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Characterization of teeth fluorescence properties due to coffee pigmentation: towards optimization of quantitative light-induced fluorescence for tooth color assessment

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ABSTRACT

The assessment of tooth color is typically performed by subjective comparison with a visual shade guide or by using objective optical techniques such as quantitative light-induced fluorescence (QLF). QLF measurements rely on the precise wavelength calibration of fluorescence excitation and emission for enhancing the contrast between the white sound tooth and stained areas. These areas may change the fluorescence emission differently depending on the color that is most absorbed by the stain on the tooth surface. Although previous studies have monitored the staining contrast generated by the consumption of beverages on teeth, the information provided is based on total intensity. However, this intensity varies from each QLF device configuration and comparison across studies may not be possible. Few studies report the wavelength-dependent characterization of the staining process, which allow the comparison on the light attenuation on specific wavelengths and can be used to design fluorescence equipment with improved contrast for the tooth color assessment. In this study, we quantified the fluorescence spectral features (fluorescence intensity, wavelength shift of the maximum intensity, full width at half maximum, and wavelength-dependent intensity attenuation) of teeth in several degrees of coffee pigmentation by using 445 nm excitation. Most of the pigmentation effect was observed on the fluorescence intensity and a linear behavior was observed for the full width at half maximum (around 11.8% increase for each pigmentation level). We characterized the fluorescence properties of each degree of pigmentation level. Both spectral features and fluorescence properties can be used to design novel fluorescence equipment capable of increasing the contrast between white and stained teeth.

Keywords: Fluorescence spectroscopy, tooth whitening, tooth color, fluorescence monitoring, optical spectroscopy, violet illumination, light-induced whitening, light-induced fluorescence.

1. INTRODUCTION

The current evaluation of teeth color is performed by subjective assessment methods such as comparison to visual stain indices¹⁻⁶, visual shade guides⁷⁻¹⁰ and calculation of the Enamel Defects Index (EDI)¹¹ and Defects of Dental Enamel index (DDE index)^{11,12} developed by the Federation Dentaire Internationale (FDI). Teeth color can also be evaluated by using objective assessment through optical techniques such as colorimetry, reflectance spectrophotometry, digital imaging analysis, and quantitative light-induced fluorescence (QLF) have been developed. The development of objective methods has been a focus of the dental and oral health international community, as subjective methods may not be reliable upon changes on lighting conditions eye fatigue^{14–16}, and professional experience. However, the perfect function of objective methods still require correction of geometrical variables (e.g. curved surfaces and optimization of position of the device^{17,18}) and optical properties (e.g. translucency¹⁹, fluorescence, and absorption of several types of pigments). In order to keep the desired contrast between stained and unstained regions, designing optimized optical equipment and following a strict protocol are essential. Fluorescence-based methods including QLF can provide increased contrast compared to colorimetric and reflectance techniques due to the easier background removal. These methods are able to achieve reliable visual shade evaluation¹³, whose efficiency depends on the selected excitation and fluorescence emission wavelengths.²⁰

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The wavelength selection is performed after characterizing the fluorescence properties of sound tooth tissues using fluorescence spectroscopy.²¹

Fluorescence spectroscopy is a non-invasive optical method with great potential for objective and real-time assessment of biological tissues. ²² Applications include surgical guidance, disease detection, treatment monitoring, and tissue characterization. ^{23–62} Characterizing tissue fluorescence properties allows the assembly of more efficient QLF and other fluorescence-based instruments, thus improving the tooth color assessment. The fluorescence characterization for building optimized equipment requires knowledge about the contribution of all the types of pigments which could stain the teeth. However, the characterization of the absorption of tooth stain pigments is still scarce and needs to be further studied.

Previous studies reported the use of fluorescence spectroscopy to characterize the autofluorescence of sound dental²⁰ dental caries⁶³, the discrimination between sound teeth and dental calculus⁶⁴, teeth with white spot, light brown and dark brown discolored enamel caries lesions²¹. On the other hand, studies about the effect of coffee, tea, wine, dark fruits and sauces on the tooth fluorescence properties is still not missing. By knowing the changes on the fluorescence properties, it is possible to design more efficient instruments for tooth color assessment and evaluate the outcome of teeth whitening treatments such as a recent approach using violet light alone.^{65–67}

In this study, we report the fluorescence properties of bovine teeth in several degrees of coffee pigmentation. The wavelength-dependent fluorescence properties were obtained between 450 nm and 700 nm by using 445 nm excitation. We calculate parameters that can characterize the fluorescence emission such as peak wavelength shift, signal broadening, changes on total and wavelength-dependent intensity.

2. METHODOLOGY

2.1 Fluorescence spectroscopy system

Our fluorescence spectroscopy system contains a 445 nm laser (BDL-445-SMC, Becker and Hickl, Berlin, Germany), a 400-µm-diameter core bifurcated fiber (BIF400-UV-VIS, Ocean Optics, Dunedin, Florida, USA) for delivering the excitation light to the sample and for collecting the fluorescence and backscattered light (figure 1). A 475 nm longpass filter was used to remove the backscattered light. Then, the fluorescence light is detected by a portable spectrometer (USB2000-FLG, Dunedin, Florida, USA) and the data is saved in a computer. More details about the system were reported elsewhere. 68,69

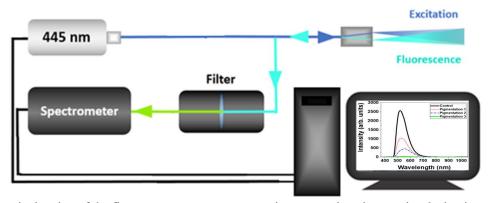


Figure 1: Schematic drawing of the fluorescence spectroscopy equipment used to characterize the bovine teeth.

2.2 Sample preparation

Bovine teeth were collected and stored in 5% thymol solution. Then, the teeth were cleaned using curettes for periodontal scaling. The dental crown of each tooth was rinsed with water, polished with pumice stone, and scrubbed by using a Robson brush. Finally, the teeth were stored at temperatures between 6 to 10 degrees Celsius until the start of the experiments. Apart from the control group, the bovine teeth were immersed in soluble instant coffee mixed in hot water at 10 mg/ml. The teeth were kept in the coffee solution during 24 hours. Pigmentation levels were categorized based on the darkness of the tooth stain.

2.3 Fluorescence measurements

The fluorescence was captured on the vestibular face of bovine incisors. At least 3 measurements were collected for each of the 5 points on the teeth surface. In order to make the average fluorescence representative of the whole tooth, the measured points were equally spaced from each other. The fiber optic probe was positioned at 90 degrees angle to the tooth surface.

2.4 Data analysis

The spectral data analysis and plotting was performed in the Origin software (OriginLab Corporation, Northampton, Massachusetts, USA). The peak wavelength shift (i.e., the wavelength shift was of the maximum intensity), the mean intensity, the full width at half maximum (FWHM) were monitored for each pigmentation level. In addition, we calculated the wavelength-dependent intensity difference between the control group and each pigmentation level from 450 nm to 700 nm. Then the effects of the coffee staining on the wavelength-dependent fluorescence intensity were calculated using the equation 1 below:

Fluorescence intensity attenuation (pigmentation level n) =
$$\frac{F_{Control\ group}(\lambda) - F_{Pigmentation\ n}(\lambda)}{F_{Control\ group}(\lambda)} \tag{1}$$

3. RESULTS AND DISCUSSION

3.1 Fluorescence spectra differences

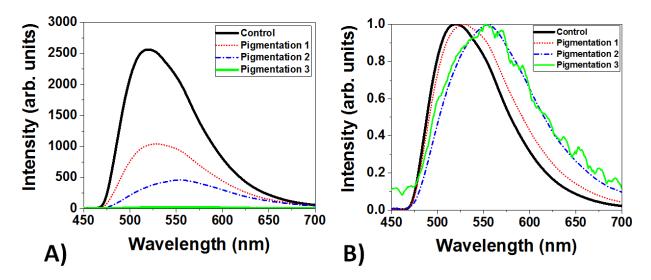


Figure 2: A) Average fluorescence spectra and B) Normalized fluorescence spectra under 445 nm excitation of bovine incisors in the control group and teeth with each pigmentation level.

Figure 2 shows the fluorescence spectra the fluorescence intensity dramatically decreases for all the wavelengths. Spectral changes such as wavelength shift and spectral broadening become clearer once the pigmentation is higher than pigmentation 2. The attenuation in intensity due to the coffee absorption shifts the normalized fluorescence intensity to longer wavelengths and broadens the normalized intensity spectrum. This effect originates from the increasing coffee absorption as wavelengths become shorter than 550 nm, i.e., from a broad wavelength-dependent coffee absorption band with peak on wavelength shorter than 475 nm.

Our results agree with previous studies reporting the intensity decrease on fluorescence and reflectance due to the tooth staining caused by beverages. ^{70–75} On the other hand, studies about wavelength-dependent fluorescence intensity

variations are scarce and limit the understanding of the tooth staining process. Based on our spectral results, the coffee absorption increases for wavelengths shorter than 550 nm and spectral features associated to this absorption can be useful to estimate the level of coffee staining on teeth.

3.2 Fluorescence spectral features of coffee absorption

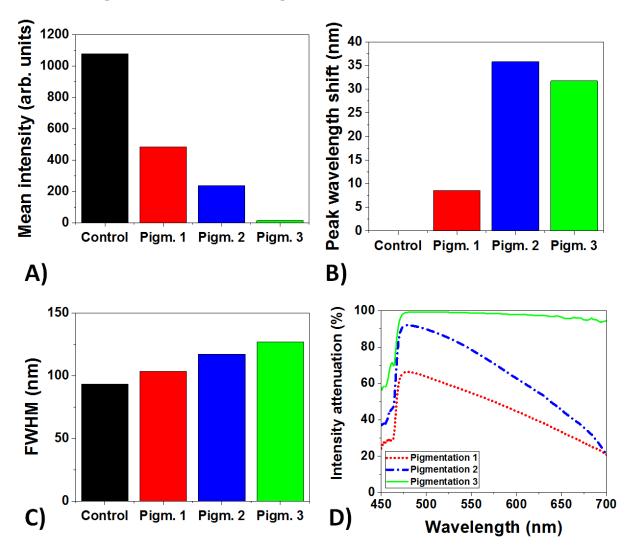


Figure 3: A) Mean fluorescence intensity between 470 nm and 700 nm, B) Wavelength shift of the maximum intensity and C) Full width at half maximum (FWHM) for the control group and each coffee pigmentation level and D) Wavelength-dependent intensity attenuation between the control group and teeth with each of the pigmentation levels.

Figure 3 shows the spectral features that can be considered on the estimation of coffee pigmentation on the tooth surface. First, the mean fluorescence intensity is the most affected parameter (figure 3A), as there is a clear attenuation as the pigmentation levels increase (54.9%, 78% and 98.7% attenuation for pigmentation levels 1, 2 and 3, respectively). On the other hand, the fluorescence intensity is difficult to be calibrated in cases when it is not possible to ensure low ambient light or when background subtraction cannot be properly performed. Second, the wavelength shift of the maximum intensity can distinguish between low and high pigmentation levels by setting a threshold of 20 nm on the wavelength shift (figure 3B). The drawback of using the wavelength shift is the non-linearity of this parameter, i.e., the wavelength shift

does not increase linearly with the pigmentation. Third, the FWHM of the fluorescence spectrum increases linearly as a function of pigmentation (figure 3C), but is not as sensitive as the first two parameters (10.8%, 25.2% and 35.9% difference for pigmentation levels 1, 2 and 3, respectively). To the best of our knowledge, previous studies on investigation of tooth staining do not describe parameters that could be used for this application other than color difference metrics, fluorescence and reflectance intensities. The parameters reported in this study can be further studied for the application of fluorescence spectroscopy on the estimation of the pigmentation level of tooth stains.

The fluorescence intensity attenuation occurs on shorter wavelengths (i.e., those closer to 475 nm). According to the measured fluorescence spectra, the peak of the coffee absorption would be close to 470 nm. However, the decrease in intensity attenuation for wavelengths shorter than 470 nm may be associated with the low fluorescence intensity in this spectral region. In this case, it is only possible to conclude the peak of coffee absorption on the tooth surface occurs on wavelengths equal or shorter than 470 nm. For attenuations close to 100% on teeth with pigmentation level 3, there may be an overestimation of the attenuation. On the other hand, the attenuation curve has the same trend for all the pigmentation levels, with a decreasing attenuation towards long wavelengths until 700 nm.

4. CONCLUSIONS

Our study quantified the fluorescence spectral features (fluorescence intensity, wavelength shift of the maximum intensity, full width at half maximum, and wavelength-dependent intensity attenuation) of teeth in several degrees of coffee pigmentation. Even though most of the pigmentation effect was observed on the fluorescence intensity, a linear behavior was observed for the FWHM (around 11.8% increase for each pigmentation level). In addition, we characterized the fluorescence properties of each degree of pigmentation level. Both spectral features and fluorescence properties can be used to design novel fluorescence equipment capable of increasing the contrast between white and stained teeth. Fluorescence spectral properties helps understanding the optical effects of the pigment accumulation on sound teeth, as well as choosing the wavelengths (or colors) were novel fluorescence or QLF equipment will operate. Future work includes determining metrics for fluorescence instrument design at specific wavelengths and further investigation of the biophotonics processes related to the light attenuation due to coffee pigmentation.

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