

## ORIGINAL ARTICLE

# The study of skin hydration, anti-wrinkles function improvement of anti-aging cream with alpha-ketoglutarate

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**Abstract**

**Introduction:** The tricarboxylic acid (TCA) cycle is a key metabolic pathway for driving the generation of mitochondrial energy in all oxidative organisms. Alpha-ketoglutarate (Alpha-KG), a precursor of glutamine, is known as a crucial intermediate of the TCA cycle and plays a pivotal role in multiple metabolic processes. As a precursor of glutamate and glutamine, AKG acts as an antioxidant agent as it directly reacts with hydrogen peroxide with formation of succinate, water, and carbon dioxide; meanwhile, it discharges plenty of ATP by oxidative decarboxylation. Several studies reported that Alpha-KG is a key participant in the detoxification of reactive oxygen species and acts as an integral part of the oxidative defense machinery. However, few studies have been reported on the efficacy of Alpha-KG in the maintenance of skin functions. This study demonstrated that Alpha-KG has beneficial effects on skin hydration and barrier function and that fermentation is an effective way to enhance the synthesis of Alpha-KG in yeast, which possesses mitochondria.

**Methods:** *Evaluation of promoting effects on epidermal keratinocyte proliferation:* Keratinocytes were incubated with a test sample, and the degree of proliferation was determined by MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] assay. *Evaluation of promoting effects on mRNA expression of genes related to skin hydration and barrier function:* Keratinocytes were incubated with a test sample, and gene expression levels of filaggrin (FLG), serine palmitoyltransferase (SPT), and involucrin (IVL) were analyzed by real-time RT-PCR. *Analysis of Alpha-KG in rice fermented liquid:* Alpha-KG in rice fermented liquid was quantitatively analyzed by capillary electrophoresis time-of-flight mass spectrometry (CE-TOF-MS). *Clinical study testing methods and VISIA testing:* After 28 days of treatment use the cream with Alpha-KG and control sample without Alpha-KG, instrumentation measurements were adopted to assess skin wrinkles, texture, elasticity, and firmness, tested by the VISIA-CR.

**Results:** Immediately after using the sample for D0, the skin wrinkles were significantly reduced by 23.64%. After using the sample for D7, the average clinical score of outer corner wrinkles was significantly reduced by 15.23%, and nasolabial groove wrinkles were significantly reduced by 25.68%. After using the sample for D56, the mean clinical evaluation score of crow's feet decreased significantly by 25.42%; the

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average score of clinical evaluation of skin firmness increased significantly by 41.40%; the skin gloss increased significantly by 28.67%.

**Conclusion:** It was demonstrated that Alpha-KG is expected to promote skin hydration and barrier function by the activation of cell proliferation and the up-regulation of mRNA expression of genes related to the maintenance of epidermal function in *in vitro* tests. In order to develop a cosmetic ingredient including Alpha-KG, we focused on fermentation with yeast as an efficient production method of Alpha-KG and found that fermentation of rice with yeast efficiently produced Alpha-KG. Taken together, it is considered that rice fermented liquid containing Alpha-KG could be a promising ingredient for skin care products. Through the clinical testing, rice fermented liquid containing Alpha-KG could be a promising cosmetic ingredient for skin care products.

#### KEYWORDS

anti-aging cream, filaggrin, involucrum, keratinocyte proliferation, rice fermented liquid, serine palmitoyl transferase,  $\alpha$ -ketoglutaric acid

## 1 | INTRODUCTION

The tricarboxylic acid (TCA) cycle is a key metabolic pathway for driving the generation of mitochondrial energy in all oxidative organisms. Alpha-ketoglutaric acid (Alpha-KG), one of the TCA cycle intermediates, is a weak acid containing carboxyl groups and a ketone group (Figure 1) and is involved in multiple metabolic processes.<sup>1</sup> A number of studies reported that Alpha-KG plays a pivotal role in the detoxification of reactive oxygen species (ROS) by improving antioxidative capacity and acts as an essential part of the oxidative defense machinery. In addition, Alpha-KG contributes to the oxidation of nutrients and the energy provision by producing plenty of ATP in the TCA cycle.<sup>2</sup> However, few studies have been reported on the efficacy of Alpha-KG in the maintenance of skin functions. Therefore, in this study, we aim to clarify whether Alpha-KG has beneficial effects on skin function and to find an effective way to enhance the production of Alpha-KG.

The TCA cycle is an essential mechanism that provides energy source via the production of ATP for cellular metabolism. We focused on among intermediate metabolites in the TCA cycle as Alpha-KG has been reported to possess many physiological functions. Alpha-KG could not only eliminate hydrogen peroxide by non-enzymatic oxidative decarboxylation in the TCA cycle but also serve as a natural scavenger of ROS by showing its antioxidative capacity.<sup>2</sup> In humans, Alpha-KG is widely used in various diseases induced by oxidative stress, such as trauma, aged diseases.<sup>3</sup> Furthermore, Alpha-KG could regulate organismal lifespan and prevent age-related

diseases by regulating cellular energy metabolism.<sup>4,5</sup> However, few reports are available on the application of Alpha-KG for the maintenance of human skin functions. Thus, we investigated the efficacy of Alpha-KG on skin function in keratinocytes. As a result, Alpha-KG promoted cell proliferation and mRNA expressions of FLG, SPT, and IVL in keratinocytes. FLG is degraded into free amino acids that contribute to generation of the natural moisturizing factors (NMFs) for maintaining epidermal hydration. Low FLG biosynthesis induces the pathogenesis of xerosis skin conditions.<sup>6</sup> SPT is the rate-limiting enzyme in the *de novo* synthesis of ceramides which are main components of intercellular lipids in the stratum corneum and play an essential role in skin barrier function.<sup>7,8</sup> IVL is a component of the epidermal cornified cell envelopes, which are significant components of the epidermal barrier.<sup>9</sup> Therefore, Alpha-KG is expected to promote skin hydration and barrier function by the activation of cell proliferation and the up-regulation of mRNA expression of these genes related to the maintenance of epidermal function. Next, we focused on fermentation with yeast as an efficient production method of Alpha-KG for developing a cosmetic ingredient containing Alpha-KG. Yeast cells have mitochondria in which the TCA cycle takes place and could boost the production of amino acids by fermentation.<sup>10,11</sup> As a starting raw material, rice was selected and fermented by yeast. Analysis showed that the rice fermented liquid contained Alpha-KG abundantly. Taken together, it was considered that fermentation is one of the best ways to produce Alpha-KG effectively and to develop beneficial ingredients in cosmetics.

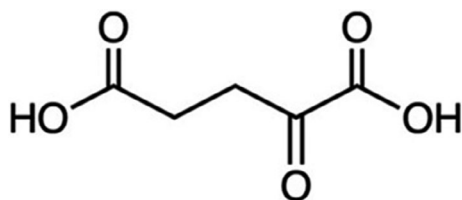


FIGURE 1 Chemical structure of Alpha-ketoglutarate

## 2 | METHODS

### 2.1 | Materials

Alpha-ketoglutaric acid (Alpha-KG; Alfa Aesar) and rice fermentation liquid fermented with *Saccharomyces veronae* were used for this study.

Normal human epidermal keratinocytes (NHEKs; KURABO) were cultured in keratinocyte basal medium (KBM) or keratinocyte growth medium (KGM) containing insulin, hydrocortisone, gentamicin/amphotericin B, and growth additives such as bovine pituitary extract and human epidermal growth factor (KURABO) at 37°C under an atmosphere of 5% CO<sub>2</sub> in air.

The skin care cream is provided by Mageline Biology Tech Co., Ltd; facial image analysis system VISIA-CR (Canfield company); facial image analysis system VISIA (Canfield company).

The sample cream (50 g cream: main ingredient: 30% rice fermented liquid(contact Alpha-KG); 1.2% sodium acrylate/sodium acryloyl dimethyl taurine copolymer, 1.5% trehalose, 1.0% phytosterol/octyl dodecanol lauroyl glutamate, and 66.3% water).

The control cream (50 g cream: main ingredient: 1.2% sodium acrylate/sodium acryloyl dimethyl taurine copolymer, 1.5% trehalose, 1.0% phytosterol/octyl dodecanol lauroyl glutamate, and 96.3% water).

## 2.2 | Evaluation of promoting effects on epidermal keratinocyte proliferation

NHEKs were seeded on a 96-well plate and cultured in KGM for overnight. The cells were then treated with or without Alpha-KG for 3 days. The promoting effects on cell proliferation were evaluated by MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] assay.

## 2.3 | Evaluation of promoting effects on mRNA expression of genes related to skin hydration and barrier function

NHEKs were seeded on a 6-well plate and cultured in KGM for overnight. The medium was changed to KBM, and cells were cultured for 24 h. The cells were then treated with or without Alpha-KG for 24 h. Total RNAs were isolated in accordance with the standard operation method. Real-time RT-PCR reactions were performed by using a Thermal Cycler Dice<sup>®</sup> Real Time System III and TaKaRa SYBR<sup>®</sup> PrimeScriptTM RT-PCR Kit (Perfect Real Time) (Takara Bio). The amount of mRNA expressions of filaggrin (FLG), serine palmitoyltransferase (SPT), and involucrin (IVL) was normalized by that of GAPDH used as an internal standard.

## 2.4 | Analysis of Alpha-KG in rice fermented liquid using CE-TOFMS

To determine and quantify Alpha-KG in rice fermented liquid, capillary electrophoresis time-of-flight mass spectrometry (CE-TOFMS) analysis was carried out using an Agilent G7100 CE system, an Agilent G6224AA LC/MSD TOF system, and an Agilent 1200 series isocratic HPLC pump. The system was controlled by Agilent

ChemStation software for CE and MassHunter software for the Agilent TOFMS.

## 2.5 | Statistical analysis

The data were statistically analyzed using parametric multiple comparison procedure of Williams by BellCurve for Excel (Social Survey Research Information). A *p*-value for rejection of less than 5% was considered statistically significant.

## 2.6 | Clinical study testing methods

Thirty healthy female subjects, aged from 25 to 45 years old, were recruited. According to the self-assessment, the facial skin was rough and loose, and the wrinkles (crow's feet) on one side of the face were graded from 1 to 3 (according to the Japanese wrinkle classification map), the fine lines on one side of the face were graded from 1 to 3 (according to the Asian skin aging map VOL.02, p46-47), the forehead wrinkles were graded from 1 to 3 (according to the Asian skin aging map VOL.02, p32-33), and the nasolabial groove wrinkles on one side of the face were graded from 1 to 2 (according to the Asian skin aging map VOL.02, p46-47). According to the atlas of Asian skin aging VOL.02 (English version), p54-55, there were no obvious redness, skin lesions, scars, etc., in the test area, 30 people were recruited, and the effective data were 30.

According to the method of use, half face use the sample cream with Alpha-KG, and the other half face use the control cream continuously for four weeks, and the clinical evaluation of the test area of the subject's face was carried out through the specified time of D0, D14, D28, D56, VISIA-CR images were collected to analyze skin wrinkles and texture, and skin elasticity and firmness were measured to evaluate whether the test samples had the effect of improving facial wrinkles and texture, and the satisfaction of the subjects during the test was recorded.

On the test day, the subjects cleaned their faces with the cleansing products provided by the laboratory and then waited for 30 minutes in the laboratory environment with a temperature of 21 ± 1°C and a relative humidity of 50% ± 5% RH. The clinical evaluation on the subjects' faces. The experimenters collected facial images of the subjects, and measured the skin elasticity and skin tightness of the subjects' cheek area. The tester distributed test samples, auxiliary samples, and usage logs.

The subjects used the test samples and the control cream according to the instructions for use for 56 days. After 14 days, 28 days, and 56 days of using samples, the patients were followed up. On the day of follow-up, the subjects cleaned their faces with the cleansing products provided by the laboratory and then waited for 30 minutes in the laboratory environment with a temperature of 21 ± 1°C and a relative humidity of 50% ± 5% RH. The clinical evaluation on the subjects' faces. The experimenters collected facial images of the subjects and measured the skin elasticity and skin tightness of the

subjects' cheek area. The subjects filled in the self-assessment questionnaire in the laboratory.

## 2.7 | Clinical evaluation

Tester evaluated the efficacy of the product according to the facial condition of the subjects.

## 2.8 | Self-assessment

The subjects evaluated themselves according to their use.

## 3 | RESULTS

### 3.1 | Promoting effects of Alpha-KG on epidermal keratinocyte proliferation

First, we examined whether Alpha-KG has an ability to promote cell proliferation in keratinocytes. Alpha-KG is a weak acid containing two carboxyl groups and a ketone group which is also called 2-ketoglutaric acid or 2-oxoglutaric acid. Alpha-KG possesses many physiological function.

Alpha-KG significantly promoted epidermal keratinocyte proliferation (Figure 2).

### 3.2 | Promoting effects of Alpha-KG on mRNA expressions of genes related to skin hydration and barrier function

To further examine whether Alpha-KG promotes the mRNA expressions of genes which relate to skin hydration and barrier function,

promoting effects of Alpha-KG on mRNA expressions of FLG, SPT, and IVL in keratinocytes were investigated. Alpha-KG significantly promoted mRNA expressions of FLG (Figure 3), SPT (Figure 4), and IVL (Figure 5).

Filaggrin (FLG) is an important molecule that connects keratin fibers in the stratum corneum of human skin. It is abundant in the epidermis and plays an important role in the barrier function of the epidermis. The encoding gene filaggrin is located in the epidermal differentiation complex. The transparent keratinous particles in the granular layer of the epidermis are the expression sites of filaggrin. Under the condition of elevated calcium ion concentration, filaggrin is dephosphorylated and then proteolytic enzyme and serine protease CAP1/Prss are hydrolyzed and separated to produce free filaggrin monomer. Under the action of transglutaminase, the filaggrin monomer cross-links the keratinous intermediate filaments in KC, making it aggregate into giant fibril fibers, so that KC can be "compressed" into keratinocytes. Filaggrin is also involved in the formation of keratinized envelope. With the help of FLG monomers, keratin fibers gather regularly to form a solid physical barrier on the outermost layer of the epidermis. Skin barrier dysfunction caused by filaggrin (FLG) gene mutations plays an important role in the pathogenesis of atopic dermatitis and is considered to be the strongest genetic risk factor.

At the addition of 200, 1000, and 5000  $\mu\text{mol/L}$  of Alpha-ketoglutaric content, Alpha-ketoglutaric acid showed a significant promoting effect on FLG mRNA expression from  $100.0 \pm 5.5$  to  $128.3 \pm 2.9$ . These results indicate that Alpha-ketoglutaric can enhance the skin barrier.

Serine palmitoyl transferase (SPT) mRNA expression represents the synthesis level and content of sphingolipids in the skin. Serine palmitoyl transferase (SPT) is a key enzyme in the pathway of sphingolipid synthesis. Serine palmitoyl transferase (SPT) is the first step to catalyze the de novo synthesis of sphingolipids, and the first rate-limiting enzyme in this pathway. It plays a pivotal role in regulating sphingolipid metabolism, and its expression level affects the

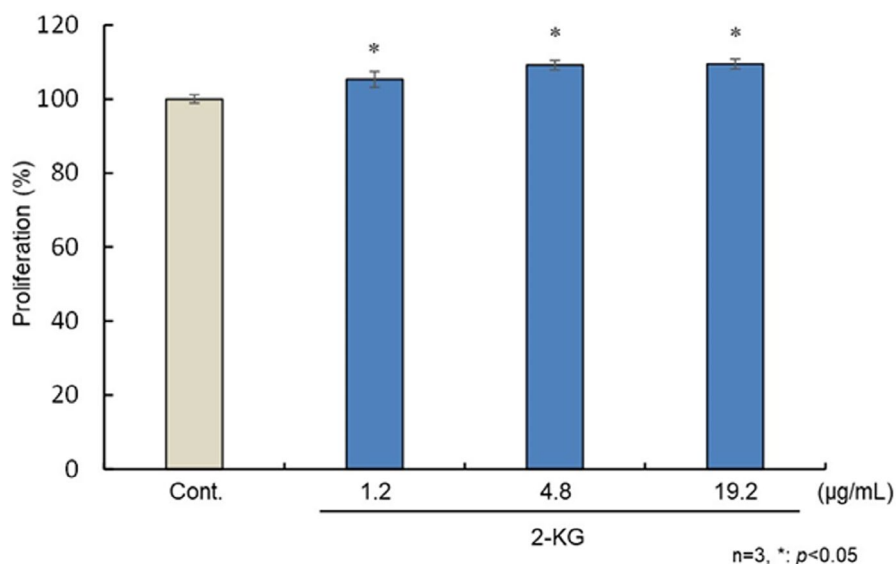


FIGURE 2 Promoting effects of a-KG on epidermal keratinocyte proliferation

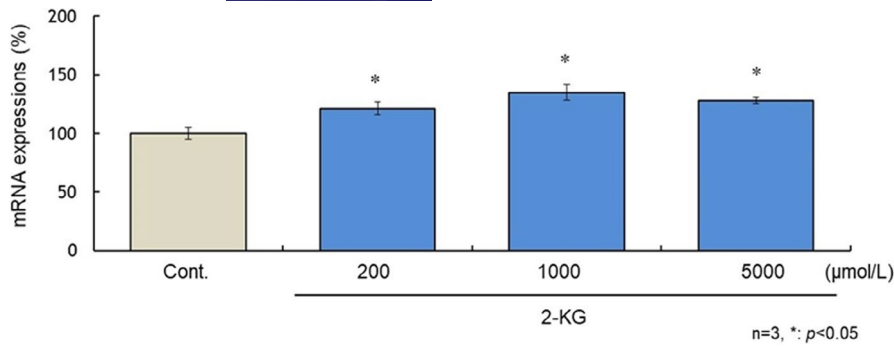


FIGURE 3 Promoting effects of a-KG on filaggrin mRNA expressions

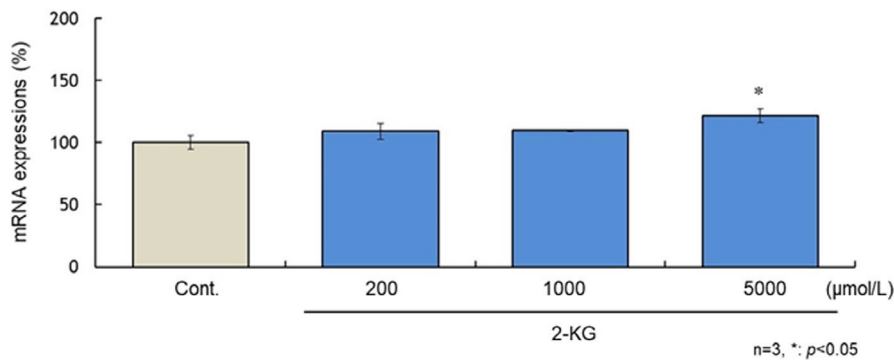


FIGURE 4 Promoting effects of a-KG on serine palmitoyltransferase mRNA expressions

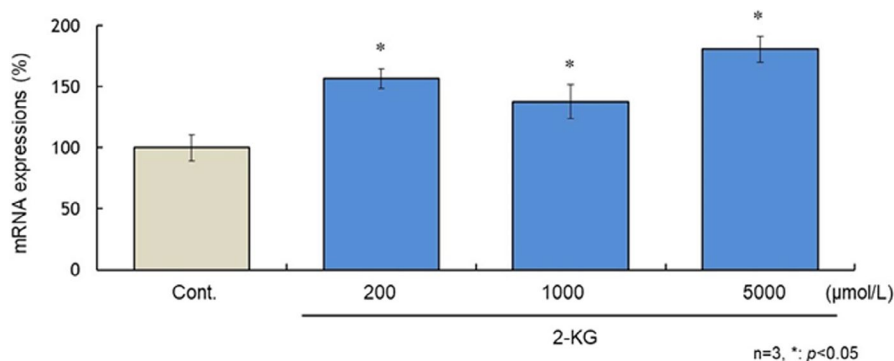


FIGURE 5 Promoting effects of a-KG on involucrin mRNA expressions

synthesis, distribution, and function of various sphingolipids in the body. Sphingolipid is an important component of the structure of biological membranes. Sphingolipids and their metabolites belong to a very important class of active molecules. They are involved in the regulation of cell growth, differentiation, senescence and programmed cell death and many other important signal transduction processes. Ceramides in the skin belong to sphingolipids. Sphingolipids mainly refer to ceramides, sphingomyelin, and glycosphingolipids. They are important components of cell membranes, which stimulate skin cell regeneration and enhance skin vitality. Ceramide is the epidermal stratum corneum. It is an important ingredient with excellent moisturizing effect that prevents moisture loss inside the skin and resists external damage.

According to Figure 4, at the addition of 200, 1000, and 5000 µmol/L of Alpha-ketoglutaric content, Alpha-ketoglutaric acid showed a significant promoting effect on SPT mRNA expression from  $100.0 \pm 5.5$  to  $121.5 \pm 5.5$ . These results indicate that

Alpha-ketoglutaric can enhance the skin barrier. The higher the expression of serine palmitoyl transferase (SPT) mRNA, the higher the synthesis level of sphingolipids in the skin, and the higher the content, hence promoting the content of ceramide in the skin and enhancing the less sensitive skin barrier.

Involucrin is a soluble cytoplasmic protein precursor of the epidermal cornified envelope that becomes cross-linked by transglutaminase during envelope assembly. Involucrin is expressed in a range of stratified squamous epithelia, including the cornea which lacks a distinct cornified layer. The protein is a useful marker of terminal differentiation. It is tightly linked to the onset of differentiation and first expressed in the immediate suprabasal layers of the epidermis. Involucrin is mapped to 1q21 chromosome, among calpactin I light chain, trichohyalin, profilaggrin, loricrin, and calcyclin.

Involucrin's structure is composed of a conserved region of about 75 amino acid residues followed by two very variable length segments that contain glutamine-rich tandem repeats. The glutamine

residues in the tandem repeats are the substrate for the transglutaminases in the cross-linking reaction. The size of the protein varies from 285 residues (in dog) to 835 residues (in orangutan).

At the addition of 200, 1000, and 5000  $\mu\text{mol/L}$  of Alpha-ketoglutaric content, Alpha-ketoglutaric acid showed a significant promoting effect on IVL mRNA expression from  $100.0 \pm 5.5$  to  $180.5 \pm 10.7$ . These results indicate that Alpha-ketoglutaric can enhance the skin barrier. The higher the expression of IVL mRNA, the better adhesion of the outer matrix to the skeletal structure in the cell, the stronger the brick wall structure of the cell, the tighter the cell and matrix link, and the better the skin barrier function.

It is concluded that Alpha-KG promotes skin hydration and barrier function by activating cell proliferation and by up-regulating mRNA expressions of FLG, SPT, and IVL in keratinocytes and that rice fermented liquid containing Alpha-KG could be a promising cosmetic ingredient for skin care products.

### 3.3 | Analysis of Alpha-KG in rice fermented liquid

As a candidate for cosmetic ingredients containing Alpha-KG, fermented materials were noticed. Five lots of rice fermentation liquid fermented with *Saccharomyces veronae* were analyzed. The analysis detected abundant Alpha-KG in the fermented liquid but not in the prefermented liquid (Table 1).

### 3.4 | VISIA testing

In Figure 6, from the analysis of VISIA image, we can see that there are 3-grade wrinkles around the eyes before treatment with the cream sample contact Alpha-KG. The wrinkles are thicker and denser. After D28 of use, the wrinkles around the eyes can be reduced to 2 grade. After D56 of use, the wrinkles around the eyes are further reduced to 1 grade, and the texture density is also further reduced. It shows that the sample has the effect of reducing wrinkles around the eyes.

In Figure 7, it can be seen from the cheek texture analysis of VISIA image that the facial pores are thicker and denser before the sample cream with Alpha-KG treatment. After D28 of using the sample cream, the facial pore texture begins to decrease from 3 grade to 2 grade, and the skin begins to become delicate. After D56 of using the sample, the facial pores become significantly smaller to 1 grade,

TABLE 1 The amount of  $\alpha$ -KG in rice fermented liquid

	The amount of $\alpha$ -KG (nmol/L)
Rice prefermented liquid	N.D.
Rice fermented liquid	$856.7 \pm 12.8$

Note: Mean  $\pm$  SE,  $n = 5$ .

Abbreviation: N.D., Not detected.

and the skin texture further decreases. It shows that the skin care product has the effect of refining pores and tightening skin.

Through the VISIA testing, the skin wrinkles were significantly reduced by 23.64% by D0. After using the sample for D7, the average clinical score of outer corner wrinkles was significantly reduced by 15.23%, and nasolabial groove wrinkles were significantly reduced by 25.68%. The mean clinical evaluation score of crow's feet decreased significantly by 25.42% after 56 days using.

### 3.5 | Clinical testing

Compared with that before using the sample, after D7 of using the sample, the average clinical score of external canthus wrinkle grade was significantly reduced by 15.23%, the average clinical score of nasolabial groove wrinkle grade was significantly reduced by 25.68%, and the average clinical evaluation score of skin firmness was significantly increased by 41.40%; the characteristic count of forehead skin wrinkles decreased significantly by 21.83%. The mean clinical evaluation score of crow's feet decreased significantly by 25.42% in subjects who used D56 continuously. It shows that the sample can reduce outer corner wrinkles and nasolabial groove wrinkles, improve skin firmness, reduce forehead wrinkles, and has good anti-aging effect.

### 3.6 | Subjects self-assessment

After D56 of using the sample, the mean self-assessment score of crow's feet decreased significantly by 25.68%; the mean value of self-assessment score decreased significantly by 18.17%; the mean self-assessment score of forehead wrinkles decreased significantly by 15.08%; the mean self-assessment score of present wrinkles decreased significantly by 24.01%; the mean self-assessment score of skin firmness increased significantly by 40.24%; the mean value of skin color self-assessment score increased significantly by 42.05%. Most of the participants thought that the cream sample was helpful to smooth fine lines, thinning pores, pulling tight and brightening complexion, and satisfied with the results of the test samples. All subjects did not have any adverse skin reactions.

## 4 | DISCUSSION

Alpha-KG is derived from natural rice fermentation broth. In relevant literature research, it has many related applications in protein synthesis, promoting collagen synthesis, immune system regulation, and delaying aging. However, there are few literature reports on its application in the field of skin care. In this study, it was applied to cream as an anti-aging active ingredient. The experimental data show that Alpha-KG has a positive effect on the expression of mRNA in filaggrin (FLG), serine palmitoyltransferase (SPT), and involucrin (IVL),



FIGURE 6 VISIA analysis of periorcular wrinkles



FIGURE 7 VISIA analysis of cheek wrinkles

which can enhance the water retention capacity of the stratum corneum and maintain the skin barrier function.

Immediately after using the sample for D0, the skin wrinkles were significantly reduced by 23.64%. After using the sample for D7, the average clinical score of outer corner wrinkles was significantly reduced by 15.23%, and nasolabial groove wrinkles were significantly reduced by 25.68%. After using the sample for D56, the mean clinical evaluation score of crow's feet decreased significantly by 25.42%; the average score of clinical evaluation of skin firmness increased significantly by 41.40%; the skin gloss increased significantly by 28.67%.

Most of the participants thought that the cream sample was helpful to smooth fine lines, thinning pores, pulling tight, and brightening complexion, and satisfied with the results of the test samples. All subjects did not have any adverse skin reactions.

To sum up, instant cream containing Alpha-KG can significantly smoothen the skin and reduce the wrinkles of the eyes and nasolabial fold. Continuous use can improve the moisture content of the skin, tighten the skin, reduce fine lines on the forehead, outer corners of the eyes, nasolabial groove, now and the whole face, and improve the skin gloss. According to the above research data, the cream containing Alpha-KG has good anti-aging effect.

## 5 | CONCLUSION

It is concluded that Alpha-KG promotes skin hydration and barrier function by activating cell proliferation and by up-regulating mRNA expressions of FLG, SPT, and IVL in keratinocytes and that rice fermented liquid containing Alpha-KG could be a promising cosmetic ingredient for anti-aging skin care products. Through the clinical

testing, rice fermented liquid containing Alpha-KG could be an anti-aging cosmetic ingredient for skin care products.

## CONFLICT OF INTEREST

I am aware that this journal requires all authors to disclose any potential sources of conflict of interest in the manuscript. There is no conflict of interest of any of the authors of this paper.

## AUTHOR CONTRIBUTIONS

Fan Yang and Ziyang Zhou designed the research study. Miao Guo contributed essential reagents and tools. Fan Yang completed the experimental testing. Zheng Zhou analyzed the testing data. Fan Yang and Ziyang Zhou wrote the paper. Thank you to Jinlong Zhang and Yang Guo who contributed to the early stages of the article. I confirm that each of the co-authors acknowledges their participation in conducting the research leading to this manuscript and has agreed to its submission to be considered for publication.

## ETHICAL STATEMENT

I confirm that all the research meets the ethical guidelines, including adherence to the legal requirements of the study country.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

## REFERENCES

1. Mailloux RJ, Bériault R, Lemire J, et al. The tricarboxylic acid cycle, an ancient metabolic network with a novel twist. *PLoS One*. 2007;2(8):e690.

2. Liu S, He L, Yao K. The antioxidative function of alpha-ketoglutarate and its applications. *Biomed Res Int.* 2018;2018:1-6. <https://doi.org/10.1155/2018/3408467>
3. Xiao D, Zeng L, Yao K, Kong X, Wu G, Yin Y. The glutamine-alpha-ketoglutarate (AKG) metabolism and its nutritional implications. *Amino Acids.* 2016;48(9):2067-2080.
4. Chin RM, Fu X, Pai MY, et al. The metabolite alpha-ketoglutarate extends lifespan by inhibiting the ATP synthase and TOR. *Nature.* 2014;510(7505):397-401.
5. Bayliak MM, Lushchak VI. Pleiotropic effects of alpha-ketoglutarate as a potential anti-ageing agent. *Ageing Res Rev.* 2021;66(suppl 1):101237.
6. Horii I, Nakayama Y, Obata M, Tagami H. Stratum corneum hydration and amino acid content in xerotic skin. *Br J Dermatol.* 1989;121:587-592.
7. Weiss B, Stoffel W. Human and murine serine-palmitoyl-CoA transferase cloning, expression and characterization of the key enzyme in sphingolipid synthesis. *Eur J Biochem.* 1997;249:239-247.
8. Tanno O, Ota Y, Kitamura N, Katsube T, Inoue S. Nicotinamide increases biosynthesis of ceramides as well as other stratum corneum lipids to improve the epidermal permeability barrier. *Br J Dermatol.* 2000;143:524-531.
9. Eckert RL, Yaffe MB, Crish JF, et al. Involucrin - structure and role in envelope assembly. *J Invest Dermatol.* 1993;100(5):613-617.
10. Moat AG, Ahmad F, Alexander JK, Barnes IJ. Alteration in the amino acid content of yeast during growth under various nutritional conditions. *J Bacteriol.* 1969;98(2):573-578.
11. Liu Q, Yu T, Campbell K, Nielsen J, Chen Y. Modular pathway rewiring of yeast for amino acid production. *Methods Enzymol.* 2018;608:417-439.

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