News Release

November 22, 2024

mandom

Mandom Discovers That Potassium Alum, Known to Be Found in Hot Spring Water, Improves the Barrier Function of the Skin

—Loricrin production and tight junction formation promoted in human epidermal keratinocytes—

Mandom Corporation (head office: Osaka City, President Executive Officer & Director: Ken Nishimura, hereinafter referred to as "Mandom") conducted joint research at the Laboratory of Advanced Cosmetic Science, Graduate School of Pharmaceutical Sciences, Osaka University, in collaboration with Professor Ken Ishii in the Department of Microbiology and Immunology, The Institute of Medical Science, The University of Tokyo (formerly Invited Project Leader of the Mockup Vaccine Project, National Institutes of Biomedical Innovation, Health and Nutrition), aiming to develop a technology for keeping the human skin healthy and beautiful. We discovered that potassium alum, known to be found in hot spring water, promotes loricrin production and tight junction formation by human epidermal keratinocytes to improve skin barrier function.

Mandom will apply the results to developing products that help create healthy and beautiful skin, with a focus on the mechanism of skin beautification by hot springs.

We presented the results at the 34th Congress of the International Federation of Societies of Cosmetic Chemists at Iguazu 2024 (IFSCC Congress), held in Iguazu, Brazil, from Monday, October 14 to Thursday, October 17, 2024.

Background of the research

Thermal skin care by warming the skin, such as through hot springs, spas, and massages, is known to be beneficial for the skin, but the mechanism of its action is unclear. We have found so far that potassium alum, found in hot spring water, activates TRPM4, a TRP channel*1 serving as a cellular sensor for warm temperatures, suppresses inflammatory signals from human epidermal keratinocytes (press release of May 28, 2019), and promotes the proliferation of epidermal keratinocytes (press release of November 30, 2023), thus demonstrating that potassium alum improves skin condition by activating TRPM4. However, the effect of potassium alum on the barrier function of the skin was unknown.

In this situation, we attempted to determine the effect of potassium alum on the skin barrier function and to analyze the mechanism involved.

1. TRPM4, the target of potassium alum, is present in the human epidermis

Although TRPM4 was known to be contained in human skin and skin cells, its spatial distribution in human skin, i.e., where it is localized, was unclear.

Therefore, we analyzed human skin samples to locate the expression sites of the TRPM4 gene, using a technique called *in situ* hybridization*2, which allows us to locate genes in biological tissue. As a result, we confirmed the wide distribution of the TRPM4 gene in the human epidermis (Figure 1). This suggests that TRPM4 is also present in actual human skin, particularly in the epidermis, and that activating epidermal TRPM4 with thermal stimulation or potassium alum may be effective in improving skin condition.

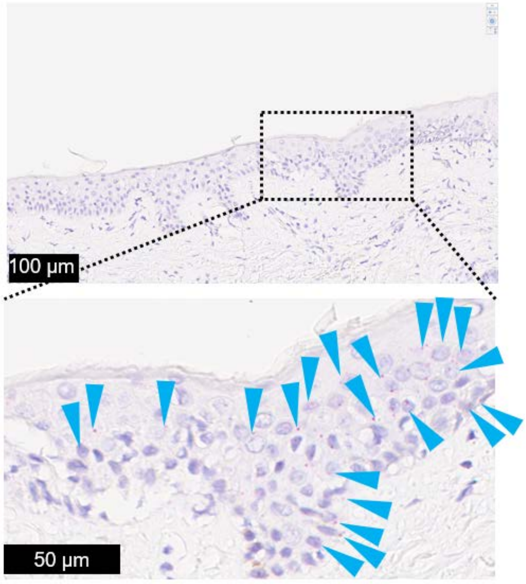

Figure 1. Expression of the TRPM4 gene in the human skin
◄: Each symbol represents a TRPM4 gene expression site.

2. The hot spring ingredient potassium alum promotes the keratinization of epidermal keratinocytes

The epidermis constantly repeats turnover process. Epidermal keratinocytes are born in the basal layer, the innermost layer of the epidermis, and pushed out by later-born cells, then migrate to the upper layers while keratinizing, eventually falling off. During this turnover process, loricrin, one of the important molecules responsible for the barrier function of the cornel layer, is produced.

When culturing a 3D epidermal model with the addition of the TRPM4 activator potassium alum, we showed that the expression level of the loricrin gene in epidermal keratinocytes significantly increased (Figure 2A). It was also found that transepidermal water loss (TEWL) decreased at the same time (Figure 2B). These results indicate that potassium alum promoted the keratinization of epidermal keratinocytes and enhanced the barrier function of the skin.

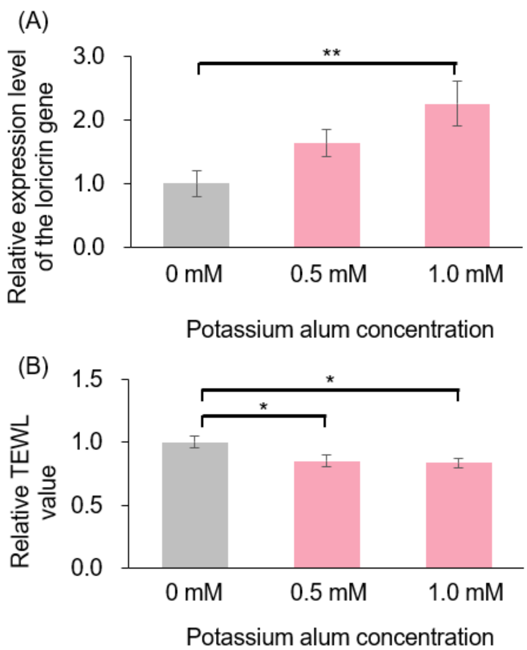

Figure 2. Effects of potassium alum on the barrier function of a three-dimensional model of the epidermis
(A) Effect on loricrin gene expression level (B) Effect on transepidermal water loss (TEWL).
* p<0.05, ** p<0.01, one-way ANOVA, Dunnett's test.

3. The hot spring ingredient potassium alum enhances the tight junction function of epidermal keratinocytes

It is known that not only keratinization but also tight junctions in the stratum granulosum play an important role in the barrier function of the skin. Tight junctions act as strong adhesives between cells, preventing the entry of irritating substances from outside the body and the evaporation of water and moisturizing components from the inside (Figure 3). When the barrier function of the skin weakens, the function of the tight junctions is impaired.

We evaluated the function of the tight junctions by adding a tracer to the 3D epidermal model and imparting a red color. The tracer is capable of freely passing through the intercellular gaps but cannot penetrate the normally functioning tight junctions. In the 3D epidermal model cultured at 37°C, the tracer cannot pass through the tight junctions, so the tracer ceases its movement at tight junction spots (shown in light blue, Figure 4A). On the other hand, in the same model cultured at 33°C, the tracer did not cease its movement at the tight junction spots but leaked out (shown in pink, Figure 4A). This indicates that the function of tight junction is impaired under low-temperature conditions. In fact, we found that the number of tight junctions in which the tracer had ceased its movement, i.e., which were functioning normally, was reduced under low temperature conditions (Figure 4B). Furthermore, when adding potassium alum, even at a culture temperature of 33°C, the tracer ceased its movement (Figure 4A), resulting in an increased number of functional tight junctions (Figure 4C). Similar results were obtained with BTP2*3, a known TRPM4 activator (Figure 4D), demonstrating that potassium alum, a hot spring ingredient and a TRPM4 activator, enhances the function of the tight junction by activating TRPM4.

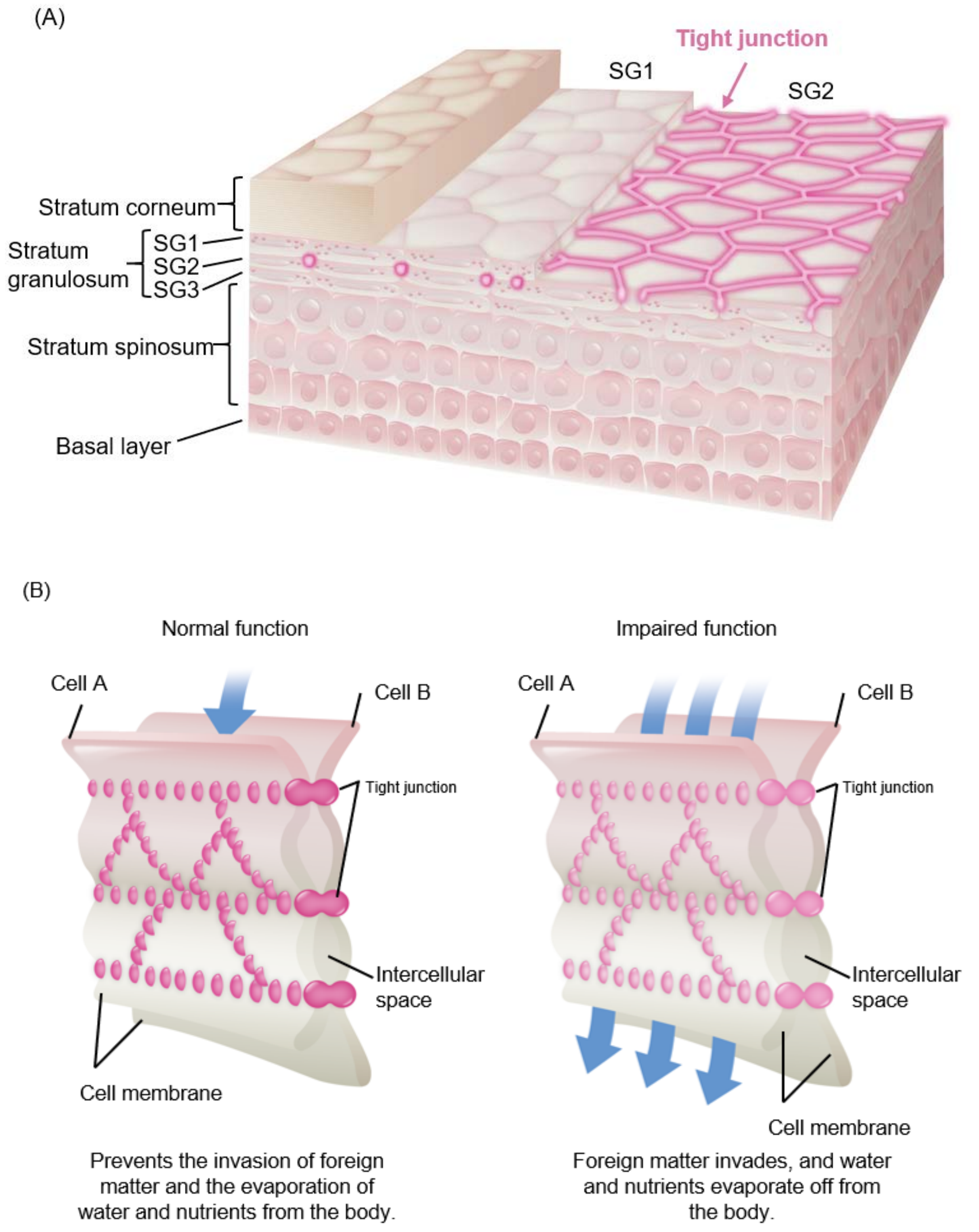

Figure 3. Schematic diagram of tight junctions
(A) Epidermal structure and tight junctions
(B) Magnified view of tight junctions

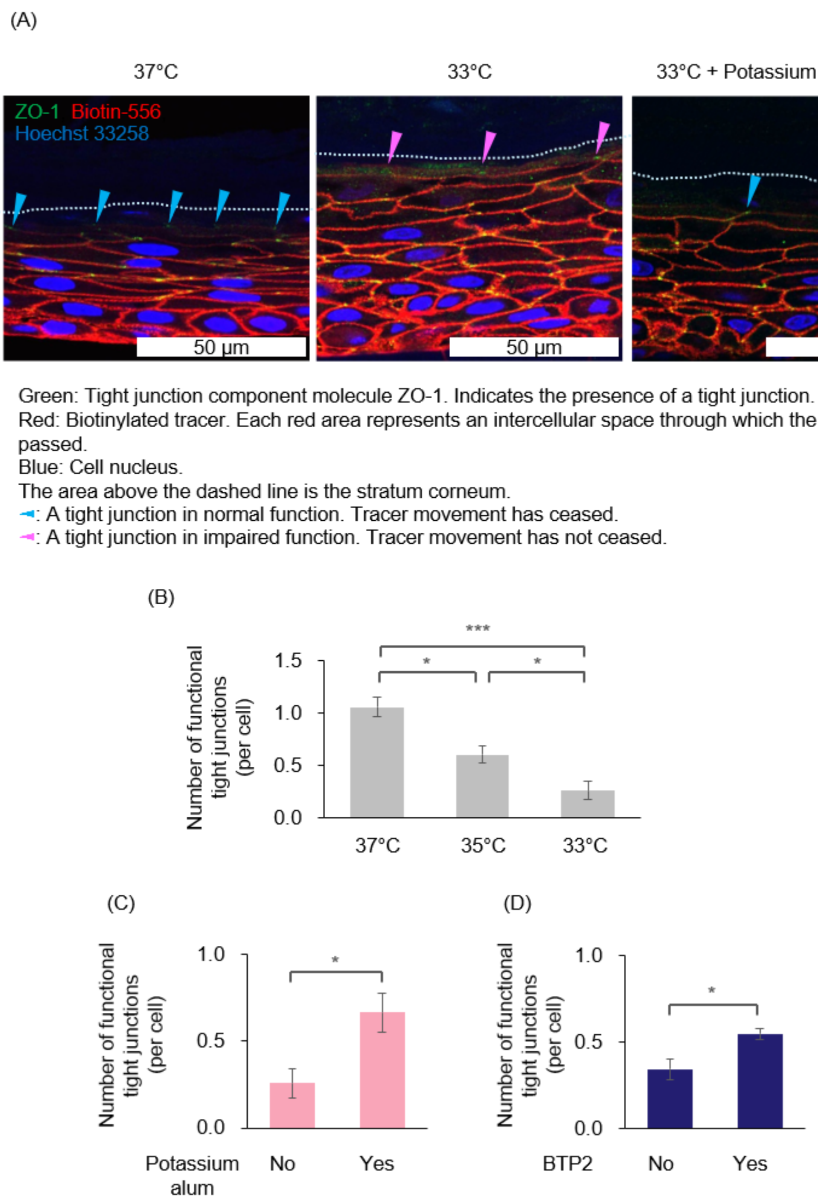

Figure 4. Effects of temperature and TRPM4 activator on tight junction formation in a three-dimensional model of the epidermis
(A) Microscopic image (B) Effect of temperature. * $p<0.05$, *** $p<0.001$, one-way ANOVA, Tukey's test. Effects of TRPM4 activators potassium alum (C) and BTP2 (D) on tight junction formation under low-temperature conditions (33°C). * $p<0.05$, Student's t-test.

4. A lotion containing potassium alum improves skin condition

In a group of subjects using a lotion containing potassium alum twice a day for four consecutive weeks, the TEWL value, an index of skin barrier function, decreased (Figure 5A), demonstrating improved skin barrier function. In another group using a control lotion, on the other hand, the TEWL value did not change (Figure 5B). Additionally, a previous study showed that corneal layer moisture contents increased with the use of a lotion containing potassium alum (published at the International Federation of Societies of Cosmetic Chemists Conference, 2019 at Milan), and similar results were obtained in the present study

(Figure 5C). In the group using the control lotion, on the other hand, the corneal moisture content decreased (Figure 5D). This is probably because the study was conducted in winter. Therefore, it is considered that skin condition can be improved by using a lotion containing potassium alum, even at low temperatures and in dry environments.

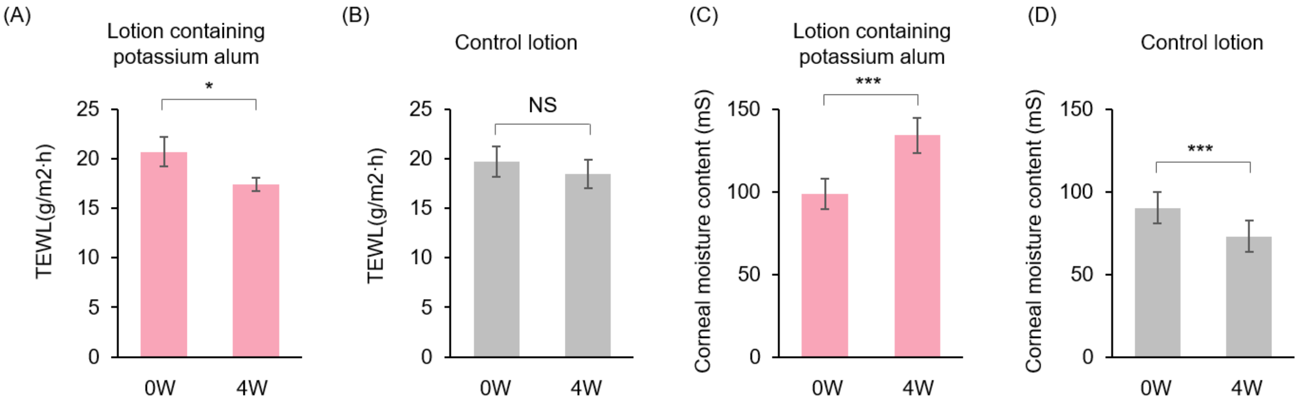

Figure 5. Effects of topical application of the lotion containing potassium alum on the skin
(A, B) Effects on transepidermal water loss (C, D) Effects on corneal moisture content
A total of 64 subjects in two groups received either the lotion containing potassium alum (A, C) or control lotion (B, D) and apply twice a day for four consecutive weeks. * $p<0.05$, *** $p<0.001$, Student's t-test.

The hot spring ingredient potassium alum activates TRPM4 to suppress inflammatory signals from epidermal keratinocytes and promotes their proliferation. Besides these effects, our study demonstrated its effect in enhancing the barrier function of the skin. This finding is expected to lead to the development of a new approach to skin care that is as effective in skin beautification as heat, using potassium alum, without actually warming the skin.

Additionally, we will proceed to investigate the actions of other hot spring ingredients on the skin and the thermal impacts on skin condition and promote the development of products that help create healthy, beautiful skin like that observed just after bathing in a hot spring.

Notes and glossary

*1 TRP = Transient Receptor Potential
A sensor belonging to the cation channel family involved in various sensory receptions that senses chemicals, temperatures, etc. and converts them into electrical signals.

*2 *In situ* hybridization
An experimental method for investigating the spatial distribution of a target gene by reacting the target gene with a probe specific to its gene sequence in a sample, such as biological tissue.

*3 A substance known to activate TRPM4.
N-[4-[3,5-Bis(trifluoromethyl)-1H-pyrazol-1-yl]phenyl]-4-methyl-1,2,3-thiadiazole-5-carboxamide