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Hygroscopicity and water-holding capacity of moisturizing agents: A single-application *in vivo* study

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Synopsis

A comparative study of four kinds of moisturizers for their hygroscopicity and water-holding capacities as measured by a skin surface hygrometer showed that all agents could improve the water content of the skin surface. The most effective agent was Eucerit, a lanolin alcohol-containing agent, and the second was a urea-containing agent. The Eucerit-containing agent also showed high stability according to the water-holding capacity patterns observed after five minutes and three hours.

INTRODUCTION

The water content of the stratum corneum plays an important role in providing the skin surface with suppleness, good function, pliability, and smoothness (1). This thin layer serves as a protective sheath that protects the body from the invasion of various kinds of external attacks (2) such as living organisms, chemical agents, and physical factors. There is a big difference between the moist and humid environment beneath the stratum corneum and the dry atmosphere outside the body, and the stratum corneum is the only layer between them. *In vitro* studies on stratum corneum showed that it is flexible as long as it contains more than 10% water (3). The water-holding property of the stratum corneum is influenced by water-soluble materials, such as free amino acids, organic acids, urea, and inorganic ions (4). Lipids in the lamellar structure in the intercellular spaces of the stratum corneum also play a part in water-holding (5,6).

Smooth and soft skin is not only good-looking but is also healthy. On the other hand, dry, scaly, or rough skin shows an unhealthy condition or underlying disease (7) and reflects low water content in the stratum corneum (3). People accept these concepts and try to use agents to improve water content and smooth their skin. There are many agents on the market that claim to smooth the skin and hold water on the skin surface. A variety of techniques for assessing skin hydration have been developed and reviewed (7–11). Most *in vivo* techniques are based on electrical measurements such as resistance, capacitance, impedance and, conductance. We try to evaluate the moisturizing agents in three aspects: 1) water content of the skin surface before and after application of moisturizers; 2) hygroscopicity, maximum water-holding ability of particular agents after hydrating the skin surface; and 3) water-holding capacity, an ability to hold water on the skin surface after a single application of water by measuring the electrical conductance that should correlate with the amount of water on the skin surface.

In this study, we used moisturizers available at the Institute of Dermatology, Bangkok, Thailand. The agents are cream base, 10% urea cream, 5% lactic acid cream, and pH5-Eucerin lotion, a commercial moisturizer, which contained Eucerit or lanolin alcohol.

MATERIALS AND METHODS

INSTRUMENTS

This study used a skin surface hygrometer model Skicon 200 (IBS Inc., Hamamatsu, Japan), which reads the conductance in terms of μS (microsiemen), which is the reciprocal value of an ohm (Ω). $1 \text{ S} = 1\ \Omega^{-1}$.

In our procedure, we measured the conductance of the skin surface in three parts (Figure 1). First we measured the water content of the skin, before water application. This parameter was called the prehydration state or baseline level. Then we applied water on the skin for ten seconds, wiped it with tissue, and immediately measured the conductance of the skin again. This parameter relates to the maximum water content that the skin can hold, called hygroscopicity. Consequently, we measured the conductance of the skin at 30-second intervals for three minutes. The pattern of conductance was called

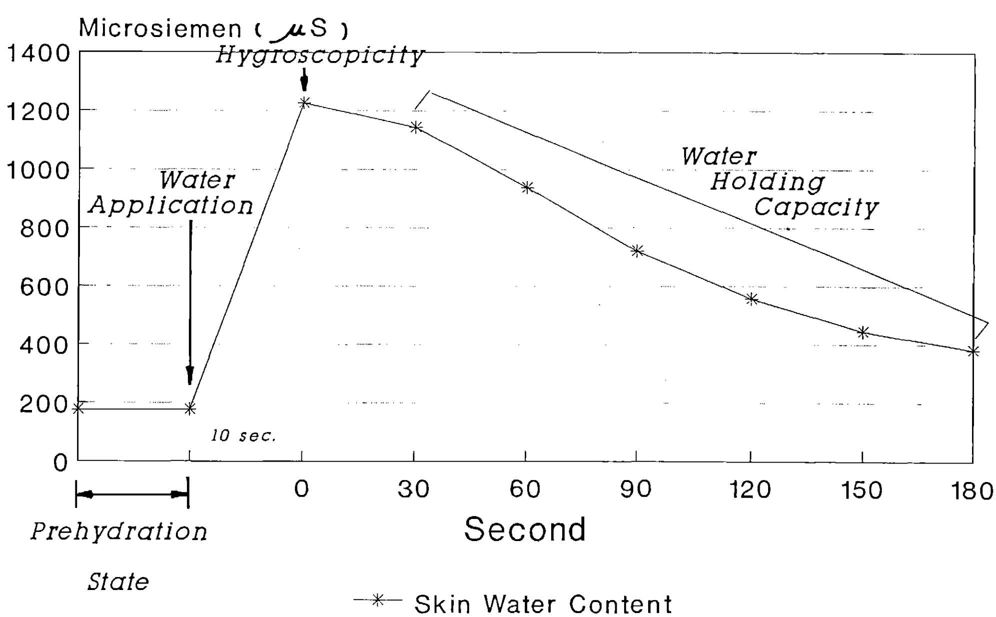

Figure 1. Standard pattern measured by Skicon 200.

water-holding capacity. This value shows the ability of the skin to hold water in each time period.

To obtain reliable results, we used the same probe and same examiner throughout the study. The examiner held the flexible cable that connects the probe with the machine, by the fingers, at about 10 cm distance from the probe, which was gently lowered on a test area to rest with its own weight (80 gm) on the skin without applying any additional manual pressure that could affect the result (7).

SUBJECTS

One hundred and eleven normal hospital personnel, from the Institute of Dermatology, Bangkok, male and female, ages 18–45 years, took part in this study.

TEST AGENTS

1. Cream base (Institute formula)
   Formulation: Stearyl alcohol 7.00%
   Cetyl alcohol 2.67%
   Sodium lauryl sulfate 0.5%
   Liquid paraffin 18.0%
   Propylene glycol 6.67%
   Methyl paraben 0.02%
   Propyl paraben 0.0006%
2. 10% Urea cream (Institute formula)
   Formulation: Urea 10% W/W
   Cream base
3. 5% Lactic acid cream (Institute formula)
   Formulation: Lactic acid 5% W/W
   Cream base
4. pH5-Eucerin lotion
   Formulation: Ammonium dihydrogen citrate 0.218 gm, ammonium monohydrogen citrate 0.382 gm, in an oil-in-water emulsion containing Eucerit (lanolin alcohol).

METHODS

In the examining room, a temperature of 24–26°C and a relative humidity of 45% was controlled throughout the study. To avoid sweating, subjects were asked to wait for at least five minutes before starting the examination. (The study was conducted in July 1991, and the average temperature was about 34°C in Bangkok; after waiting, the subjects would feel cool and not sweat in the examining room.) The test procedure was divided into two parts:

1. Prehydration state, hygroscopicity, and water-holding capacity of normal skin. We established a base level, or prehydration level, of the water content of the skin surface at the flexor surface of both forearms, 5 cm below the antecubital fossa, by taking three recordings and using an average result. After drops of water were applied to the examination site for ten seconds and wiped off by tissue paper, conductance was measured immediately after wiping, giving the maximum water content or hygroscopicity. Then the measurement was repeated at intervals of 30 seconds for three minutes, measuring water-holding capacity (7).

2. *Prehydration state, hygroscopicity, and water-holding capacity on moisturizer-applied skin (immediate and delayed observation).* We selected two square areas, 5×5 cm^2 each, at the middle part of both forearms; the upper line of the square was 5 cm below the antecubital fossa. Using the syringe, we applied 10% urea cream on the first square of the left forearm, 2 μl/cm^2, and spread it evenly by a glass rod. After waiting five minutes, we observed an immediate result by measuring the water content of the prehydration state (baseline). Then we applied water for ten seconds and removed it carefully, trying not to remove the agent. Then hygroscopicity and water-holding capacity were measured in the same manner as in the first test procedure. The same method was used to measure other agents: cream base was applied on another square area of the left forearm, and 5% lactic acid cream and pH5-Eucerin were applied on square areas of the right forearm. Prehydration state, hygroscopicity, and water-holding capacity were measured in the same manner (Figure 2). After that, we observed the delayed result by measuring the prehydration state, hygroscopicity, and water-holding capacity of all four agents three hours after application.

STATISTICS

The Kruskal-Wallis test and least significant difference (LSD) analysis were used to compare the results and analyze data.

RESULTS

There were 109 volunteers, 24 males and 85 females, ages 20–44 (average age, 30.96), who completed the experiment. Two women were excluded from the test because they were pregnant and hormonal changes might have interfered with the result.

WATER CONTENT OF NORMAL SKIN

We found that the prehydration state, hygroscopicity, and water-holding capacity of the normal skin of both forearms were approximately the same and did not have any statistical difference. The water content of the skin increased sharply after water appli-

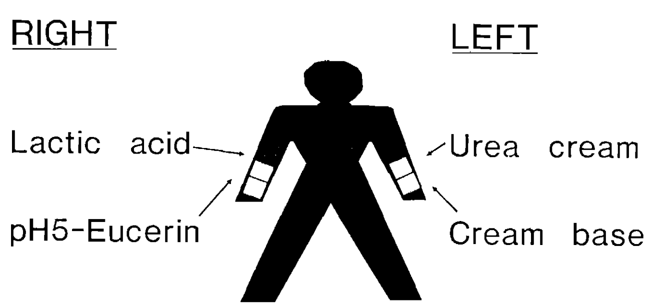

Figure 2. Site of application of testing agents.

cation, showing high hygroscopicity. Most of this increase was lost within 30 seconds, followed by a gradual return to a prehydration level in 180 seconds (Figure 3).

WATER CONTENT OF THE SKIN SURFACE AFTER APPLICATION OF MOISTURIZERS

From statistical analysis, we found that the distribution pattern of the data was not in a normal distribution. Therefore, the analysis system that we used to analyze the data is a non-parametric test (Kruskal-Wallis test) to show the differences between the groups.

From the Kruskal-Wallis test it was shown that the differences between the groups had a statistical significance of $P < 0.00001$. Although the Kruskal-Wallis test can test the hypothesis that there is at least one group that differs from the others, it is unable to determine which specific group has a real difference in relation to another. Therefore, the least significant difference (LSD) analysis can be useful to determine comparatively how different each group is from another, or, in other words, we can rank the groups according to the value of interest by comparing the value of one group to that of another pair, by pairs, until every pair is analyzed.

We then used a numeric number to replace the testing agents:

1 = normal skin as a control
2 = 10% urea cream
3 = cream base
4 = 5% lactic acid cream
5 = pH5-Eucerin

Five minutes after the application of agents we used the least significant difference (LSD)

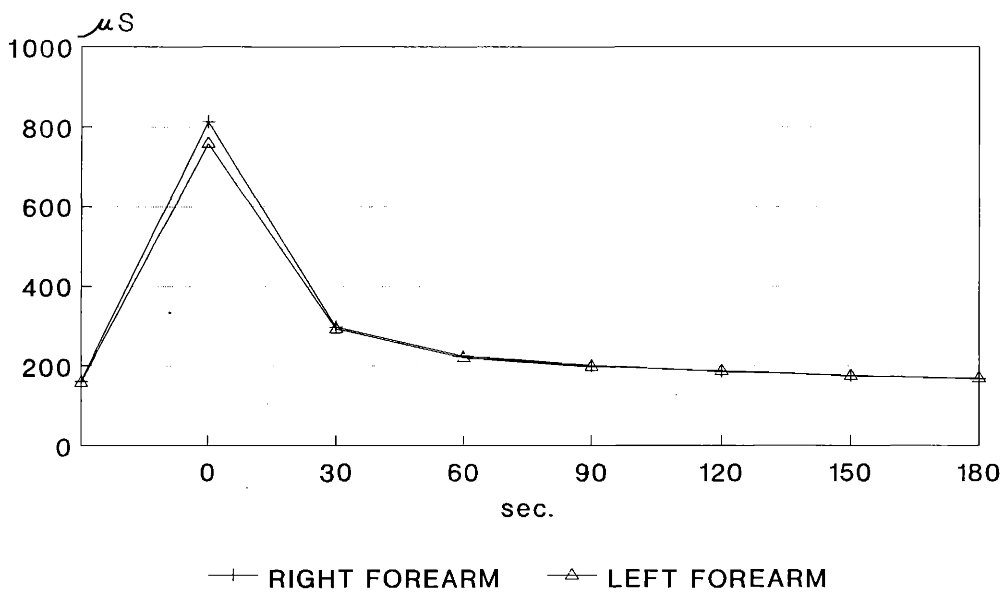

Figure 3. Hygroscopicity and water-holding capacity pattern of pretreated normal skin, right and left forearm.

analysis in order to rank and compare the results between the group at level P = 0.05 (Figure 4).

Three hours after the application of agents we also used the least significant difference (LSD) analysis in the same manner at level P = 0.05 (Figure 5).

We found that conductance of the prehydration state of normal skin is about 180 μS. After hydrating, normal skin showed high conductance around 800 μS, but it declined rapidly within 30 seconds and returned to the prehydratic level in 180 seconds. This result implies that the normal skin surface has an ability to absorb water but cannot retain it for long, or that it has a low water-holding capacity (Figure 3).

Observation at five minutes after application of the agents (Figure 4) showed that the baseline water content of all agents was better than with control normal skin. PH5-Eucerin and 10% urea cream had the highest level of water content among the agents, followed by 5% lactic acid cream and cream base. After applying drops of water on the testing areas and wiping them off in ten seconds, pH5-Eucerin obtained the highest hygroscopicity, followed by 10% urea cream, 5% lactic acid cream, and cream base that had the same level of hygroscopicity. The water-holding capacity of all agents declined in a slower manner than with control normal skin, and at 180 seconds the water content of 10% urea cream, pH5-Eucerin, and 5% lactic acid cream was more than the cream base and control normal skin levels. The water-holding capacity of 10% urea cream, 5% lactic acid cream, and cream base declined in a slower manner than in PH5-Eucerin, and 10% urea cream was the slowest.

From observation three hours after application (Figure 5), we found that at the prehydration state, pH5-Eucerin had the highest water content among all agents and that every agent had a lower water content when compared to the same agent five minutes after application. The water content levels of 5% lactic acid cream, 10% urea cream,

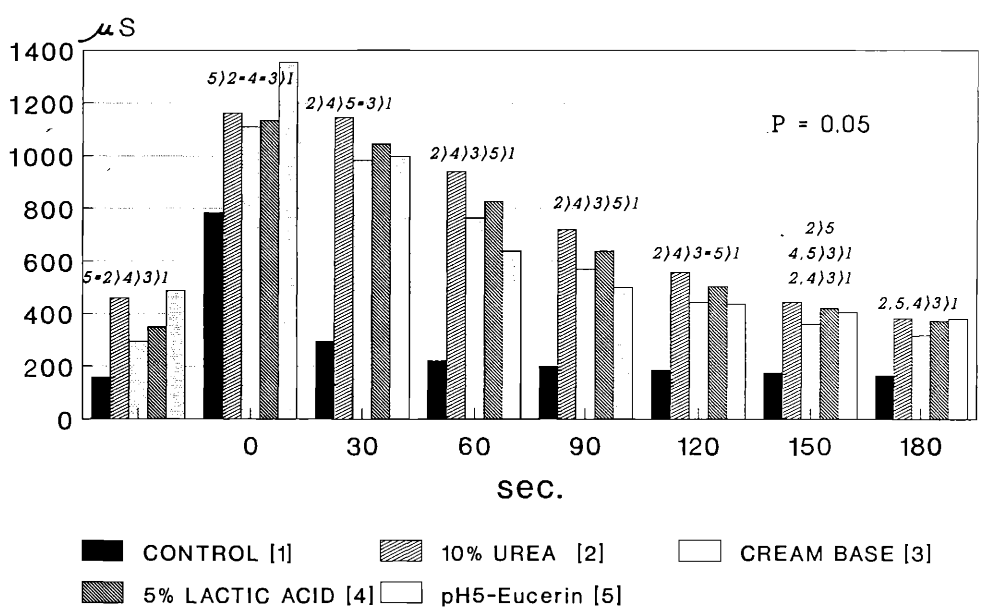

Figure 4. Hygroscopicity and water-holding capacity pattern of observation five minutes after application.

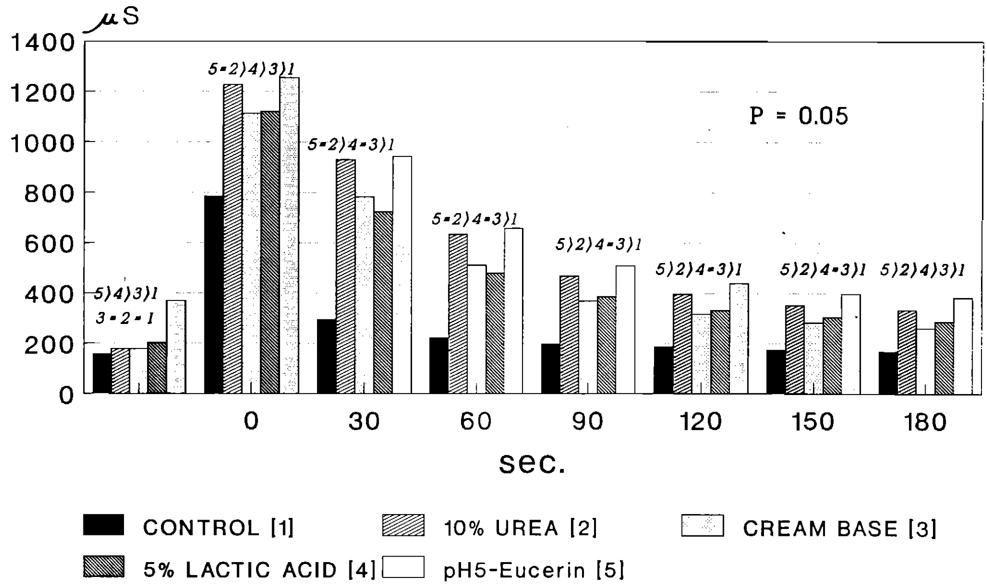

Figure 5. Hygroscopicity and water-holding capacity pattern of observation three hours after application.

cream base, and control normal skin were nearly the same at the prehydration state. After applying and wiping off water, pH5-Eucerin and 10% urea cream both showed the highest hygroscopicity, and both of them showed the same water content level at the beginning of the water-holding capacity period. But after one minute, pH5-Eucerin obtained higher efficacy than 10% urea cream (Figure 6). The water-holding capacity of all agents dropped faster than at five minutes after application (Figure 7), except for

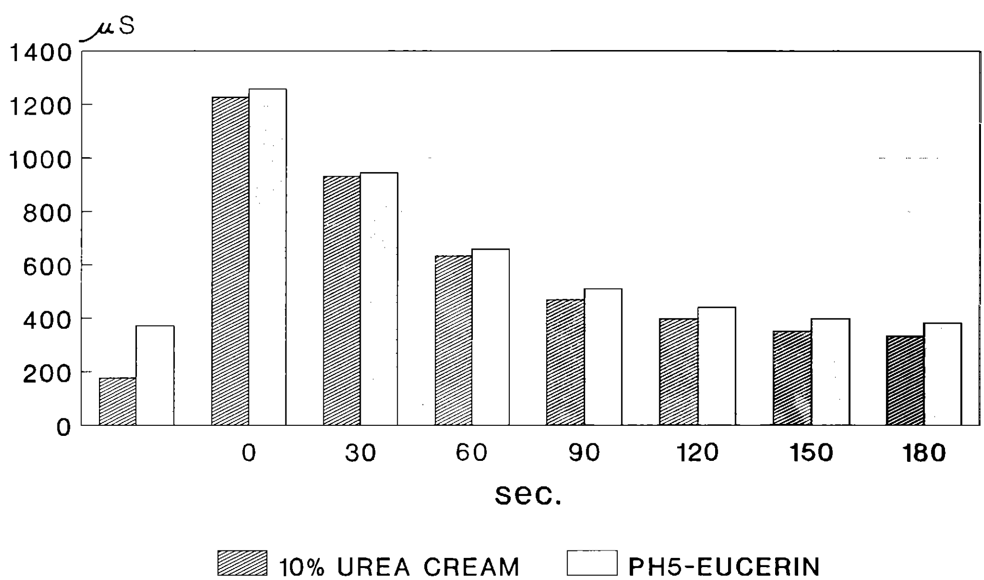

Figure 6. Comparison of results of 10% urea cream and pH5-Eucerin at observation three hours after application.

pH5-Eucerin, which retained the ability to hold water at nearly the same level. At 180 seconds, all agents still had water content levels higher than the normal skin control (Figure 5).

DISCUSSION

The water content of stratum corneum in our test is chiefly concerned with the amount of the rapidly gained and lost "bound water" in stratum corneum (12). The baseline conductivity of normal skin is around 180 μS, which is higher than in other reports (7). This could be due to the climate in Thailand, which is warm and humid. Normal skin has a high hygroscopicity but a low water-holding capacity, and water content drops sharply in 30–60 seconds.

This experiment, like previous reports (13–18), showed that moisturizers can increase the water content of the prehydration state and improve water-holding capacity. Comparing water content at the prehydration state at five minutes and three hours after application of moisturizers, we found that the water content of the latter is less than that of the former. There are many reasons to explain this event: perhaps the properties of the agents are not stable due to degradation, or when we wiped off water, part of the agents may have also been removed. pH5-Eucerin showed the highest water content, meaning that this agent is more stable and longer-lasting. We were disappointed that we could not continue the experiment longer than three hours because the volunteers were not available. This meant that we could not establish when the water content of all treatments dropped to the level of normal skin.

Among these agents, pH5-Eucerin obtained the highest hygroscopicity, both at five

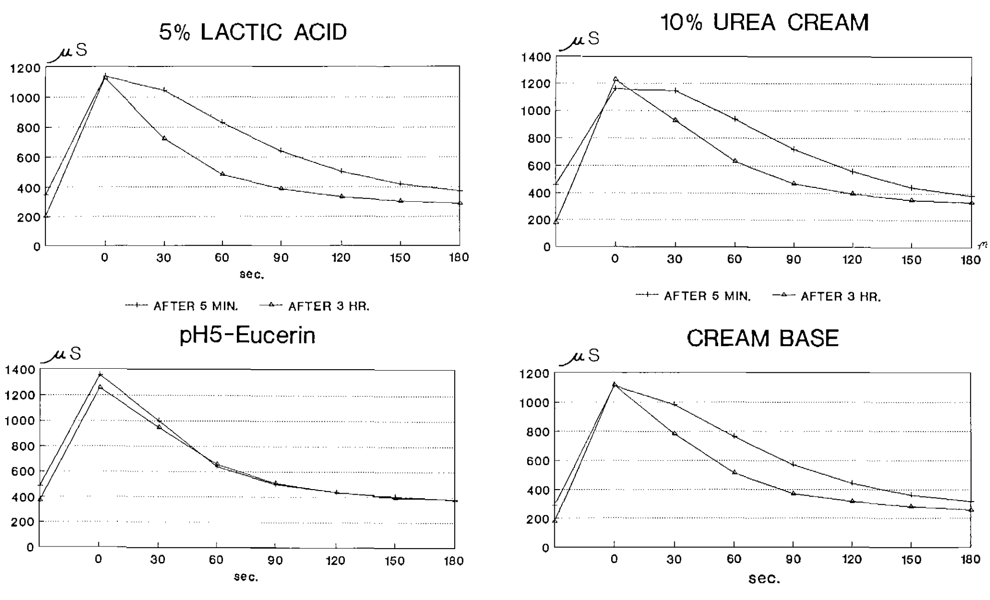

Figure 7. Hygroscopicity and water-holding capacity of all agents, comparing observations five minutes and three hours after application.

minutes and three hours after application of agents, and 10% urea cream was second in rank. For the water-holding capacity aspect, at five minutes after application 10% urea cream was the most efficient agent. It sustained a high water content, which declined slowly. At the same time, pH5-Eucerin seemed to drop faster than the rest, but at the end there were no differences among all agents in the water-content aspect. When compared to three hours after application, the water-holding capacity of pH5-Eucerin declined at nearly the same rate after five minutes observation. On the other hand, other agents showed lower water-holding capacity than pH5-Eucerin.

We concluded that pH5-Eucerin is more effective than other agents because

1. The conductance of the prehydration state of pH5-Eucerin is higher than others at observation both five minutes and three hours after application. This reflects the higher water content on the skin surface.
2. pH5-Eucerin can obtain highest hygroscopicity at observation both five minutes and three hours after application.
3. For the water-holding capacity aspect, pH5-Eucerin obtains nearly the same rate of decrement at observation five minutes and three hours after application. This means that pH5-Eucerin still has high stability after three hours application.

Among the institute formulas, 10% urea cream was the most effective agent, but urea is one of the popular agents used in commercial preparation, and we feel that it is worthwhile to prescribe it for patients with pathological dry skin conditions (19).

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