Novel second skin technology by Direct-Electrospinning method with adhesive primer to achieve superior makeup appearance and longevity

Asami, Nobuyuki[1*]; Park, Soyoung[1]; Okiyama, Natsuko[1]; Nishino, Ken[1]; Igarashi, Takanori [2]; Takimoto, Baku[3]; Miyazaki, Yukihiro[1]; Yamamoto, Takamasa[1]

[1] Makeup Product Research, Kao Corporation, Kanagawa, Japan; [2] Skincare Product Research, Kao Corporation, Tokyo, Japan; [3] Analytical Science Research, Kao Corporation, Tokyo, Japan

* Nobuyuki Asami, 5-3-28, Kotobuki-cho, Odawara, Kanagawa, 250-0002, Japan
+81-(0)465-34-6478, email: asami.nobuyuki@kao.com

Abstract

Background: Skin has a barrier function and a social function, e.g. aesthetic appearance. Recently, technology to form a single ultra-thin film on the skin, termed second skin, has been expected to go beyond conventional cosmetic products, and the second skin is reported to have highly effective barrier functions. However, the applicability of the second skin on makeup is limited owing to noticeable film deterioration, such as peeled edges and wrinkles. To address these issues, we aimed to develop a novel second skin technology applicable to makeup for the first time. Next, we evaluated the makeup effects of the developed technology. We then attempted to determine the mechanisms behind these effects.

Methods: We developed a technology utilising the direct-electrospinning (D-ES) method, wherein an ultra-thin film is formed by electrospinning fine fibres directly onto the skin using a portable device, after applying an adhesive primer.

Results: (1) The new technology formed an unnoticeable ultra-thin film with sufficient durability. (2) Superior makeup effects of smooth appearance, long-lasting performance, and high removability, as well as anti-pollution and moisturising effects, were seen. (3) SEM observations clarified three unique structures: (a) the film-bridged skin grooves, (b) pigments existing inside the fine fibre network, and (c) skincare and makeup formulations existing separately inside the film. These results indicate that these structures led to superior makeup and skincare effects.

Conclusion: Combining the D-ES method and the primer enabled makeup using the second skin for the first time, which resulted in superior cosmetic effects.

Keywords: second skin; direct electrospun fine fiber; adhesive primer; smooth and lasting makeup; makeup removability; barrier function

1. Introduction

Human beings have acquired various skin functions through the evolutionary process. One of them is a barrier function that protects from excess water loss and unwanted external irritants through a dense internal structure consisting of stratum corneum cells and intercellular lipids [1]. Skin also has a social function e.g. a unique aesthetic appearance, by having a rough surface structure with grooves and pores, as well as an internal structure with coloured melanin inside translucent epidermis [2]. Skincare and makeup products have long been developed to improve these skin functions.

Recently, a technology to form a single ultra-thin film, termed second skin, has attracted attention in the cosmetic field [3]. This technology is expected to overcome the limitation of skincare and makeup effects of conventional products by controlling the film density, surface roughness, and colourants distribution. As for skincare, it has been reported that the second skin technology, such as cross-linked polymers [4], polymer composites [5] and electrospinning sheets [6], enhances the skin functions such as moisturisation and protection against pollution.

However, for makeup, the applicability of second skin technology to improve aesthetics is limited. This is because deterioration of the film is an obstacle in its practical use. Specifically, because it is a single film, noticeable edges and wrinkles are likely to occur during the application. In addition, it is very difficult to have sufficient durability for long-term wear under a non-woven fabric mask which many people currently use to prevent COVID-19.

To address the issues of noticeable defects and low durability, we aimed to develop a novel second skin technology which is suitable for makeup for the first time. To overcome the defects, we have focused on the direct-electrospinning (D-ES) method [7]. Because this technology forms an ultra-thin film by electrospinning fine fibres directly onto the skin using a portable device, the film is expected to have no noticeable edges or wrinkles during application. Furthermore, to make this film sufficiently durable against the mask, we also tried to develop a primer formulation that increased adhesion to the skin by applying it before D-ES.

We evaluated this technology as follows: First, we evaluated whether the technology was suitable for facial makeup by overcoming film deterioration. Then, we evaluated the makeup effects of the developed film combined with the primer and foundation as well as its barrier function. Furthermore, we analysed the mechanisms behind the makeup effects of this technology.

2. Materials and Methods

2.1. Material

Creation of the film using the D-ES method

A polymer solution for forming an ultra-thin film using electrospinning was newly developed by dissolving polyvinyl butyral in ethanol with additives such as polyol and oils, based on past knowledge [8]. In subsequent experiments, 1 mg/cm^2 of the polymer solution was electrospun directly onto the skin or substrate using a portable device [9].

Primer formulation

A primer formulation to improve the durability of the D-ES film was newly developed by mixing a water-resistant polymer emulsion with additives, such as polyols and thickeners, in water. Then, 1.5 mg/cm^2 of the primer was spread onto the skin or substrate.

2.2. Evaluation of the film with the primer for practical use

To confirm the suitability of the developed technology for makeup, the following evaluations were carried out immediately after application and after long-time wear.

2.2.1 Observation of the film state immediately after application

In vivo test

After a commercial toner and lotion were applied on the faces of eight Asian female volunteers (ages: 20s–50s), the primer was applied only on their half faces, and the film was created using D-ES method on their entire faces. To visualise the film, whereas the toner and lotion contained no ultraviolet (UV) absorbent, a UV absorbent was added to the polymer solution for this experiment. The photographs and UV images were taken using VISIA Evolution (Canfield Scientific Inc.).

Height image observation

For the precise analysis, a height image of the film was obtained. The polymer solution was sprayed directly onto a silicon wafer. For comparison, a ready-made electrospun sheet with the same polymer solution was pasted, and the same solution was spread using a finger. The height images of the samples were obtained using a laser microscope (VR-5000, Keyence).

2.2.2. Durability evaluation of the film against friction of mask

In vivo test

After the in vivo test described in section 2.2.1, the volunteers put on a non-woven fabric mask and spent their time as usual for 8 hours. To quantitatively analyse the durability of the film against the mask, the film area was visualised with VISIA UV imaging, and the remaining rates at 6 and 8 hours were calculated as follows:

$$(Remaining\ rate\ \%) = \frac{Film\ Area\ @\ 6\ or 8\ hours}{Film\ Area\ @\ 0\ hour} \times 100$$

In vitro adhesion force measurement

A polymer solution was electrospun on artificial leather (Laforet S2923, Okamoto Shinwa Co., Ltd.) with and without the primer to form a film sample with a size of 1.5 cm × 12 cm. After drying for 1 hour, a peeling test was conducted using a tensile tester (Tensilon, ORIENTEC Co., Ltd.) at a speed of 300 mm/min to measure the adhesion force of the film [10]. The tests were conducted more than three times.

2.3. Evaluation of the cosmetic effects of the primer and film

To investigate the cosmetic effects of the developed technology, the following evaluations were performed:

2.3.1. Appearance evaluation

In vivo test

A commercial toner, a commercial UV lotion and the developed primer were applied on the entire faces of eight Asian female volunteers (ages: 30s–50s), followed by the D-ES film application. Finally, a commercial foundation was applied. The makeup appearance was compared to that without the primer and film.

In addition to the questionnaire administered to the volunteers, photographs of the appearance were taken using VISIA, and deep neural network-based artificial intelligence (AI) analysis [11] was also utilised on the VISIA images to evaluate the smoothness.

For optical analysis, photographs of 5 cm × 5 cm on the cheek were captured using a camera (D300s, Nikon). The light source and the camera were arranged in a horizontal plane. The detection angle was fixed at 0° or –45°, while the angle of incidence was set from –75° to 75°. The standard deviation (SD) of the pixel values in the central area of 1 cm × 1 cm of the image was calculated.

The distribution of the foundation on the skin was observed using a microscope (RH-2000, Hirox). To emphasise the distribution of the foundation colourants, two polarising filters and a specially designed multi-bandpass filter were attached to the microscope [12, 13].

2.3.2. Lasting performance

In vivo test

After a commercial toner and UV lotion were applied to the entire faces of eight Asian female volunteers (ages: 20s–50s), the primer and the film created using D-ES were applied only on half of their faces. Finally, a commercial foundation was applied. They wore a non-woven fabric mask and spent their day as usual.

In addition to the questionnaire administered to the volunteers, photographs were taken using a camera (D7100, Nikon) at 6 and 8 hours. VISIA images were analysed using the AI system [14].

Photographs of the mask after 8 hours were also captured with a camera (EOS R5, Canon). The image of the mask was analysed to calculate the colour transfer rate to the mask as follows:

$$(colour\ transfer\ rate\ \%) = \frac{Colour\ transfer\ area\ @\ 8hours}{Area\ of\ mask} \times 100$$

2.3.3. Makeup removability

In vivo test

After the 8-hour wearing test described in section 2.3.2., the film was peeled off, and the skin was observed using a microscope in the same way as described in section 2.3.1. For comparison, a microscope image was also obtained after the conventional makeup was wiped off using a commercial makeup-remover wipe.

In vitro test

The primer and the film created using D-ES were applied on a green artificial skin plate (Bioskin, Beaulax Co., Ltd). Finally, a commercial foundation was applied. The a* value of the skin plate was measured before and after peeling, and the removal rate was calculated as follows:

$$(removal\ rate\ \%) = \left\{1 - \frac{\Delta a^*_{before\ peeling-modeled\ skin}}{\Delta a^*_{after\ peeling-modeled\ skin}}\right\} \times 100$$

2.3.4. Transepidermal water loss (TEWL) test

In vivo test

The primer and the film created using D-ES were applied in the range of 3 cm × 3 cm of bare skin on the inner forearm of eight Asian volunteers (ages: 20s–50s). TEWL was measured using VapoMeter (Delfin Technologies) 30 minutes and 6 hours after the application. The measurement was carried out in the environment of 22°C and 50 %RH at least 20 minutes after the acclimation.

2.3.5. Anti-pollution test

In vitro test

A commercial lotion, the primer, and the film created using D-ES were applied to artificial leather (Laforet). Graphite (2 mg/cm^2) of was uniformly applied on top. The microscopic image (VHX-600, Keyence) was then used to calculate L*, a*, and b*. The removal rate of pollutant by peeling off the film was calculated as follows:

$$(removal\ rate\ \%) = \left\{1 - \frac{\Delta E_{before\ peeling-articial\ leather}}{\Delta E_{after\ peeling-artificial\ leather}}\right\} \times 100$$

$$\Delta E = \sqrt{\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}}$$

The tests were conducted three times. Both sides of the peeled film were observed by scanning electron microscope (SEM, TM3030Plus, Hitachi).

2.4. SEM observation to reveal the mechanisms behind the makeup performances.

The surface and internal structures of the films on Aluminum and polyimide substrates were observed with SEM (JSM 7200F, JEOL). A cryostat-cross section polisher (IB-19520 CPP, JEOL) was used for the creation of a cross-section.

2. 5. Statistical analysis

Differences between groups, expressed as mean ± standard error (SE), were evaluated using a paired *t*-test or Welch's *t*-test. A probability of $p<0.05$ was considered significant (* $p<0.05$; ** $p<0.01$; *** $p<0.001$).

3. Results

3.1 Establishment of the novel second skin technology

3.1.1. Observation of the film state immediately after application

In vivo test

To evaluate whether the D-ES film was suitable for makeup, the film state was observed after D-ES application. As shown in Figure 1(a), the film was invisible, and there were no noticeable defects, such as edges or wrinkles in the film. This result was the same as that obtained for the film with the primer.

Height image observation

The results of the height images are shown in Figure 1(b). While the pasted ready-made film and the film of the same solution spread with a finger had noticeable edges and wrinkles, the D-ES film was flat without any defects. This flatness of the film was consistent with the result on the face.

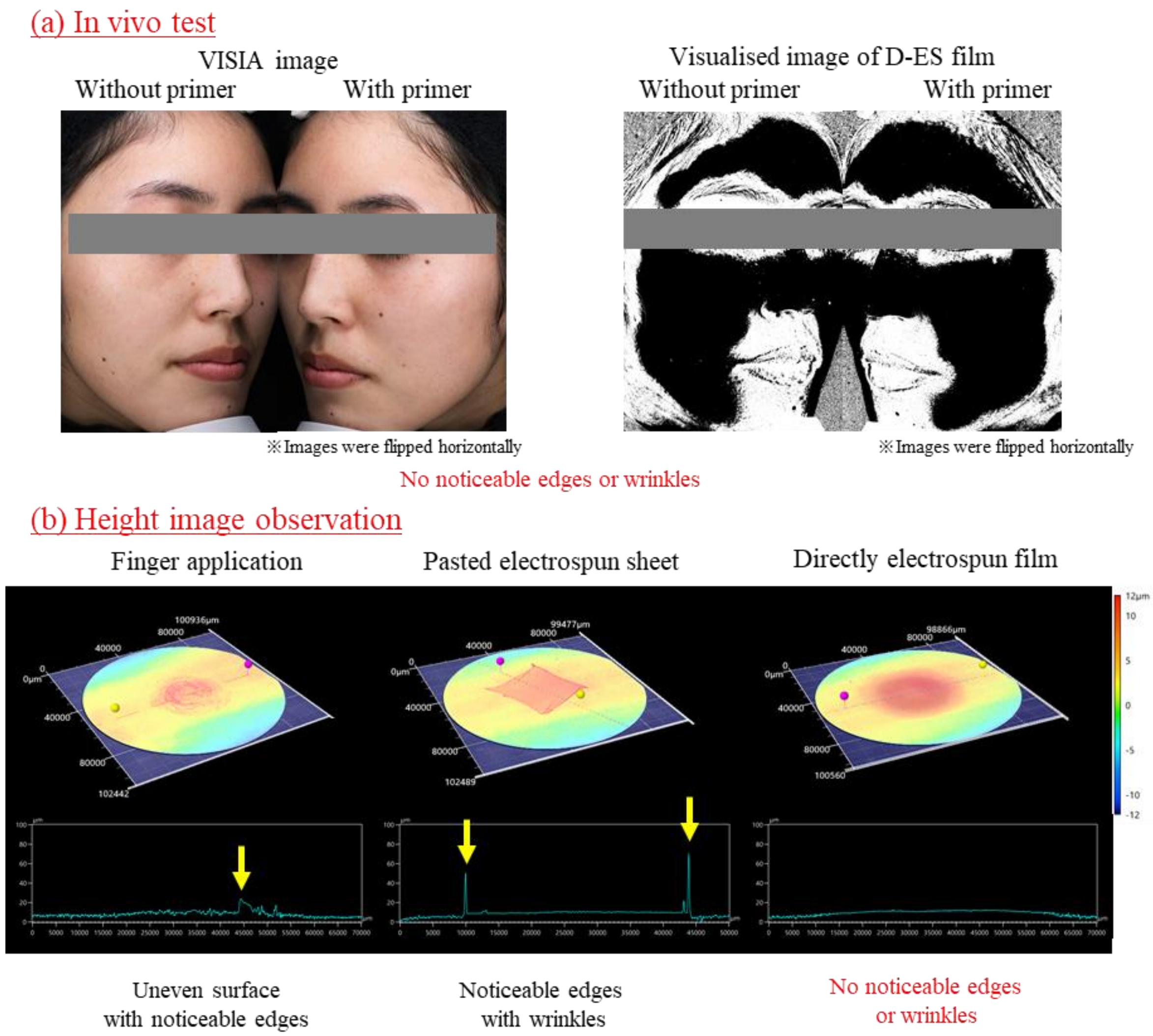

Figure 1: Observation of the film state immediately after direct-electrospinning (D-ES) application. (a) VISIA image (coloured) and the visualised film image by VISIA-UV (black-and-white) after applying a commercial toner and lotion without UV absorbent, followed by the application of the film containing UV absorbent by D-ES method. (b) In vitro height image of direct-electrospun film on silicon wafer obtained by laser microscope and the comparison of the film spread with a finger and the pasted ready-made electrospun film.

3.1.2. Durability of the film with the primer

In vivo test

To evaluate the durability of the film against the mask, the volunteers wore the film and spent their time wearing a mask. Figure 2(a) shows that the peeled edge of the film occurred without the primer at 8 hours, indicating that the film without the primer was not sufficiently durable against the mask. On the other hand, when primer was applied below the film, no noticeable edges were observed, with only a slight invisible edge in the thinned part.

The average remaining rates of the film at 6 and 8 hours in Figure 2(b) prove that the remaining rates were significantly higher with the primer. This result indicates that the primer increased the durability of the D-ES film to a practical level.

Adhesion force measurement

The adhesion force between the film and substrate was measured in vitro. Figure 2(c) reveals that while the adhesion force without the primer was minimal, the force was increased by the developed primer. These results indicates that the increased adhesion by the primer improved the durability against the mask.

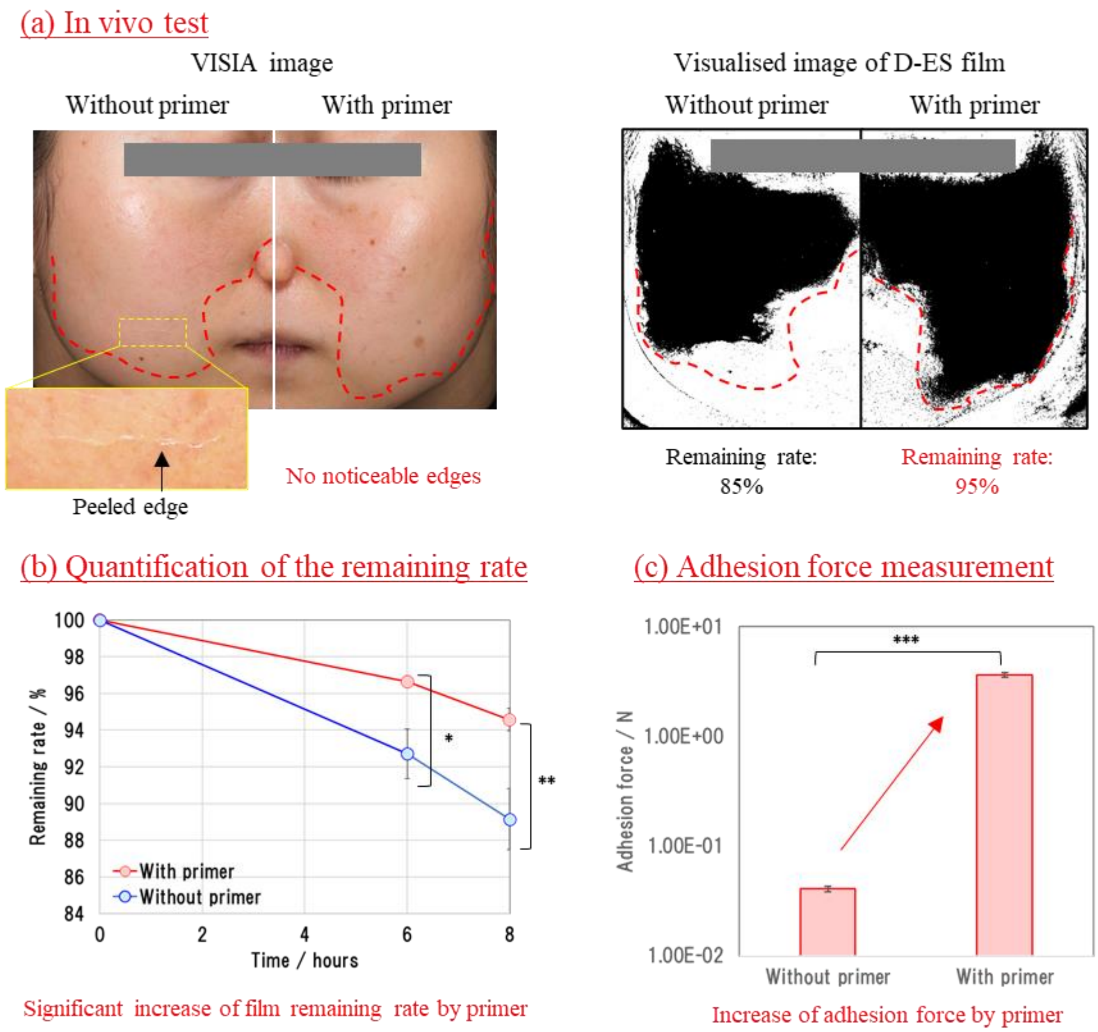

Figure 2: Evaluation of the durability of the D-ES film with primer. (a) VISIA image (coloured) and visualised film image (black-and-white) after an 8-hour in vivo wearing test with a non-woven fabric mask. The red dashed line represents the area of the film immediately after the application. Yellow dashed square represents the noticeable peeled edge of the film without primer. (b) Average remaining rates of the film after an 8-hour wearing test with eight Asian female volunteers. The difference with and without the primer was evaluated by the paired *t*-test. (c) Adhesion force measured by a peeling test with and without the primer. The difference with and without the primer was evaluated by the Welch's *t*-test.

3.2. Evaluation of the film with the primer on the cosmetic effects

To characterise the makeup effects of the technology combined with foundation, appearance, lasting performance, and removability were evaluated, as well as its moisturising and anti-pollution effects.

3.2.1. Makeup appearance

In vivo test

The results of the questionnaire shown in Figure 3(a) indicates that the appearance became smoother by using the primer and film.

In the VISIA image in Figure 3(b), no noticeable defects were observed in the film even after applying the foundation. Furthermore, AI analysis proved that the smoothness score was higher than that of the foundation alone, especially in the high parts of the cheeks.

Next, to verify the makeup smoothness more quantitatively, photographs of 1 cm × 1 cm skin on the cheeks were analysed. Figure 3(c) shows that the SD without the primer and film was higher around the specular reflection angle, indicating that the uneven surface of the skin resulted in the unevenness of the reflected light intensity. However, it was proven that the presence of the primer and film decreased the SD from a wide range of angles of incidence with both detection angles, especially around the specular angle. This indicates that the primer and film increased the smoothness of skin, which was consistent with the results of the questionnaire and AI analysis.

Furthermore, the microscope image in Figure 3(d) shows that the foundation colourants on the film were more uniform without aggregation in the grooves and pore than in the case of the foundation alone. The results indicates that a uniform foundation distribution was the reason for the smooth appearance.

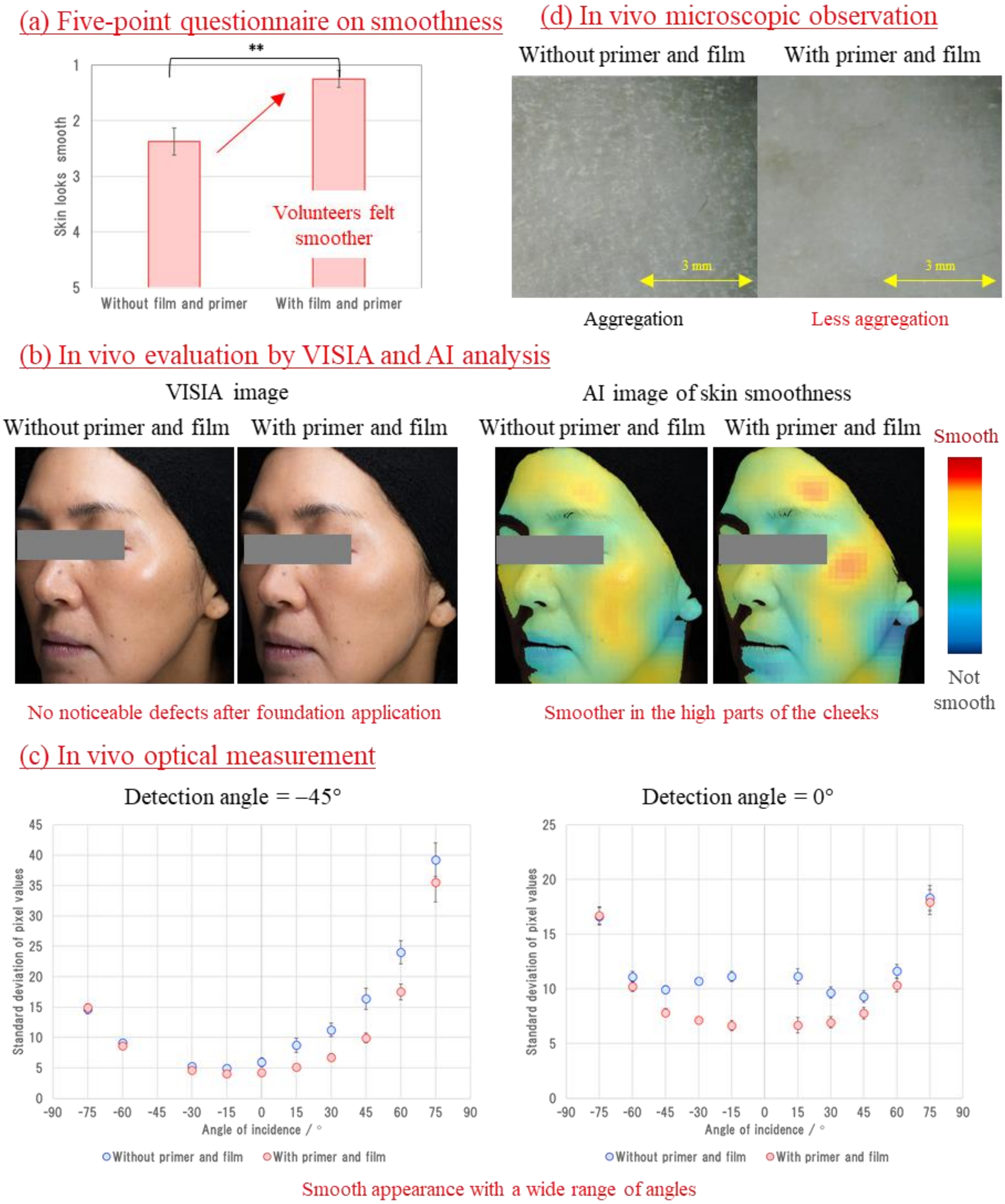

Figure 3: Evaluation of the makeup appearance of foundation with the primer and D-ES film. (a) Results of the five-point questionnaire (1: agree very much, 2: agree, 3: neither, 4: not agree, and 5: not agree at all) answered by the volunteers on the skin smoothness. The difference was evaluated by the paired *t*-test. (b) VISIA (left) and AI images of the skin smoothness (right). (c) Dependence of the skin smoothness on the angles of incidence and detection by the optical measurement. The y axis represents the standard deviation of the pixel values of the image. The difference was evaluated by the paired *t*-test. (d) Foundation colourants distribution image obtained by the microscope with and without the primer and D-ES film

3.2.2. Lasting performance of foundation

In vivo test

The questionnaire results in Figure 4(a) show that the lasting performance of the foundation was better with this technology than with the foundation alone, especially in terms of oily sheen and colour transfer to the mask.

To further investigate details of makeup smudges, photographs taken with the camera are shown in Figure 4(b). Regarding the conventional makeup, oily sheen on the side of the nose and colour transfer on the area in contact with the mask were observed. However, with this technology, colour transfer was suppressed after 8 hours, with less oily shine.

For further evaluation, VISIA images were obtained and analysed using the AI system. Figure 4(c) shows that the makeup smudge score decreased with the technology compared to the foundation alone, especially from the side of the nose to the cheek area, as well as the forehead.

Figure 4(d) and (e) show that, although colour transfer occurred slightly near the nose, the colour transfer to the mask was significantly suppressed with this technology. This result was also consistent with the result that foundation transfer was less on the face.

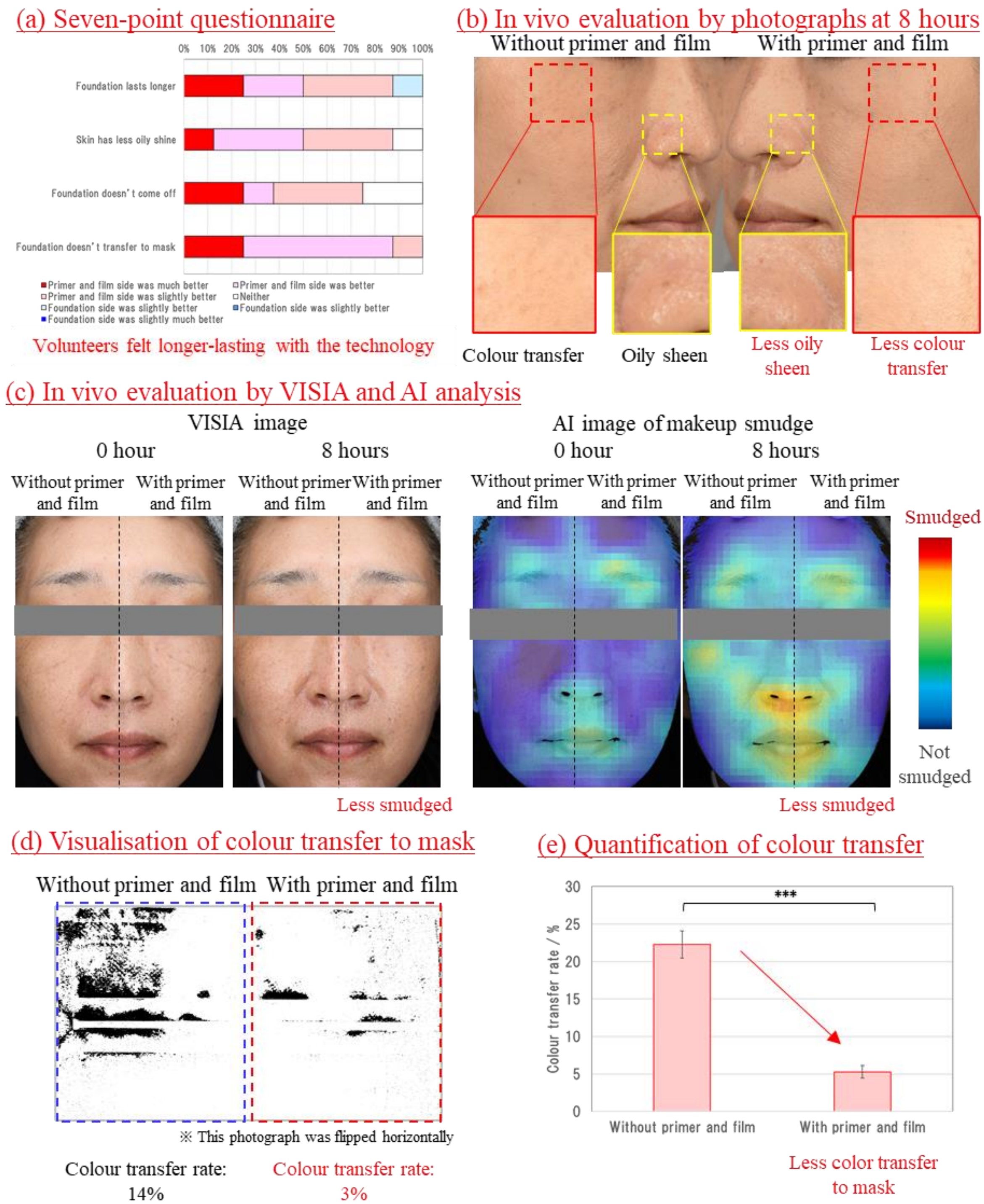

Figure 4: Evaluation of the long-lasting performance of foundation with the primer and D-ES film. (a) Results of seven-point questionnaire on the lasting performance of foundation with and without the primer and the film. (b) Photographs after spending 8 hours with a non-woven fabric mask. (c) VISIA images (left) and the makeup smudge images visualised by AI analysis (right) of foundation alone (left side of face) and the developed technology (right side of face) (d) Visualised image of foundation colourant on the mask after 8 hours. (e) Average colour transfer rate on the mask among eight Asian female volunteers. The difference with and without the primer was evaluated by the paired *t*-test.

3.2.3. Makeup removability

In vivo test

Figure 5(a) shows that almost no foundation colourant remained on the skin after peeling off the film. In contrast, when the conventional makeup was removed using a commercial makeup-remover wipe, the foundation remained in the pore. This result indicates that the technology had a higher foundation removability than the conventional makeup method.

In vitro test

To calculate the removal rate quantitatively, an in vitro experiment was conducted. Figure 5(b) and (c) show that almost all the foundation colourants were removed by peeling off the film. This result was consistent with the in vivo results.

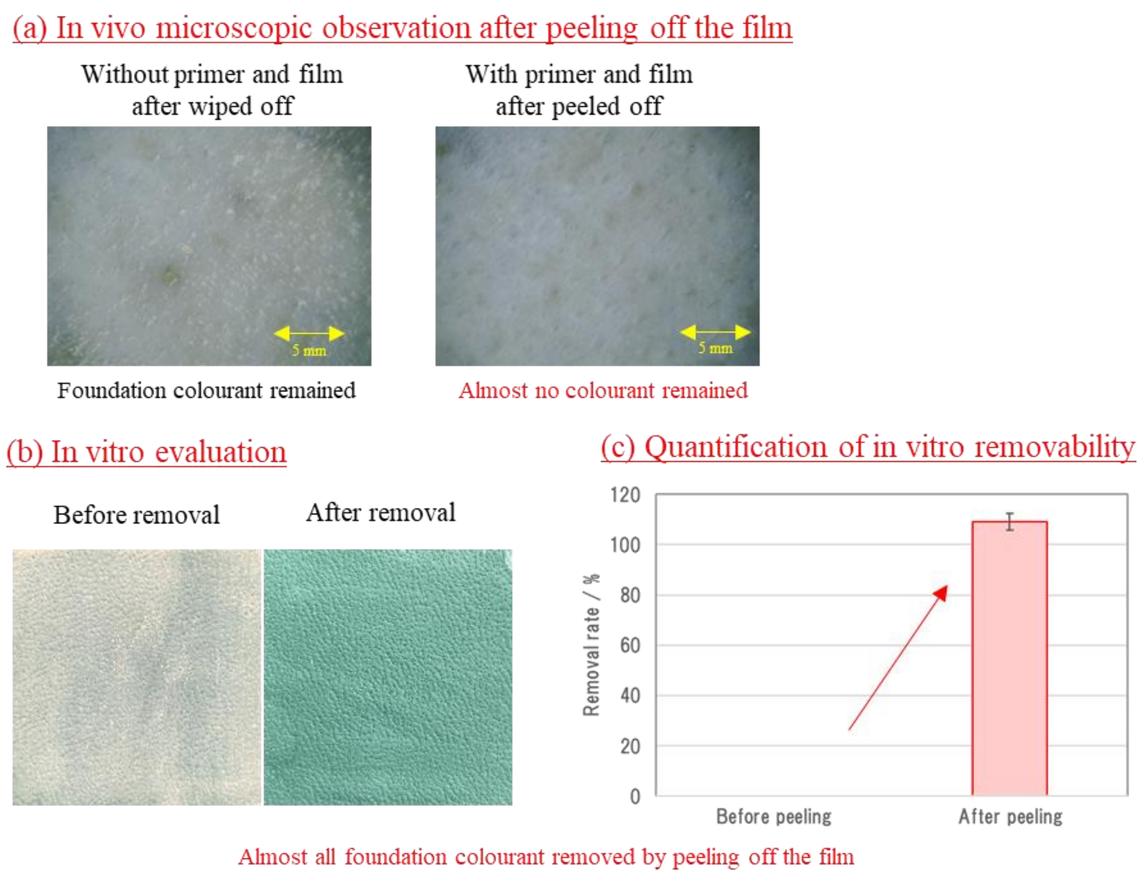

Figure 5: Evaluation of the makeup removability with the primer and D-ES film. (a) Microscopic observation on the skin. Foundation was wiped off by a commercial makeup remover sheet (left) and the foundation was removed by peeling the film at 8 hours (right). (b) In vitro evaluation of removability after peeling the film below foundation. (c) The calculated removal rate by peeling.

3.2.4. Anti-pollution effect

In vitro test

Figure 6(a) shows that the removal rate of external pollution particles was almost 100% after peeling off the film. In addition, the SEM images in Figure 6(b) reveal that pollutants were observed on the outer side, while they were not observed on the skin side. The results clarified that the film prevented external pollutants from directly touching the skin.

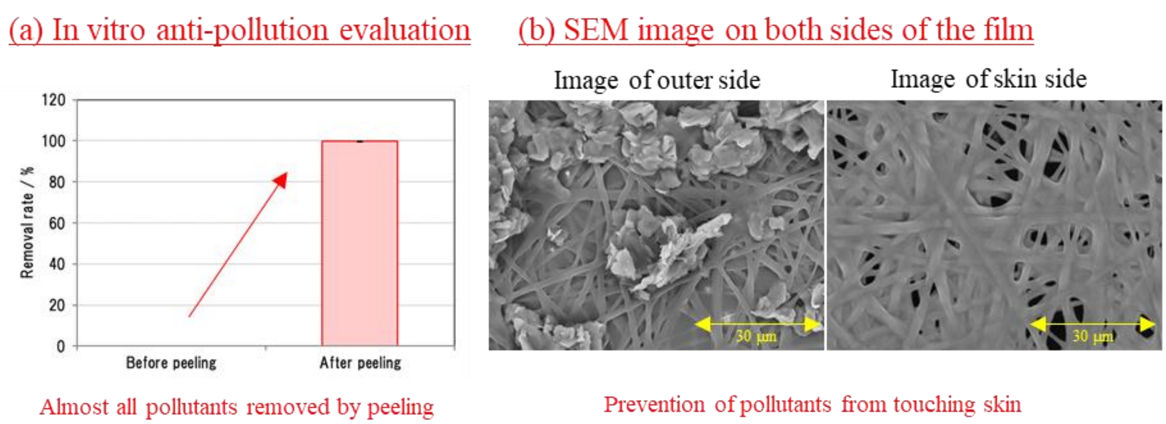

Figure 6: Evaluation of the anti-pollution effect of the primer and D-ES film. (a) In-vitro removal rate of pollutants by peeling of the film. (b) SEM image on outer side (left) and skin side (right) of the peeled film.

3.2.5. Moisturising effect

In vivo test

Figure 7 shows that the TEWL value with the primer and film was significantly decreased 30 minutes and 6 hours after application than bare skin. In other words, the primer and film protected the skin from excess water loss over time.

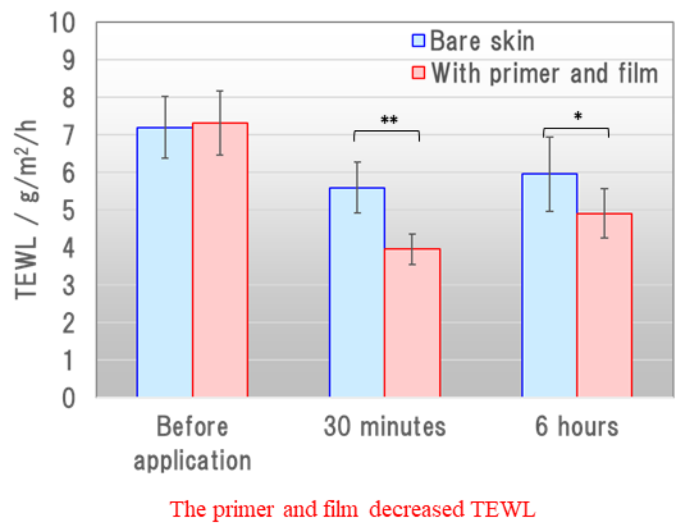

Figure 7: Evaluation of the moisturising effect of the primer and D-ES film. TEWL values measured on the forearm skin with and without the primer and film 30 minutes and 6 hours after application. The difference with and without the primer and film was evaluated by the paired *t*-test.

3.3. SEM observation to reveal the mechanisms behind the makeup performances.

To elucidate the mechanisms underlying the superior cosmetic effects, structural analysis was performed using SEM. Figure 8 shows that our novel technology had three unique structures:

The first unique structure was that the film formed a single film which bridged over a groove like a hammock. This indicates that the technology prevented the foundation particles from falling into the skin grooves and pores, resulting in a smoother appearance than the foundation alone.

The second was the structure of a film with fine fibres which retained the foundation particles inside the network. This result indicates that the network prevented colour transfer. It was also indicated that this network structure was expected to spread the sebum uniformly with a high capillary force, reducing oily sheen over time.

The third was the distribution of the foundation in the upper layer of the film. This result indicates that the foundation particles did not directly contact the skin, leading to higher removability. This structure was also considered to be the same mechanism for the anti-pollution effect.

These results indicates that superior cosmetic effects can be achieved using our technology with three unique structures.

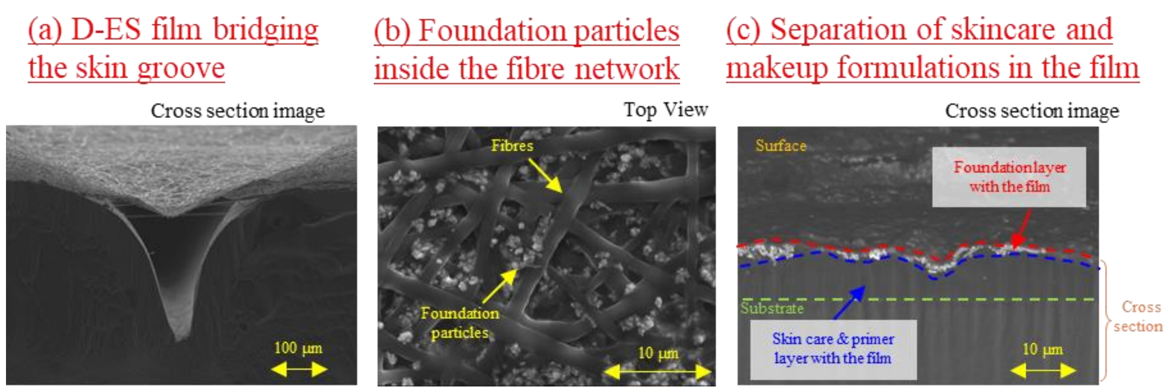

Figure 8: SEM images of the developed technology. (a) Cross section image of D-ES film bridging over the groove of an Aluminum substrate. (b) Surface image of the D-ES film with a commercial foundation on an Aluminum substrate. (c) Cross section image of a commercial lotion, the primer, the D-ES film and a commercial foundation on the polyimide substrate.

4. Discussion

4.1. Novel approaches toward expansion of second skin technology

In this study, we established a novel second skin technology that is suitable for facial makeup by preventing noticeable deterioration during application and long-term wear. This technology has brought about a new breakthrough in expanding the application area of the second skin technology as follows:

4.1.1. Unnoticeable film forming technology using D-ES

The previous second skin technologies were advanced by spreading a formulation [15] or pasting a sheet [5] to form an ultra-thin film on the skin. However, these methods tend to result in noticeable surface roughness, edges, or wrinkles as shown in Figure 1(b).

On the other hand, we have focused on a new method of electrospinning fine fibres directly onto the skin to form a uniform single film without any noticeable edges or wrinkles, even on the curved surface of facial skin. This knowledge is important for the applicability of second skin technology to other complex body parts such as elbow and knees where skin troubles such as atopic skin tend to occur [16].

4.1.2. Two-step application for sufficient durability

Previous second skin technologies have not used different approaches for film formation and adhesion. However, we applied an adhesive primer as a separate formulation, resulting in sufficient durability for practical use.

When we try to expand the second skin technology to other body parts, the required adhesion force to achieve sufficient durability of an ultra-thin film may depend on the body parts. To control the adhesion force for each body part, separating the forming and adhesion functions of the film can be an efficient approach.

4.2. The dawn of a new era of superior cosmetic effects

Next, we clarified the superior makeup effects, such as smoothness, lasting performance, and removability of the novel makeup technology, compared to conventional cosmetics, in addition to its anti-pollution and moisturising effects. These results can lead to a paradigm shift in cosmetic effects by overcoming the various limitations of the viewing angle, type of products, external friction, location, and combination of other cosmetics, as follows:

4.2.1. Enhancement of smoothness of makeup from anywhere with any product type

The results revealed that the conventional foundation had a large angle dependence on smoothness, but our technology had less angle dependence, indicating a smoother appearance from any viewing angle.

Furthermore, the results revealed that this technology upgraded the smoothness of the combined commercial foundation. In other words, this approach can be combined with customers' favourite cosmetics to improve smoothness. Moreover, this technology can be applicable not only to foundations but also to other makeup products, such as eye makeup, which are applied to areas where wrinkles are often prominent.

4.2.2. Enhancement of lasting property of makeup against external friction

Owing to the increased wearing rate of masks to prevent COVID-19, colour transfer to a mask has become a significant issue [17]. The results prove that this technology improved the lasting performance of the combined foundation. This result indicates that this makeup technology can also expand the application area to body areas with bruised and uneven skin tones, by reducing colour transfer to clothing.

4.2.3. Makeup removal in any circumstances

In conventional cosmetics, it is common to remove foundations using makeup removers, and the development of various cleansing agents has been promoted. However, our technology made it possible to remove the colourants of the foundation from skin by peeling off the film. The removability of the other components in the foundation needs further confirmation, but the makeup can be removed without a makeup remover or water, indicating easy removal regardless of the situation and location.

4.2.4. Expansion of anti-pollution technology with any cosmetics

A major approach to the anti-pollution effect is to control the repulsive force between the skin and external particles [18], which may be affected by other makeup applied on top. However, our results revealed that the film prevented external pollutants from directly touching the skin. This indicates that the anti-pollution effect can be achievable even with other products.

4.3. Evolution of second skin film to mimic real skin

The SEM observation revealed the following three unique structures of the obtained film: (1) a bridge structure that covered pores and grooves, (2) a network structure in which fibres retained the foundation particles, and (3) a layered structure in which skincare and makeup formulations existed separately inside the film. Through the elucidation of the mechanisms, we have brought a new perspective on controlling cosmetic films to mimic real skin from the following perspectives:

4.3.1. Precision control of uneven surface structure

The developed film achieved smoothness by bridging the roughness of the skin like a hammock, which is not achievable by conventional cosmetic products. Furthermore, our technology reduced the roughness, not making it completely flat, while pasting a ready-made film may form a completely flat structure. This result indicates that this technology can not only reduce the surface roughness but also maintain the skin aesthetics caused by roughness.

4.3.2. Pigment distribution control using the internal network structure

The results proved that the foundation particles were retained in the fine fibre network. This means that the colourants were present inside the translucent film, similar to real skin in which the coloured melanin is inside the translucent epidermis. These results also indicate that this technology can maintain the skin aesthetics caused by the internal structure.

4.3.3. Amalgamation of skincare and makeup through a layered structure and beyond

The results showed that the film held skincare and makeup formulations separately inside itself, which is difficult to be achieved by conventional cosmetics. This means that skincare and makeup formulations can fulfil their effects without disturbing each other.

This may expand the scope of cosmetic research. Since the electrospinning technology is expected to have therapeutic effects on atopic skin [5] and wounds [19], our technology can combine a medical formulation under the film to cover them with makeup without disturbing the medical effect.

We believe that the developed technology will lead to further evolution of the second skin technology to improve skin function.

5. Conclusion

We developed a novel second skin technology to address the issues of film deterioration, by combining the D-ES method with the adhesive primer formulation. This technology achieved no noticeable defects during application and sufficient durability against the mask. In addition, this technology has also achieved superior smooth appearance, lasting performance, and removability of makeup, as well as moisturising and anti-pollution effects. SEM observations clarified (a) the structure that bridged the pores and skin grooves, (b) the network structure in which the fibre retained the foundation particles, and (c) the structure in which the skincare and makeup formulations existed separately inside the film. These three unique structures may be the reason for their superior cosmetic effects.

6. Acknowledgments

Authors appreciate Tatsuya Yoshino for the development of the D-ES polymer solution, Tomomi Hagio and Muneki Sakata for the development of the primer, Asuka Imai for fruitful discussion, and Keiko Yoda, Koko Higashio and Haruka Nakashima for data acquisition.

7. Conflict of Interest Statement

NONE.

8. References

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