Near infrared imaging for measuring and visualizing skin hydration. A comparison with visual assessment and electrical methods

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Abstract. A near infrared (NIR) multispectral imaging method for measuring skin hydration has been applied in a clinical study for estimating skin hydration effects of skin moisturizers and cleansers. The method has been compared to the commercially available, standard electrical methods for evaluating changes in skin hydration based on conductance and capacitance measurements. All of the instrumental methods have been compared to the visual assessment of skin dryness. It has been shown that the NIR imaging method is capable of detecting changes in skin hydration induced by skin moisturizers and cleansers. A large positive hydration change upon treatment by a moisturizing cream was easily detected by all three instrumental methods and by the expert visual assessment of dryness. The imaging technique is rapid, noncontact and noninvasive, and has the additional important advantage of showing the degree of hydration as a function of location, for rapid assessment of change in hydration. There was a clear difference between the instrumental methods when the induced changes were not as great as that from the moisturizing cream. The imaging technique showed more sensitive discrimination between treatments and control, and strong correlation to visual appearance of dryness. © 2005 Society of Photo-Optical Instrumentation Engineers. [DOI: 10.1117/1.1922347]

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1 Introduction

Hydration of the stratum corneum (SC) is a key factor in skin health. It is well known that optimal hydration of the SC is required to maintain flexibility and to facilitate the enzymatic reactions that drive SC maturation.1 Decreased water content impairs the natural desquamation process and leads to the formation of dry scaly skin.² Moisturizing products are typically used to relieve the symptoms of dry skin and the efficacy of these products is determined in part by evaluating their ability to improve skin hydration. In recent years, the moisturization efficacy of cosmetic products has been evaluated based on noninvasive methods that measure changes in the electrical properties of skin. In these methods, the state of skin hydration is determined through correlation with the electrical properties of skin such as conductance and capacitance that change with the water content of skin.^{3–5} While it is widely accepted that these electrical measures directly correlate with skin hydration, there are concerns about several confounding factors such as the effect of other polar molecules in skin, temperature, and surface roughness on the electrical measurements.^{5,6} Additionally, it is evident from the literature that substances or treatments that interact with the keratin-

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water network of the SC can change the electrical properties of skin without actually altering water content.⁵ Commercially available instruments generally use rigid probes in contact with skin where the area of contact with skin is dependent on the applied pressure, thereby introducing an element of operator dependency on the estimation of skin hydration.

Near infrared (NIR) spectroscopic methods, on the other hand, do not suffer from these drawbacks and can be used to directly measure the absorption of NIR light by water in living tissue from its reflectance spectrum. A number of intense absorption bands exist in the NIR spectral region of the skin spectrum that are due predominantly to absorption by water. Figure 1 compares the spectra of water and *in vivo* skin, showing the same major absorption bands at approximately 960, 1200, and 1450 nm. One of many advantages of using NIR is the linear correlation between the NIR absorption intensity and the concentration of water in skin over a wide dynamic range of water concentration. All Richards are also less susceptible to interfering factors such as the deposition of the electrical conducting materials from moisturizing and cleansing products.

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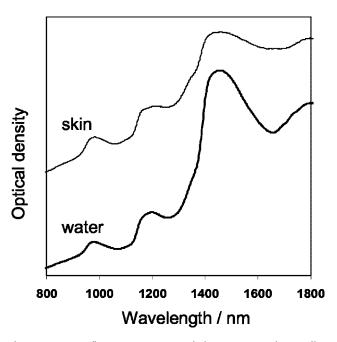


Fig. 1 Near-IR reflectance spectrum of skin in-vivo with an offset spectrum of water for comparison. The water sample contained 12% milk for increased scattering to mimic skin reflectance.

Imaging techniques can also measure skin hydration quantitatively. In addition, it provides a visualization tool that can aid in assessing consumer perception and preference with respect to skin moisturization technologies. Various microscopic skin imaging techniques, including confocal microscopy, fluorescence imaging, and polarized microscopy have been used to measure changes in skin hydration following various skin treatments. Near infrared reflectance-based imaging using a digital camera and tunable filter provides the most direct measure of the overall skin moisture content on the length scale relevant to consumers' perception of the skin condition. It has recently been shown to be able to distinguish between different levels of skin hydration under treatments with a skin moisturizer and artificial dehydration using a solvent. 10–14

The infrared imaging technique does not provide depthresolved information on skin hydration and instead, measures the overall skin hydration. It is well known that near infrared light penetrates deep into the skin, although the depth of penetration is greatly reduced at the water absorption bands compared to that at other wavelengths. Confocal Raman spectroscopy has been recently shown to be a useful tool for measuring the water content of skin as a function of depth.¹⁵

While it is known that skin moisturizers increase SC hydration, it is not clear whether cleansers marketed to have moisturizing properties are able to significantly impact skin hydration levels. Previous evaluations of skin hydration changes using electrical methods have shown decreases in skin hydration inconsistent with measured improvements in visual dryness in the same tests, when evaluating moisturizing skin cleansers.

In the present work, a clinical study was conducted to explore the utility of the NIR imaging method for estimation of skin hydration effects of skin moisturizers and cleansers and to compare this approach to the commercially available elec-

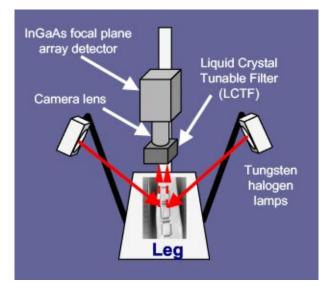


Fig. 2 A sketch of the NIR imaging system. The setup was tilted back so that the camera pointed to the imaging subject horizontally when the outer lower leg was the test area.

trical methods for evaluating changes in skin hydration. In this context, the instrumental methods including the NIR imaging technique, the standard conductance and capacitance methods are compared to the visual assessment of dryness—which is a gross but consumer-relevant evaluation of skin moisturization.

2 Experiment

2.1 The NIR Imaging System

The previously developed, noncommercial near infrared imaging system and its use for skin hydration measurement have been described in detail. 12-14 The system is schematically shown in Fig. 2 and briefly described in the following. It consists of two main components, a digital camera with a 12-bit indium-gallium arsenide (InGaAs) sensor (Sensors Unlimited, Inc., Princeton, NJ) and a liquid-crystal tunable filter (LCTF) (Cambridge Research and Instrumentation, Inc., Woburn, MA). The LCTF was mounted in front of a standard photographic objective lens. It operates over the same wavelength range as the camera sensor (960-1700 nm) and has a nominal bandpass of 6 nm. A filter wheel with a set of fixed narrow bandpass filters have also been used in the place of LCTF. Custom software was written using the LabView programming environment (National Instruments, Austin, TX) to synchronize the camera and the tunable filter (or filter wheel) and to provide an interface for images to be acquired and displayed in near-real time. The software was modified from the previous version^{10–14} to adapt to the new computer operating system and some changes in the newer camera settings and the filter-wheel controller.

In the feasibility study reported previously, ^{12–14} each image set contained 54 images spanning the wavelength range from 1100 to 1630 nm in 10 nm increments. Acquisition time for the entire data set took over 60 s. In addition, the 1450 nm band was shown to be the only band that could be used to detect a change in hydration in the superficial layer of the



Fig. 3 The marking of the test sites. The leg was placed behind a window for NIR imaging measurement; the white labels were placed in the gaps separating test sites prior to collecting images.

skin.¹⁰ Therefore, the NIR images collected in our current study were only taken across the most intense water band at seven preselected wavelengths accessible by the imaging system. These preselected wavelengths used in this study were 1280, 1390, 1410, 1430, 1450, 1470, and 1620 nm. It took approximately 12 s to collect the entire set of seven images using ten individual 16.67 ms exposures summed for each of the seven wavelength images. A reference image set of a Kodak white card was also taken using the same measurement parameters.

2.2 Clinical Design

A 6 h time-moisturization test¹⁶ of a single application of product was performed on the outer lower legs of female subjects. An informed consent was obtained from each subject and the study procedure was IRB-approved prior to the start of the study.

Before the start of the product application, all subjects entered a 5 day in-home conditioning phase. They were asked to use a mild cleansing bar for all general cleansing. Subjects were also asked to refrain from using any other skin products (creams, lotions, moisturizers, sunscreens, etc.) and washing appliances (sponge, wash cloth, loofah, etc.) on or near their lower legs. Upon completion of the conditioning phase, 14 subjects remained who met the inclusion/exclusion criteria and had dryness scores of 1.0–2.0 (see later for the grading scale) and erythema scores no greater than 2.0 on their lower legs entered the 6 h time moisturization test.

Each subject had their outer lower leg divided and marked with a skin marker pen into three-test sites of approximately 6 (height)×7 (width) cm² each, as depicted in Fig. 3. Four treatments—moisturizing cream, water, traditional body wash, and premium moisturizing body wash (see Sec. 2.5 later for description) were applied in a rotated, balanced, fashion across the six test sites (upper, middle, and lower on both left and right legs). Each leg also had an untreated control site. Study clinicians then performed a single application of 0.20 mL of a liquid cleanser to each test site for a 30 s wash, 15 s rinse, and pat dry wash regime. For moisturizer, 0.20 mL

Table 1 Clinical grading of skin dryness.

Grade	Observations
0	No flakes
1	Slightly flaking/uplifting of flakes (patchy) and/or powdered appearance)
2	Moderate flaking/uplifting flakes (uniform) and/or slight scaling
3	Severe flaking/uplifting, uplifting of scales, and/or slight fissuring
4	Severe scaling/uplifting scales with severe fissuring/cracking

was glove-rubbed into the test site until completely absorbed. A similar procedure was followed for the water application.

The following assessments of skin condition were made at base line (just prior to the product application), and 1, 3, and 6 h postproduct application: (1) expert visual evaluation of dryness on each test site; (2) NIR imaging of each leg using the above NIR imaging system; and (3) skin conductance and capacitance measurements on each test site.

2.3 Visual Assessment and Instrumental Measurement

A trained expert evaluator performed a visual assessment of dryness for each test site. They were each graded on the scale of 0–4 in 0.5 increments (nine-point grading scale) at each of the four time points. Table 1 shows the grading scale for visual dryness. A reduction in dryness score means improved skin condition. After the baseline visual grading, the subjects were acclimatized in a temperature (21±1 °C) and humidity (38%±5% relative humidity) controlled room for 20 min prior to the baseline instrument measurements. The subjects then stayed in the room for the duration of the study.

Following visual assessment, NIR images were taken of each leg with the leg placed behind a window, as shown in Fig. 3. There were four imaging sessions at four time points specified earlier and two sets of images in each session (one from the left leg and one from the right leg).

Conductance measurements were taken using a Skin Hygrometer Skicon 200 with MT8-C probe. Capacitance measurements were taken using a Courage-Khazaka Corneometer-CM 820PC.⁵ An increase in probe reading's values for each instrument corresponds to an increase in skin hydration. Three replicate readings were taken for each probe at each test site at all measurement time points. All electrical measurements were made immediately after the NIR imaging sessions.

2.4 Data and Image Analysis

The image data were processed using in-house routines written in Matlab (The MathWorks, Natick, MA). The first step in the processing sequence was to convert the raw reflection intensity data to optical density format. This consisted of first calculating the ratio of the pixel intensity in the subject to the intensity of the corresponding pixel in the image of the reference card, and then taking the negative logarithm of this ratio.

This process is equivalent to the conversion of a single beam spectrum to an absorbance spectrum in infrared spectroscopy. Further processing was performed on optical density data.

Before calculating the O-H absorption intensity, the spectrum at each pixel was modified by subtracting a linear background drawn between the spectral points at 1280 and 1620 nm. The integrated O-H absorption intensity was calculated as the sum of the intensity values at the remaining five wavelengths near the band center at 1450 nm. These corrected intensity values were used to generate a false color image referred to as the hydration image.

To visualize the changes in skin hydration upon product treatment, a difference image was generated by subtracting the hydration image before product application (baseline) from the post-application image, a procedure similar to that used in the literature. $^{10-14}$

A rectangle area in the center of the test site was used to obtain the average pixel intensity, which is reported as the NIR score for the test site. The change-from-baseline (CFB) scores were obtained by subtracting the base line NIR score from the scores of the post-product application. A small change in the untreated control site was also subtracted from those of the treated site within each image. A positive change upon product treatment corresponds to an increase in water absorption and, hence, increased skin hydration, while a negative change indicates just the opposite.

For the conductance and capacitance data, the average responses were obtained from three replicate readings.

Within-product and between-products comparisons were performed based on the changes in the visual dryness, NIR scores, conductance and capacitance readings of the post-product applications from their corresponding baseline values. Comparisons were made independently at each time point and no temporal correlation was taken into account. The Wilcoxon Signed Rank test¹⁷ with the Pratt–Lehman adjustment¹⁸ for ties was used for the nonparametric variables of the visual grades and t test for the parametric variables of the instrumental measurements. Based on the study design, the α level of 0.10 was chosen for the analysis of statistical significance. Statistical analysis was performed using JMP (version 5, SAS) and Clinskin, a SAS-based in-house data analysis package.

2.5 Materials

Four products were used in this study: moisturizing cream, water (control), and two liquid cleansers. The moisturizing cream was a commecially available product. The liquid cleansers consisted of two main types of marketed liquid body washes, a traditional body wash (low to no emollients), and a premium moisturizing body wash (high level of emollients). Examples of the main ingredients from each liquid cleanser type are shown as follows: traditional body wash (TBW): water, sodium laureth sulfate, cocamidopropyl betaine, sodium lauryl sulfate, fragrance, thickener salts, preservatives, color, and moisturizing body wash (MBW): water, sunflower oil (emollient), sodium laureth sulfate, cocamidopropyl betaine, glycerin, petrolatum, foam booster, fragrance, thickener salts, preservatives.

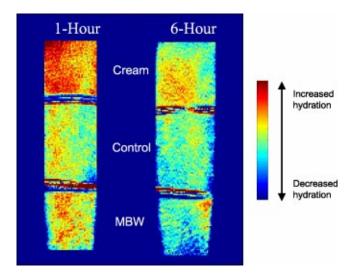


Fig. 4 Sample NIR difference images showing the CFBs in O–H absorption intensity on the same leg at 1 h (left panel) and 6 h (right panel) post-product applications.

3 Results and Discussion

3.1 *Visualizing Hydration Changes*

An example of the difference hydration images is shown in Fig. 4. These false color images represent the changes in skin hydration on the same leg between 1 h (left panel) and 6 h (right panel) after product applications and their baseline image, respectively.

The 1 h post-treatment image clearly indicates increased water content in both moisturizing cream treated site (upper site) and MBW treated site (bottom site), compared to the control site in the middle. However, the increased water content at the MBW site disappeared mostly by 6 h after treatment, while the cream site still maintained a significant level of increased water content, although reduced from the 1 h level. This is expected because the moisturizing cream is a leave-on product that usually provides a longer-lasting benefit than a wash-off product (MBW, a cleanser).

3.2 Hydration Changes from Baseline as Measured by all Four Evaluation Methods

The average responses over all subjects (N=14) from visual evaluation, NIR imaging, conductance, and capacitance are compared in Fig. 5. The error bar indicates the standard error of the mean (SEM). The mean differences of the dryness scores in Fig. 5(a) are provided for descriptive purposes.

Each evaluation method detected different changes in skin hydration upon product application. The visual evaluation [Fig. 5(a)] showed reduced skin dryness, i.e., improved skin hydration, for all products at all three time points except for water. For the moisturizing cream, the skin hydration improvements were all significant ($p \le 0.001$) at all three time points. For MBW, the changes were also detected from the corresponding baseline evaluation with p values equal to 0.08, 0.01, and 0.15 at the 1, 3, and 6 h time points, respectively. For TBW, the positive change was significant at the 1 h time point (p = 0.01) and insignificant at other two time points (p > 0.1). For water, a statistically insignificant increase in

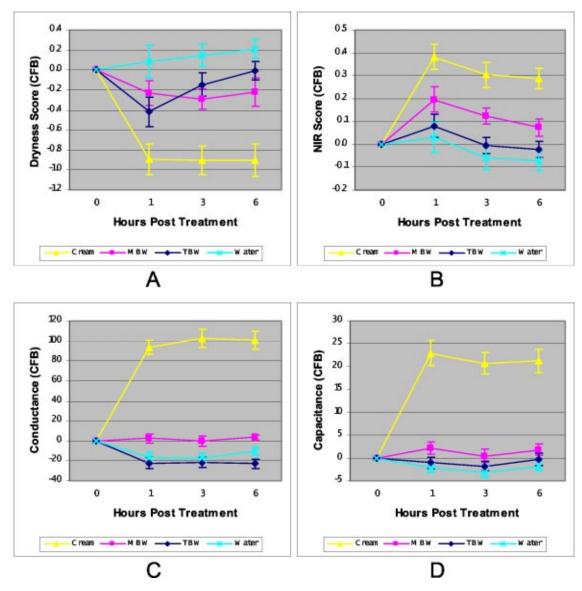


Fig. 5 Time responses as measured by each technique: (a) visual evaluation of dryness; (b) the NIR imaging technique; (c) the conductance measurement by Skicon; and (d) the capacitance measurement by Corneometer. The error bars indicate the standard error of the mean (±SEM). See text for details on statistical significance of each measurement point.

dryness was observed in the first 3 h (p>0.1) with a significant increase in dryness 6 h after product application (p=0.06).

The NIR imaging method gave a very similar result [Fig. 5(b)]. For the moisturizing cream, an increased skin hydration was measured at all three time points ($p \le 0.001$). For MBW, a positive change was observed with the p values equal to 0.004, 0.004, and 0.07 at the 1, 3, and 6 h time points, respectively. For TBW and water, the changes in skin hydration were all small with p = 0.13 for TBW at 1 h time point and 0.11 for water at 6 h time point. These results are in general agreement with the observations from the expert evaluation of dryness.

However, the two electrical methods [Figs. 5(c) and 5(d)] showed less consistent results with each other and with the expert evaluation of dryness with the exception for the moisturizing cream. For the cream, the values of both electrical

properties increased significantly ($p \le 0.001$) upon product application at all measurement time points. For MBW, both methods showed no significant changes (p > 0.1) in their measured properties at any time point, while the visual evaluation and NIR both showed a positive benefit. For TBW and water, both conductance and capacitance were reduced at all time points. The changes in conductance [Fig. 5(c)] were all significant ($p \le 0.02$) for both TBW and water at all three time points. However, the change in capacitance [Fig. 5(d)] were only significant (p < 0.05) for water at the 1 and 3 h time points and insignificant (p > 0.10) for water at the 6 h time point and for all TBW measurements.

Clearly, all these evaluation methods are able to detect a large positive hydration benefit provided by the moisturizing cream. For body wash cleansers, the induced effects are smaller. While both body washes seem to have a similar effect via visual assessment with a slightly longer lasting benefit from MBW, clearly the MBW is seen to be more hydrating by the NIR measurement. This is consistent with the fact that the MBW contains a high level of moisturizing emollients that are delivered on to the skin during wash. It is interesting that washing with water alone leads to a low level dehydrating effect and this can be attributed to the fact that some of skin's natural moisturizers may be leached away by water.

3.3 Product Differentiation

Product comparisons were made between all possible pairs of the products tested independently at each time point. Figure 5 gives a qualitative view of how products are differentiated by each of the four techniques. Statistically, the moisturizing cream stands in its own class, providing a large positive hydration benefit, as measured by all four methods.

The expert evaluation of dryness separated water from the cleansers, but only showed an insignificant difference between MBW and TBW. The NIR method placed TBW and water in the same category and separated MBW by itself at the 3 and 6 h time points.

The conductance measurement ranked the products in a similar way, placing the moisturizing cream as the best to provide a moisturizing benefit and MBW better than TBW and water. However, this method ranked TBW and water directionally in the reversed order.

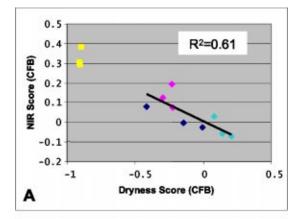
The capacitance measurement provides the least product differentiation. It could only differentiate the moisturizing cream from the other three products and somewhat differentiated between MBW and water at the 1 h time point. At all other time points, there is no product differentiation between MBW, TBW, and water.

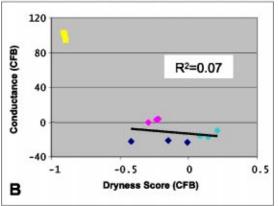
Therefore, for this set of products all measurement techniques are clearly in good agreement in distinguishing moisturizing cream from the cleansers, but only the imaging technique ranked the cleansers and water in the same way as the visual assessment.

3.4 Comparing the Instrumental Methods for Evaluating Skin Hydration

The results from the three instrumental techniques have been compared earlier in two ways, the within-product difference, i.e., the difference between the individual treatments and their corresponding baseline, and the between-product difference for product differentiation. In both cases, all methods detected the large difference in skin hydration induced by the moisturizing cream. However, they showed different abilities in detecting smaller changes from other products. In particular, the NIR imaging technique clearly detected a significant hydration benefit from MBW, while the capacitance method showed a positive benefit qualitatively without a statistical significance. The conductance method showed no benefit at all. In ranking product performance, the imaging and capacitance methods showed the same order, but the conductance had two products in the reversed order. The statistical significance in ranking the product is also different between these instrumental methods.

The visual assessment of dryness is relevant to consumers' perceived status of their skin health. Therefore, it is important to evaluate the relative usefulness and sensitivity of the instrumental assessment of the skin hydration by comparing them





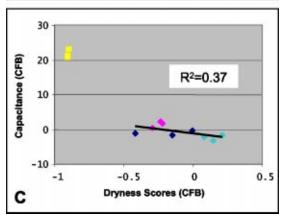


Fig. 6 The correlation between instrumental evaluations and visual assessment of skin hydration; (a) NIR scores versus visual grades, (b) conductance versus visual grades, and (c) capacitance versus visual grades. The data points have been color coded to match those in Fig. 5. The solid straight line was fitted with all points except the yellow ones of the moisturizer.

with the visual grading. As stated earlier, the nonparametric treatment was applied to the visual grades, while the parametric treatment was applied to the instrumental data. However, it is also within common practice to use parametric methods for the grading data with nine-point ordinal scales. In fact, they gave essentially the same statistical comparisons of the product performance from the visual grades in this study. Therefore, the mean values from the visual grades are used for a correlation analysis with the instrumental data.

Figure 6 shows the correlation between the visual dryness

and the instrumental measures of skin hydration, color coded to match those in Fig. 5. The linear fit was performed on the data points excluding those of the moisturizer. It is evident that the NIR responses correlate well with the clinical grades [Fig. 6(a)] with $R^2 = 0.61$ and this correlation seems to fit the large responses from the moisturizer nicely (if extrapolating the fitted line), indicating a wide range of agreement between these two evaluation methods. The conductance showed no correlation with the clinical grades [Fig. 6(b)] with $R^2 = 0.07$. The capacitance displayed a weak correlation with the clinical grades [Fig. 6(c)] with $R^2 = 0.37$. Neither electrical method indicated that the large responses from the moisturizing cream fit the pattern of the responses from the cleansers.

It should be pointed out that the correlations presented in Fig. 6 were not intended as predictive of one measure from another. They do, however, show some important differences between the instrumental methods. The visual grading and NIR imaging measure hydration related properties of the skin in a similar way, while the same is not true for the electrical methods. The NIR imaging technique is shown to be a better tool for evaluating skin hydration as related to the visual appearance of the skin than are the electrical measures.

4 Conclusion

A large positive hydration change upon treatment by the moisturizing cream was observed and easily differentiated from the effect of the cleansers by all four techniques used in this study. However, there was a clear difference between instrumental methods when the induced changes were not as great as that from the moisturizing cream. The results observed from the NIR imaging technique are more consistent with those of the visual grades of dryness.

The current application of the NIR imaging technique has shown that it is capable of detecting changes in skin hydration induced by skin moisturizers and cleansers. The technique is rapid, noncontact, and noninvasive, and has the additional important advantage of showing the degree of hydration as a function of location, for rapid assessment of change in skin hydration.

Conductance and capacitance-based methods undoubtedly measure quite different aspects of the skin condition. Compared to these electrical methods and previously reported probe-based NIR methods, the imaging technique provides many advantages for objective assessment of skin hydration. It showed more sensitive discrimination between treatments and control and strong correlation to visual appearance of skin dryness.

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