

# Association between tongue coating thickness and ultraviolet fluorescence in patients with functional dyspepsia

## A prospective observational study

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### Abstract

The aim of this study was to examine the correlation between the tongue coating thickness (TCT) and ultraviolet (UV) fluorescence and propose a new method for the estimation of TCT using a computerized tongue image acquisition system (CTIS).

In this prospective and observational single-center study, we acquired tongue images under visible light and near-UV light for 60 patients with functional dyspepsia. Tongue images were acquired twice within a 30-minute interval to assess the reliability of CTIS. Then, the tongue coating was scraped and weighed to derive the wet weight of the tongue coating (WWTC). The percentage of the tongue coating area was calculated from the tongue images acquired under visible light. Mean color values (mCVs) for the UV fluorescence of the dorsal surface of the tongue were also computed.

The reliabilities of the derived mCVs and percentage of the tongue coating area were acceptable (intraclass correlation coefficients, 0.907–0.947). The mCVs were more strongly correlated with WWTC than with the area, with mCV of modified lightness showing the strongest association ( $r=0.785$ ,  $P<.01$ ). Finally, we suggested an estimation model for TCT based on the results.

The results of this study suggest that both UV fluorescence of the dorsal tongue and the distribution area of tongue coating are useful parameters for the quantitative assessment of tongue coating. We believe that these findings will contribute to the development of a clinically useful CTIS.

**Abbreviations:** ANOVA = one-way analysis of variance, B = blue, CTIS = computerized tongue image acquisition systems, G = green, ICCs = intraclass correlation coefficients, mCV = mean color value, R = red, TCM = traditional Chinese medicine, TCT = tongue coating thickness, TKM = traditional Korean medicine, UV = ultraviolet, WWTC = wet weight of tongue coating.

**Keywords:** tongue coating, tongue coating thickness, tongue diagnosis, ultraviolet fluorescence, wet weight of tongue coating

### 1. Introduction

Detection of tongue abnormalities is one of the most important diagnostic methods in traditional Korean medicine (TKM) and traditional Chinese medicine (TCM).<sup>[1]</sup> It is believed that changes in the tongue body and tongue coating status are indicators of

disease, because the tongue is connected to internal organs and reflects internal health. Attachments formed on the lingual papillae indicate conditions related to fluid and humor in the body, and in particular, the digestive system. In particular, the tongue coating thickness (TCT) is considered an important

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The research followed guidelines of the Declaration of Helsinki and Tokyo for humans. This study was approved by the Korean Food and Drug Administration and the Institutional Review Board of the Kyung Hee University Korean Medicine Hospital (IRB no. KOMCIRB-2013-01). All participants received information about the purpose of this study and provided written informed consent before enrollment.

The data used in the analyses are available on request by contacting the corresponding author.

The authors declare that there are no conflicts of interest.

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indicator of physiological and pathological conditions of qi.<sup>[2,3]</sup> Moreover, assessment of TCT is used in the field of dentistry, because tongue coating is not only a cause of halitosis but also a risk factor for aspiration pneumonia in edentate elderly individuals.<sup>[4]</sup>

To date, various methods have been used for the evaluation of TCT.<sup>[5]</sup> The conventional method used widely in TKM and TCM involves the classification of TCT into 3 categories on the basis of naked eye observations and the intuitive judgment of practitioners: no coating, thin coating, and thick coating.<sup>[6,7]</sup> Other proposed methods include quantitative estimation of tongue coating via assessment of the degree of visibility of the pink tongue body or tongue coating discoloration.<sup>[8,9]</sup> However, these methods can be affected by the practitioner's inexperience, subjectivity, and various environmental factors; furthermore, the criteria for differentiation are ambiguous.<sup>[10]</sup> One study also proposed a direct method for TCT evaluation, wherein the tongue coating is scraped and weighed.<sup>[11]</sup> The validity and reliability of the method involving weighing the tongue coating were not demonstrated sufficiently because of inconsistencies in scraping and the long reformation time after scraping.<sup>[12]</sup> Nevertheless, this method of weighing the tongue coating has been regarded as the most suitable approach because it is the most direct and feasible method.<sup>[5]</sup>

To overcome the limitations of the conventional method, a variety of computerized tongue image acquisition systems (CTISs) have been designed to obtain digital tongue images and objectively assess characteristics such as color and texture.<sup>[13–16]</sup> Several studies have also analyzed the tongue coating area and developed diagnostic criteria for TCT.<sup>[2,6,17]</sup> However, the efficacy of TCT evaluations based only on the percentage of the tongue coating area remains controversial because of lack of sufficient correlation between the tongue coating area and the directly weighted tongue coating.<sup>[17]</sup>

As ultraviolet (UV) fluorescence in the root and center of the tongue under Wood lamp light was reported,<sup>[18]</sup> it has been well known that tongue coating emits weak green or red fluorescence when illuminated by near-UV light.<sup>[16–21]</sup> The intensity of UV fluorescence of the tongue coating (hereafter referred to as tongue coating UV fluorescence) varies according to TCT.<sup>[18]</sup> However, few reports on the relationship between TCT and tongue coating UV fluorescence are available, whereas several studies on TCT evaluation under visible light are available.<sup>[22–24]</sup> The hypothesis of the current study was that TCT is strongly related to the intensity of tongue coating UV fluorescence calculated from tongue images obtained under near-UV LED light. Therefore, in the present study, we investigated the relationship between the TCT and tongue coating UV fluorescence and suggested a new method for the estimation of TCT based on the tongue coating UV fluorescence.

## 2. Methods

### 2.1. Ethics and study design

This prospective and observational study was conducted at Kyung Hee University Korean Medicine Hospital in Seoul, Republic of Korea (ClinicalTrials.gov, NCT01864837) and approved by the Korean Food and Drug Administration and the Institutional Review Board of the Kyung Hee University Korean Medicine Hospital (IRB no. KOMCIRB-2013-01).

A schematic block diagram of the study design is presented in Fig. 1. The study was conducted in accordance with the Good Clinical Practice guidelines and adhered to the principles of the Declaration of Helsinki. All participants received information about the purpose of this study and provided written informed consent before enrollment.

### 2.2. Study participants

All participants were recruited through posters that were displayed around the university and hospital. Their tongue images obtained under near-UV LED light were evaluated. According to the TKM and TCM theory, tongue coating is related to the digestive system; therefore, this study focused on patients with functional dyspepsia for more than 6 months. Sixty participants above 20 years of age who fulfilled the Rome III criteria for functional dyspepsia were included. The exclusion criteria were as follows: pregnancy, mental disorders, severe systemic organ diseases, and geographic tongue. Details of the inclusion and exclusion criteria have been described in a previous study.<sup>[25]</sup>

### 2.3. Procedures

All participants were asked to visit the hospital on 2 occasions during the study period. At the first visit, we provided instructions and explained all necessary precautions to minimize the influence of multiple factors. The participants were requested to avoid food and liquid intake for at least 4 hours before the experiments and refrain from brushing their teeth and tongues, which can affect the tongue coating status. Considering the possibility of diurnal variation in the TCT, we recommended that they visit between 10 and 12 AM for imaging of the tongue.

At the second visit, tongue images of all participants were captured using CTIS. Tongue images under white LED light were obtained first. Then, images under near-UV light were obtained using a CTIS. Before tongue images were acquired, the participants exercised several times by opening their mouths and extruding their tongues such that the entire tongue was visible. To assess the reliability of the CTIS, images were obtained again in the same manner after 30 minutes.

After obtaining the second tongue image, we measured the wet weight of tongue coating (WWTC) according to the method by Yaegaki and Sanada.<sup>[11]</sup> Briefly, we swabbed the dorsal surface of the tongue using a gauze, carefully scraped the surface using a scraper, collected tongue coating on a slide glass, and measured WWTC using an electronic scale (mg).

### 2.4. Measurement device and outcome parameters

**2.4.1. Tongue image acquisition.** Details of the CTIS used in the present study have been described in our previous study.<sup>[25]</sup> Figure 2 shows the system and the acquisition of a tongue image. Tongue images were first obtained under white LED (LWH5000, Luxpia, Suwon, Korea) light, following which they were obtained under near-UV light (T5F36, Seoul Optodevice, Ansan, Korea) using a CTIS. For uniform illumination of the dorsal surface of the tongue, 12 white LED lamps (dia., 5 mm; double peak wavelengths, 470 and 580 nm) and 40 near-UV LED lamps (dia., 5 mm; peak wavelength, 365 nm) were arranged in concentric circles around the camera, as shown in Fig. 2B. A double light diffusion plate was placed in front of the white LED lamps.

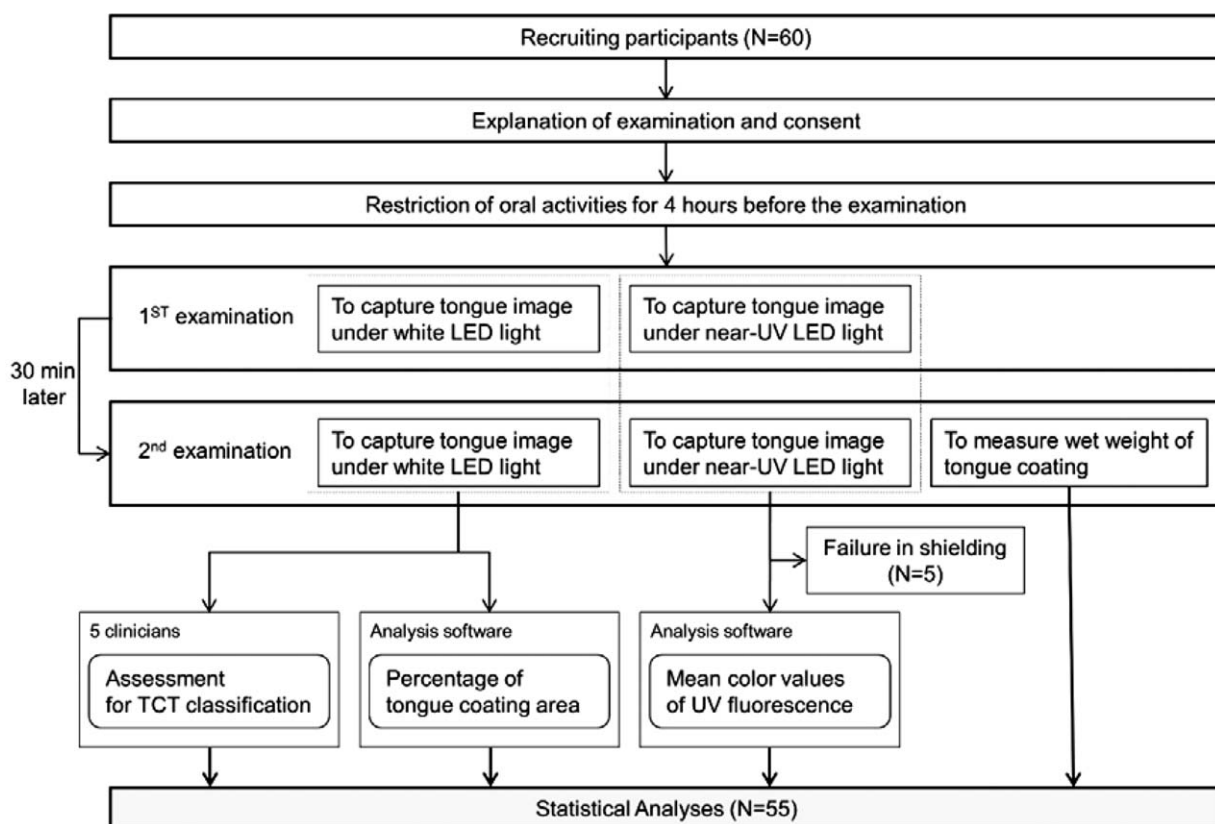


Figure 1. Block diagram depicting the study design. TCT = tongue coating thickness; UV = ultraviolet.

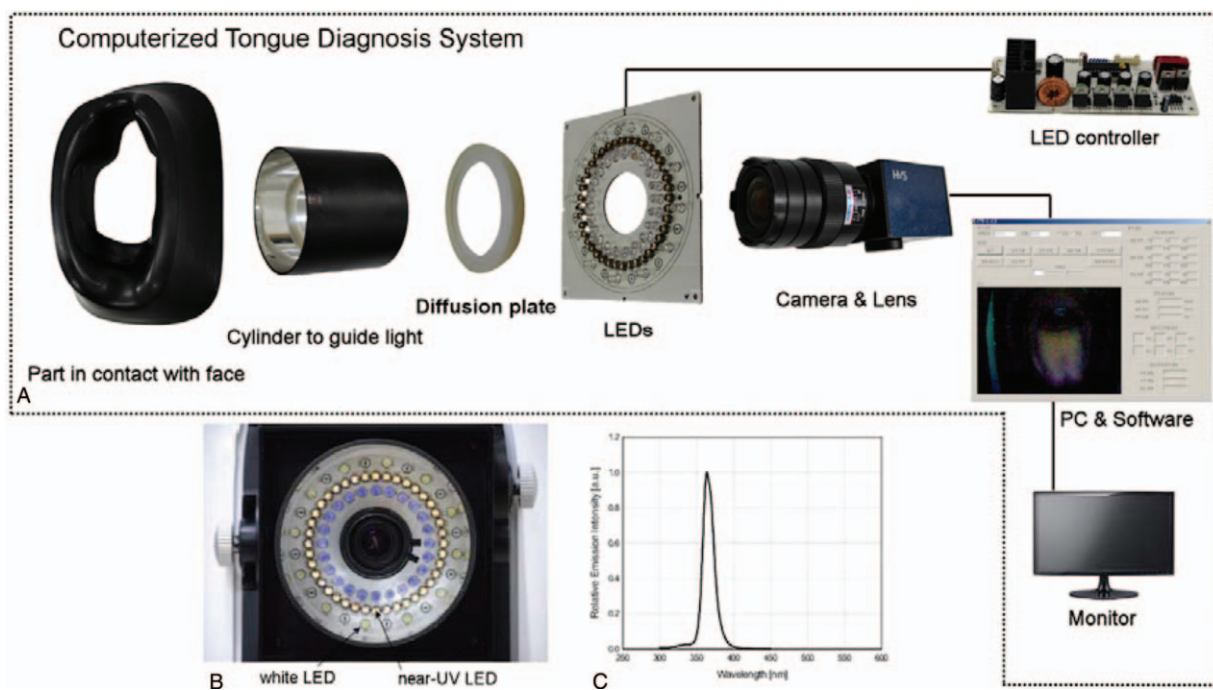


Figure 2. Schematic of the computerized tongue image acquisition system (CTIS) used in the present study. (A) Major components of the CTIS: part in contact with the face, cylinder to guide light, LEDs, LED controller, camera, lens, PC, monitor, and software; (B) Circular configuration with the LED illuminator; the ultraviolet (UV) LEDs are turned ON; (C) Wavelength of the UV light.

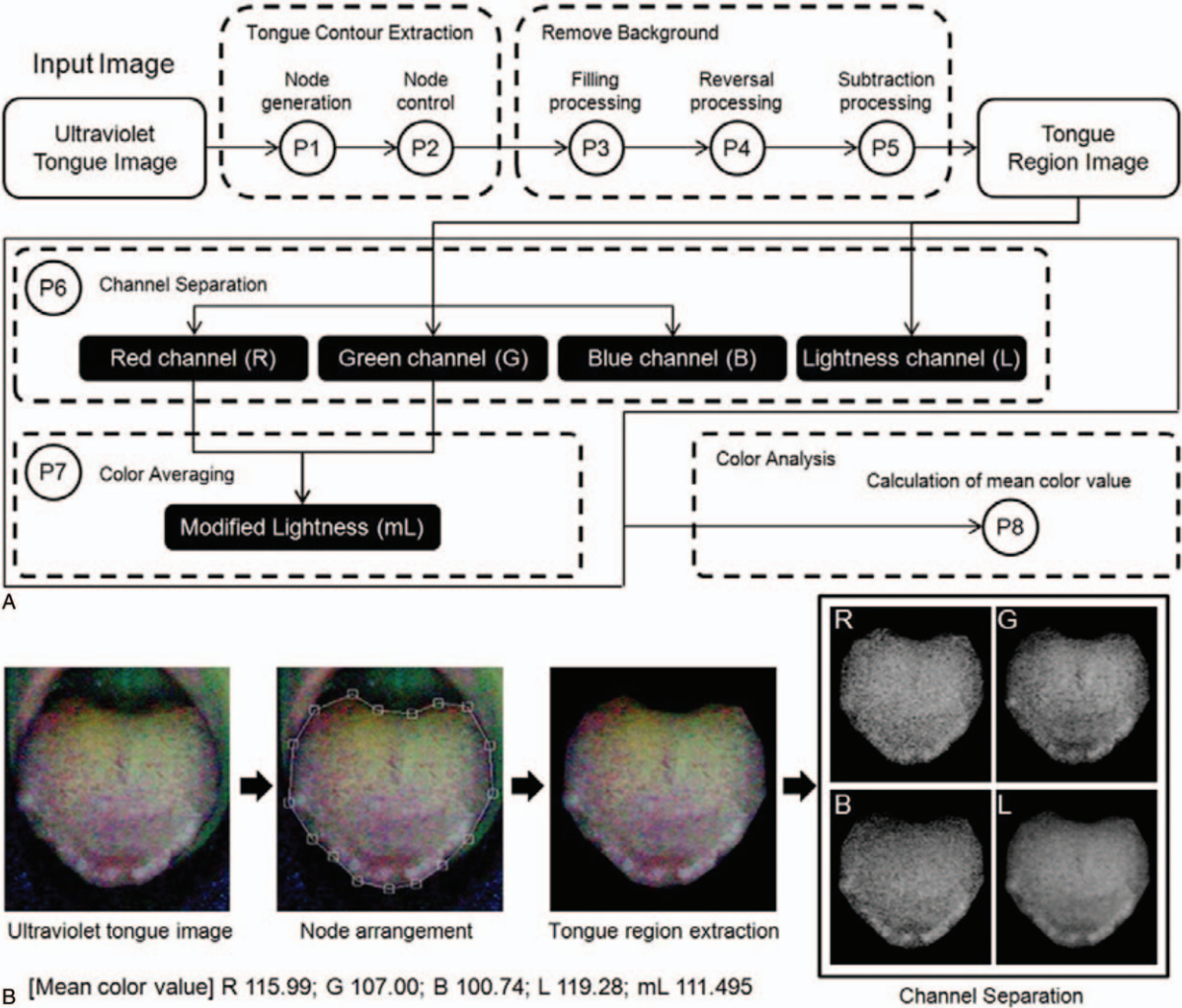


**2.4.2. Assessment of tongue coating thickness.** For TCT assessment using the conventional method, the tongue images obtained under white LED light were displayed in random order on a 50-inch color monitor. Five TKM practitioners with clinical experience of 5 years or more assessed TCT independently and simultaneously and classified it as no coating, a thin coating, or a thick coating. All discrepancies among the assessors occurred near the boundary between the no coating and the thin coating or between the thin coating and the thick coating. In case of discrepancy, the decision of the majority was considered final.

The tongue images obtained under white LED light were also used for calculating the percentage of the tongue coating area, while those obtained under near-UV LED light were used for measuring the tongue coating UV fluorescence. The percentage of the tongue coating area is defined as the percent rate of the number of pixels in the tongue coating region with respect to the number of pixels in the entire tongue region. We extracted the tongue coating region from the tongue region using differences in the CIE *a\** values between the tongue body and tongue coating.

The segmentation algorithm has been described in detail in our previous study.<sup>[2,25]</sup>

**2.4.3. Analysis of ultraviolet fluorescence using the tongue images.** We analyzed UV fluorescence of the tongue coating using the tongue images obtained under near-UV light. Figure 3 presents the algorithm for color analysis. First, we extracted the tongue region from the tongue image. Then, we created a layer on the tongue image and generated some nodes on it, following which the operator dragged and dropped the nodes on the outline of the tongue using a mouse. The remaining region excluding the selected tongue region was eliminated. A channel is a grayscale image made of only one of primary colors; the image obtained under near-UV light was a combination image of RGB channels. Therefore, the image with the eliminated background was separated into 3 individual channels: red (R), green (G), and blue (B). Subsequently, the mean color value (mCV) of each color channel was calculated as the average grayscale value of the pixels in the tongue area. The lightness channel in the HSL color



**Figure 3.** (A) Algorithm for the color analysis of tongue images under near-ultraviolet (UV) light; (B) Schematic diagram for calculating the mean color value (mCV) of each channel. After the node arrangement processes, the tongue image with the eliminated background is separated into three channels (red, green, and blue). The lightness channel is also extracted. Modified lightness is an average of the red and green channels.

model was also separated from the selected tongue region, and the mCV of the lightness channel was calculated according to the following definition of lightness:

$$\text{Lightness} = \frac{\max(R, G, B) + \min(R, G, B)}{2} \quad (1)$$

In Eq. (1),  $\max(R, G, B)$  and  $\min(R, G, B)$  represent the maximum and minimum of values in parentheses, respectively.  $R$ ,  $G$ , and  $B$  represent the  $R$ ,  $G$ , and  $B$  values of each pixel in the tongue area. Considering that the dorsum of the tongue emitted green and red light under near-UV light and that the blue image sensor recorded not only the fluorescence but also the reflected near-UV light, we removed the blue value from the definition of lightness in order to modify the mCV of the lightness channel as an indicator of the tongue UV fluorescence. The definition of modified lightness was as follows:

$$\text{Modified lightness} = \frac{R + G}{2} \quad (2)$$

mCV of modified lightness was also calculated according to Eq. (2).

## 2.5. Statistical analysis

The general characteristics of the participants are presented as descriptive statistics. We calculated intraclass correlation coefficients (ICCs) for mCVs and the percentage of the tongue coating area in order to assess the reliability of CTIS. The mCVs and the percentage of the tongue coating area were compared among the TCT groups classified using the conventional method using one-way analysis of variance (ANOVA) and Duncan multiple range test. Shapiro–Wilk test and Levene test were performed to assess the normality of data distribution and the equality of variances for these 2 parameters among the TCT groups. If the data were not normal distributions, they were compared by Kruskal–Wallis test and Mann–Whitney  $U$  test. We calculated Pearson correlation coefficients to analyze the relationships among the percentage of the tongue coating area, mCVs of UV fluorescence, and WWTC. Subsequently, we performed regression analyses to establish estimation models for TCT. All statistical analyses were performed using IBM SPSS Statistics 21 (IBM Corporation, NY) and SAS 9.4 PROC NLIN (SAS Institute Inc., NY). A  $P$  value  $< .05$  was considered statistically significant.

## 3. Results

### 3.1. Study participants

The general characteristics of the study participants are summarized in Table 1. There was no adverse event during the study period. The data for 5 participants for whom external light could not be blocked were excluded from the final analysis.

**Table 1**  
General characteristics of the study participants (N = 55).

Variables, unit	
Sex (male: female) (%)	10 (18.2): 45 (81.8)
Age (mean $\pm$ standard deviation), y	47.51 $\pm$ 11.46
Height (mean $\pm$ standard deviation), cm	159.95 $\pm$ 5.97
Body weight (mean $\pm$ standard deviation), kg	58.68 $\pm$ 10.07

### 3.2. Reliability of the computerized tongue image acquisition system

The ICC values for assessing the reliability of CTIS are summarized in Table 2. ICCs for mCVs and the percentage of the tongue coating area ranged from 0.907 to 0.947 and indicated an acceptable reliability for all parameters.<sup>[26,27]</sup>

### 3.3. Comparison of mean color values and the percentage of the tongue coating area among the tongue coating thickness groups classified by the conventional method

For all variables except for mCV of R channel in the thin coating group, the  $P$  values resulting from Shapiro–Wilk test and Levene test were  $> .05$ . mCVs of  $G$ ,  $B$ , lightness, and modified lightness were significantly lower for the thin coating group than for the thick coating group, with no significant differences between the no coating and thin coating groups. In contrast, there was no significant difference between the thin coating and thick coating in the R channel, while there was a significant difference between the thin coating and no coating. This implies that the R channel can be more sensitive in cases of less tongue coating, while the others can be more sensitive when tongue coating is extensive. There were significant differences among all 3 groups with regard to the percentage of the tongue coating area. The high sensitivity of the percentage of the tongue coating area for the TCT classification may be caused by the fact that it was derived from the same tongue images under white LED light. The results are presented in Table 3.

### 3.4. Regression analyses for the estimation of tongue coating thickness

**3.4.1. Simple correlation.** Pearson correlation coefficients for the relationships among mCVs, WWTC, and the percentage of the tongue coating area are presented in Table 4. All mCVs were more strongly correlated with WWTC than with the area, with the mCV of modified lightness showing the strongest association with WWTC. Increased keratinization of the tongue papillae often appears as a thin tongue coating, although it cannot be scraped.<sup>[28]</sup> This error may have contributed to the relatively weak correlation between the percentage of the tongue coating area and WWTC.

**Table 2**  
Reliability of the mean color values (mCVs) and percentage of the tongue coating area.

	ICC	95% CI	Level of reliability
mCV of R channel	0.913	0.855–0.948	Acceptable
mCV of G channel	0.935	0.890–0.942	Acceptable
mCV of B channel	0.928	0.879–0.957	Acceptable
mCV of lightness channel	0.942	0.903–0.966	Acceptable
mCV of modified lightness	0.947	0.911–0.969	Acceptable
Percentage of tongue coating area	0.907	0.840–0.946	Acceptable

Percentage of tongue coating area is the percentage rate of the area of tongue coating with respect to the gross area of tongue.

CI = confidence interval, ICC = intraclass correlation coefficient, mCV = mean color value calculated as average grayscale value of the pixels in tongue area of each color channel; R channel, a grayscale image made of only red of the primary colors; G channel, a grayscale image made of only green of the primary colors; B channel, a grayscale image made of only blue of the primary colors; lightness channel, a grayscale image made of only lightness calculated from the primary colors.

**Table 3**  
Comparison of the mean color values (mCVs) and percentage of the tongue coating area among patients with no coating, a thin coating, and a thick coating.

	No coating N=12	Thin coating N=35	Thick coating N=8
mCV of R channel*	109.45±12.27 <sup>†</sup>	119.96±14.63	130.00±19.29
mCV of G channel <sup>‡</sup>	100.67±11.12 <sup>a</sup>	107.32±17.85 <sup>a</sup>	127.44±21.35 <sup>b</sup>
mCV of B channel <sup>‡</sup>	102.27±10.29 <sup>a</sup>	112.07±15.11 <sup>a</sup>	126.67±15.56 <sup>b</sup>
mCV of lightness channel <sup>‡</sup>	104.51±10.03 <sup>a</sup>	113.33±15.41 <sup>a</sup>	128.32±16.94 <sup>b</sup>
mCV of modified lightness <sup>‡</sup>	105.06±10.61 <sup>a</sup>	113.64±15.43 <sup>a</sup>	128.72±19.36 <sup>b</sup>
Percentage of tongue coating area <sup>‡</sup>	22.21±12.55 <sup>a</sup>	41.88±8.57 <sup>b</sup>	62.59±13.94 <sup>b</sup>

The results are expressed as mean±standard deviation.  
CI=confidence interval, ICC=intraclass correlation coefficient, mCV=mean color value calculated as average grayscale value of the pixels in tongue area of each color channel; R channel, a grayscale image made of only red of the primary colors; G channel, a grayscale image made of only green of the primary colors; B channel, a grayscale image made of only blue of the primary colors; lightness channel, a grayscale image made of only lightness calculated from the primary colors.  
\*  $P<.05$ ; Kruskal–Wallis test was used to compare mCVs of R channel among three groups. mCV of R channel was compared with each other by Mann–Whitney  $U$  test.  
<sup>†</sup>  $P<.05$ , no coating versus thin coating, and no coating versus thick coating; there is no significant difference between thin coating and thick coating.  
<sup>‡</sup>  $P<.01$ ; One-way ANOVA was used to compare mCVs except for R channel and the percentage of the tongue coating area among groups. Duncan test was used for post-hoc analysis (a, b, c: Subset for  $\alpha=.05$ ).

**3.4.2. Regression model.** We assumed that the relationship between WWTC and the mCV of modified lightness (mCV<sub>mL</sub>) could be described by a logistic-curve (pearl-curve) usually modeled by the log-transformed linear regression model (Eq. 3) and the nonlinear regression model (Eq. 4). Each basic equation is represented below, where  $K$  denotes the maximum value of possible WWTCs, and  $\beta_0$  and  $\beta_1$  are, respectively, the intercept and the regression coefficient in the log-transformed linear regression.  $\varepsilon$  is a random error.

$$\ln\left(\frac{k}{WWTC} - 1\right) = \beta_0 + \beta_1 mCV_{mL} + \varepsilon \tag{3}$$

$$WWTC = \frac{K}{1 + \exp(\beta_0 + \beta_1 mCV_{mL})} + \varepsilon \tag{4}$$

In the log-transformed linear regression model,  $K$  is preset as 250 (250=90+2×80) before estimating the regression coefficients, where 90 and 80 indicate the mean and standard deviation of WWTC, respectively, according to the study by Lundgren et al.<sup>[28]</sup> We estimated the coefficients for the nonlinear regression model with  $K$  calculated by the nonlinear least squares method. The estimates of parameters and the standard errors are given in Table 5. We noted that the results of the log-transformed

regression model depended crucially on the choice of  $K$ , whereas the nonlinear regression estimation provided optimal estimates of  $K$  as well as the regression coefficients for the given data.  
To measure the goodness of fit of each model, the mean squared error (MSE) was employed as follows:

$$MSE = \frac{1}{n - p} \sum_{i=1}^n (\circ y_i - y_i)^2 \tag{5}$$

Here,  $y_i$  and  $\circ y_i$  represent the observed WWTC and predicted WWTC based on the estimated model, respectively.  $n$  and  $p$  represent the sample size and the number of parameters to be estimated in the models, respectively. For this criterion, a smaller value results in a better model. Nonlinear regression showed a smaller MSE compared with log-transformed regression. This implies that, for any given mCV of modified lightness, WWTC can be finally predicted by Eq. (6).

$$WWTC(mg) : \circ y = \frac{210.5}{1 + \exp(8.03 - 0.0576 \cdot mCV_{mL})} \tag{6}$$

The scatter diagram of the estimation model is presented in Fig. 4.

4. Discussion

In the present study, we investigated differences in tongue coating UV fluorescence according to the tongue coating classification using the conventional method and suggested a new method for the estimation of TCT based on the tongue UV fluorescence. The results revealed that both UV fluorescence of the dorsal tongue and the distribution area of the tongue coating are useful parameters for the quantitative assessment of tongue coating.

**Table 5**  
Regression models for the wet weight of the tongue coating.

Parameter	Log-transformed regression		Nonlinear regression	
	Estimate	Standard error	Estimate	Standard error
$K$	250	—	210.5	61.826
Intercept ( $\beta_0$ )	9.887	0.798	8.03	1.374
Modified lightness ( $\beta_1$ )	−0.0692	0.0069	−0.0576	0.0141
Mean squared error	687.85		598.78	

**Table 4**  
Relationships among the mean color values for the dorsal tongue ultraviolet (UV) fluorescence, wet weight of the tongue coating, and percentage of the tongue coating area.

mCV	WWTC	Percentage of tongue coating area
R channel	0.721*	0.480*
G channel	0.767*	0.526*
B channel	0.663*	0.596*
Lightness channel	0.759*	0.559*
Modified lightness	0.785*	0.531*

mCV=mean color value calculated as average grayscale value of the pixels in tongue area of each color channel; WWTC=wet weight of tongue coating measured by scrapping and weighing tongue coat; R channel, a grayscale image made of only red of the primary colors; G channel, a grayscale image made of only green of the primary colors; B channel, a grayscale image made of only blue of the primary colors; lightness channel, a grayscale image made of only lightness calculated from the primary colors.  
\*  $P<.01$ .

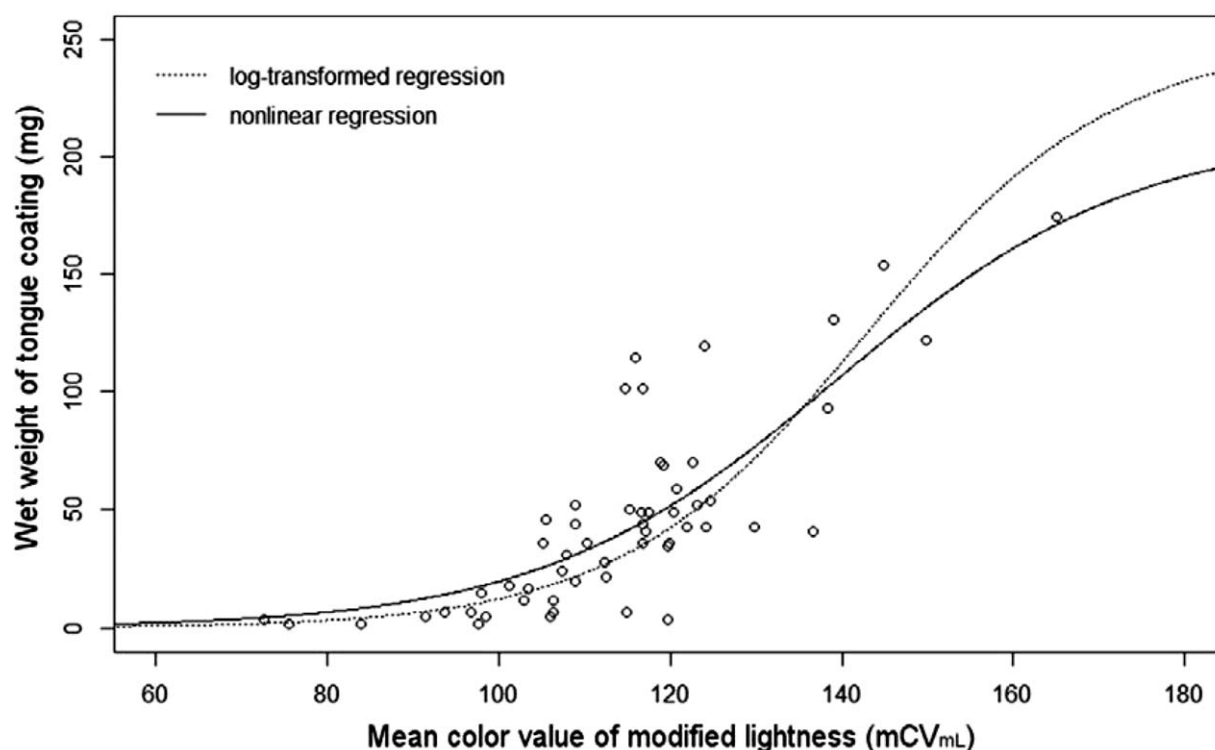


Figure 4. Regression models for the wet weight of the tongue coating.

TCT is a term used to represent the degree of tongue coating in accordance with the amount of tongue coating in TKM.<sup>[5]</sup> TCT evaluation methods can be categorized as intuitive and objective. The intuitive methods use naked eye observations to classify TCT into 3 to 5 grades according to the severity<sup>[7,29–31]</sup> or distribution area of tongue coating.<sup>[32,33]</sup> However, some researchers reported that the reliability of TCT classification for conventional diagnosis was very low and argued that inadequate operational definitions of the tongue characteristics were a major cause.<sup>[10]</sup> Other methods involve segmentation of the dorsum of the tongue into sections for rating the severity of tongue coating.<sup>[8,9,23,34]</sup> In 1 study,<sup>[9]</sup> the dorsum of the tongue was divided into 6 sections, and tongue coating in each section was scored on a 3-point scale (0, no coating; 1, light coating; and 2, severe coating). In another study,<sup>[8]</sup> the dorsum was divided into 9 sections for scoring the tongue coating.

The objective methods for the TCT evaluation involve weighing of the scraped tongue coating and measurement of the distribution area. Although measurement of the tongue coating weight is the most direct method, it is rarely used in the clinical field because patients may gag during the scraping procedure. In a systematic review of the TCT evaluation methods, weighing method was set as a gold standard in order to assess the accuracy of the other methods.<sup>[5]</sup> Recently, criteria for differentiation were developed through analyses of the consistency between the results obtained by well-trained clinicians and those based on the distribution area of tongue coating on the dorsum.<sup>[6]</sup> However, the area method cannot be expected to be highly accurate because it does not take into account the thickness of the tongue coating.

With regard to the severity of tongue coating, it is important to establish a gold standard method based on the above techniques.

In our previous study,<sup>[17]</sup> we noted that the correlation between the percentage of the tongue coating area and WWTC was not very strong ( $r=0.442$ ). This implied that the severity of tongue coating could not be accurately estimated from the distribution area, despite the fact that the distribution area well represents TCT in conventional tongue diagnosis.

The fluorescence phenomenon in the oral cavity has been described since the 1920s.<sup>[35]</sup> An observational study conducted in the 1950s mentioned that the dorsal surface of the tongue emits green or red light under near-UV excitation, and that the fluorescence intensities are associated with TCT.<sup>[18]</sup> In the present study, mCVs were significantly higher for the thick coating group than for the no coating group, confirming the results of the pioneer study. Nevertheless, the high standard deviations for mCVs countervailed differences in mCVs between the thin and no coating groups.

It is widely accepted that fluorescence is mainly caused by the metabolic process of microorganisms, endogenous porphyrins, and recently ingested food mixed in tongue coating.<sup>[18,36]</sup> A study<sup>[20]</sup> reported that fluorescence spectra acquired from the dorsum of dog tongues peaked at 500 to 520 nm (green) and approximately 700 nm (red) under near-UV light, while another<sup>[19]</sup> reported that the dorsum of the human tongue also emitted fluorescent light as a double peak under an excitation wavelength of 405 nm. Here, the wavelengths of the primary and secondary peaks were approximately 520 (green) and 636 (red) nm. Furthermore, the intensity of fluorescence at the lateral border of the tongue was the lowest in the oral cavity; tongue coating rarely exists at the tip and lateral borders of the tongue even if it covers the dorsum of the tongue widely. These findings indicate that tongue coating is closely associated with green and red fluorescence. The camera's blue image sensor detects the primary



peak to some extent and also detects the near-UV light reflected from the dorsal surface of the tongue. Therefore, the blue channel can contain light information that is not related to fluorescence. In the present study, WWTC was more strongly correlated with mCVs of the green and red channels than with the mCV of the blue channel. These findings were consistent with those of previous studies.<sup>[19,20]</sup> Therefore, we excluded mCV of the blue channel from the estimation model for TCT.

We could not separate the tongue coating area from the total tongue area because the contour of the tongue coating area was not clear under near-UV light. We believe this could be a reason for the strong collinearity between the percentage of the tongue coating area and mCVs in the estimation model. Therefore, the percentage of the tongue coating area was also excluded from the estimation model.

Some limitations of the study should be considered when interpreting the results. First, when tongue images are obtained under near-UV light, external light should be blocked as much as possible; however, we failed to block out the external light in five participants with small, round faces. Second, the range of WWTC values was not wide enough to set an indisputable maximum value (*K*). Under certain conditions such as awakening in the morning, WWTC may be considerably higher than the maximum weight estimated in the estimation model. Therefore, incorporation of a tongue image obtained early in the morning may have allowed generation of a better model. Third, the tongue itself can also emit fluorescence. Although the fluorescence intensity of the tongue itself is lower than that of the tongue coat, this may have reduced the accuracy of the method used in this study.

To the best of our knowledge, this is the first study investigating the relationship between the tongue UV fluorescence and TCT. Considering the significance of TCT in TKM, tongue coating should be quantitatively assessed in terms of not only the distribution area but also the amount of tongue coating. Our results suggest that the dorsal tongue UV fluorescence as well as the distribution area of tongue coating should be considered in the quantitative evaluation of tongue coating. We believe that these findings will contribute to the development of a clinically useful CTIS.

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