

Wash or wipe? A comparative study of skin physiological changes between water washing and wiping after skin cleaning

著者	Ogai Kazuhiro, Matsumoto Masaru, Aoki Miku, Ota Reina, Hashimoto Kanae, Wada Risa, Kobayashi Masato, Sugama Junko
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Complete List of Authors:	Ogai, Kazuhiro; Kanazawa University, Wellness Promotion Science Center Matsumoto, Masaru; Kanazawa University, Faculty of Health Sciences, Institute of Medical, Pharmaceutical and Health Sciences; Kanazawa University, Wellness Promotion Science Center Aoki, Miku; Kanazawa University, Department of Clinical Nursing, Division of Health Sciences, Graduate School of Medical Sciences; Kanazawa University, Wellness Promotion Science Center Ota, Reina; Kanazawa University, Department of Nursing, School of Health Sciences, College of Medical, Pharmaceutical and Health Sciences Hashimoto, Kanae; Kanazawa University, Department of Nursing, School of Health Sciences, College of Medical, Pharmaceutical and Health Sciences Wada, Risa; Kanazawa University, Department of Nursing, School of Health Sciences, College of Medical, Pharmaceutical and Health Sciences Kobayashi, Masato; Kanazawa University, Wellness Promotion Science Center Sugama, Junko; Kanazawa University, Advanced Health Care Science Research Unit, Innovative Integrated Bio-Research Core, Institute for Frontier Science Initiative (InFiniti); Kanazawa University, Faculty of Health Sciences, Institute of Medical, Pharmaceutical and Health Sciences; Kanazawa University, Wellness Promotion Science Center
Keywords:	skin physiological function, transepidermal water loss, skin hydration, skin pH, ceramide, cleaning agent, water washing, wiping

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5 **1 Wash or wipe? A comparative study of skin physiological changes between water washing**
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7 **2 and wiping after skin cleaning**
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11 4 Kazuhiro Ogai^{1,*}, Masaru Matsumoto^{1,2}, Miku Aoki^{1,3}, Reina Ota^{4,§}, Kanae Hashimoto^{4,§},
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13 5 Risa Wada^{4,§}, Masato Kobayashi¹, and Junko Sugama^{1,2,5}
14
15

16
17 7 ¹Wellness Promotion Science Center, Institute of Medical, Pharmaceutical and Health Sciences,
18
19 8 Kanazawa University, 5-11-80 Kodatsuno, Kanazawa, Ishikawa 9200942, Japan
20

21 9 ²Faculty of Health Sciences, Institute of Medical, Pharmaceutical and Health Sciences,
22
23 10 Kanazawa University, 5-11-80 Kodatsuno, Kanazawa, Ishikawa 9200942, Japan
24

25 11 ³Department of Clinical Nursing, Division of Health Sciences, Graduate School of Medical
26
27 12 Sciences, Kanazawa University, 5-11-80 Kodatsuno, Kanazawa, Ishikawa 9200942, Japan
28

29 13 ⁴Department of Nursing, School of Health Sciences, College of Medical, Pharmaceutical and
30
31 14 Health Sciences, Kanazawa University, 5-11-80 Kodatsuno, Kanazawa, Ishikawa 9200942,
32
33 15 Japan
34

35 16 ⁵Advanced Health Care Science Research Unit, Innovative Integrated Bio-Research Core,
36
37 17 Institute for Frontier Science Initiative (InFiniti), Kanazawa University, 5-11-80 Kodatsuno,
38
39 18 Kanazawa, Ishikawa 9200942, Japan
40

41 19 [§]These authors equally contributed to this work.
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44

45 21 * All correspondence should be addressed to:
46

47 22 Kazuhiro Ogai
48

49 23 Wellness Promotion Science Center, Institute of Medical, Pharmaceutical and Health Sciences,
50
51 24 Kanazawa University, 5-11-80 Kodatsuno, Kanazawa, Ishikawa 9200942, Japan
52

53 25 Tel.: +81-76-265-2590
54

55 26 E-mail: kazuhiro@staff.kanazawa-u.ac.jp
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5 **Abstract**
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7 **Background/purpose:** Presently, skin-cleaning agents that claim to be removed by water or
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9 wiping alone are commercially available and have been used for the purpose of bed baths.
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11 However, there is a lack of knowledge on how water washing and wiping differently affect skin
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13 physiological functions or ceramide content. The aim of this study was to compare the effects of
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15 water washing and wiping on skin physiological functions and ceramide content.
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17 **Methods:** Three kinds of the cleaning agents with different removal techniques (i.e., water
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19 washing and wiping) were used in this study. Skin physiological functions (i.e., transepidermal
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21 water loss, skin hydration, and skin pH) and skin ceramide content were measured before and
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23 after seven consecutive days of the application of each cleaning agent.
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25 **Results:** No significant differences in skin physiological functions or ceramide content were
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27 observed between water washing and wiping.
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29 **Conclusion:** Cleaning agents that claim to be removed by water washing or wiping do not affect
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31 skin physiological functions or ceramide content by either removal method.
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38 **Running Head**

39 How does washing or wiping affect skin after cleaning?
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43 **Key words**

44 skin physiological function; transepidermal water loss; skin hydration; skin pH; ceramide;
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46 cleaning agent; water washing; wiping
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49 **1. Introduction**

50 Bathing is one of the most fundamental activities to keep our body clean. It usually involves
51 washing away dirt and sweat from the skin with lukewarm water, cleansing oily dirt with soap
52 or a cleaning agent, rinsing the soap/cleaning agent off, and drying. These processes are not
53 only a cleaning procedure but also have beneficial effects on the prevention of several kinds of
54 diseases (1, 2) and the eradication of bacterial skin colonization (3, 4).

55 Bed baths are a type of bathing technique in which the caregiver wipes the patient's
56 skin by means of a soft, wet towel with or without soap. This technique is particularly effective
57 for patients who are bedridden and/or receiving home-care services. When soap is used, the
58 caregiver is required to purge the remaining soap from the skin by water washing. Because soap
59 remnants may irritate the skin (5) and cause deterioration in skin physiology (6), plenty of water
60 and a basin are required to remove remnant soap on the skin. It is, however, a burdensome task
61 for caregivers, particularly at the bedside and at home (7). In addition, such water washing is not
62 available when water usage is restricted, for example, after natural disasters.

63 As an alternative to the traditional bed bath with soap and water, cleaning agents that
64 can be removed just by wiping and without water have been introduced. Such
65 "removable-by-wiping" cleansing is considered to be comparable to the traditional
66 soap-and-water bed bath (8), less invasive (9), and more cost effective (10). Because wiping can
67 cleanse the skin as well as water washing can, there is a growing acceptance of
68 "removable-by-wiping" cleaning agents in both clinical and home-care settings (8, 10).

69 Although such "removable-by-wiping" agents claim that both water washing and
70 wiping can be interchangeably used for removal, one question remains: how do the different
71 removal techniques (i.e., water washing and wiping) affect skin physiology? Considering the
72 fact that wiping cannot completely remove soap on the skin (11) and that residual soap irritates
73 and causes skin deterioration (5, 6), we need to know whether wiping alone can maintain skin
74 integrity comparable to water washing.

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5 75 Furthermore, because the cleaning agents are amphiphilic (i.e., have an affinity for
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7 76 both water and lipids), skin lipids, particularly ceramides, may be ablated during wiping.
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9 77 Ceramides are the main components of lipids in the stratum corneum (SC) and are responsible
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11 78 for skin barrier function. Depletion of ceramides leads to skin dryness (12). Therefore, we also
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13 79 need to know how wiping without water affects skin ceramide retention.
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15 80 To address these questions, the aim of this study was to reveal the effect of different
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17 81 removal techniques (i.e., water washing and wiping) on skin physiological functions [e.g., skin
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19 82 pH, hydration, and transepidermal water loss (TEWL)] and ceramide content in the SC.
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23 84 **2. Materials and methods**

24 85 *2.1. Ethical consideration*

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27 86 This study was approved by the Medical Ethics Committee of Kanazawa University (approval
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29 87 number: HS27-9-1) and was performed in accordance with the Declaration of Helsinki.
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32 89 *2.2. Cleaning agents*

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35 90 Three kinds of cleaning agents, A, B, and C, were chosen based on their market share and
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37 91 availability in Japan. Since the aim of this study was not product evaluation, the name and
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39 92 manufacturer of each agent are withheld. According to each manufacturer's instructions, all of
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41 93 the cleaning agents could be removed by both water washing and wiping. Each of the cleaning
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43 94 agents had different properties; agent A was a creamy foam (Fig. 1A), agent B was an airy foam
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45 95 (Fig. 1B), and agent C was a cream (Fig. 1C). The contents of each cleaning agent are shown in
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47 96 Table 1.
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50 98 *2.3. Participants and allocation*

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53 99 In this study, 15 healthy participants (age: 21–22 years) were recruited and provided written
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55 100 informed consent. The participants were randomly divided into three groups corresponding to
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5 101 the three cleaning agents (see Section 2.2) with five participants to each agent.
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9 103 *2.4. Experimental setup*

10 104 The experimental setup of cleaning/removal was as follows (Fig. 2). First, the washing area was
11 105 defined as a circle with a radius of 3.5 cm, centered 10 cm distal to the cubital fossa (red circle,
12 106 Fig. 2A). The trained researcher applied the “protective cleaning method” [Fig. 2B, (13)] on
13 107 both forearms of each participant for 30 s. The amount of each cleaning agent was as follows: 2
14 108 mL for agents A and B, and 0.6 g for agent C.
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21 109 After the washing procedure, the cleaning agent on the right arm was removed by
22 110 applying one liter of lukewarm water ($40 \pm 1^\circ\text{C}$) to the washed region in 20 s, followed by
23 111 gentle tapping (not wiping) five times with a gauze (Cueb CARE Gauze; Koshiya Medical Care
24 112 Corp., Ishikawa, Japan). The cleaning agent on the left arm was wiped off five times in a
25 113 clockwise direction by means of a gauze. In both methods, the gauze was changed with a new
26 114 one for every tap or wipe.
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33 115 After removal of the cleaning agent, the washed regions were covered with gauze
34 116 and a film dressing (AIRWALL Fuwari; Kyowa Ltd., Osaka, Japan) to avoid external
35 117 disturbance. In addition, the participants were requested not to dip the washed regions into
36 118 water (e.g., bathing, washing dishes, etc.). The overall cleaning/removal procedure was
37 119 performed once a day for seven consecutive days by the trained researcher (Fig. 2C).
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46 121 *2.5. Measurement of TEWL, skin hydration, and skin pH*

47 122 To test the differences of skin physiological changes between water washing and wiping, skin
48 123 physiological functions and skin ceramide content were measured before and after the
49 124 cleaning/removal experiment (Fig. 2C). The TEWL, skin hydration, and skin pH were measured
50 125 using Vapometer (Delfin Technologies Ltd, Kuopio, Finland) (14), Corneometer[®] CM 825
51 126 (Courage+Khazaka electronic GmbH, Cologne, Germany) (15), and skin-pH-meter PH 900
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5 127 (Courage+Khazaka electronic GmbH) (16), respectively, at a specific area on the skin (Fig. 2A).
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7 128 Skin physiological functions were measured by one trained researcher in an air-conditioned
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9 129 room (25 ± 2 °C, $50 \pm 10\%$ relative humidity). Each measurement was performed in triplicate
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11 130 and the average of three data was used for the analysis.
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14 132 *2.6. Semi-quantification of skin ceramide content*

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17 133 The skin ceramide content was measured as described previously (17) with some modifications.
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19 134 In brief, the cells of the SC (2nd to 5th layers) were collected by applying an adhesive tape (1.4
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21 135 cm \times 4.2 cm; #08380; A-ONE G. K., Tokyo, Japan) to the skin (Fig. 2A). The collected tape
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23 136 was divided into two parts, one for analysis of ceramide content and one for determination of
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25 137 total protein content as a surrogate index of total SC (18).
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28 138 For the analysis of ceramide content, lipids, including ceramides, were extracted
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30 139 from the SC cells using 200 μ L of methanol with sonication for 30 min (ASU-2; AS ONE Corp.,
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32 140 Osaka, Japan). The extracted lipid solution was spotted on a polyvinylidene difluoride
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34 141 membrane (Immobilon-P®; Merck Millipore Corp., Hesse, Germany), followed by a dot-blot
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36 142 analysis using mouse IgM anti-ceramide antibody (ALX-804-196; 1:10 dilution; Enzo Life
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38 143 Sciences, Inc., NY, USA) and horseradish peroxidase (HRP)-conjugated anti-mouse IgM
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40 144 antibody (ab97230; 1:1000 dilution; Abcam plc., Cambridge, UK). The ceramide signals were
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42 145 developed by the Clarity Western ECL Substrate (Bio-Rad Laboratories, Inc., CA, USA) and
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44 146 captured by the C-DiGit blot scanner (LI-COR, Inc., NE, USA). The intensity of each ceramide
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46 147 spot was calculated with Image Studio™ Lite software (version 4.0; LI-COR, Inc).
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48 148 The total SC amount for the normalization of the ceramide content was estimated by
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50 149 the total protein amount collected by the tape stripping (18). The collected tapes were
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52 150 individually soaked in 100 μ L of 1 M NaOH with vigorous shaking for 30 min, followed by
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54 151 neutralization with 100 μ L of 1M HCl. The total protein amount was then determined by a
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56 152 spectrophotometer (680 XR; Bio-Rad Laboratories, Inc.) at 595 nm with the Quick Start™
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5 153 Bradford Protein Assay (#5000205JA; Bio-Rad Laboratories, Inc.). The normalized relative
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7 154 amount of skin ceramide was then calculated by simply dividing the raw value of the ceramide
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9 155 signal by the protein amount of the corresponding sample.
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12 157 2.7. Statistics

13 158 Data are expressed as means and standard deviations (SD). Comparison of the baseline data
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15 159 between the right (i.e., water washing) and left (i.e., wiping) forearms was performed using
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17 160 Welch's *t*-test. Two-way analysis of variance of mixed design [time (pre- versus
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19 161 post-experiment) × removal technique (water washing versus wiping)] was used for interaction
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21 162 analysis. A *P*-value of <0.05 was considered statistically significant. All statistical analyses were
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23 163 performed with the SPSS Statistics software (version 23; International Business Machines Corp.,
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25 164 NY, USA).
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31 166 3. Results

32 167 The baseline (i.e., pre-experiment) data of the right (water washing) and left (wipe) forearms
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34 168 were not significantly different in all participant groups (Table 2), which enabled direct
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36 169 comparison between the right and left arms. Tables 3, 4, and 5 show the pre-/post-results of the
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38 170 skin physiological functions (TEWL, skin hydration, and skin pH) and normalized skin
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40 171 ceramide content with agents A, B, and C, respectively. There were no interactions between the
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42 172 removal technique and change in skin physiological functions for the cleaning agents used in
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44 173 this study. Figure 3 summarizes skin physiological functions and ceramide content. In the agent
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46 174 B group, a statistically significant increase in skin hydration ($P = 0.0059$) and decrease in skin
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48 175 pH ($P = 0.0015$) were observed between pre- and post-experiment, regardless of water washing
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50 176 or wiping. The ladder plots of the individual data are shown in Figure S1.
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55 178 4. Discussion

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5 179 In this study, we tested the effect of different removal techniques (i.e., water washing and
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7 180 wiping) of several cleaning agents on skin physiological function. In addition, we evaluated
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9 181 ceramide content in the SC, which affects skin barrier function. Because it is known that
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11 182 cleaning agent residues (containing several kinds of surfactant) can deteriorate skin
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13 183 physiological function, it is important to know how removal methods affect the skin.

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15 184 Even if a single application of the irritant (i.e., cleaning agent ingredients) may cause
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17 185 skin reactions, such as erythema, barrier function disruption, or reduction of skin hydration, it is
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19 186 relatively easy for users to discontinue use because such irritations are usually severe enough to
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21 187 cause apparent drawbacks (5, 6, 19, 20). The problem is that mild but repetitive application of
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23 188 such agents may cause low but sustained irritation of the skin (5, 19, 21, 22). In this study,
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25 189 therefore, we evaluated changes in skin physiological function after repetitive use of cleaning
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27 190 agents. In addition, we focused on the “removable-by-wiping” cleaning agents because of their
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29 191 potential usefulness and cost-effectiveness for home-care services.

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31 192 Because the residue of cleaning agents on the skin is reportedly greater in wiping
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33 193 compared to water washing (23), it is speculated that just wiping may not be a sufficient
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35 194 removal technique and that cleaning agent remnants may deteriorate skin physiological function
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37 195 in the wiping group compared to water washing. However, in this seven-day experiment with
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39 196 three kinds of cleaning agents, none produced adverse effects on skin physiological function or
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41 197 ceramide content, regardless of water washing or wiping (Tables 3–5). In the agent C group, one
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43 198 participant was excluded because of significant erythema. However, this was caused by the film
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45 199 dressing, which was used as a cover of the region of interest, and not due to the cleaning agent
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47 200 or the removal technique, as the covered areas of both forearms (i.e., wiping and water washing)
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49 201 were affected. In sum, removal of cleaning agents by just wiping has comparable effects on skin
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51 202 physiological functions and skin ceramide content to water washing.

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53 203 Significant changes in skin hydration and pH were observed with cleaning agent B
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55 204 (Fig. 3). Although we do not have any solid reason for this, specific component(s) of agent B
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5 205 may have affected skin physiological functions, particularly skin hydration and skin pH. For
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7 206 example, triethanolamine (TEA)-cocoyl hydrolyzed collagen, which was only found in agent B,
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9 207 has been patented worldwide for its mildness (24); however, this cannot fully explain the
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11 208 observed effects on the skin.

12
13 209 There are two major limitations in this study. One is the small number of participants.
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15 210 As we wanted to test three kinds of cleaning agents, we had no choice but to limit the number of
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17 211 participants assigned to each agent. However, the small sample size makes it difficult to
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19 212 generalize the results of this study. The other limitation is the age of the participants. The study
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21 213 participants were all young (21 or 22 years); therefore, care must be taken with the elderly who
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23 214 are more susceptible to external insult.
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28 **5. Conclusion**

29
30 217 In this study, we compared the effects of removal techniques (i.e., water washing and wiping) of
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32 218 cleaning agents on skin physiological functions (e.g., TEWL, skin hydration, and skin pH) and
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34 219 skin ceramide content. As a result, we did not find any differences between water washing and
35
36 220 wiping on skin physiological functions and skin ceramide content.

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38 221 Cleaning agents that claim to be removed by both water and just wiping produce no
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40 222 differences in skin physiological function or ceramide content depending on which removal
41
42 223 method is used.
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7 **232 Conflict of interest**

8
9 233 The authors have declared that no competing interests exist.
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5 299 **Figure legends**
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7 300 Figure 1 Cleaning agents used in this study. (A), (B), and (C) correspond to the agents A, B, and
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9 301 C, respectively.
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13 303 Figure 2 Experimental setup. (A) Regions of washing (red circle), skin pH measurement (blue
14
15 304 circle), and skin TEWL, hydration, and ceramide content measurement (green circle) are shown.
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17 305 (B) “Protective cleaning method” used in this study. (C) Time course of this study.
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20
21 307 Figure 3 Changes in skin physiological functions and skin ceramide content before and after
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23 308 seven-day cleaning by water washing and wiping. Black squares denote the water washing
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25 309 group, whereas red circles denote the wiping group.
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29 311 Figure S1. Detailed ladder plots of Figure 3.
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Table 1 Ingredients of each cleaning agent used in this study

Agent A	Agent B	Agent C
Aqua	Chlorhexidine digluconate	Aqua
Butylene glycol	Polysorbate-60	Ethylhexyl palmitate
Lauramine oxide	TEA-cocoyl hydrolyzed collagen	Butylene glycol
Polyglyceryl-10 laurate	Cetyl alcohol	Dipropylene glycol
Octyldodeceth-20	Alcohol denat.	Squalene
Decyl glucoside	Aqua	Macadamia oil
Dipotassium glycyrrhizate		Jobba oil
Cetyl-PG hydroxyethyl palmitamide		Carbomer
Alcohol		Acrylates/C10-30 alkyl acrylate crosspolymer
		Ascorbyl tetraisopalmitate

In order of appearance.

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Table 2 P-values of the Welch's t-test between right and left forearms at the baseline

	Agent A group	Agent B group	Agent C group
Participants, <i>n</i>	5	5	4 [†]
TEWL	0.36	0.71	0.53
Skin hydration	0.78	0.70	0.88
Skin pH	0.74	0.48	0.58
Skin ceramide content	0.58	0.76	0.98

TEWL: Transepidermal water loss.

Refer to Tables 3, 4, and 5 for the actual data.

[†]Five participants were recruited, and one participant dropped out due to the reaction against film dressing.

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Table 3 Changes of skin physiological functions and ceramide content before and after using agent A (n = 5)

	Removal technique	Pre	Post	P-value for interaction
TEWL, g/m ² /h	water washing	3.72 (0.26)	3.81 (0.58)	0.35
	wiping	3.73 (0.33)	3.39 (0.70)	
Skin hydration, a.u.	water washing	31.98 (12.52)	31.35 (5.65)	0.93
	wiping	30.64 (10.54)	30.55 (4.58)	
Skin pH	water washing	4.85 (0.31)	4.71 (0.36)	0.78
	wiping	4.95 (0.52)	4.73 (0.40)	
Skin ceramide content, ×10 ⁶ a.u.	water washing	1.83 (0.51)	3.49 (3.55)	0.17
	wiping	2.73 (1.32)	1.56 (1.26)	
Mean (SD).				

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Table 4 Changes of skin physiological functions and ceramide content before and after using agent B (n = 5)

	Removal technique	Pre	Post	P-value for interaction
TEWL, g/m ² /h	water washing	4.02 (0.30)	3.85 (0.46)	0.96
	wiping	4.11 (0.63)	3.91 (0.54)	
Skin hydration, a.u.	water washing	26.87 (4.85)	35.05 (6.00)	0.64
	wiping	26.95 (4.43)	37.63 (7.56)	
Skin pH	water washing	5.22 (0.22)	4.69 (0.34)	0.41
	wiping	5.19 (0.35)	4.43 (0.34)	
Skin ceramide content, ×10 ⁶ a.u.	water washing	2.25 (1.88)	1.50 (0.92)	0.44
	wiping	3.02 (3.16)	1.29 (0.96)	

Mean (SD).

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Table 5 Changes of skin physiological functions and ceramide content before and after using agent C ($n = 4^{\dagger}$)

	Removal technique	Pre	Post	P-value for interaction
TEWL, g/m ² /h	water washing	4.03 (0.75)	3.43 (0.81)	0.71
	wiping	3.68 (0.58)	3.33 (0.69)	
Skin hydration, a.u.	water washing	26.79 (2.63)	30.41 (6.42)	0.72
	wiping	25.60 (5.04)	30.79 (6.05)	
Skin pH	water washing	4.92 (0.22)	4.86 (0.13)	0.19
	wiping	5.18 (0.32)	4.75 (0.17)	
Skin ceramide content, $\times 10^6$ a.u.	water washing	1.29 (0.77)	1.15 (0.32)	0.36
	wiping	0.97 (0.93)	1.45 (0.36)	

Mean (SD).

[†]Five participants were recruited, and one participant dropped out due to the reaction against film dressing.

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Figure 1 Cleaning agents used in this study. (A), (B), and (C) correspond to the agents A, B, and C, respectively.

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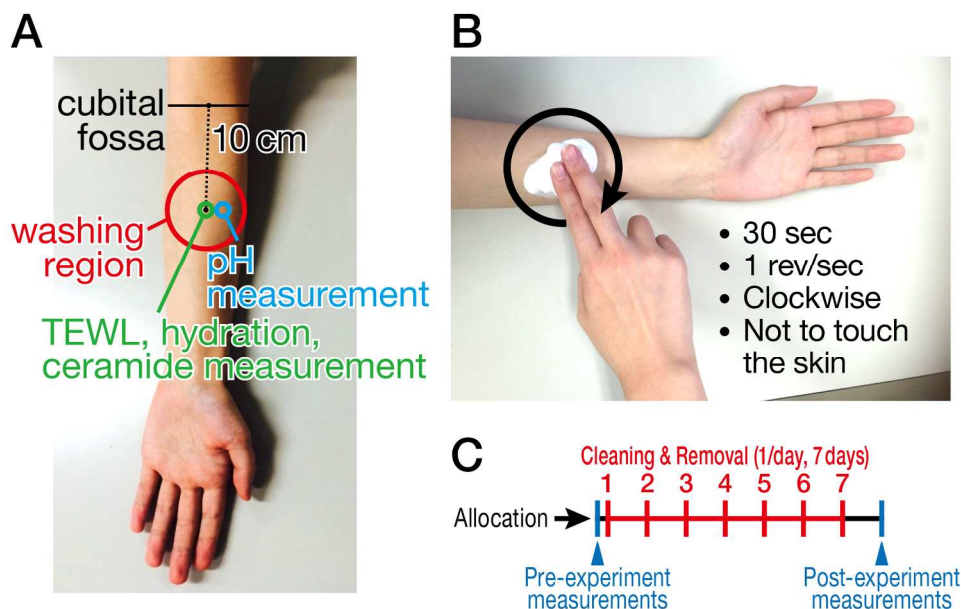


Figure 2 Experimental setup. (A) Regions of washing (red circle), skin pH measurement (blue circle), and skin TEWL, hydration, and ceramide content measurement (green circle) are shown. (B) "Protective cleaning method" used in this study. (C) Time course of this study.

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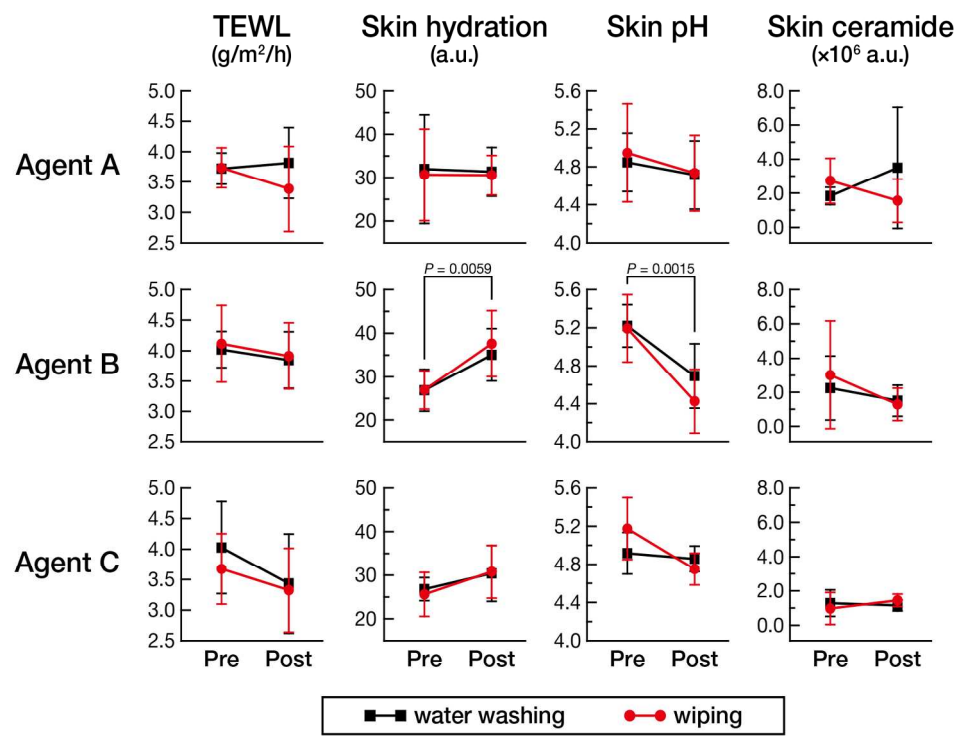


Figure 3 Changes in skin physiological functions and skin ceramide content before and after seven-day cleaning by water washing and wiping. Black squares denote the water washing group, whereas red circles denote the wiping group.

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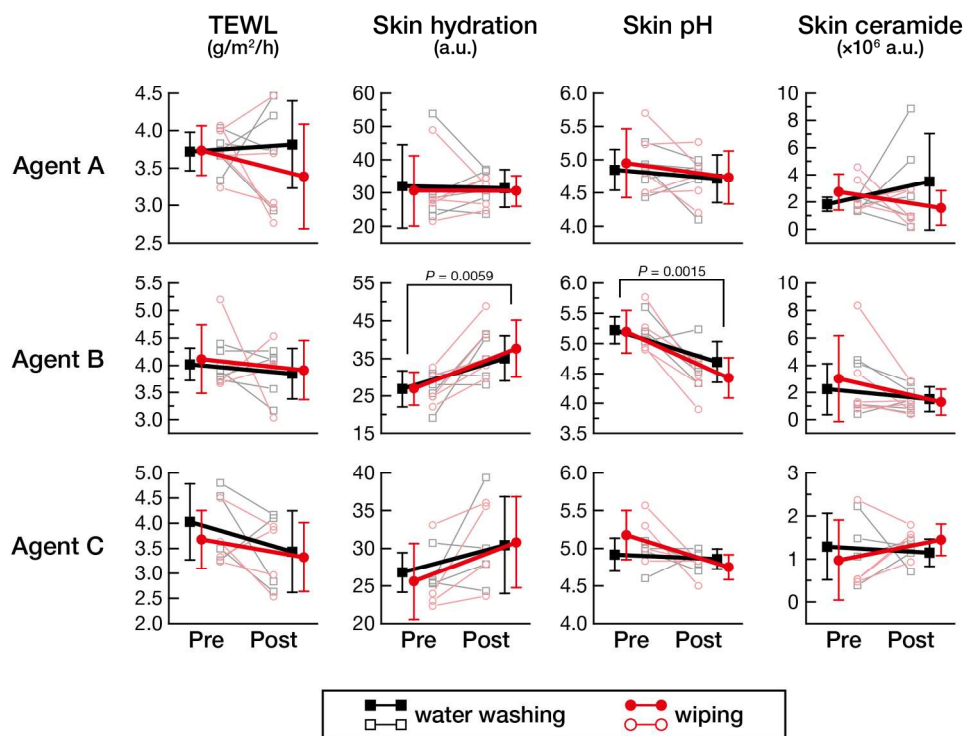


Figure S1. Detailed ladder plots of Figure 3.

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