SHORT REPORTS

Differences in the Skin Surface pH and Bacterial Microflora Due to the Long-term Application of Synthetic Detergent Preparations of pH 5.5 and pH 7.0

Results of a Crossover Trial in Healthy Volunteers

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Skin cleansing preparations consisting of identical synthetic detergents but differing in pH-value (pH 5.5 and 7.0) were applied twice daily on the forehead and forearm of healthy volunteers in a randomized crossover trial. The skin surface pH was found to be significantly higher when the neutral preparation had been used, as was the propionibacterial count (p<0.05). The number of propionibacteria was significantly linked to the skin pH. Hence even minor differences in the pH of skin cleansing preparations seem to be of importance for the integrity of the skin surface. This should be taken into account when planning the formulation of optimal skin care products.

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Repeated washing of normal human skin with (alkaline) soap or (acidic) synthetic detergent solutions affects both the skin surface pH and its bacterial microflora (1). Similar effects as with soap were also found after using alkaline synthetic detergents (2). These findings also substantiate our opinion that different washing habits can influence the biology of the skin surface, which has previously been questioned (3).

There is currently a debate about whether cleansing preparations based on synthetic detergents should be acidic or neutral. Neutral preparations may be less irritating. Yet so far only commercial preparations with differing chemical composition have been assessed (4). To ascertain whether the wanted effects of synthetic detergent preparations on the skin surface will still be encountered if the pH is 7.0 instead of 5.5, we performed a controlled trial using preparations with identical active ingredients.

MATERIAL AND METHODS

Study population
Six male and 4 female healthy volunteers were enrolled after written informed consent. Their ages ranged between 23 and 34 years (mean 28 years). Five individuals were allotted to a group called A, starting with the application of the synthetic detergent preparation characterized by pH 5.5; the others to group B using a preparation of pH 7.0 first.

Cleansing agents
Both liquid synthetic detergent preparations contained identical proportions of water, nipa-laureth-sulfate and cocamidepropylbetaine; potassium coco-hydrolysed animal protein; PEG-7 glyceryl cocoate; cocamidepropyl lauryl ether; PEG-10 olive oil; sodium lactate; perfume, benzyl alcohol and methylisothiazolinone; hydrogenated cocoglyceride and tocopherol; niacin amide and pyridoxine hydrochloride, biotin and amino acids; disodium EDTA; dye C.I. 47005, C.I. 61570; BHT and citric acid and ascorbic palmitate (in descending quantitative order). The pH-difference of the preparation was due to a different amount of sodium hydroxide added for adjustment. Both preparations were supplied by Dr. Schäferbock from Sebapharma, Boppard, FRG, preparation A in fact represented a previous commercial formula (Sebamed® flüssig).

pH-determination
For skin surface pH-determination, the flat glass electrode developed by Ingold and evaluated by Schirren (5) was used (GlassElektrode 403-S7, Ingold-Meßtechnik, Steinbach, FRG) being connected to a precision pH-MV-Meter (pH 521, WTW, Weilheim, FRG).

The values given are means of three consecutive measurements. Technical details of application corresponded to the procedure as described by Arbenz (6).

Investigation of the bacterial flora
Sampling of bacterial flora specimens was based on the detergent scrub method (7). The methods for bacterial culture and identification are described in greater detail elsewhere (2).
Definite identification of coagulase-negative staphylococci and propionibacteria was based on colony morphology, Gram stain, and their biochemical reaction patterns; with staphylococci the plasma coagulase test was also used.

**Cleansing procedure**

Throughout the trial period each volunteer washed the skin of the forehead (median line) and the proximal part of the flexor side of one forearm b.i.d. (in the morning and in the evening) for a period of one minute each. The choice of the appropriate dilution was up to the individual—it should reflect his normal habits. One minute after application the test sites were rinsed with plain tap water.

**Time course of investigations**

During the first 3 days all volunteers continued cleansing their skin as previously. Both cutaneous pH and bacterial flora, however, were investigated at the forehead and on the forearm (days 1 to 3 and days 1 and 3 resp.). During the next 4 weeks either preparation A or B was used. During a further 4 weeks the preparation not applied so far was used. The whole crossover design was based on a random plan. At about the middle of the application interval, i.e. at least 6 h after the last application of the synthetic detergent preparation, both skin surface pH and micro-flora were analysed as at the start on every 7th day (day 10 and so on).

**Mathematical and statistical analysis**

All figures included in the following text show mean as well as maximum and minimum values; both represent standard deviation. The "±" signs stand for the median. Its 95% confidence range is represented by a blank space. Time is always given in days, pH values in units, bacterial counts as logarithm of colony-forming units (CFU). In the graphs, continuous lines connect data concerning volunteers starting with preparation A, data representing group B are connected by dotted lines. For statistical analysis, Student's t-test for the comparison of two independent samples was used, the correlation coefficient was determined according to Bravais & Pearson (8).

**RESULTS**

**pH values**

During the initial period of the trial the pH values of forehead and forearm skin amounted to 4.4 to 5.7 and 4.3 to 5.8. No major difference was found between the two groups of panelists.

In the panelists using the neutral preparation first (group B) the skin surface pH rose markedly from day 4 to day 21. After the switch to preparation A, being acidic, there was a sharp decline to lower pH values lying in the range found at the start. This applies both to the forehead and to the forearm (Fig. 1). Correspondingly, the skin surface pH increased markedly when the volunteers using group A changed from preparation A to preparation B. The skin surface pH values were significantly lower in the panelists using the pH 5.5 preparation on the forehead on days 17, 24, and 31, on the forearm on day 24. During the period from days 4 to 31 the mean skin surface pH was lower in the group of volunteers using the acidic preparation by 0.38 units at the forehead and by 0.46 at the forearm. During the consecutive 4-week trial period, these values amounted to 0.55 and 0.67 units correspondingly.

**Bacterial counts**

Counts of coagulase-negative staphylococci tended to be somewhat higher while preparation B was used (data not shown). The difference between the two groups of volunteers, however, was never significant, either on the forehead or on the forearm. Propionibacterial counts increased markedly while preparation B was being used and decreased correspondingly after the switch to preparation A. Precisely the opposite was true when the preparations were used in
converse order (Fig. 2). On the forearm this phenomenon was even more marked than on the forehead. Thus at the former site the differences were significant on days 17, 24, 31, 45, 52, and 59.

Interdependence of bacterial counts and pH values
The number of coagulase-negative staphylococci was significantly correlated with the skin pH on the forehead, but not on the forearm. The r-value, however, was comparatively low at both sites, amounting to 0.315 and 0.155, respectively. Propionibacterial counts at both sites were linked to the skin pH in a significant manner, the r-value also being comparatively low (0.385) on the forehead and somewhat higher (0.439) on the forearm.

DISCUSSION
Major differences in the pH of otherwise similar synthetic detergent preparations for skin cleansing have recently been demonstrated to influence both the skin surface pH and the bacterial micro-flora, both on the forehead and on the forearm of healthy individuals (2). The results of the present investigation support the previous findings. In addition, they show that even minor differences, of the order of a single pH unit, have a correspondingly marked differential effect on both skin surface pH and micro-flora. Although the differences are not especially remarkable when synthetic detergent preparations of pH 5.5 and pH 7.0 are used in a controlled fashion, as compared with the application of preparations of pH 5.5 and pH 8.5, the effects are in principle the same: using a less acidic synthetic detergent preparation makes the skin relatively more alkaline and thus favours propionibacterial growth.

High propionibacterial counts on the skin may be linked to adolescent acne (9). Moreover, even those who do not share this opinion consider the pH to be one of the ecological factors which might modulate the acne-inducing potential of propionibacteria (10). This has to be considered when planning the formulation of optimum preparations for cleansing normal human skin. Whatever the influence of the pH value of the skin-cleansing preparation on its cutaneous irritancy (4), one must not ignore the definite dependence of the behaviour of the skin pH and micro-flora in the long-term.

REFERENCES