

Detection and visualization of intracutaneous allergic type-specific elements using long-wavelength near-infrared hyperspectral imaging

Ken Nishino¹, Toshiharu Fujiyama², Hideo Hashizume³ and Shigeki Nakauchi¹

¹Department of Computer Science and Engineering, Toyohashi University of Technology, Toyohashi, Japan,

²Department of Dermatology, Hamamatsu University School of Medicine, Hamamatsu, Japan and ³Division of Dermatology, Shimada Municipal Hospital, Shimada, Japan

Background/purpose: This study aimed to develop a method for the assessment of allergic dermatitis by using the long-wavelength near-infrared spectrum (more than 1000 nm) to detect intracutaneous allergic type-specific elements. Such a method was realized by establishing a spectral classifier for the spectra of type I and type IV allergic dermatitis reactions.

Methods: Near-infrared spectral images of histamine-induced cutaneous reaction (type I) and contact hypersensitivity erythema elicited by squaric acid dibutylester (SADBE; type IV) were obtained, and the absorption spectra of normal and inflamed skin were extracted from these spectral images. A spectral classifier was established from these training datasets, and it was then applied to two test cases, red flare by methyl nicotinate (normal) and metal allergy (type IV).

Results: The spectral classifier established by canonical discriminant analysis (CDA) achieved very accurate detection (normal: 87.67%, type I: 87.00%, type IV: 98.5%). Furthermore, the test cases were also correctly classified: the red flare induced by methyl nicotinate was categorized as normal skin and the metal allergy was categorized as a type IV allergic reaction.

Conclusions: These results suggest a possible application of near-infrared spectral imaging to the assessment of allergic dermatitis.

Key words: allergic dermatitis – visualization – hyperspectral imaging – near-infrared

© 2012 John Wiley & Sons A/S

Accepted for publication 26 April 2012

ACCORDING TO the Gell and Coombs classification system, allergic dermatitis can be categorized as either type I (immediate type) or type IV (delayed type) based on its mechanism of development (1). However, it is difficult to determine the causative type of hypersensitivity response by the visible information such as color and morphological characteristics of the affected area. Dermatologists therefore make a diagnosis using a combination of visual inspection, a medical interview, palpation, and their clinical experience, and they must sometimes rely on the results of skin biopsy, an invasive method. However, a skin biopsy requires anesthesia and carries the risks of infection and scar formation. A noninvasive inspection providing an objective evaluation index would therefore be highly beneficial as an alternative or precursor to a skin biopsy.

Hyperspectral imaging has been rapidly evolving in recent years as a nondestructive

method for measuring the grossly invisible physical and chemical properties of objects. The light reflected from an object's surface is known to contain a great deal of information about its source. Many applications of spectroscopy and spectral analysis have been reported. A hyperspectral image, which is an image that has spectral data for each pixel, allows such spectroscopy and spectral analysis research to expand from a single-point measurement to visualization of the distribution of measured properties. Most recently, image sensors for long-wavelength near-infrared light (e.g., MCT, InGaAs) and hyperspectral imaging devices using these sensors have been developed. This wavelength range is especially effective for measuring the composition of an object because it includes the absorption spectra of molecular vibrations. Therefore, this technique is a promising new nondestructive visualization method that could be applied in a wide variety of fields. In this study,

long-wavelength near-infrared spectral imaging was used to evaluate allergic dermatitis.

The nondestructive and noncontact nature of optical measurements has long attracted research on their applications to biological targets. Hyperspectral imaging has the same advantages and has been reported to be useful for evaluating dermatitis. Human skin pigments have spectral absorption properties in the visible range that produce human skin color (2). The saturation levels of melanin and hemoglobin are the main contributors to skin color, and the red flare and melanin pigmentation developed by concentration of these pigments are actually quite important in dermatological diagnosis. In order to measure these human skin pigments, Tsumura et al. proposed a method for visualizing the distributions of melanin and hemoglobin from cross-polarized RGB images taken with polarization films installed on both the RGB camera and the illuminant (3). In contrast, dermoscopy, which is a form of microscopy used for contact measurement, is well-known as a powerful medical technique for capturing images of the subsurface anatomic structures of the epidermis and papillary dermis. This technique greatly aids in the diagnosis of skin diseases, especially melanoma, because the morphology of the skin lesion as characterized by the distributions of its pigments is readily discerned in the dermoscopic image (4–7). The spectrophotometric intracutaneous analysis (SIA) scope was developed as a medical instrument combining dermoscopy and spectral imaging technologies (8). This technique allows spectrophotometric analysis of the dermoscopic features and visualization of internal melanin and hemoglobin distribution and collagen holes. The SIA scope is more sensitive than dermoscopy alone for detecting melanoma. On the other hand, spectrophotometric assessments of allergic dermatitis have been performed not only in the visible wavelength range for the quantification of oxyhemoglobin pigments but also in the near-infrared wavelength range for the evaluation of water content. Detection and quantification of the water contained in an object using near-infrared spectroscopy is a conventional technique, as infrared and near-infrared reflectance spectra are normally strongly affected by the spectral absorption properties of the hydroxyl group. Therefore, there are many reports on the estimation of

water content *in vivo* and the applications thereof, including the evaluation of lung edema, detection of interactions between atopic skin and pharmaceutical additives, assessment of the nasal mucosa for swelling, and measurement of the water content in the stratum corneum (9–14). For the assessment of allergic dermatitis, Stamatas et al. proposed a method for evaluating erythema and edema in which oxyhemoglobin and water content distributions were visualized by visible and near-infrared spectral imaging (15–17).

However, the earlier studies allowed only the evaluation of red flare and swelling, which can be assessed by visible inspection and palpation. Therefore, the previously proposed techniques have not attempted to determine the type of allergic response (immediate or delayed), a key element in dermatologic diagnosis that at present can often be obtained only by a skin biopsy.

The purpose of this study was to develop a noninvasive method for assessing allergic dermatitis by using the long-wavelength near-infrared spectrum to detect intracutaneous allergic type-specific elements. As mentioned above, in general type I and type IV allergic dermatitis reactions have internal pathological differences as described in Fig. 1. In particular, the erythema and edema in type I reactions are developed by IgE, mast cells, and particular chemical mediators, while type IV reactions are induced by macrophage and lymphocyte infiltration. Our plan to realize this measurement method was to develop a spectral classifier for the long-wavelength near-infrared spectra of the three

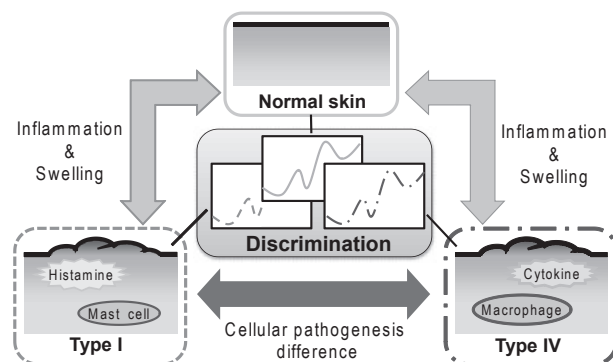


Fig. 1. Differences between normal skin and two types of allergic dermatitis. Both type I and type IV dermatitis produce red flare and swelling. In contrast, the internal compositions of these types of dermatitis, such as inflammatory agents and mediators, are different. This study aims to develop a spectral classifier for the long-wavelength near-infrared spectra of the three skin conditions.

skin conditions shown in Fig. 1: normal skin and types I and IV allergic dermatitis. Briefly, near-infrared spectra are affected by the absorption properties of molecular vibrations of not only hydroxyl groups but also hydrocarbons. Furthermore, long-wavelength near-infrared spectra are very suitable for analyzing absorption properties as there is less light scattering in this range than at shorter wavelengths (18). Long-wavelength near-infrared spectra of allergic dermatitis lesions may therefore contain information that is useful for discriminating among the different types. However, the spectral differences caused by oxyhemoglobin accumulation and swelling alone will not suffice for the differentiation of type I and type IV dermatitis, as both types involve edema and erythema. For these reasons, any possible spectroscopic method for discriminating between these forms of dermatitis must rely on detection of intracutaneous allergic-type specific targets (e.g., accumulation of macrophages) that could aid in the differential diagnosis of allergic dermatitis.

In this study, a near-infrared spectral classifier was established using the spectral dataset of artificially-induced allergic dermatitis, and this classifier was then applied to the assessment of a metal allergy patch test. The patch test for metal allergy is often used in clinical practice because it is one of the few effective noninvasive inspection methods for the assessment of type IV dermatitis. However, it is not easy to make a correct diagnosis based on the patch test because the diagnosis relies on visual information such as the shape, color, and size of the flare reaction. In practice, there are occasionally false reactions, either false-positive reactions induced by the irritant reagent or false-negative reactions with inflammatory cell infiltration in the absence of red flare. Therefore, a method for detecting and classifying allergic dermatitis lesions would be a powerful tool for the evaluation of patch test results.

Materials and Methods

In this study, near-infrared spectra of erythematous artificially-produced urticarial (type I) and contact hypersensitivity (type IV) lesions were measured, and a classifier of near-infrared spectra in three skin conditions, two erythematous lesions and normal skin, was established to detect the spectral differences caused by the

distinct immune responses. Previously existing techniques were not able to classify these spectra because these techniques focused only on the inflammation and swelling, whereas differential diagnosis of urticaria and contact hypersensitivity by near-infrared spectra will probably require detection of spectral differences caused by the specific mediators of the immune responses.

Near-infrared spectral images of four cases of artificially produced reddened skin were obtained in this study.

Case 1: Contact hypersensitivity induced by squaric acid dibutylester (SADBE)

Case 2: Urticaria induced by a histamine prick test

Case 3: Flare reaction induced by methyl nicotinate

Case 4: Contact hypersensitivity induced by a metal allergy patch test

Near-infrared spectral images from case 1 and case 2 were used as the training dataset for establishing the classifier. In this study, CDA was used as the classification method. Case 3 and case 4 were used as the test cases. The flare reaction induced by methyl nicotinate should be classified as 'normal skin' because it is not an allergic reaction. Case 4 should also be classified as contact hypersensitivity.

Hyperspectral image measurement conditions

Near-infrared hyperspectral image

Figure 2 shows the near-infrared spectral imaging system that was used to obtain the hyperspectral images of allergic dermatitis. A near-infrared line-scan hyperspectral camera, Spectral Camera SWIR (Specim) is mounted in the system, and the measurement target is placed on the movable plate under the camera. This camera has a 2-dimensional MCT sensor and can acquire an image in which the axes are position in the scanning area and wavelength. The spectral dataset of a linear scanning area obtained by measurement over a definite period of time can be reconstructed as a hyperspectral image because the measurement area plate slides with a constant velocity. The wavelength range for the measurement was 1000–2400 nm with 10 nm steps, the image resolution was 0.2 mm/pixel, the width of the scanning area was 320 pixels, and the height of the image was

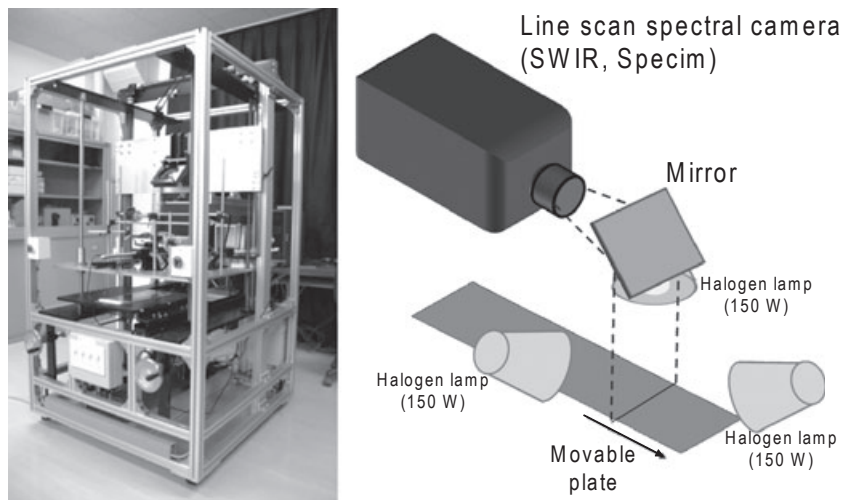


Fig. 2. Near-infrared hyperspectral imaging system. Photos of imaging system (left) and the measurement geometry (right). A near-infrared line-scan spectral camera SWIR (Specim) is mounted in the system and the measurement target is placed on the movable plate under the camera.

defined as up to the size of the measurement target. Three 150-W halogen lights were installed around the scanning area. The camera was calibrated using the Spectralon white reference.

Visible hyperspectral image

To determine the relationships between the detection results and accumulation of oxyhemoglobin in the lesions, hyperspectral images in a range of visible wavelength were obtained in cases 2, 3 and 4. A multispectral imaging system, Nuance (CRi, Vis), was used for these measurements. The wavelength range of the hyperspectral image was 420–720 nm in 5 nm steps. A 300-W halogen light was used as the illuminant and the measurement was performed with 45/0 degree geometry. The measurement results are reported in the Discussion section.

Clinical protocol

Contact hypersensitivity induced by SADBE

The induction of contact hypersensitivity by SADBE was performed on the left forearm of a 24-year-old man. First, sensitization was performed by applying SADBE diluted to 1% with acetone to the subject's left forearm. The size of the application area was 3 cm × 10 cm. One hour after the SADBE application, the application area was washed. Six hours later, a slight red flare had developed, and this progressed to intense irritation after 12 h. Twenty-four hours after application, the irritation had disappeared and pigmentation had appeared. A papular

rash developed 9 days after the sensitization. The papular rash then spread over the application area, and edema developed. Near-infrared hyperspectral images were acquired on 1, 4 and 5 days after the edema development.

Histamine-induced urticaria

The histamine prick tests were performed on the right forearms of three healthy adult men (1 aged 20–30 and 2 aged 30–40), and hyperspectral images of the resulting urticaria were obtained. The prick test patch Prick-Lancett (EWO Care AB, Gislaved, Sweden) was used with the histamine diluted to 1 mg/mL. Ten minutes after the prick test, an approximately 5-mm wheal surrounded by erythema developed. Both wheal and erythema disappeared within an hour. Therefore, hyperspectral images were measured 30 min after the application of the prick test.

Rubefaction by methyl nicotinate

A methyl nicotinate solution was applied to 2 square areas (1.5 cm × 1.5 cm) on the right forearm in order to produce a red flare. This experiment was performed on a 20–30-year-old male. In this rubifying procedure, methyl nicotinate solution was used at a concentration of 10%. The hyperspectral image was measured 10 min after the application

Metal allergy

The metal allergy patch test was applied to the right forearm of a healthy adult male (Fig. 3). Sixteen kinds of metal allergen from Torii

Pharmaceutical Co., Ltd. (Tokyo, Japan) were used in this test. Patch tests with 17 substances (16 allergens and Vaseline) were left in contact with the arm for 48 h. The substances included are described in Table 1.

Results

Measurement results

Reflectance spectra of both allergic dermatitis responses and normal skin were extracted from the measured hyperspectral images of cases 1 and 2 (contact hypersensitivity and histamine-induced urticaria), and spectral datasets were constructed. For each image, masks of dermatitis and normal skin area were created manually. The average spectra of the measured absorbance spectra sets are shown in Fig. 4. Here, the multiplicative scattering correction (MSC) was applied on the reflectance domain with using the average of all extracted spectra as the reference. As shown in the figure, the absorbance spectrum of the SADBE-induced lesion is obviously different from that of the others. The cause of the spectral difference may be the severity of the edema rather than differences in the immune reaction, as the severities of the two dermatitis responses were clearly different.

Development of the classifier

The purpose of this experiment was to obtain indices for independently evaluating the histamine-induced urticaria and SADBE-induced contact hypersensitivity. To that end, CDA was

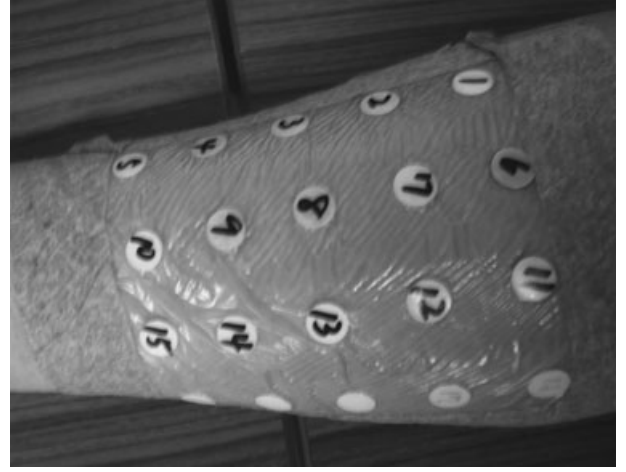


Fig. 3. Metal allergy patch test.

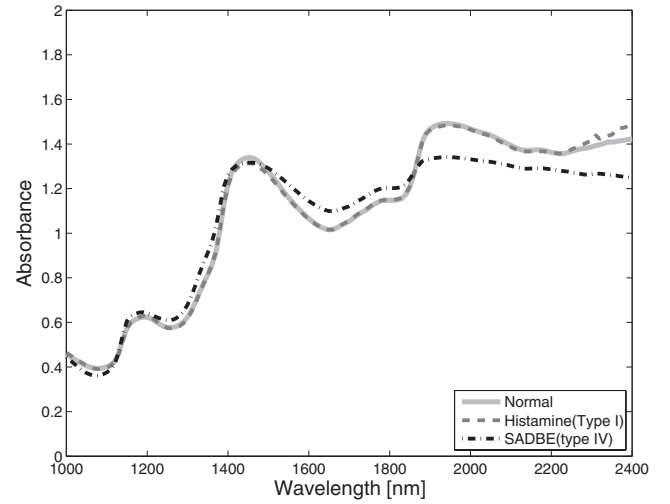


Fig. 4. Absorbance spectra of three skin conditions: contact hypersensitivity induced by SADBE, urticaria induced by histamine prick test, and normal skin.

TABLE 1. Description of the metal allergy patch test

No.	Substance
1	Aluminum chloride 2%
2	Chloroauric acid 0.2%
3	Stannic chloride 1%
4	Ferric chloride 2%
5	Chloroplatinic acid 0.5%
6	Palladium chloride 1%
7	Indium trichloride 1%
8	Indium tetrachloride 1%
9	Cobalt chloride 2%
10	Potassium bichromate 0.5%
11	Chromium sulfate 2%
12	Copper sulfate 1%
13	Nickel sulfate 5%
14	Zinc chloride 2%
15	Manganese chloride 2%
16	Silver bromide 2%
17	Vaseline

performed on the near-infrared spectra of both dermatitis lesions and normal skin. The CDA creates a discriminant space based on maximizing the variations between subgroups. Therefore, if it is possible to classify these three conditions by projection of the near-infrared spectra to the canonical discriminant space, the canonical discriminant scores will allow independent evaluation of these dermatitis responses. In other words, such a result suggests that the discriminant vector detects some sort of spectral difference caused by the different immune reactions.

Computational results

To construct the training datasets, 500 spectra were extracted from both of normal skin area

and allergic dermatitis lesion in a hyperspectral image using boot-strap resampling. In total, 6000 spectra were extracted from all images (1500 spectra of urticaria, 1500 spectra of contact hypersensitivity and 3000 spectra of normal skin) to construct the training dataset. The validation of the accuracy of the CDA was performed by n -fold cross-validation. The spectral datasets consist of spectra extracted from six hyperspectral images (three of urticaria and three of contact hypersensitivity). Therefore, the datasets were divided into six groups according to the hyperspectral images, and n -fold cross-validation was performed using $n = 6$. Here spectral wavelength range was limited to 1250–1870 nm because it showed the highest discriminant accuracy through the all-possible search.

The canonical score map obtained by the CDA is shown in Fig. 5. The ovals in the figure show the equiprobability ellipses, the shafts of which were defined by the standard deviation under the 2-dimensional normal probability distribution assumption.

As shown in Fig. 5, the spectral sets of the three conditions are clearly separated in the canonical discriminant space. Note that the directions of the score shifts from normal skin to hypersensitivity and normal skin to urticaria are nearly orthogonal. This result indicates that the canonical discriminant space allows the creation of independent evaluation indices for each dermatitis response. To obtain the evaluation indices, a quadratic Bayesian classifier for

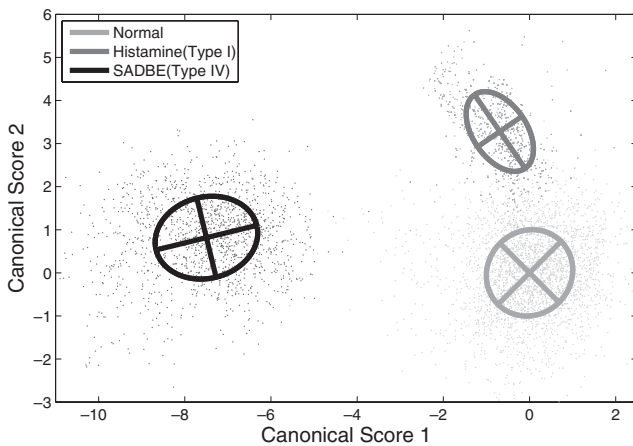


Fig. 5. Canonical score map. Canonical scores of all spectra were computed using the canonical discrimination function and the scores of each skin type were plotted on the canonical space. The ovals in the figure show the equiprobability ellipses, the shafts of which were defined by the standard deviation under the 2-dimensional normal probability distribution assumption.

these three skin conditions was created on this space. The rates of correct classification are shown in Table 2. These were computed using 6-fold cross-validation.

Visualization results

The developed canonical discrimination function was applied to the spectral images to visualize their dermatitis lesions. Here, the near-infrared hyperspectral images used to construct training datasets were also used for visualization.

The visualization results are shown in Figs. 6 and 7. The posterior probabilities of dermatitis defined by the canonical score distributions in Fig. 5 were computed for all pixels of the spectral images, and the dermatitis lesions were colored. An MSC and masking of the background were applied as preprocessing steps, and a moving average filter (15 by 15 pixels) was applied to the canonical score map. In these figures, a probability of contact hypersensitivity is colored red, a probability of urticaria is colored green, and other areas are not colored. As shown in Figs. 6 and 7, the dermatitis lesions were detected perfectly.

Classification and visualization of test cases

As described above, near-infrared spectra of normal skin, skin with hypersensitivity, and urticaria can be classified very accurately by the system we developed. We suspected that the discrimination function detected some pathological difference between these dermatitis reactions other than inflammation and swelling. Therefore, two additional cases, rubefaction by methyl nicotinate and a metal allergy test, were used to support this hypothesis.

Rubefaction by methyl nicotinate

Methyl nicotinate induces a flare reaction with increased blood flow, but no edema is formed

TABLE 2. Rates of correct skin condition classification

Skin condition	Correct rate (%)
Normal skin	87.67
Histamine (Type I)	87.00
SADBE (Type IV)	98.53
Average	90.22

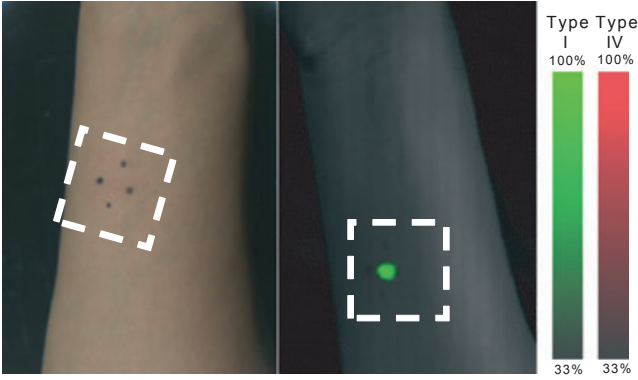


Fig. 6. Visualization result of histamine-induced urticaria. The left image shows the RGB color image and the right the detection result. The color image was computed from the spectral image, which was measured by the Nuance (CRi, Vis) multispectral imaging device. The dermatitis lesion is indicated by the rectangle.

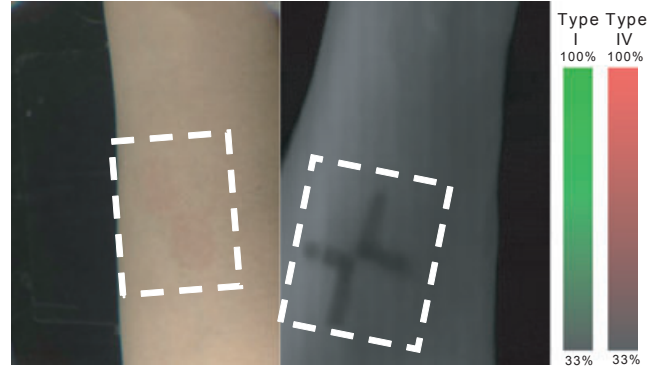


Fig. 8. Visualization result of red flare reaction. The left image shows the RGB color image and the right the detection result. The color image was computed from the spectral image at visible wavelength. Flare reaction areas are indicated by the rectangle. The black line in the detection result was the marker of the flare activation spot.

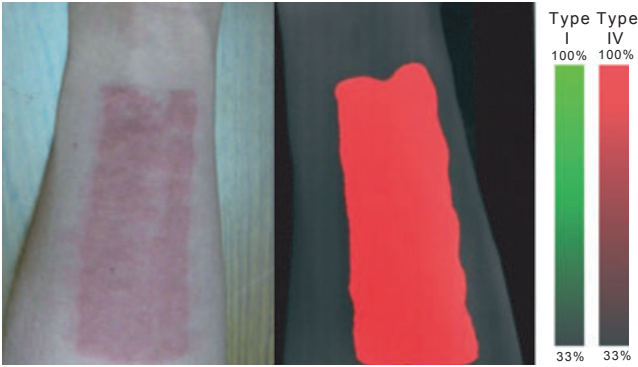


Fig. 7. Visualization result of SADBE-induced contact hypersensitivity. The left image shows the RGB color image and the right the detection result. The color image was computed from the spectral image, which was measured by the Nuance (CRi, Vis) multispectral imaging device.

and no inflammatory cytokines are released, so this lesion should be classified as normal skin.

To confirm the accuracy of the discrimination function for this skin condition, the canonical discrimination function (developed above) was applied to the near-infrared spectral image of rubefaction by methyl nicotinate.

The visualization result is shown in Fig. 8. The bottom figure shows the RGB image of the red flare. In the measurement in the near-infrared domain, black markers were applied to clarify the areas of red flare. As shown in Fig. 8, no area was classified as dermatitis.

Metal allergy

Finally, the discrimination function was performed on the spectral image of a metal allergy test, and the activated skin lesions were visual-

ized. Although the measurements were performed on two subjects, one showed only a weak allergic reaction, and therefore the spectral image of only the strongly reacting subject was used. The spectral image was measured 30 min after the patch test was removed. The visualization result is shown in Fig. 9.

As shown in the figure, three areas (= three different substances), those for cobalt chloride, nickel sulfate and zinc chloride, were detected as allergic dermatitis, which is in the same category as the SADBE-induced contact hypersensitivity. This is the correct classification result as a metal allergy is also a contact hypersensitivity reaction. In addition, the irritated spot indicated in Fig. 9 was not detected as an allergy. These results indicate that the discrimination function



Fig. 9. Visualization result of metal allergy patch test. The left image shows the RGB color image and the right the detection result. The color image was computed from the spectral image, which was measured by the Nuance (CRi, Vis) multispectral imaging device. The rectangle indicates the area of detection and color. Arrows indicate the irritated spot produced by Indium trichloride (No. 7 in Table 1).

might well detect spectral differences caused by some difference in cellular pathogenesis such as an accumulation of macrophages.

Discussion

As described in the first section, earlier studies of the evaluation of erythema focused on the spectral absorption properties of oxyhemoglobin and hydroxyl groups. However, these absorptions only allow the evaluation of visible elements such as inflammation and swelling. In contrast, our results described above suggest that the discrimination function that we developed may detect an internal component of allergic dermatitis. Therefore, in this section we compare the visualization maps to oxyhemoglobin and water content images to discuss what target was actually detected.

Comparison with oxy-hemoglobin accumulation

To determine the relationship between the detection results and the accumulation of oxy-hemoglobin, hyperspectral images of cases 2, 3 and 4 were transformed into an $L^*a^*b^*$ color space, as the a^* value is related to hemoglobin content (19). A D65 spectrum illuminant was used for the color signal computation. The a^* channel images are shown in Fig. 10. In these images, the skin area with accumulation of oxyhemoglobin appears light. As described in Figs. 8 and 10, accumulation of oxyhemoglobin induced by methyl nicotinate did not affect the output of the discrimination function. In addition, in the case of metal allergy only three patches were detected as contact hypersensitivity even though irritation with erythema appeared in many spots. Furthermore, in the case of histamine-induced urticaria, only the edematous area was detected as allergic dermatitis, while the inflammation around the edema was not detected. Based on these results, the developed discrimination function detects spectral changes by edema rather than by erythema (accumulation of oxyhemoglobin).

Comparison with a water content image

The results above indicate that the discrimination function used spectral characteristics of edema for discrimination between urticaria and

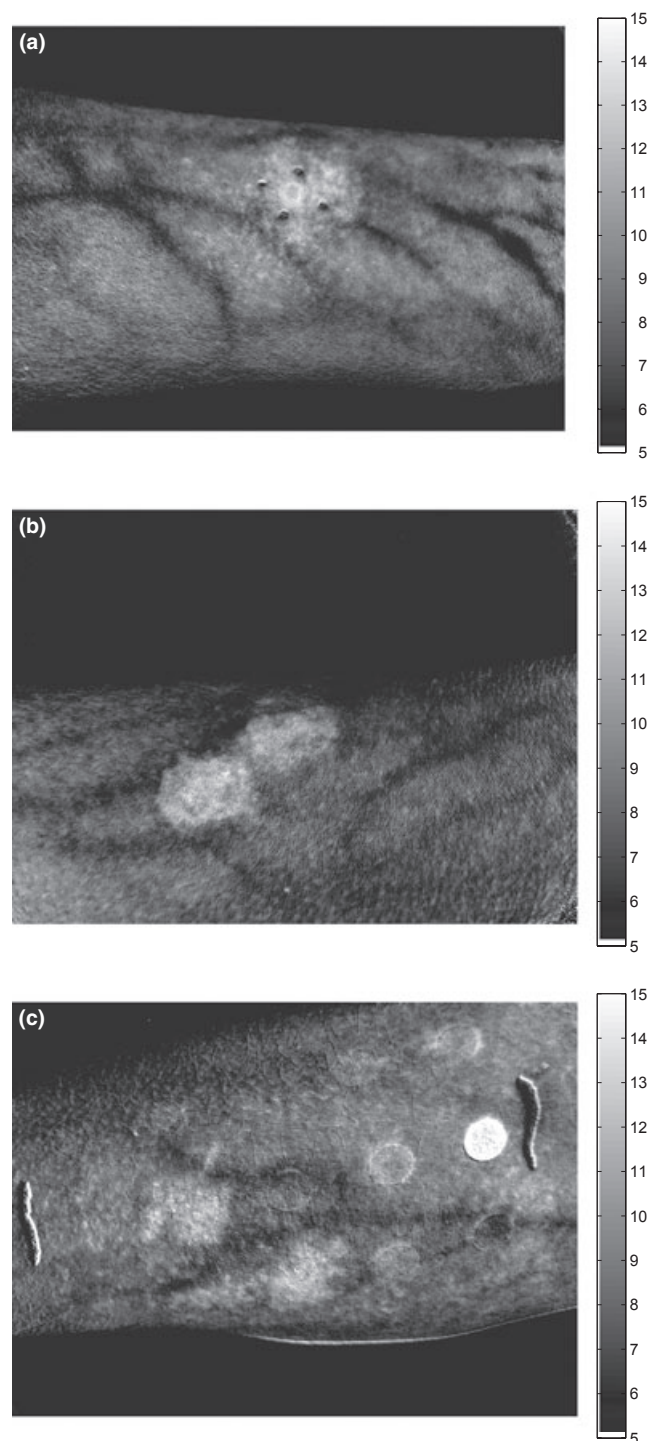


Fig. 10. The a^* images of (a) Urticaria induced by a histamine prick test, (b) flare reaction induced by methyl nicotinate, and (c) contact hypersensitivity induced by a metal allergy patch test.

contact hypersensitivity. As mentioned from the previous study, the absorption properties of the hydroxyl group, i.e., water content, figure strongly in the detection of the edema of allergic dermatitis (15, 17). Therefore, water content images were computed from the near-infrared

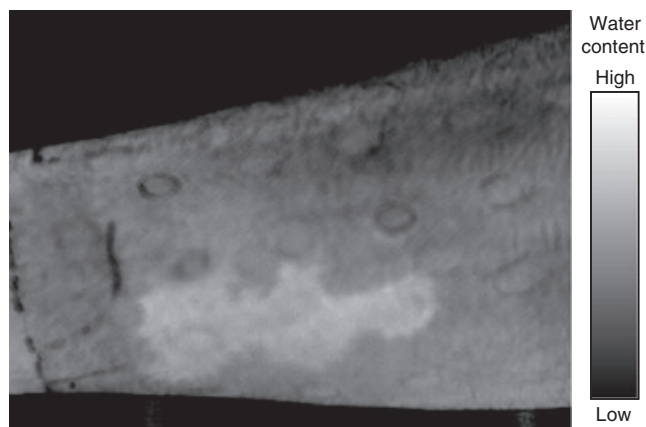


Fig. 11. Water content image of metal allergy. The water content was computed by the subtraction of the absorbances at 1450 and 1060 nm (20). The area of highest water content does not match the detection result.

spectral images to compare the allergic dermatitis visualization results and the corresponding water content distributions. In this analysis, a computation method for human skin moisture, subtraction of the spectral images at 1450 and 1060 nm, was used for the comparison because of the wavelength range of the spectral image (20). The computational water content image in the case of metal allergy is shown in Fig. 11. As shown, the distribution of the water content is obviously different from the visualization result in Fig. 9. Especially, No. 15 (manganese chloride) had high water accumulation even though it was not allergic reaction because there was no oxy-hemoglobin concentration [Fig. 10(a)]. This result suggests that our method detected some sort of allergic type-specific element that was neither hemoglobin pigment nor water content described in previous studies. However, more experiments and discussions are required to identify the detected element.

Conclusions

A long-wavelength near-infrared spectral imaging technique was applied to the assessment of allergic dermatitis in an attempt to detect and visualize intracutaneous differences caused by

the activation mechanisms of different types of allergic dermatitis. The assessment method was established based on the spectral classification of three skin conditions, normal skin, and types I and IV allergic dermatitis (Fig. 1). Near-infrared spectral images were measured over a wavelength range of 1000–2400 nm for four skin conditions: contact hypersensitivity induced by SADBE (type IV), urticaria induced by a histamine prick test (type I), flare reaction induced by methyl nicotinate (normal skin), and contact hypersensitivity induced by a metal allergy patch test (type IV). Training datasets for establishing the spectral classifier were extracted from the images of SADBE-induced contact hypersensitivity and histamine-induced urticaria (Fig. 4). The canonical scores of these spectral datasets computed by the established canonical discrimination function made separate clusters as shown in Fig. 5. The discrimination accuracies were also significantly high (Table 2), and the detected dermatitis lesions in the visualization results largely matched the macroscopic findings (Figs. 6 and 7). Finally, this method was applied to two test cases (flare reaction induced by methyl nicotinate and metal allergy) with correct results for both classification and visualized lesions (Fig. 9). Furthermore, an irritated spot (false-positive reaction) was not detected. It is apparent that the discrimination function detected some sort of internal allergic type-specific element that was neither hemoglobin pigment nor water content. These results suggest the potential utility of near-infrared spectral imaging for the assessment of allergic dermatitis. Specifically, this technique may be a useful tool for assisting the evaluation of metal allergy patch tests, as it can be difficult for the human eye to detect the false-positive/negative reactions. However, more subjects and cases should be evaluated to form a solid conclusion and develop a diagnostic tool. In our future work, we will apply this technique to other cases of allergic dermatitis to identify the detected substance.

References

1. Gell PGH, Coombs RRA. Clinical aspects of immunology. Oxford: Blackwell Science, 1963: 575–596.
2. Angelopoulou E. The reflectance spectrum of human skin. GRASP Laboratory. Department of Computer and Information Science, University of Pennsylvania, Technical Report MS-CIS-99-29 1999.
3. Tsumura N, Kawabuchi M, Haneishi H, Miyake Y. Mapping pigmentation in human skin from a multi-channel visible spectrum image by inverse optical scattering tech-

- nique. *J Imag Sci Technol* 2001; 45: 444–450.
4. Bafounta M, Beauchet A, Aegerter P, Saiag P. Is dermoscopy (epiluminescence microscopy) useful for the diagnosis of melanoma? Results of a meta-analysis using techniques adapted to the evaluation of diagnostic tests. *Arch Dermatol* 2001; 137: 1343–1350.
5. Ruocco E, Argenziano G, Pellacani G, Seidenari S. Noninvasive imaging of skin tumors. *Dermatol Surg* 2004; 30: 301–310.
6. Esmaeili A, Scope A, Halpern A, Marghoob A. Imaging techniques for the in vivo diagnosis of melanoma. *Semin Cutan Med Surg* 2008; 27: 2–10.
7. Iyatomi H, Oka H, Celebi M, Oga-wa K, Argenziano G, Soyer H, Koga H, Saida T, Ohara K, Tanaka M. Computer-based classification of dermoscopy images of melanocytic lesions on acral volar skin. *J Invest Dermatol* 2008; 128: 2049–2054.
8. Moncrieff M, Cotton S, Claridge E, Hall P. Spectrophotometric intracutaneous analysis: a new technique for imaging pigmented skin lesions. *Br J Dermatol* 2002; 146: 448–457.
9. Shibata S, Ohdan H, Noriyuki T, Yoshioka S, Asahara T, Dohi K. Novel assessment of acute lung injury by in vivo near infrared spectroscopy. *Am J Res Crit Care Med* 2009; 160: 317.
10. Dreassi E, Ceramelli G, Fabbri L, Vocioni F, Bartalini P, Corti P. Application of near-infrared reflectance spectrometry in the study of atopy part 1. Investigation of skin spectra. *Analyst* 1997; 122: 767–770.
11. Dreassi E, Ceramelli G, Mura P, Perruccio P, Vocioni F, Bartalini P, Corti P. Near-infrared reflectance spectrometry in the study of atopy part 2. Interactions between the skin and polyethylene glycol 400, isopropyl myristate and hydrogel. *Analyst* 1997; 122: 771–776.
12. Corti P, Ceramelli G, Dreassi E, Njine M. Near infrared reflectance spectroscopy in the study of atopy part 3. Interactions between the skin and fomblins. *Analyst* 1998; 123: 2313–2317.
13. Hampel U, Schleicher E, Wustenberg E, Huttenbrink K. Optical measurement of nasal swellings. *IEEE Trans Biomed Eng* 2004; 51: 1673–1679.
14. Rigal JD, Losch M, Bazin R, Camus C, Sturelle C, Descamps V, Leveque J. Near infrared spectroscopy: a new approach to the characterization of dry skin. *J-Soc Cosmet Chem* 1993; 44: 197–209.
15. Stamatas G, Zmudzka B, Kollias N, Beer J. Non-invasive measurements of skin pigmentation in situ. *Pigment Cell Res* 2004; 17: 618–626.
16. Meglinski I, Matcher S. Quantitative assessment of skin layers absorption and skin reflectance spectra simulation. *Physiol Measur* 2002; 23: 741–753.
17. Stamatas G, Kollias N. In vivo documentation of cutaneous inflammation using spectral imaging. *J Biomed Opt* 2007; 12: 051603.
18. Gemert MV, Jacques S, Sterenborg H, Star W. Skin optics. *IEEE Trans Biomed Eng* 1989; 36: 1146–1154.
19. Jung B, Choi B, Durkin A, Kelly K, Nelson J. Characterization of port wine stain skin erythema and melanin content using crosspolarized diffuse reflectance imaging. *Lasers Surg Med* 2004; 34: 174–181.
20. Iwasaki H, Miyazawa K, Nakauchi S. Visualization of the human face skin moisturizing-ability by spectroscopic imaging using two near infrared bands. In Eighth International Symposium on Multispectral Color Science, San Jose, CA, USA, January 2006: 606203-1–606203-8.

Address:
 Ken Nishino
 Department of Computer Science and
 Engineering
 Toyohashi University of Technology
 Toyohashi
 Japan
 Tel: +81-532-44-6765
 Fax: +81-532-44-6651
 e-mail: nishino06@vpac.cs.tut.ac.jp