A Novel Technology in Mild and Moisturizing Cleansing Liquids

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Skin cleansers have evolved from merely cleansing to providing mildness and moisturizing benefits as well. Alkyl carboxylate, commonly known as soap, is the prototypical surfactant used in skin-cleansing soap bars; however, the superior mildness of syndet bars over soap bars is well documented in the literature. Harsh surfactants in cleansers can cause damage to skin lipids and proteins, leading to after-wash tightness, dryness, barrier damage, irritation, and even itch. The structure of synthetic surfactants is often tailored to minimize damage to the stratum corneum. A significant breakthrough in cleansing came with the introduction of syndet bars containing sodium cocoyl isethionate as the cleanser and long-chain fatty acids as the moisturizing agents. Current liquid cleansers use a combination of anionic and amphoteric surfactants to reduce protein damage and skin irritation potential of anionic surfactants. These combinations can still cause skin dryness, and this article indicates that this may be due to the interaction of surfactants with skin lipids. The combination of anionic and amphoteric surfactants can result in increased damage to lipids, even though their skin irritation potential is reduced considerably. Skin dryness is addressed in current moisturizing cleanser systems with the use of emollients, such as petrolatum and triglyceride oils. Typically, higher levels of petrolatum are used to increase moisturization by occlusion, with some moisturizers having a petrolatum content as high as 50% to 60% by weight. A novel approach to skin moisturization involves using a combination of lipids, natural oils, and humectants, supplemented with occlusives. In this article, we describe the efficacy of a new moisturizing body wash technology, with sodium cocoyl isethionate as the primary surfactant and fatty acids and triglyceride oils as the emollients.

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Cleansers are designed to remove oil, dirt, sweat, and sebum from skin through the action of surfactants. Surfactants aid in the uplifting of dirt and solubilization of oily soils and help promote normal exfoliation and skin rejuvenation. Repetitive washing with soaps and washing solutions results in after-wash tightness, skin dryness, stratum corneum (SC) barrier damage, erythema, irritation, and itch. The extent to which cleansers can cause such damage depends on the nature of the surfactants and the cleansing conditions. Designing mild and moisturizing cleansers requires an in-depth understanding of cleanser interactions with skin.

The SC is the outermost layer of the skin and the body's interface with the external environment. One of the key functions of the SC is to regulate loss of water from the body and maintain steady-state equilibrium with the environment. Water content of the SC has a significant impact on its tactile, optical, and barrier properties, including its biologic functions. Day-to-day challenges, such as changes in humidity, temperature, and daily cleansing, can affect skin's barrier properties and its ability to hold water. Current moisturizing body wash technologies have a combination of anionic and amphoteric surfactants as the cleanser base, along with high levels of oils and occlusives as moisturizers. High concentrations of occlusives found within cleansers are not efficiently absorbed by skin, and a significant amount is often lost during the wash period. A more effective way to moisturize is to minimize cleanser damage using mild surfactants while supplementing the cleanser with a combination of efficient natural emollient oils, lipids, occlusives, and humectants. This allows skin to maintain its natural moisture while oils and lipids penetrate rapidly into deeper layers and help skin improve its barrier and water-retention properties. In this article, the science underpinning the current technologies is briefly reviewed, and the development of a new moisturizing body wash technology and its in vivo performance are presented.

BACKGROUND

The SC contains 2 distinct structural components: the corneocytes and the intercellular lipids. The "bricks-and-mortar" model is often used to describe the organization of these components within the SC, where the flat corneocytes are the "bricks" and the lipid matrix represents the "mortar."[1] The upper layers of the SC are made up of approximately 65% to 75% proteins.[2] Keratin is the major protein in the SC. Other proteins include glycoproteins, which are structurally incorporated into the cornified envelope to provide cohesion by binding homophilically with proteins on adjacent cells, and hydrolytic enzymes involved in the desquamatory process necessary for corneocyte exfoliation from the surface of the skin. Water content of the SC is modulated by natural moisturizing factors, which include short-chain amino acids. Cleanser surfactants can bind to SC proteins, leading to transient swelling and hyperhydration under wash conditions. Transmission and cryo-scanning electron microscopy have confirmed that extended water exposure leads to extensive disruption of the SC structure.[3] Hyperhydration occurring during regular wash and after extended water exposure is followed by shrinking as water evaporates, leading to drying stresses.[4,5] Swelling facilitates the penetration of surfactants into deeper layers, possibly leading to a biochemical response such as irritation and itch.[2] Because surfactant binding reduces the skin's ability to retain water, skin often returns to a state of lower hydration after washing.[6] Cleansers can also lead to a reduction in natural moisturizing factors, consequently changing skin elasticity within minutes of washing.[7,8] Surfactant binding to SC proteins can cause significant protein denaturation, leading to barrier damage. Studies have shown that the tendency of surfactants to cause protein denaturation is related to the head-group size, with larger head groups causing less damage.[9] Anionic surfactants have a greater tendency to cause protein denaturation than amphoteric or nonionic surfactants. One of the common approaches to reducing surfactant-induced protein damage in current liquid cleansers is to increase the size of the polar group of the surfactant and to use a combination of anionic and amphoteric surfactants.[9-11]

Skin lipids, which compose approximately 10% to 15% of the SC, are also susceptible to surfactant-mediated damage. Lipids within the SC include approximately 47% ceramides, 24% cholesterol, 11% fatty acids, and 18% cholesterol esters.[12] In the SC, the physical conformation of the intercellular lamellar lipids provides a tight and semipermeable barrier to the passage of water through the tissue.[12] Surfactant removal of lipid components can alter the optimum levels required to maintain a healthy SC. Some studies suggest that surfactants above their critical micelle concentration cause delipidation of the SC by solubilization of the lipids in surfactant micelles.[6,13] Lipid damage can also be caused by the adsorption and intercalation of surfactants into SC lipid bilayers, resulting in its increased permeability and even bilayer destabilization.[14] Washing skin with a liquid cleanser base (anionic-amphoteric surfactant mix without any moisturizing ingredients) can reduce the level of fatty acids and cholesterol in the skin after a single wash.[15] Transmission electron micrographs of human skin washed ex vivo under exaggerated conditions have shown that nonionic surfactant-based cleansers alter the lipid region to a greater extent than do mild cleansing bars with sodium cocoyl isethionate (SCI) as the surfactant.[15] Previous studies have not, however, examined the impact of lipid-damaging surfactants on skin condition in vivo.

The most basic cleanser, soap, is created by a heating process called saponification, which occurs when an alkali and a long-chain fat compound are combined, producing a fatty acid salt that exhibits anionic detergent properties under high pH conditions (approximately pH 10).[16] Given their excellent foaming and lathering characteristics, anionic surfactants are typically used as primary surfactants in cleansers, whereas nonionic surfactants are used less often because of their poor lathering

characteristics.[2] In addition to soaps, synthetic anionic surfactants (syndets), such as alkyl ether sulfate, alkyl acyl isethionates, alkyl phosphates, alkyl sulfosuccinates, and alkyl sulfonates, are used in cleansers.[2] Liquid cleansers often have a combination of anionic and amphoteric surfactants. Both syndet bars and liquid cleansers have evolved to contain moisturizing lipids, emollients, occlusives, and humectants that offer skin care benefits beyond cleansing.

The first breakthrough in mild cleansing came with the introduction of the syndet bars,[17] which have mild sodium alkyl isethionate as the synthetic surfactant. The mildness of isethionate has been attributed to its larger polar head group, with a lower charge density and its ability to function as a cleanser under neutral pH conditions. The moisturizing ingredient used in one of the most widely used syndet bars is stearic acid. Previous studies[18] have shown that fatty acids from syndet bars deposit on skin during washing, penetrating into deeper layers and possibly replacing the fatty acids during wash. In addition, it has been suggested that the fatty acids in the bar act as a buffer or "sacrificial lipid" against lipid extraction by cleanser surfactants by presaturating the surfactant micelles, thus minimizing the surfactant-mediated depletion of skin lipids.[2] Liquid cleansers and shower gels use a combination of anionic and amphoteric surfactants to reduce their protein denaturation and skin-irritation potential. Unlike bars, current moisturizing shower gels do not contain high levels of long-chain fatty acids or other lipids to minimize damage and moisturize skin. Instead, they contain moisturizing oils, such as petrolatum and triglycerides, and humectants, such as glycerol. Petrolatum provides an occlusive barrier, reducing water evaporation from skin. Liquid triglyceride oils, such as sunflower or soybean oil, are effective in rapidly penetrating into crevices and cracks and providing moisturization to deeper layers. Humectants, such as glycerol, can further help skin retain moisture[19]; however, deposition and retention of glycerol from cleansers is a challenge because of its high water solubility. The recent trend in moisturizing body wash technologies has been toward a significant increase in the petrolatum levels to as high as 60% by weight.[20] This approach provides occlusive moisturization but does not protect the SC and underlying skin from damage. Although higher levels of petrolatum lead to higher deposition on skin, higher levels of petrolatum are also lost during the wash, making this an inefficient process. Further increases in petrolatum levels to enhance moisturization to a greater degree are impractical because there are formulation constraints and decreased cleanser efficacy. More importantly, a significant amount of the occlusive is lost in washing.

A radically different approach to achieving higher moisturization is to increase the mildness of the cleanser surfactant base and use minimum levels of a combination of moisturizing ingredients that will enhance skin's natural ability to hold water. Indeed, these formulations could certainly be supplemented with occlusives if necessary. In the present study, we sought to determine how mild surfactants could be further modified to reduce their protein and lipid damage potential and moisturize skin using a combination of lipids and triglyceride oils.

MATERIALS AND METHODS

Materials

Specific surfactants tested included sodium laurate (SL), SCI, alkyl polyglucoside (APG), sodium laureth sulfate (SLES), and cocamidopropyl betaine (CAPB). Other chemicals used included soybean oil, glycerin, cow fatty acid, zein powder, stearic acid, phosphatidyl choline, phospatidic acid, and cholesterol.

Zein Solubilization Assay

The zein solubilization assay provides an in vitro measure of protein solubility and, consequently, protein damage. Zein is a model corn protein with limited solubility in water. The ability of surfactants to denature and solubilize zein has been linked to their skin-irritation potential. In this study, zein dissolution solubility in surfactant solutions was used as a measure of protein denaturation potential of the surfactant. Zein powder was mixed with 5 wt % surfactant solution for 24 hours. Solutions were then filtered using a nylon membrane. Dissolved zein was separated from the undissolved materials and the in-solution zein concentration was determined using UV absorbance.

Charge Density and Micellar Charge

Higher charge density of the micelle is linked to increased harshness of the surfactant toward proteins.[20] Previous studies have shown that there is a correlation between micelle charge density and zeta potential with the denaturation of zein protein.[20] The zeta potential of the micelle was taken as a measure of the micelle charge density. Zeta potential was measured at room temperature using a zeta potential analyzer.[20,21]

Lipid Solubility/Extraction Assay

The ability of surfactants to extract medium-chain lipids, such as stearic acid and cholesterol, was assessed by determining the solubility of stearic acid and cholesterol separately in a 5 wt % surfactant solution. Excess cholesterol or stearic acid was contacted with 5 wt % surfactant solution, and the ingredients were mixed at

room temperature (23°C–25°C). Samples were stored for 3 weeks for further equilibration. Excess lipids separated at the bottom, and the supernatant was analyzed using gas chromatography or high-performance liquid chromatography for determining the concentration of cholesterol and stearic acid solubilized by the micelle.

Liposome Solubilization Assay

The ability of surfactants to alter a lipid bilayer was assessed using a liposome assay as previously reported in the literature.[22,23] Specifically, permeability changes to the liposome structure caused by surfactants were detected by monitoring red shift of a UV probe present as an integral part of the membrane. Both the concentration at which the liposome destruction begins to occur and the magnitude of UV signal change were used as indicators of surfactant damage to the bilayer. The previously mentioned liposome test could not be carried out for fully formulated systems because they were turbid. A modified liposome test, described by Pashkovski et al,[24] was used for evaluating the samples.

Clinical Studies

All clinical studies were reviewed and approved by an institutional review board. Following are descriptions of protocols for various clinical studies.

Forearm Controlled Application Test and Transepidermal Water Loss Assessments

The forearm controlled application test was used to determine the skin-drying potential of selected single surfactants.[25] Participants were men and women between the ages of 18 and 65 years. This was a randomized, double-blind study consisting of a 5-day conditioning phase and a 5- to 12-day application phase. For certain studies, the standard forearm controlled application test methodology was modified for screening purposes, and the application phase lasted only 2 days. During the conditioning phase, participants were requested not to use moisturizers on their arms and to cleanse with single surfactants formulated into typical body wash formulations using 12 wt % surfactant and a typical polymeric thickener, Jaguar C-162. Polymer-thickened water was used as a control.

During the product-application phase, qualified participants had up to 4 test sites identified and marked on each of their volar forearms, for a total of up to 8 test sites per subject. Each day of the product-application phase consisted of 3 wash sessions (with 2 successive washes per session) a minimum of 2 hours apart. During the product-application phase, visual evaluations of dryness and erythema were conducted prior to the first wash of each wash session and a minimum of 2 hours after the last wash session of each day (total of 8 evaluations). Instrumental measurements of transepidermal water loss (TEWL) were conducted following the visual evaluations at baseline (before the first wash) and a minimum of 2 hours after the last wash. TEWL was measured using a DermaLab TEWL probe. Visual assessment of dryness was rated on a scale of 0 to 6, where 0=none and 6=severe.

Normal Home-Use Study

A randomized, single-blind study was conducted with 65 female participants aged 31 to 65 years who were in general good health. Within this group, 45 were white (69%), 11 were African American (17%), and 9 were Hispanic (14%). At-home use wash conditions were employed rather than exaggerated conditions typically used in clinical cleansing tests.

Clinical dryness and erythema were determined, with the treatment phase lasting 3 weeks. The results at the end of weeks 1 and 3 were recorded. Expert clinical dryness and erythema were rated on a scale of 0 to 4, where 0=none and 4=severe. Consumer self-perception data were collected from questionnaires asking participants to score significant improvements in skin "feel," dryness, and appearance following a 1- and 3-week treatment phase.

Statistical Analysis

A paired *t*-test to assess change from baseline was conducted. Differences were considered significant at $P \leq .05$.

RESULTS AND DISCUSSION

Cleanser interaction with skin proteins can negatively affect skin hydration and viscoelasticity.[5,7,8,26] To determine the surfactant-mediated damage potential of cleanser surfactants to the proteins found within the upper layers of the SC, protein denaturation and SC swelling analyses were conducted. Nonionic APG as well as anionic SL and SCI surfactants were evaluated, with SCI and SL use resulting in an increase in protein denaturation potential and corneocyte swelling relative to water (data not shown). These data are consistent with previous studies showing that the irritation potential of surfactants on SC protein follows the well-known order, namely, anionic surfactants are more irritating than amphoteric surfactants, which are more irritating than nonionic surfactants.[2] Amphoteric surfactants generally show good skin compatibility and can decrease the skin-irritation potential of harsher anionic surfactants when used in combination with them.[11,27] Previous studies have shown that, at a constant level of sodium lauryl sulfate, increasing concentrations of amphoteric CAPB result in a CAPB concentration-dependent reduction in the percentage of SC swelling.[4]

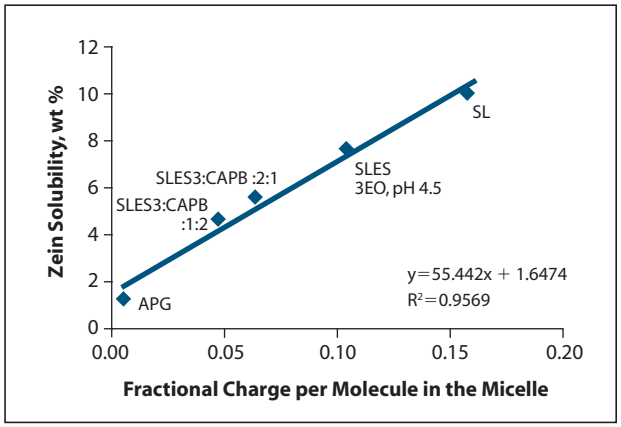

Figure 1. Correlation with micellar diffuse layer charge. Surfactant at 5%>10 critical micellar charge in all cases. APG indicates alkyl polyglucoside; SLES:CAPB, sodium laureth sulfate:cocamidopropyl betaine; SLS, sodium lauryl sulfate; SL, sodium laurate.

In addition, other studies have shown that one of the factors responsible for the mildness or harshness of a surfactant is the charge density of the surfactant polar head group.[2,28-30] We hypothesized that the tendency of surfactants to cause protein denaturation would be related to charge density of protein-bound, micellelike aggregates. Micelle charge density for selected surfactants was calculated and graphed as a function of percent zein solubility. Protein damage did increase with micelle charge density (Figure 1), confirming that amphoteric surfactant CAPB is milder toward SC proteins than anionic surfactants SL and sodium lauryl sulfate. Mildness of SCI compared with SL is also consistent with its lower charge density compared with SL. Taken together, these data indicate that nonionic surfactants are significantly milder on SC proteins than anionic surfactants. Although the conclusion that lowering the charge density of the surfactant will lower the protein damage potential of surfactants is not novel, these data quantify the hitherto hypothesized relationship between surfactant head-group charge density and protein denaturation potential of surfactants and provide a simple method of assessing the damage potential of mixed surfactant systems. Furthermore, these results suggest that micelle charge density can be used as an irritation "ruler" for surfactant-mediated protein damage.

Surfactant interactions with skin lipids have been extensively studied; however, the impact of such interactions on cleanser mildness and the mechanisms by which surfactants interact with lipids and cause skin damage have yet to be fully established.[29-31] Fatty acids and cholesterol are more vulnerable to solubilization than ceramides.[32] Consequently, the use of a cleanser without any moisturizing ingredients can reduce levels of fatty acids and cholesterol in skin by solubilization. Intercalation of the surfactant into a lipid layer can also cause barrier perturbation even without significant extraction of lipids. In vitro assessment of the lipid damage potential of surfactants includes the ability of surfactants to disrupt model liposomes[14] or dissolve fatty acids and cholesterol in surfactant micelles. The effect of various surfactants on a model lipid bilayer membrane was measured in a liposome-damage assay. Results given in Figure 2 show that APG, CAPB, and SL have a higher potential to damage the liposome consisting of phosphatidyl choline, phosphatidic acid, and cholesterol than SCI. Although the specific effects of these surfactants are still being defined, we hypothesized that the high liposome-damage potential of APG and CAPB was related to their large head-group size and their ability to accommodate solubilized molecules in the micelle compared with SCI. Although SL has a compact head group, its high pH (approximately pH 10) compared with SCI (approximately pH 7) can impact bilayer stability.[33] The ionization of the fatty acids and the resultant increase in the electrostatic repulsion in the bilayer leads to an increased fluidity and lipid-extraction potential.[33] Additional in vivo studies will help to determine which of these metrics provides the strongest correlation to surfactant mildness.

Clinical dryness levels caused by APG, SCI, SL, SLES, and CAPB surfactant usage were assessed in patients on days 5 and 12. By day 12, both nonionic APG and anionic SL resulted in increased skin dryness compared with anionic SCI (Figure 3). In contrast to minimal APG-mediated protein damage,[2] APG-mediated lipid damage (Figure 2) and in vivo dryness (Figure 3) were significant. Similarly, CAPB, which causes minimal

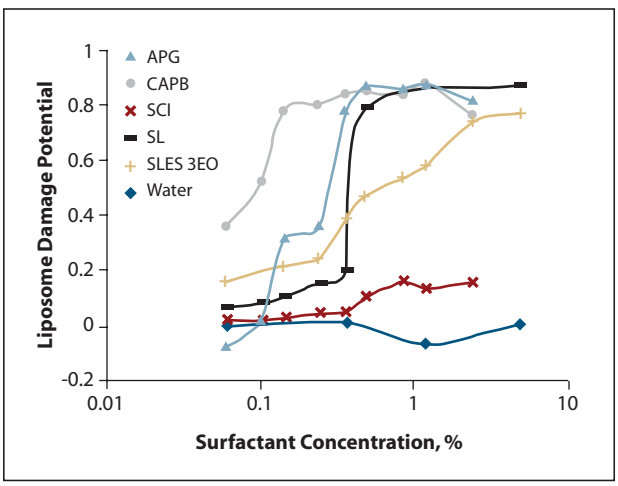

Figure 2. Liposome damage potential of various surfactant solutions (5 wt %) and water plotted as percent dissolved per wt % surfactant. APG indicates alkyl polyglucoside; CAPB, cocamidopropyl betaine; SCI, sodium cocoyl isethionate; SL, sodium laurate; SLES, sodium laureth sulfate.

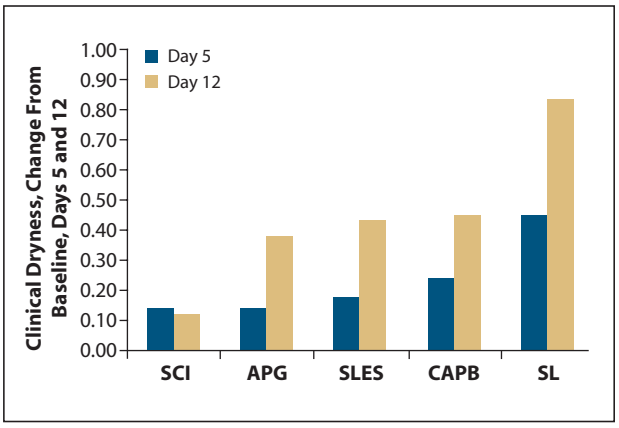

Figure 3. In vivo clinical dryness as assessed by forearm controlled application test after 5 and 12 days. Surfactant 12 wt %. SCI indicates sodium cocoyl isethionate; APG, alkyl polyglucoside; SLES, sodium laureth sulfate; CAPB, cocamidopropyl betaine; SL, sodium laurate.

protein swelling but significant liposome damage, also resulted in high in vivo skin dryness. Interestingly, SL, which damages both proteins[2] and liposomes (Figure 2), led to the highest degree of in vivo dryness among these surfactants (Figure 3). The day 5 dryness scores for APG, CAPB, and SL indicate that APG and CAPB cause less dryness in the SC than SL. These data, in combination with the protein and lipid damage potential, suggest that the impact of surfactants that damage both proteins and lipids on in vivo skin dryness is more rapid and severe than that of surfactants that damage only lipids. The data from day 12 suggest that the lipid-damaging surfactants also cause significant dryness. Importantly, the surfactant SCI is the least drying among these surfactants because it causes minimal damage to both proteins and liposomes (Figures 2 and 3). These results clearly demonstrate that cleanser formulations that only consider potential SC protein damage might negatively impact SC lipids and cause skin dryness.

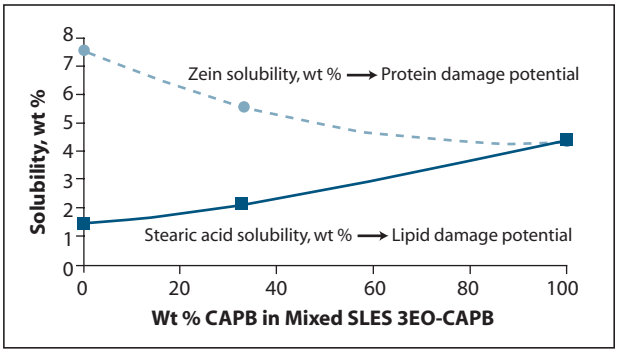

Figure 4. A comparison of the protein-damage potential (zein solubility) and lipid-extraction potential (stearic acid solubility) of sodium laureth sulfate (SLES), triethoxylate, and cocamidopropyl betaine (CAPB) mixture as a function of the wt fraction CAPB to total surfactant.

The use of a combination of anionic and amphoteric surfactants in liquid cleansers indicates that previous efforts in creating mild cleansers have focused mostly on reducing protein damage. The relative effects of the addition of an amphoteric surfactant (CAPB) to anionic surfactant SLES on protein and lipid damage are shown in Figure 4. Although the addition of CAPB resulted in lower protein solubility by decreasing micelle charge density (Figure 1), there was an increase in stearic acid solubility and, consequently, the potential of the mixture to damage lipids (Figure 4). These results reinforce the importance of balancing both protein and lipid damage to maximize the overall mildness of a cleanser.

The data from Figures 2 and 3 indicate that SCI was milder toward both proteins and lipids. Therefore, in designing a new mild cleanser base, SCI was used as the primary surfactant. As mentioned earlier, the excess stearic and palmitic acids contained within syndet bars may act as a buffer against skin lipid extraction, contributing to syndet bar mildness. Inclusion of such fatty acids in body wash may also enhance their mildness and moisturization benefits. In this context, we sought to determine if the addition of fatty acids of varying chain lengths would affect protein or lipid damage potential of SCI alone or SCI with increasing weight concentrations of C10, C12, C14, C16, and C18 fatty acids. Protein solubility and the minimum concentration required to destabilize liposomes were determined. Results obtained show that C16 and C18 fatty acids were more effective in reducing both the zein solubility and liposome damage than the shorter-chain fatty acids, even at lower concentrations (Figure 5). Thus, addition of long-chain fatty acids can be expected to increase the mildness of SCI toward both proteins and lipids, and the present results provide new mechanistic insights into the mildness of the syndet bar over soap.

The SCI in syndet bars is obtained by a direct esterification of fatty acids and isethionate (known as directly esterified fatty isethionate [DEFI]). The data in Figure 5 were obtained using only surfactants and fatty acids. To determine whether the addition of fatty acid improved the mildness of isethionate in a typical fully formulated system, prototype formulations were prepared with SCI as the primary surfactant. Prototypes with and without fatty acids were evaluated using in vitro and clinical assays (Figure 6). These formulations did not contain other emollients or moisturizers. Results were also obtained for a marketed high–emollient-containing body wash with petrolatum as the moisturizer. As shown in Figure 6A, the SCI-based prototype without fatty acid showed a lower liposome damage potential than the high-emollient body wash. The addition of C16 and C18 fatty acids significantly reduced the damage potential even

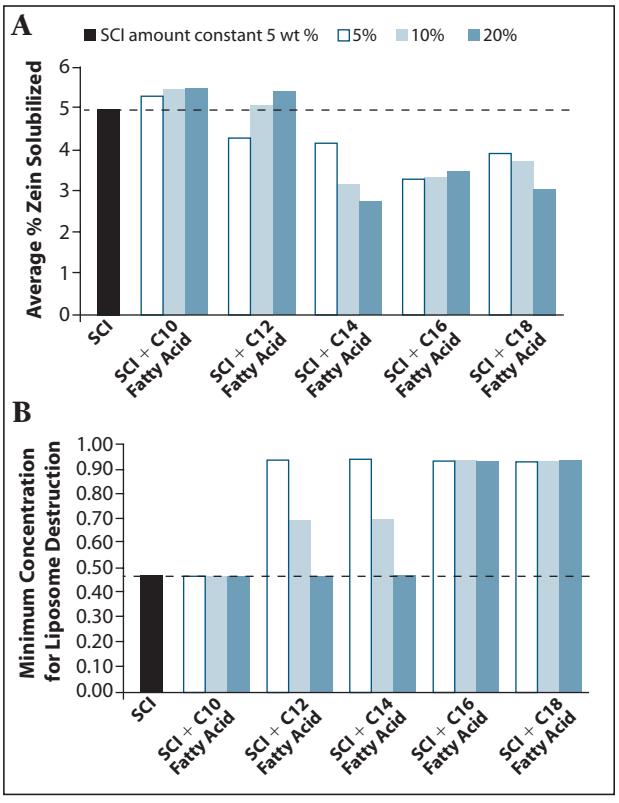

Figure 5. The protein- and lipid-damage potential of sodium cocoyl isethionate (SCI) as determined by zein solubility (A) and minimum concentration for liposome damage (B).

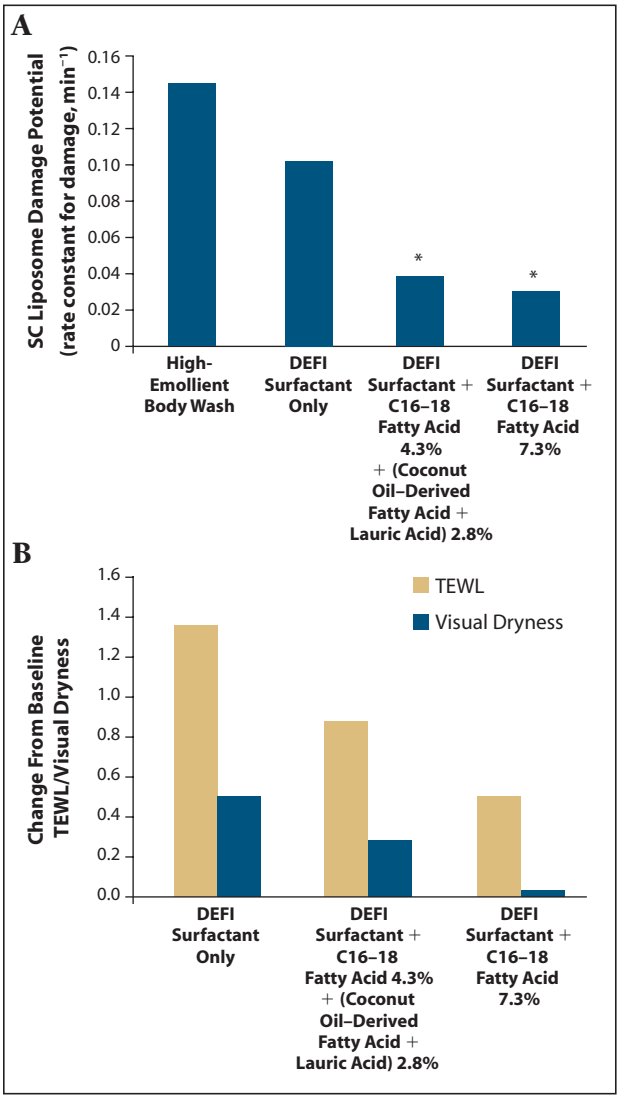

Figure 6. Stratum corneum liposome damage potential (A) and change from baseline in transepidermal water loss (TEWL) and visual dryness scores of a high-emollient body wash (B). Asterisk indicates $P<.05$.

further as a function of fatty acid concentration. Clinical dryness and TEWL were determined following a 2-day use of SCI prototypes with and without different combinations of fatty acids (Figure 6B). The addition of fatty acids to the SCI prototype reduced dryness and lowered TEWL. Thus, a cleanser containing SCI as the primary surfactant along with specific long-chain fatty acids can lower the lipid-damage potential of the system; this, in turn, is reflected as a significant reduction in skin dryness and barrier damage in a standard clinical test.

Surfactant interactions with proteins are nonspecific in nature[29,30]; therefore, we hypothesized that surfactants that are milder toward proteins would be milder toward enzymes in general. This was tested in a clinical study using 2 benchmarks of cleanser mildness: a soap bar and a syndet bar.[34] The activity of beta glucocerebrosidase, an enzyme involved in ceramide synthesis, decreased upon use of a soap bar (with SL as the surfactant) for a week, whereas the activity increased markedly when the participants were switched to a milder SCI-based syndet bar.[34] Thus, the mildness of the cleanser base allows the skin's biology to function optimally, leading to a healthy SC.

A moisturizing body wash is expected to not only have a mild cleanser base but to also deposit and deliver moisturizing ingredients to skin during cleansing. In addition to fatty acids, other types of moisturizing ingredients, such as humectants, glycerol, triglyceride oils, and petrolatum, can be added. Previous studies have shown that polar oils, such as triglyceride oils, can rapidly penetrate the upper layers of the SC to aid in reinforcing the barrier and lowering surfactant binding to the SC. Therefore, we hypothesized that the addition of a triglyceride oil, such as soybean oil, to a DEFI cleanser might provide moisturization and increase the cleanser mildness. Results from a cumulative in vivo patch test using identical DEFI-based formulations (with soybean oil versus petrolatum) shown in Figure 7A indicate that the addition of soybean oil resulted in decreased irritation compared with petroleum jelly. The addition of the polar triglyceride oil has an effect

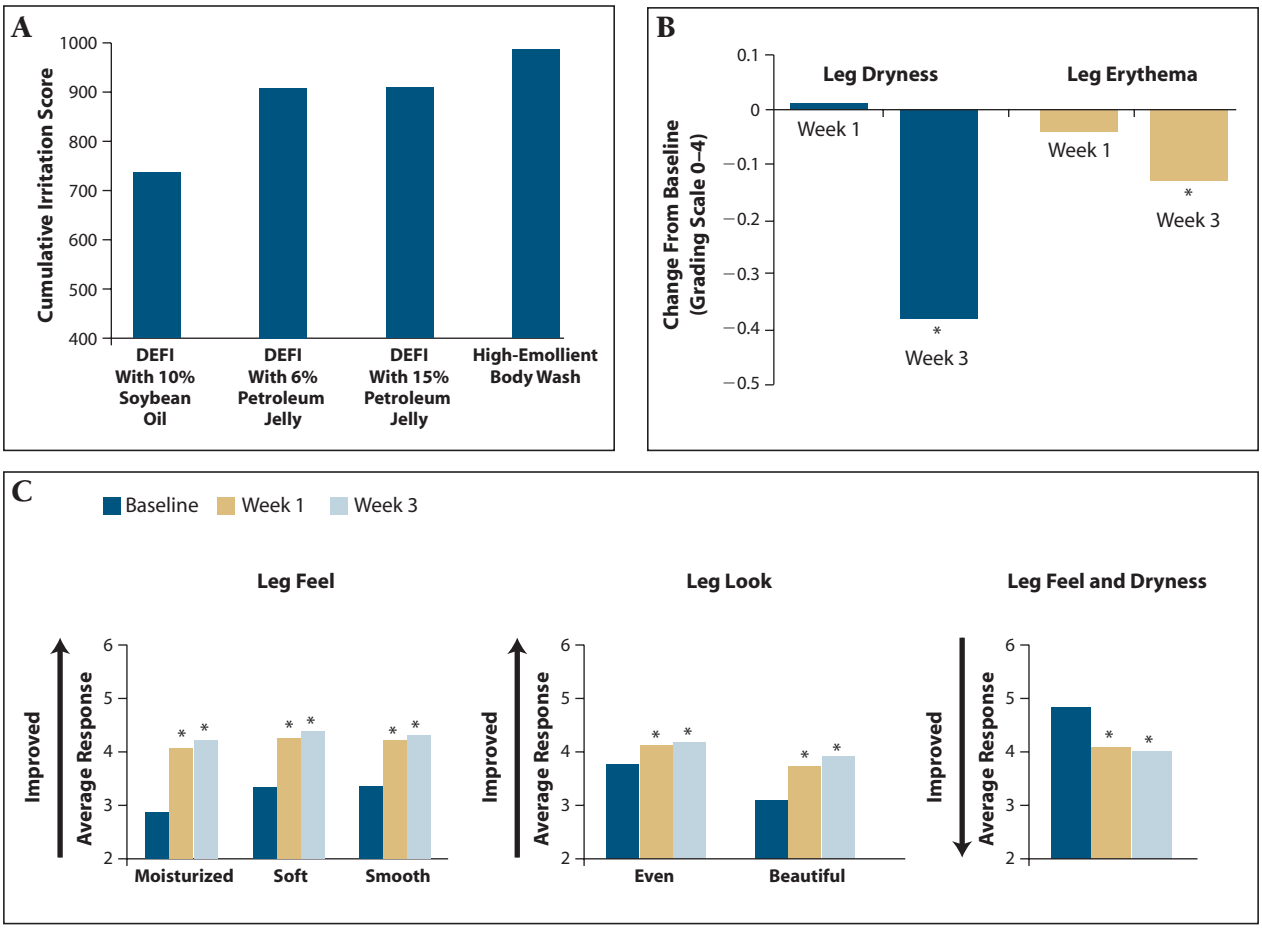

Figure 7. The cumulative irritation score of a high-emollient body wash and esterified fatty isethionate (DEFI) with soybean oil or petroleum jelly (A), change from baseline in leg dryness and leg erythema scores with DEFI + 10% soybean oil (B), consumer self-perception questionnaire scores of leg feel, leg appearance, and leg dryness with DEFI + 10% soybean oil (C). Asterisk indicates $P<.05$.

on the irritation potential of the DEFI-based cleanser similar to data reported for the SLES betaine base.[35] Our ongoing spectroscopic studies suggest that the mechanism by which polar oils reduce surfactant binding to proteins is by blocking their protein interaction sites (S. Mukherjee, unpublished data, 2009).

Clinical and consumer studies have been conducted to assess the effects of the new DEFI-based body wash on skin condition, and the results will be reported separately in forthcoming publications (V. Foy, K. Vetro, S. Zhang, et al, unpublished data, 2008). In the current study, clinical metrics, such as visual clinical dryness and erythema, were determined on the legs of participants after 1 and 3 weeks of DEFI use (Figure 7B). There was a significant reduction from baseline in visual dryness and erythema after 3 weeks of DEFI cleanser use, indicating that the regular use of the new DEFI moisturizing body wash will improve skin condition. In this study, participants also answered a self-perception questionnaire, and the results show that participants perceived improved moisturization as early as week 1 (Figure 7C). Participants were also presented with images of pairs of legs, which included their own legs as well as those of other participants in the study. One image was taken at baseline and the second taken at week 1 or week 3 in a randomized manner. Participants were asked to indicate which of the 2 images looked less dry. Seventy-six percent of all participants preferred the week-3 leg photographs over baseline. Figure 8 shows samples of such leg images for 2 participants representing 1 extreme case of improved skin condition and 1 case of average change. Taken together, these data indicate that a liquid cleanser based on a mild DEFI surfactant in combination with fatty acids and triglyceride oils provides mild cleansing and moisturization.

CONCLUSION

Since their inception, skin cleansers have continued to evolve from basic agents for the removal of soil, dirt, and bacteria to also providing mildness and moisturizing benefits. Soap-based products offered improved cleansing

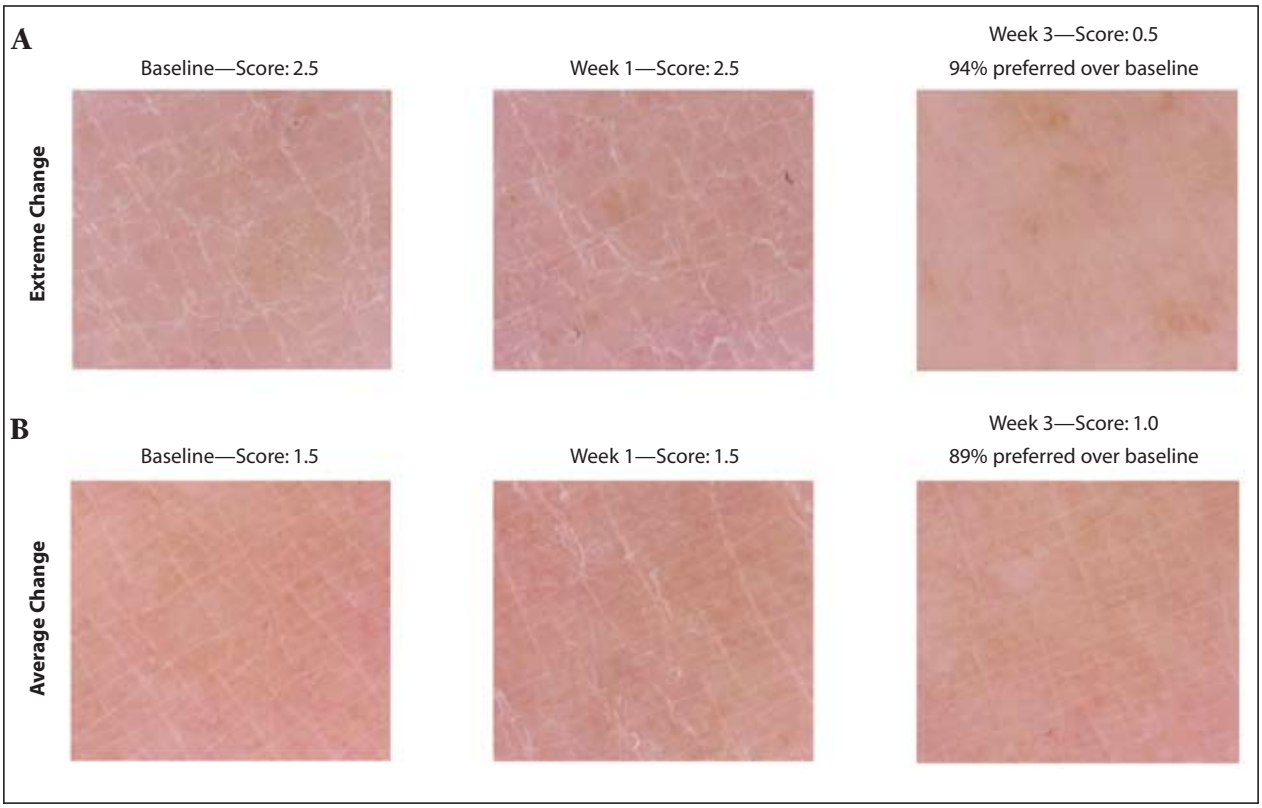

Figure 8. Sample leg images of a 27-year-old white patient (A) and 58-year-old white patient (B).

over mechanical methods of water alone but could irritate and dry skin. Bars based on synthetic detergents that offer improved skin compatibility compared with soap have become available during the last several decades. The variety of body washes has been growing with increasing popularity as the processing of liquid systems makes it possible to select milder surfactants and surfactant mixtures. A combination of an anionic and an amphoteric surfactant has become the typical formulary system for liquid cleansers because of the ability of the amphoteric surfactant to lower the skin-irritation potential of the anionic surfactant. However, these cleanser bases are still drying to skin. Liquid technology also allows more efficient deposition and delivery of beneficial agents onto skin from a wash-off system, and this alleviates the symptoms of dryness and moisturizes the skin. The recent trend in moisturizing body wash technology has been the use of increasingly higher levels of occlusives, such as petrolatum, to maximize moisturization.

The present work details a different approach to increasing moisturization by minimizing damage to both skin proteins and lipids within the SC. Our system provides moisturization by maintaining and enhancing the skin's ability to hold moisture and by supplementing it with lipids, oils, and occlusives. The present study shows that minimizing damage to proteins involves lowering surfactant micelle charge density and that this is the mechanism by which an amphoteric surfactant reduces the irritation potential of an anionic surfactant. Furthermore, the addition of long-chain fatty acids lowers the tendency of a surfactant system to damage lipid membranes. This is perhaps due to their ability to act as a buffer against micelle lipid extraction by micelles and to deposit and replenish some of the fatty acid lipids lost during cleansing. Our studies also show that polar moisturizing oils, such as triglyceride oils, also reduce surfactant binding to proteins and in turn make the cleansers milder toward skin. These actions allow skin to maintain its moisture and biologic functions.

The deposition and delivery of humectants, triglyceride oils, and occlusives can further enhance moisturization. These data have led to the creation of a new body wash technology, consisting of DEFI as a mild surfactant combined with fatty acids and triglyceride oils as moisturizers. In vivo patch studies show that the new system is less irritating than the current high-emollient body washes. A 3-week, normal-use clinical study confirmed that the new technology reduced the visible signs of dryness and increased moisturization. Developing cleansers that effectively deliver moisturizing benefits is a technical challenge, requiring the

deposition of skin agents that are normally removed by cleansers under wash-off conditions. Cleanser systems that provide additional skin care benefits as outlined previously will result in novel technologies and products.

REFERENCES

1. Elias PM. Epidermal lipids, barrier function, and desquamation. *J Invest Dermatol*. 1983;80(suppl):44s-49s.
2. Ananthapadmanabhan KP, Moore DJ, Subramanyan K, et al. Cleansing without compromise: the impact of cleansers on the skin barrier and the technology of mild cleansing. *Dermatol Ther.* 2004;17(suppl 1):16-25.
3. Warner RR, Stone KJ, Boissy YL. Hydration disrupts human stratum corneum ultrastructure. *J Invest Dermatol*. 2003;120:275-284.
4. Rhein LD, Robbins CR, Fernee K, et al. Surfactant structure effects on swelling of isolated human stratum corneum. *J Soc Cosmet Chem.* 1986;37:125-139.
5. Wilhelm KP, Cua AB, Wolff HH, et al. Surfactant-induced stratum corneum hydration in vivo: prediction of the irritation potential of anionic surfactants. *J Invest Dermatol*. 1993;101:310-315.
6. Imokawa G, Akasaki S, Minematsu Y, et al. Importance of intercellular lipids in water-retention properties of the stratum corneum: induction and recovery study of surfactant dry skin. *Arch Dermatol Res*. 1989;281:45-51.
7. Prottey C, Ferguson T. Factors which determine the skin irritation potential of soaps and detergents. *J Soc Cosmet Chem*. 1975;26:29-46.
8. Kawai M, Imokawa G. The induction of skin tightness by surfactants. *J Soc Cosmet Chem.* 1984;35:147-156.
9. Piérard GE, Goffin V, Piérard-Franchimont C. Corneosurfametry: a predictive assessment of the interaction of personal-care cleansing products with human stratum corneum. *Dermatology*. 1994;189:152-156.
10. Faucher JA, Goddard ED. Interaction of keratinous substrates with sodium lauryl sulfate: I. sorption. *J Soc Cosmet Chem.* 1978;29:323-337.
11. Domínguez JG, Balaguer F, Parra JL, et al. The inhibitory effect of some amphoteric surfactants on the irritation potential of alkylsulfates. *Int J Cosmet Soc*. 1981;3:57-68.
12. Rawlings AV, Scott IR, Harding CR, et al. Stratum corneum moisturization at the molecular level. *J Invest Dermatol*. 1994;103:731-741.
13. Inoue I, Miyakawa K, Shimozawa R. Interaction of surfactants with vesicle membrane of dipalmitoylphosphatidylcholine. effect on gel-to-liquid-crystalline phase transition of lipid bilayer. *Chem Phys Lipids*. 1986;42:261-270.
14. de la Maza A, Coderch L, Lopez O, et al. Permeability changes caused by surfactants in liposomes that model the stratum corneum lipid composition. *J Am Oil Chem Soc*. 1997;74:1-8.
15. Misra M, Ananthapadmanabhan KP, Hoyberg K, et al. Correlation between surfactant-induced ultrastructural changes in epidermis and transepidermal water loss. *J Soc Cosmet Chem.* 1997;48:219-234.
16. Draelos ZD, ed. *Moisturizers in Cosmetic Dermatology.* 2nd ed. New York, NY: Churchill-Livingstone; 1995.
17. Frosch PJ, Kligman AM. The soap chamber test. a new method for assessing the irritancy of soaps. *J Am Acad Dermatol*. 1979;1:35-41.
18. Ananthapadmanabhan KP, Subramanyan K, Rattinger GB. Moisturizing cleansers. In: Leyden LJ, Rawlings AV, eds. *Skin Moisturization*. New York, NY: Marcel Dekker, Inc; 2002:406-432. Cosmetic Science & Technology Series; vol. 25.
19. Fluhr W, Bornkessel A, Berardesca E. Glycerol – just a moisturizer? biological and biophysical effects. In: Loden M, Maiback HI, eds. *Dry Skin and Moisturizers.* Boca Raton, FL: CRC Press; 2006:227-244.
20. Lips A, Ananthapadmanabhan KP, Vethamuthu M, et al. Role of surfactant micelle charge in protein denaturation and surfactant-induced skin irritation. In: Rhein LD, Schlossman M,O'Lenick A, et al, eds. *Surfactants in Personal Care Products and Decorative Cosmetics*. Boca Raton, FL: CRC Press; 2006:177-187. Surfactant Science Series; vol 135. 3rd ed.
21. Loeb AL, Overbeek JTG, Wiersema PH, eds. *The Electrical Double Layer Around a Spherical Colloid Particle*. Cambridge, MA: The MIT Press; 1961.
22. Kolusheva S, Shahal T, Jelinek R. Peptide-membrane interactions studied by a new phospholipid/polydiacetylene colorimetric vesicle assay. *Biochemistry*. 2000;39:15851-15859.
23. Evrard D, Touitou E, Kolusheva S, et al. A new colorimetric assay for studying and rapid screening of membrane penetration enhancers. *Pharm Res*. 2001;18:943-949.
24. Pashkovski E, Ngankam PA, Ananthapadmanabhan KP, et al. A new liposome assay for determining lipid damage potential of cleansers. Poster presented at: the 67th Annual Meeting of the American Academy of Dermatology; March 6-10, 2009; San Francisco, CA.
25. Ertel KD, Keswick BH, Bryant PB. A forearm controlled application technique for estimating the relative mildness of personal cleansing products. *J Soc Cosmet Chem*. 1995;46:67-76.
26. Imokawa G, Sumura K, Katsumi M. Study on skin roughness caused by surfactants: II. correlation between protein denaturation and skin roughness. *J Am Oil Chem Soc*. 1975;52:484-489.
27. Uehara M, Takada K. Use of soap in the management of atopic dermatitis. *Clin Exp Dermatol*. 1985;10:419-425.
28. Ananthapadmanabhan KP, Yu KK, Meyers CL, et al. Binding of surfactants to stratum corneum. *J Soc Cosmet Chem*. 1996;47:185-200.
29. Imokawa G. Surfactant mildness. In: Rieger MM, Rhein LD, eds. *Surfactants in Cosmetics*. New York, NY: Marcel Dekker; 1997:427-471. Surfactant Science Series; vol 68. 2nd ed.
30. Rhein LD. In vitro interactions: biochemical and biophysical effects of surfactants on skin. In: Rieger MM, Rhein LD, eds. *Surfactants in Cosmetics*. New York, NY: Marcel Dekker; 1997:397-425. Surfactant Science Series; vol 68. 2nd ed.
31. Rawlings AV, Watkinson A, Rogers J, et al. Abnormalities in stratum-corneum structure, lipid-composition, and desmosome degradation in soap-induced winter xerosis. *J Soc Cosmet Chem.* 1994;45:203-220.
32. Subramanyan K, Wong J, Ananthapadmanabhan KP, et al. Deposition of lipids from personal wash cleansers. Poster presented at the 21st IFSCC Conference; September 11-14, 2000; Berlin, Germany.
33. Ananthapadmanabhan KP, Lips A, Vincent C, et al. pH-induced alterations in stratum corneum properties. *Int J Cosmet Sci.* 2003;25:103-112.
34. Aho S, Harding C, Meyers CL, et al. Soap versus non-soap surfactants: effect on the epidermal activity of β-glucocerebrosidase [abstract]. *J Am Acad Dermatol*. 2006;54(suppl):AB83.
35. Turkoglu M, Sakr A. Evaluation of irritation potential of surfactant mixtures. *Int J Cosmet Sci.* 1999;21:371-382.