ± 3.14	21.8 ± 7.44	18.6 ± 4.06	χ^2 = 5.57; *p* = 0.06	20.0 ± 4.22	18.9 ± 4.25	18.4 ± 4.13	χ^2 = 2.23; *p* = 0.20
SCH	46.8 ± 8.80	48.6 ± 11.6	47.7 ± 12.8	χ^2 = 0.43; *p* = 0.81	44.3 ± 7.37	51.8 ± 11.6*	49.7 ± 9.82*	χ^2 = 7.54; *p* = 0.023*
R5 (SE)	0.66 ± 0.11	0.69 ± 0.10*	0.67 ± 0.10	χ^2 = 3.57; *p* = 0.17	0.66 ± 0.12	0.69 ± 0.11*	0.72 ± 0.12*	χ^2 = 8.27; *p* = 0.016*
R7 (SVE)	0.48 ± 0.10	0.50 ± 0.09	0.48 ± 0.08	χ^2 = 5.81; *p* = 0.06	0.49 ± 0.10	0.51 ± 0.11	0.52 ± 0.11*	χ^2 = 5.54; *p* = 0.06
	≥46 years, *n* = 40 (M4; F36)							
	Placebo, *n* = 19 (M2; F17)				PHGG, *n* = 21 (M2; F19)			
	T0	T6	T12	RM × Time	T0	T6	T12	RM × Time
TWEL	18.8 ± 2.91	18.5 ± 3.21	16.7 ± 2.67	χ^2 = 3.07; *p* = 0.22	19.1 ± 3.54	16.0 ± 3.31*†	16.1 ± 3.52*	χ^2 = 12.7; *p* = 0.002*
SCH	49.5 ± 6.01	49.4 ± 9.64	51.7 ± 7.55*	χ^2 = 0.40; *p* = 0.82	50.0 ± 5.87	51.7 ± 7.96*	53.4 ± 8.99*	χ^2 = 8.33; *p* = 0.016*
R5 (SE)	0.55 ± 0.06	0.56 ± 0.09	0.56 ± 0.08	χ^2 = 0.54; *p* = 0.77	0.54 ± 0.08	0.58 ± 0.09*	0.58 ± 0.08*	χ^2 = 7.69; *p* = 0.021*
R7 (SVE)	0.41 ± 0.04	0.41 ± 0.07	0.41 ± 0.06	χ^2 = 0.37; *p* = 0.83	0.39 ± 0.06	0.42 ± 0.07*	0.42 ± 0.06*	χ^2 = 5.09; *p* = 0.08

RM, repeated measure (within group); *Significant *p*≤0.05 (from baseline); †Between group significance *p*≤0.05.

and nutrient delivery to the skin, thereby potentially impacting the formation and turnover of collagen, and resulting in a paler or darker complexion. Such pallor might be particularly noticeable in areas with thinner skin, such as the face (i.e., cheeks). Furthermore, the correlation coefficients of parameters R2, R5, R6, and R7 with TEWL and SCH values improved with PHGG dietary fiber intake when compared to placebo, illustrating that the cheek SE ameliorated. However, due to the cross-sectional nature of the results, no casual inference could be reached about any of the identified associations. However, any alteration of elasticity parameters has a detrimental effect on skin biomechanical features.

Different skin types and ages might react differently to winter weather. For instance, mature skin may experience a pronounced decline in elasticity and color changes. Consequently, as people age, the skin's ability to retain moisture and the regulated TEWL might decline due to an impairment in natural moisturizing factors.[73–75] Previous studies have also explored and compared the TEWL reliance on SCH by age range.[76] The present investigation also demonstrated a difference between the subjects stratified by age into ≤45 years of age and ≥46 years of age groups following PHGG dietary fiber intake compared to the placebo. Such stratified age groups were chosen because of the significant changes reported in perimenopause women, particularly dry and rough skin, sagging skin, and complexation changes caused by a drop in estrogen levels, which leads to reduced formation and repair of collagen and elastin in the dermal connective tissues.[77–79] This explains why the water received by the stratum corneum from the underlying tissue via diffusion and TEWL to the environment through evaporation, as well as the extent of SCH with the main barrier located near the stratum corneum base, were equilibrated differently by the study's stratified age groups. The diffusion coefficient for stratum corneum water may rise with stratum corneum water content in the PHGG dietary fiber compared to placebo response in a distinct way in stratified age groups. It was evident that subjects of ≥46 years of age group had substantially higher assessed SCH levels. The findings suggest that situations characterized by increased SCH and PHGG dietary fiber consumption might not necessarily account for higher TEWL levels as compared to placebo. Wherein, skin dryness during the winter season (i.e., a decrease in skin hydration) caused by a drop in collagen and lipid content may have increased extensibility and hence decreased SE.

A significant change in the MI at the cheek in the current study most likely reflects a change in the oxygenation level of red blood cells rather than a change in melanin content. The expected decrease in MI following an increase in $HbSO_2$-index in the PHGG dietary fiber intake group compared to placebo during winter conditions demonstrated a substantial drop in hemoglobin index with PHGG dietary fiber intake. Overall, the results indicated that PHGG dietary fiber intake significantly increased skin brightness (L*) while decreasing the redness (a*) of the CIE L*a*b* system, which is mostly determined by melanin content in the epidermis (Table 4). In addition to a significant positive association between brightness (L*) and SCH, there was a significant negative correlation between redness (a*) and MI with SCH in PHGG dietary fiber intake group compared to placebo. Further, a significant negative relationship for Hb-index and a significant positive correlation for $HbSO_2$-index with SCH were noticed. In contrast, the PHGG dietary fiber intake group showed a significant positive correlation for redness (a*), as well as a significant positive relationship for Hb-Index and a significant negative correlation for $HbSO_2$-index with TEWL when compared to the placebo group.

The impact of the Hb-index on redness (a*) appears to be especially important in assessing changes in facial color due to variations in cutaneous circulation during the cold environment.[80,81] Because hemoglobin and melanin are the principal light absorbers in the skin, redness results from a locally elevated concentration of hemoglobin. As a result, the Hb-index is a dermal microvasculature indication that contributes to general skin color changes in the winter, with red being the predominant hue for oxygenated hemoglobin. Blue skin coloring, which is usually caused by a shortage of oxygen absorption in the blood (i.e., oxygen saturation), could be restored to normal levels by consuming PHGG dietary fiber during the winter season. Dietary fiber PHGG consumption was found to lower MI values when compared to placebo, regardless of the CIE L* a* b* coordinates for defining and/or quantifying skin color. Whereas the L* a*, and b* parameters are not always constant and can vary depending on the skin condition. The significant difference in ΔL* and Δa* values measured simultaneously at the cheek and adjacent under-eye area indicates appearance improvement, confirming the efficacy of PHGG dietary fiber consumption compared to placebo for normal skin functions during the winter season (Fig. 3). It is noteworthy to mention that color is easy to perceive but difficult to assess objectively. These findings demonstrate that, depending on the degree and severity, skin

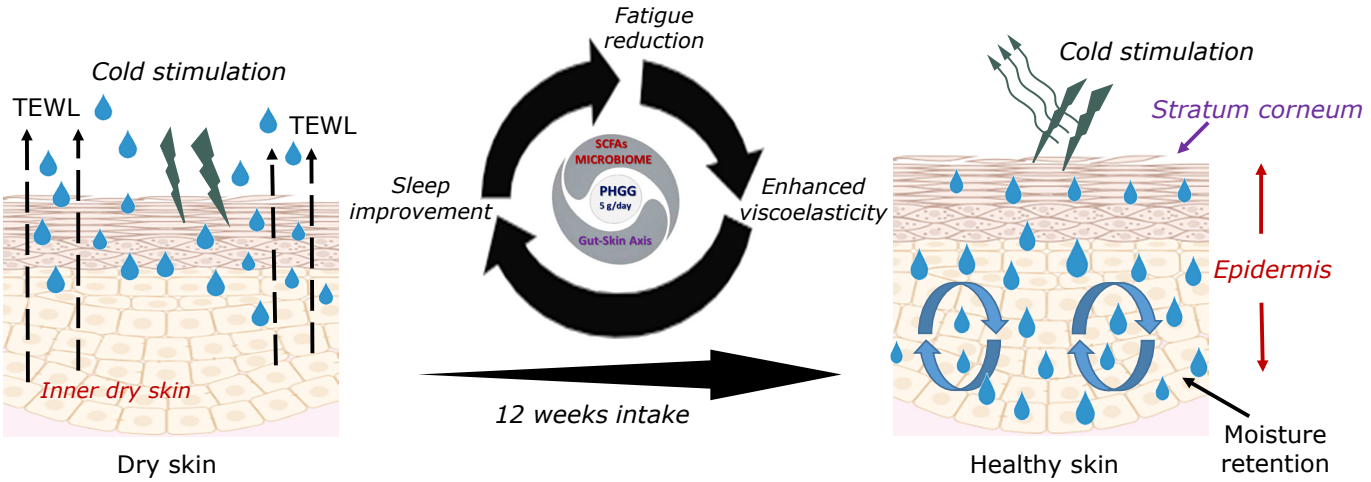

Fig. 7. An overview of schematic representation for improved stratum corneum hydration and reduction of trans-epidermal water loss (TEWL) with PHGG dietary fiber consumption during winter season. It is hypothesized that short-chain fatty acids (SCFAs) formed by bacteria in the gut during the fermentation of PHGG help in relieving skin inflammation caused on by seasonal environmental changes by modifying mitochondrial metabolism and function. PHGG dietary fiber consumption is suggested to prevent or restore dry skin conditions.

color during winter conditions is influenced by the dynamics of cutaneous circulation caused by capillary blood vessel shrinkage when the skin is exposed to a cold environment.

The human body interprets variation in skin temperature as a signal to conserve energy and heat to prevent dehydration of interior tissues by efficiently preserving skin moisture content. Dietary fibers regulate skin moisture in winter through complicated and numerous mechanisms. The transient decrease in TEWL rates could be either due to the apparent restoration of barrier function in a non-energy-dependent manner or due to the restricted exposure of deeper, hydrated tissues, which subsequently re-equilibrates the water gradient in proportion to the water diffusivity in the stratum corneum, resulting in seemingly normal TEWL rates.[20] Furthermore, adequate water accessibility at the skin surface could have a substantial impact on the skin microbiota. Short-chain fatty acids (SCFAs) are gut microbe metabolites produced by the dietary fiber fermentation in the colon that can reduce inflammatory skin conditions by acting on immune cells in the dermis and epidermis via keratinocyte-derived immune mediators.[18,42,82] Recent investigations have demonstrated that SCFAs can enhance the skin barrier and reduce skin inflammation by modifying mitochondrial metabolism and function.[83,84] The bacterially generated SCFAs metabolites produced during PHGG dietary fiber fermentation contribute to skin inflammation relief caused by environmental changes throughout the winter season by preventing or restoring disrupted and dry skin conditions.

The most notable effects were increased SCH, reduced TEWL, and improved SE measures. An overview of the schematic representation for improved SCH and reduction of TEWL with PHGG dietary fiber consumption during the winter season is shown in Fig. 7. It is hypothesized that SCFAs formed by bacteria in the gut during the fermentation of PHGG help in relieving skin inflammation caused by seasonal environmental changes. Thus, PHGG dietary fiber consumption is suggested to prevent or restore dry skin conditions. Further, to determine whether these findings were meaningful to the study subjects, a specific questionnaire was designed to correlate objective and subjective outcomes by covering several aspects related to the improvement of overall skin conditions with PHGG dietary fiber consumption during the cold environment in the winter season.

The results of the self-assessment questionnaires revealed an overall substantial perceived improvement in the PHGG dietary fiber group at the end of the treatment. One of the study's findings is that sleep deprivation affects several readily observable facial characteristic cues that may be connected to fatigue, particularly fatigue evaluations being strongly related to cues involving the face. Perceived fatigue, facial dullness, roughness, and other factors may also influence the skin's appearance. It is worth noting that the role of sleep in the appearance and function of the skin has received remarkably little attention, even though skin blood flow increases during sleep.[85–89] Sleep deprivation causes changes in skin color, wrinkles, and dark circles beneath the eyes, but the exact mechanism is unknown. A healthy attractive face has a certain degree of redness, which indicates increased vasodilation and vascularization.[90] In general, lower humidity, which affects the pH balance in the skin causes poor skin hydration, resulting in itchy, flaky, and dry skin. Such conditions also have an impact on skin microcirculation, ultimately limiting blood flow during the winter season and causing facial redness or skin rashes due to capillary fragility. Adequate dietary fiber consumption preserves moisture in skin stratum corneum layers, presumably increasing the thickness of the stratum corneum layer as a defensive mechanism, and inhibits water loss from the skin during dry conditions. As a result, the hydrated cells in the basal layer of the epidermis are exposed to relatively less harsh winter environmental conditions and hence create less pigment, resulting in lighter-colored skin during the winter season.

The study's potential strength is its quantitative, state-of-the-art methodology, cutting-edge approach, as well as the accurate, trustworthy, and repeatable technology applied to assess study outcomes. The equipment and procedures employed in this study were selected for their robustness. The study also considers the relatively broad age range of the subjects. However, a major limitation of the study could be the inclusion of predominately female subjects (87%). In addition, we did not incorporate clinical measurement of SCFAs metabolites, hyaluronic acid, or skin microbiomes in this investigation. According to one study, hyaluronic acid can help reduce TEWL, retain and redistribute water within the epidermis, maintain skin integrity, and improve skin barrier structure and function.[91] Furthermore, the placebo effects observed here were related to the subject's unconscious lifestyle changes throughout the study period, demonstrating the importance of the placebo control in accurately assessing the effect of PHGG dietary fiber.

doi: 10.3164/jcbn.24-69

Conclusions

The present study results confirmed that the improvement in skin conditions during the winter season could be addressed using soluble PHGG prebiotic dietary fiber, which can restore skin hydration, reduce TEWL, and improve skin texture and elasticity parameters. These findings, we believe attest to the efficacy of PHGG dietary fiber as cosmeceutical. The study ran from January to April 2023, and the expected seasonal weather changes resulted in a variation in temperature and relative humidity. Nevertheless, the findings of this study confirmed the anticipated outcomes, particularly the notable facial cues identified by the VAS questionnaire might be strongly associated with fatigue and sleep deprivation. As a result, a fiber-rich diet has been correlated with enhanced skin texture, smoothness, and skin moisture. This can help to offset the dullness and roughness that are often experienced in the winter season. Furthermore, the relationship between skin moisture, TEWL, and the elasticity characteristics of the skin has been proven to be mediated by a variety of physiological mechanisms. The primary cause for the significant improvement in skin viscoelastic parameters noticed in the present study is the management of skin moisture levels as well as the reduction in TEWL with PHGG dietary fiber ingestion. In conclusion, PHGG improves skin health by delivering vital nutrients that maintain skin cell structure, regulating skin moisture levels, and minimizing adverse events associated with skin inflammation. These mechanisms work together to improve skin SCH, viscoelasticity, and color during the winter season. Subject satisfaction with efficacy reflected these encouraging and enthusiastic findings, and the PHGG dietary fiber was well tolerated, with no adverse events witnessed during the study.

Therefore, PHGG dietary fiber provides consumers with an entirely safe and effective approach to counteracting skin-related conditions in cold weather during the winter season. Worldwide, the PHGG dietary fiber has been offered as a food for more than two decades with no severe adverse events or side effects identified. PHGG was officially approved as a Generally Recommended As Safe (GRAS) material by the Food and Drug Administration of the United States of America as well as other regulatory bodies across the world. In conclusion, a larger clinical trial with an adequate ratio of both genders is warranted to help confirm and support the findings of the current study. Further evidence and multicenter studies may provide much-needed responses to comprehend the use of dietary nutraceutical interventions for skin health in humans.

Author Contributions

Conceptualization, MPK, AA, and MO; methodology, AA, MPK, and SM; software, MPK; validation, AN, MO, and SM; formal analysis, AN, and MPK; investigation, AN, AA, MPK, and MO; resources, NS, and MO; data curation, MPK, AN, AA, and SM; writing – original draft preparation, MPK; writing – review and editing, MPK, AA, SM, MO, AN, and NS; visualization, AA, MO, and MPK; supervision, AA, AN, MPK, and NS; project administration, AN, AA, and NS; funding acquisition, NS. All authors have read and agreed to the published version of the manuscript after receiving acceptance from the research division.

Acknowledgments

TES Holding Co., Ltd., Tokyo, Japan, and Ueno-Asagao Clinic, Tokyo, Japan were responsible for the study design, and data collection as per protocol, and provided the final study report. The authors also acknowledge the helpful support and kind cooperation from the colleagues at Taiyo Kagaku Co., Ltd., Mie, Japan during this investigation. The authors appreciate the staff at TES Holdings Co., Ltd., and Ueno-Asagao Clinic, Tokyo, Japan, who performed the present study.

No external funding was received for this research. This study was fully supported by Taiyo Kagaku Co., Ltd., Japan. The study was performed by TES Holding Co., Ltd., Tokyo, Japan, at their research facility at Ueno-Asagao Clinic, Tokyo, Japan, and received research funding under the contract supported by Taiyo Kagaku Co., Ltd., Japan.

Institutional Review Board Statement

The study was conducted in accordance with the Declaration of Helsinki. The study protocol was initially reviewed and approved by the ethical committee expert group of Taiyo Kagaku Co., Ltd., Japan, and subsequently approved by the institutional review board of the Ueno Asagao Clinic Ethics Review Committee and TES Holding Co., Ltd., Japan [Approval No. HR-2023-TYC03 (2022-43); date: 14, December 2022]. Finally, the protocol was registered at the University Hospital Medical Information Network-Clinical Trial Registry (UMIN-CTR; Trial tracking identification: UMIN000049980; date: 6, January 2023).

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article. There were no particular conflicts of interest, referring that MPK, AA, SM, MO, and NS were employees of Taiyo Kagaku Co., Ltd., Japan.

References

1 Rodan K, Fields K, Majewski G, Falla T. Skincare bootcamp: the evolving role of skincare. *Plast Reconstr Surg Glob Open* 2016; **4**: e1152.

2 De Pessemier B, Grine L, Debaere M, Maes A, Paetzold B, Callewaert C. Gut–skin axis: current knowledge of the interrelationship between microbial dysbiosis and skin conditions. *Microorganisms* 2021; **9**: 353.

3 Oliveira R, Ferreira J, Azevedo LF, Almeida IF. An overview of methods to characterize skin type: focus on visual rating scales and self-report instruments. *Cosmetics* 2023; **10**: 14.

4 Carvalho MJ, Oliveira ALS, Santos Pedrosa S, Pintado M, Pinto-Ribeiro I, Madureira AR. Skin microbiota and the cosmetic industry. *Microb Ecol* 2023; **86**: 86–96.

5 Flores-Balderas X, Peña-Peña M, Rada KM, *et al*. Beneficial effects of plant-based diets on skin health and inflammatory skin diseases. *Nutrients* 2023; **15**: 2842.

6 Cao C, Xiao Z, Wu Y, Ge C. Diet and skin aging—from the perspective of food nutrition. *Nutrients* 2020; **12**: 870.

7 Ogorevc J, Geršak G, Novak D, Drnovšek J. Metrological evaluation of skin conductance measurements. *Measurement* 2013; **46**: 2993–3001.

8 Park EH, Jo DJ, Jeon HW, Na SJ. Effects of winter indoor environment on the skin: unveiling skin condition changes in Korea. *Skin Res Technol* 2023; **29**: e13397.

9 Jang IS, Choi J, Han J, Kim EJ, Lee HK. Differences between cheeks and dorsal hands skin properties during skiing in a cold and windy environment. *J Cosmet Dermatol Sci Appl* 2017; **7**: 27–33.

10 Song EJ, Lee JA, Park JJ, *et al*. A study on seasonal variation of skin parameters in Korean males. *Int J Cosmet Sci* 2015; **37**: 92–97.

11 Parke MA, Perez-Sanchez A, Zamil DH, Katta R. Diet and skin barrier: the role of dietary interventions on skin barrier function. *Dermatol Pract Concept* 2021; **11**: e2021132.

12 Parker ER. The influence of climate change on skin cancer incidence—a

review of the evidence. *Int J Womens Dermatol* 2021; **7**: 17–27.
13 Voegeli R, Rawlings AV, Seroul P, Summers B. A novel continuous colour mapping approach for visualization of facial skin hydration and transepidermal water loss for four ethnic groups. *Int J Cosmet Sci* 2015; **37**: 595–605.
14 Voegeli R, Rawlings AV, Summers B. Facial skin pigmentation is not related to stratum corneum cohesion, basal transepidermal water loss, barrier integrity and barrier repair. *Int J Cosmet Sci* 2015; **37**: 241–252.
15 Palma L, Marques LT, Bujan J, Rodrigues LM. Dietary water affects human skin hydration and biomechanics. *Clin Cosmet Investig Dermatol* 2015; **8**: 413–421.
16 Kikuchi K, Masuda Y, Yamashita T, Kawai E, Hirao T. Image analysis of skin color heterogeneity focusing on skin chromophores and the age-related changes in facial skin. *Skin Res Technol* 2015; **21**: 175–183.
17 Kikuchi K, Tagami H. Stratum corneum dynamic hydration test. *Agache's Measuring the Skin: Non-invasive Investigations, Physiology, Normal Constants (2nd ed.)*. Cham: Springer International Publishing, 2015; 341–354.
18 Kapoor MP, Yamaguchi H, Ishida H, Mizutani Y, Timm D, Abe A. The effects of prebiotic partially hydrolyzed guar gum on skin hydration: a randomized, open-label, parallel, controlled study in healthy humans. *J Funct Foods* 2023; **103**: 105494.
19 Pinson EA. Evaporation from human skin with sweat glands inactivated. *Am J Physiol-Legacy Content* 1942; **137**: 492–503.
20 Chilcott RP, Dalton CH, Emmanuel AJ, Allen CE, Bradley ST. Transepidermal water loss does not correlate with skin barrier function *in vitro*. *J Invest Dermatol* 2002; **118**: 871–875.
21 Mayrovitz HN. Transepidermal water loss and stratum corneum hydration in forearm versus hand palm. *Skin Res Technol* 2023; **29**: e13218.
22 Camilion JV, Khanna S, Anasseri S, Laney C, Mayrovitz HN. Physiological, pathological, and circadian factors impacting skin hydration. *Cureus* 2022; **14**: e27666.
23 Bolke L, Schlippe G, Gerß J, Voss W. A collagen supplement improves skin hydration, elasticity, roughness, and density: results of a randomized, placebo-controlled, blind study. *Nutrients* 2019; **11**: 2494.
24 Chakkalakal M, Nadora D, Gahoonia N, *et al*. Prospective randomized double-blind placebo-controlled study of oral pomegranate extract on skin wrinkles, biophysical features, and the gut-skin axis. *J Clin Med* 2022; **11**: 6724.
25 Shoji K, Kameda A, Furuichi K. Effects of milk amazake on skin elasticity, hydration, and transepidermal water loss: an 8-week double-blind, randomized, controlled trial. *J Oleo Sci* 2023; **72**: 329–335.
26 Hakozaki T, Takiwaki H, Miyamoto K, Sato Y, Arase S. Ultrasound enhanced skin-lightening effect of vitamin C and niacinamide. *Skin Res Technol* 2006; **12**: 105–113.
27 Genovese L, Corbo A, Sibilla S. An insight into the changes in skin texture and properties following dietary intervention with a nutricosmeceutical containing a blend of collagen bioactive peptides and antioxidants. *Skin Pharmacol Physiol* 2017; **30**: 146–158.
28 Henning SM, Guzman JB, Thames G, *et al*. Avocado consumption increased skin elasticity and firmness in women—a pilot study. *J Cosmet Dermatol* 2022; **21**: 4028–4034.
29 Nagata C, Nakamura K, Wada K, *et al*. Association of dietary fat, vegetables and antioxidant micronutrients with skin ageing in Japanese women. *Br J Nutr* 2010; **103**: 1493–1498.
30 Ideta R, Sakuta T, Nakano Y, Uchiyama T. Orally administered glucosylceramide improves the skin barrier function by upregulating genes associated with the tight junction and cornified envelope formation. *Biosci Biotechnol Biochem* 2011; **75**: 1516–1523.
31 Miyanishi K, Shiono N, Shirai H, Dombo M, Kimata H. Reduction of transepidermal water loss by oral intake of glucosylceramides in patients with atopic eczema. *Allergy* 2005; **60**: 1454–1455.
32 Dreher ML, Davenport AJ. Hass avocado composition and potential health effects. *Crit Rev Food Sci Nutr* 2013; **53**: 738–750.
33 Shin D, Lee Y, Huang YH, *et al*. Probiotic fermentation augments the skin anti-photoaging properties of Agastache rugosa through up-regulating antioxidant components in UV-B-irradiated HaCaT keratinocytes. *BMC Complement Altern Med* 2018; **18**: 196.
34 Draelos ZD. Nutrition and enhancing youthful-appearing skin. *Clin Dermatol* 2010; **28**: 400–408.
35 Bickers DR, Athar M. Oxidative stress in the pathogenesis of skin disease. *J Invest Dermatol* 2006; **126**: 2565–2575.
36 Richelle M, Sabatier M, Steiling H, Williamson G. Skin bioavailability of dietary vitamin E, carotenoids, polyphenols, vitamin C, zinc and selenium. *Br J Nutr* 2006; **96**: 227–238.
37 Thakkar SK, Moodycliffe AM, Richelle M. Carotenoids and skin. In: Pappas A, ed. *Nutrition and Skin: Lessons for Anti-Aging, Beauty and Healthy Skin*, New York: Springer, 2011; 59–78.
38 Coates M, Lee MJ, Norton D, MacLeod AS. The skin and intestinal microbiota and their specific innate immune systems. *Front Immunol* 2019; **10**: 2950.
39 Wallen-Russell C. Is there a relationship between transepidermal water loss and microbial biodiversity on the skin? *Cosmetics* 2019; **6**: 18.
40 Xiao X, Hu X, Yao J, *et al*. The role of short-chain fatty acids in inflammatory skin diseases. *Front Microbiol* 2023; **13**: 1083432.
41 Lee HJ, Kim M. Skin barrier function and the microbiome. *Int J Mol Sci* 2022; **23**: 13071.
42 Chen YE, Fischbach MA, Belkaid Y. Skin microbiota–host interactions. *Nature* 2018; **553**: 427–436.
43 Kapoor MP, Juneja LR. Partially hydrolyzed guar gum dietary fiber. In: Cho SS, Samuel P, eds. *Fiber Ingredients: Food Applications and Health Benefits*, Boca Raton: CRC Press, 2009; 70–120.
44 Ohashi Y, Sumitani K, Tokunaga M, Ishihara N, Okubo T, Fujisawa T. Consumption of partially hydrolysed guar gum stimulates Bifidobacteria and butyrate-producing bacteria in the human large intestine. *Benef Microbes* 2015; **6**: 451–455.
45 Takagi T, Naito Y, Higashimura Y, *et al*. Partially hydrolysed guar gum ameliorates murine intestinal inflammation in association with modulating luminal microbiota and SCFA. *Br J Nutr* 2016; **116**: 1199–1205.
46 Takahashi T, Tokunaga M, Okubo T, Ozeki M, Kapoor MP, Yasukawa Z. Lower rate of water absorption of an oral rehydration solution with partially hydrolyzed guar gum in conscious rats. *Nutrients* 2022; **14**: 4231.
47 Rao TP, Hayakawa M, Minami T, *et al*. Post-meal perceivable satiety and subsequent energy intake with intake of partially hydrolysed guar gum. *Br J Nutr* 2015; **113**: 1489–1498.
48 Kapoor MP, Sugita M, Fukuzawa Y, Okubo T. Impact of partially hydrolyzed guar gum (PHGG) on constipation prevention: a systematic review and meta-analysis. *J Funct Foods* 2017; **33**: 52–66.
49 Rosli D, Shahar S, Manaf ZA, *et al*. Randomized controlled trial on the effect of partially hydrolyzed guar gum supplementation on diarrhea frequency and gut microbiome count among pelvic radiation patients. *J Parenter Enteral Nutr* 2021; **45**: 277–286.
50 Niv E, Halak A, Tiommny E, *et al*. Randomized clinical study: partially hydrolyzed guar gum (PHGG) versus placebo in the treatment of patients with irritable bowel syndrome. *Nutr Metab (Lond)* 2016; **13**: 10.
51 di Lorenzo R, Bernardi A, Grumetto L, *et al*. Phenylalanine butyramide is a new cosmetic ingredient with soothing and anti-reddening potential. *Molecules* 2021; **26**: 6611.
52 Salem I, Ramser A, Isham N, Ghannoum MA. The gut microbiome as a major regulator of the gut-skin axis. *Front Microbiol* 2018; **9**: 1459.
53 Kapoor MP, Koido M, Kawaguchi M, *et al*. Lifestyle related changes with partially hydrolyzed guar gum dietary fiber in healthy athlete individuals—a randomized, double-blind, crossover, placebo-controlled gut microbiome clinical study. *J Funct Foods* 2020; **72**: 104067.
54 Okubo T, Ishihara N, Takahashi H, *et al*. Effects of partially hydrolyzed guar gum intake on human intestinal microflora and its metabolism. *Biosci Biotechnol Biochem* 1994; **58**: 1364–1369.
55 Abe A, Morishima S, Kapoor MP, *et al*. Partially hydrolyzed guar gum is associated with improvement in gut health, sleep, and motivation among healthy subjects. *J Clin Biochem Nutr* 2023; **72**: 189–197.
56 Goodyear MD, Krleza-Jeric K, Lemmens T. The declaration of Helsinki. *BMJ* 2007; **335**: 624–625.
57 Buysse DJ, Reynolds CF 3rd, Monk TH, Berman SR, Kupfer DJ. The Pittsburgh sleep quality index: a new instrument for psychiatric practice and research. *Psychiatry Res* 1989; **28**: 193–213.
58 Léger D, Gauriau C, Etzi C, *et al*. "You look sleepy…" The impact of sleep restriction on skin parameters and facial appearance of 24 women. *Sleep Med* 2022; **89**: 97–103.
59 Takiwaki H, Miyamoto H, Ahsan K. A simple method to estimate CIE-L*a*b* values of the skin from its videomicroscopic image. *Skin Res Technol* 1997; **3**: 42–44.

doi: 10.3164/jcbn.24-69

60 Kapoor MP, Sugita M, Kawaguchi M, *et al*. Influence of iron supplementation on fatigue, mood states and sweating profiles of healthy non-anemic athletes during a training exercise: a double-blind, randomized, placebo-controlled, parallel-group study. *Contemp Clin Trials Commun* 2023; **32**: 101084.
61 Sato J, Katagiri C, Nomura J, Denda M. Drastic decrease in environmental humidity decreases water-holding capacity and free amino acid content of the stratum corneum. *Arch Dermatol Res* 2001; **293**: 477–480.
62 Sato J, Denda M, Chang S, Elias PM, Feingold KR. Abrupt decreases in environmental humidity induce abnormalities in permeability barrier homeostasis. *J Invest Dermatol* 2002; **119**: 900–904.
63 Tsukahara K, Hotta M, Fujimura T, Haketa K, Kitahara T. Effect of room humidity on the formation of fine wrinkles in the facial skin of Japanese. *Skin Res Technol* 2007; **13**: 184–188.
64 Berardesca E, Maibach HI. Transepidermal water loss and skin surface hydration in the non invasive assessment of stratum corneum function. *Derm Beruf Umwelt* 1990; **38**: 50–53.
65 Grubauer G, Elias PM, Feingold KR. Transepidermal water loss: the signal for recovery of barrier structure and function. *J Lipid Res* 1989; **30**: 323–333.
66 Rawlings AV, Matts PJ. Stratum corneum moisturization at the molecular level: an update in relation to the dry skin cycle. *J Invest Dermatol* 2005; **124**: 1099–1110.
67 Huang HC, Chang TM. Ceramide 1 and ceramide 3 act synergistically on skin hydration and the transepidermal water loss of sodium lauryl sulfate-irritated skin. *Int J Dermatol* 2008; **47**: 812–819.
68 Japan Meteorological Agency/Ministry of Land, Infrastructure, Transport and Tourism, Japan. https://www.jma.go.jp/. Accessed 26 Jun 2024.
69 Miyazaki K, Masuoka N, Kano M, Iizuka R. Bifidobacterium fermented milk and galacto-oligosaccharides lead to improved skin health by decreasing phenols production by gut microbiota. *Benef Microbes* 2014; **5**: 121–128.
70 Grice K, Sattar H, Sharratt M, Baker H. Skin temperature and transepidermal water loss. *J Invest Dermatol* 1971; **57**: 108–110.
71 Cunliffe WJ, Burton JL, Shuster S. The effect of local temperature variations on the sebum excretion rate. *Br J Dermatol* 1970; **83**: 650–654.
72 Ryu HS, Joo YH, Kim SO, Park KC, Youn SW. Influence of age and regional differences on skin elasticity as measured by the Cutometer®. *Skin Res Technol* 2008; **14**: 354–358.
73 Hori M, Kishimoto S, Tezuka Y, *et al*. Double-blind study on effects of glucosyl ceramide in beet extract on skin elasticity and fibronectin production in human dermal fibroblasts. *Anti-Aging Med* 2010; **7**: 129–142.
74 Firooz A, Sadr B, Babakoohi S, *et al*. Variation of biophysical parameters of the skin with age, gender, and body region. *ScientificWorldJournal* 2012; **2012**: 386936.
75 Cua AB, Wilhelm KP, Maibach HI. Elastic properties of human skin: relation to age, sex, and anatomical region. *Arch Dermatol Res* 1990; **282**: 283–288.
76 Luebberding S, Krueger N, Kerscher M. Skin physiology in men and women: *in vivo* evaluation of 300 people including TEWL, SC hydration, sebum content and skin surface pH. *Int J Cosmet Sci* 2013; **35**: 477–483.
77 Loutfy I, Abdel Aziz F, Dabbous NI, Hassan MH. Women's perception and experience of menopause: a community-based study in Alexandria, Egypt. *East Mediterr Health J* 2006; **12 Suppl 2**: S93–S106.
78 Leitch C, Doherty V, Gebbie A. Women's perceptions of the effects of menopause and hormone replacement therapy on skin. *Menopause Int* 2011; **17**: 11–13.
79 Calleja-Agius J, Brincat M. The effect of menopause on the skin and other connective tissues. *Gynecol Endocrinol* 2012; **28**: 273–277.
80 Takiwaki H, Miyaoka Y, Kohno H, Arase S. Graphic analysis of the relationship between skin colour change and variations in the amounts of melanin and haemoglobin. *Skin Res Technol* 2002; **8**: 78–83.
81 Ohshima H, Oyobikawa M, Tada A, *et al*. Melanin and facial skin fluorescence as markers of yellowish discoloration with aging. *Skin Res Technol* 2009; **15**: 496–502.
82 Keshari S, Balasubramaniam A, Myagmardoloonjin B, Herr DR, Negari IP, Huang CM. Butyric acid from probiotic *Staphylococcus epidermidis* in the skin microbiome down-regulates the ultraviolet-induced pro-inflammatory IL-6 cytokine via short-chain fatty acid receptor. *Int J Mol Sci* 2019; **20**: 4477.
83 Trompette A, Gollwitzer ES, Yadava K, *et al*. Gut microbiota metabolism of dietary fiber influences allergic airway disease and hematopoiesis. *Nat Med* 2014; **20**: 159–166.
84 Trompette A, Pernot J, Perdijk O, *et al*. Gut-derived short-chain fatty acids modulate skin barrier integrity by promoting keratinocyte metabolism and differentiation. *Mucosal Immunol* 2022; **15**: 908–926.
85 Van Someren EJW. Sleep propensity is modulated by circadian and behavior-induced changes in cutaneous temperature. *J Therm Biol* 2004; **29**: 437–444.
86 Benca RM, Obermeyer WH, Thisted RA, Gillin JC. Sleep and psychiatric disorders: a meta-analysis. *Arch Gen Psychiatry* 1992; **49**: 651–668.
87 Sundelin T, Lekander M, Kecklund G, Van Someren EJ, Olsson A, Axelsson J. Cues of fatigue: effects of sleep deprivation on facial appearance. *Sleep* 2013; **36**: 1355–1360.
88 Kräuchi K, Wirz-Justice A. Circadian clues to sleep onset mechanisms. *Neuropsychopharmacology* 2001; **25 (5 Suppl)**: S92–S96.
89 Shahid A, Shen J, Shapiro CM. Measurements of sleepiness and fatigue. *J Psychosom Res* 2010; **69**: 81–89.
90 Stephen ID, Coetzee V, Law Smith M, Perrett DI. Skin blood perfusion and oxygenation colour affect perceived human health. *PLoS One* 2009; **4**: e5083.
91 Sundaram H, Mackiewicz N, Burton E, Peno-Mazzarino L, Lati E, Meunier S. Pilot comparative study of the topical action of a novel, crosslinked resilient hyaluronic acid on skin hydration and barrier function in a dynamic, three-dimensional human explant model. *J Drugs Dermatol* 2016; **15**: 434–441.