Direct Spectrometry: A New Alternative for Measuring the Fluorescence of Composite Resins and Dental Tissues

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Clinical Relevance

The study presents a new tool to establish a future fluorescence table of dental tissues and composites, similar to the color tables that are currently commercially available.

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SUMMARY

The aim of this study was to evaluate the fluorescence intensity of different composite resins and compare those values with the fluorescence intensity of dental tissues. Different composite resins were used to make 10 discs (2 mm in depth and 4 mm in diameter) of each brand, divided into groups: 1) Z (Filtek Z350, 3M ESPE), 2) ES (Esthet-X, Dentsply), 3) A (Amelogen Plus, Ultradent), 4) DVS (Durafill-VS, Heraeus Kulzer) with 2 mm composite resin for enamel (A2), 5) OES ([Esthet-X] opaque-OA [1 mm] + enamel-A2 [1 mm]); 6) ODVSI ([Charisma-Opal/Durafill-VSI], opaque-OM (1 mm) + translucent [1mm]), and 7) DVSI ([Durafill- VSI] translucent [2 mm]). Dental tissue specimens were obtained from human anterior teeth cut in a mesiodistal direction to obtain enamel, dentin, and enamel/dentin samples (2 mm). The fluorescence intensity of specimens was directly measured using an optic fiber associated with a spectrometer

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(Ocean Optics USB 4000) and recorded in graphic form (Origin 8.0 program). Data were submitted to statistical analysis using Dunnet, Tukey, and Kruskall-Wallis tests. Light absorption of the composite resins was obtained in a spectral range from 250 to 450 nm, and that of dental tissues was between 250 and 300 nm. All composite resins were excited at 398 nm and exhibited maximum emissions of around 485 nm. Fluorescence intensity values for all of the resins showed statistically significant differences (measured in arbitrary units [AUs]), with the exception of groups Z and DVS. Group DVSI had the highest fluorescence intensity values (13539 AU), followed by ODVS (10440 AU), DVS (10146 AU), ES (3946 AU), OES (3841 AU), A (3540 AU), and Z (1146 AU). The fluorescence intensity values for the composite resins differed statistically from those of dental tissues (E=1380 AU; D=6262 AU; E/D=3251 AU). The opacity interfered with fluorescence intensity, and group Z demonstrated fluorescence intensity values closest to that of tooth enamel. It is concluded that the fluorescence intensity values were significantly different among the composite resins and compared with dental tissues. The direct spectrofluorimetric method represents a tool for evaluating the fluorescence of composite resins.

INTRODUCTION

Personal satisfaction with one's smile may provide physical and mental well-being, primordial facts for attaining the state of health defined by the World Health Organization. The search for excellence in dentistry has favored the development of resin composites. Consequently, various types and commercial brands of composites have appeared on the market with the promise of optical properties, including fluorescence, opacity, opalescence, and translucence, similar to those of dental tissue. However, the chemical substances responsible for these properties and their concentrations in the different modalities of opaque and translucent resins and enamel are not detailed by the manufacturers.

Studies on the fluorescent phenomena involving teeth and restorative materials have shown that during the day ultraviolet (UV) radiation makes teeth appear whiter and shinier. ¹⁻⁴ This occurs because of the state of excitation of the atoms of the tooth structure, which, when they return to a state of less excitation, emit light in the visible

spectrum between 400 and 450 nm, a range characteristic of blue light. 5-7

The greater the quantity of UV light falling on the tooth surface, the greater the emission of fluorescence. Therefore, dental fluorescence becomes more evident at sea level, in the mountains, or in rooms with artificial UV light (black light). Thus, artificial materials, such as composite resins and ceramics, that do not have adequate fluorescence appear as black holes or voids in these environments.

Dental fluorescence intensity is attributed to the organic components that are photosensitive to the UV spectrum, which is why dentin presents greater fluorescence intensity than enamel. Dentin fluorescence is attributed to tryptophan and hydroxypyridine.^{4,8}

Vanini⁹ also demonstrated that a greater extent of mineralization provides a lower level of fluorescence, this being another reason why dentin would be more fluorescent than enamel. According to Dickson and others, ¹⁰ dentin fluorescence is four times greater than that of enamel, and the amelodentinal limit shows no fluorescence.

Apparently, the fluorescence of composites does not follow the same model as that of dental tissues. ¹¹ For composite resins, the superficial layers would be more relevant indicators of fluorescence properties that do not occur in the tooth, in which the more fluorescent dentin has a lower chroma. ¹² The final balance of dental fluorescence would be the sum of the enamel and dentin fluorescence. ¹³

The natural fluorescence of teeth is an important characteristic that must be reproduced in composite resin restorations to provide vitality and luminosity; it is dependent on the tooth, the restorative material, and the duration of exposure to UV light, which may occur under natural daylight or artificial light, such as that of fluorescent lamps, flashes, or the black light of nightclubs. ^{1,7,14-16} The behavior of dental tissues exposed to light has always been a complicating factor for adequate esthetic restorations, as the dental structure is polychromatic and exhibits different tonalities when light falls on it in different ways. ¹¹

A composite resin restoration must replace the lost dental structure so that it blends with the surrounding structures. Ideal restorative materials must have fluorescent properties similar to those of natural teeth. ¹⁷ If there is an absence of fluorescence, the esthetic qualities of a restoration will suffer, predominantly under UV lighting conditions. However, little is known about the extent to which base

Material/Equipment	Composition	Lot Number	Manufacturer
Esthet-X	BisGMA, modified urethane, BisEMA,TEGDMA, aluminum borosilicate fluoride glass, silanized barium	Enamel (A2): 893479	Dentsply
		Dentin (A2-0): 064644B	
Durafill VS	BisGMA, UDMA, TEGDMA, highly dispersed silicon dioxide, splinter polymer	Enamel (A2): 010213	Heraeus Kulzer
		Enamel (I): 010140	
Filtek Z-350	BisGMA, UDMA, BisEMA, TEGDMA, nanosilica filler, agglomerates of zirconia/silica particles	Enamel (A2): N125240	3M ESPE
Amelogen Plus	BisGMA, barium boron aluminosilicate glass particles	Enamel (A2): B3SH8	Ultradent
Charisma Opal	BisGMA, TEGDMA, barium aluminum fluoride glass, dispersive silicon dioxide	Dentin (OM): 010022	Heraeus Kulzer
Light Polymerizer	Light-emitting diode		Emitter- Schuster
Spectrometer Ocean Optics USB 4000			Toshiba

dimethylacrylate.

composites affect the final fluorescence of restorations and their relationships with the neighboring dental tissues. Therefore, the authors were motivated to seek details about the real properties of the materials available on the market with regards to fluorescence and to gain further knowledge about the phenomenon of fluorescence.

The aim of the present study was to evaluate the difference in fluorescence between several brands of composite resins, and combinations of those brands, on opacity and translucence, and to compare those results with the fluorescence of isolated dental tissues (enamel, dentin, and enamel/dentin) as measured by direct spectrophotometry. The hypotheses tested were as follows: 1) there would be no difference between the fluorescence of dental tissues and the tested composite resins, 2) the composites would not exhibit differences in fluorescence intensity, and 3) direct spectrophotometry would not be effective for measuring the fluorescence of composite resins and dental tissues.

METHODS AND MATERIALS

The study was approved by the Research Committee at the Sao Jose dos Campos School of Dentistry, UNESP- Univ. Estadual Paulista (Protocol 038/2009-PH/CEP).

The materials and equipment used to conduct this study are listed in Table 1, along with the respective lot numbers and manufacturers. The following brands of composite resin were used to compare the levels of fluorescence with those of a human tooth: Filtek Z350 (3M ESPE, St Paul, MN, USA); Esthet-X (Dentsply International, York, PA, USA); Durafill

VS (Heraeus Kulzer, Heraeus GmbH, Hanau, Germany), Amelogen Plus (Ultradent, South Jordan, UT, USA), and Charisma Opal (Heraeus Kulzer, Heraeus GmbH, Hanau, Germany).

A total of 70 specimens were made, divided into seven groups (n=10 each). The first four groups (n=40) were fabricated using only composite resins for enamel, in shade A2: 1) Z (Filtek Z350, 3M ESPE); 2) ES (Esthet-X, Dentsply); 3) A (Amelogen Plus, Ultradent); 4) DVS (Durafill-VS, Heraeus Kulzer). The other three groups (n=30) were fabricated using combinations of opaque, enamel, and translucent composites: 5) OES ([Esthet-X] opaque-OA [1 mm] + [Esthet-X] enamel-A2 [1 mm]); 6) ODVSI ([Charisma-Opal] opaque-OM [1 mm] + [Durafill-VSI] translucent [1 mm]); and 7) DVSI ([Durafill-VSI] translucent [2 mm]).

The specimens were obtained using a nonstick metal matrix and were standardized at 2 mm in depth and 4 mm in diameter. A polyester matrix strip was placed over the composite resin and pressed with a glass slide to provide smooth, compact, standardized specimens. The composite resin was inserted in a 1-mm increment and was polymerized with a light-emitting diode (LED) emitter (Schuster, Santa Maria, Brazil) that presented 750 mW/cm² of power for 40 seconds in contact with the polyester matrix strip.

All composite resin specimens were attached to glass slides per group of resin with the polymerized surface up, using an ethyl cyanoacrylate adhesive (Super Bonder, Henkel, Düsseldorf, Germany), to keep groups in individual supports and to have the fluorescence intensity measured in the same surface

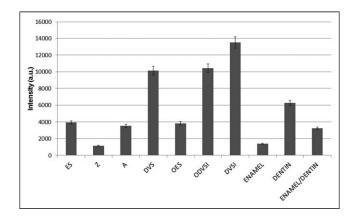


Figure 1. Column graph (mean ± standard deviation) of the fluorescence intensity values (AU) according to the different types of composite resins versus control groups (dentin, enamel and enamel/dentin).

of each specimen. These glass slides were immersed in artificial saliva at $37^{\circ}\mathrm{C}$ for 24 hours. The artificial saliva was prepared according to the method of Gohring et al. ¹⁸ using 4.8 g HCl, 3.4 g NaCl, 0.2 g MgCl₂, 0.4 g CaCl₂, 0.4 g KSCN, 1.4 g H₂KPO₄, 0.2 g H₃BO₃, and 0.4 g CHNaO₃.

Ten sound anterior human teeth, extracted for periodontal reasons, were used for comparison of the fluorescence levels of enamel, dentin, and enamel/dentin and for comparison of the fluorescence of the composite resins. The teeth were obtained following a protocol approved by the university's ethical research committee.

The initial fluorescence was recorded directly on the surface of the whole tooth using fiber optics associated with a USB 4000 spectrometer (Ocean Optics, Dunedin , FL, USA). After this initial measurement, enamel and dentin cylinders were obtained using a trephine bur (4 mm in internal diameter). To obtain 1-mm dentin specimens, the enamel of the cylinders was removed. The same was done to obtain 1-mm enamel specimens, where the dentin was removed. For that, the enamel or dentin surfaces were polished in a polishing device (DP-10, Panambra Industrial e Técnica, São Paulo, Brazil) using a sequence of 600 and 1200 grit aluminum oxide abrasive disks (Extec, Enfield, CT, USA). All specimens were stored in artificial saliva¹⁸ at 37°C, up to the time of fluorescence measurement.

The composite resin specimens were excited using an ultraviolet LED appliance with a peak centered at 398 nm. A xenon ion source (Model PX- 2), coupled to a bifurcated optical fiber connected to the spectrometer, was used to measure the fluorescence absorp-

tion and detection. The values obtained were reproduced in graphs on a computer using the Origin 8.0 program (OriginLab Corporation, Northampton, MA, USA). The fluorescence intensity values were located in the visible light spectrum between 450 nm and 700 nm.

Statistical Analysis

Dunnet, Tukey, and Kruskall-Wallis tests were performed at a level of significance of 5%.

RESULTS

The mean fluorescence intensity values of the composites, the combinations with opaque and translucent composites, and dental tissues are shown in Figure 1.

Regarding absorption measurements, there were some differences among the analyzed composites. From a general aspect, the composites had absorptions between 250 and 450 nm (Figure 2), and there was a significant difference among the composites. In the Esthet-X group, composites with different degrees of opacity had a small difference from 250 to 300 nm, probably due to the composition of Esthet-X OA2 compared with Esthet-X A2. For the Durafill VS group, when combined with Charisma Opal there was no difference in absorption, whereas the Durafill VSI group only showed a difference between 250 and 350 nm, exhibiting greater absorption. These differences may be attributed to the compositions of the composite resins, which varied according to their brand.

Based on the maximum absorption peaks, the emission spectra of all of the composite resins studied had maximum emissions at approximately 485 nm (Figure 3).

In the Z-350 group, there was a plateau during emission between 488 nm and 517 nm, probably due to the structural characteristics of this resin. In the Esthet-X group, in addition to a maximum emission at 485 nm, the appearance of a shoulder was noted at 520 nm, possibly for the same reason as the previous group. The composition of Esthet-X OA2 (opaque) did not alter the spectral profile; it only favored an increase in emission.

In the case of the Durafill VS group, there was an increase of approximately 35% in its emission compared with the Durafill VSI group. Moreover, Durafill VSI resin alone presented the highest emission, with a peak at 484 nm. In the Amelogen Plus group, in addition to the maximum emission at

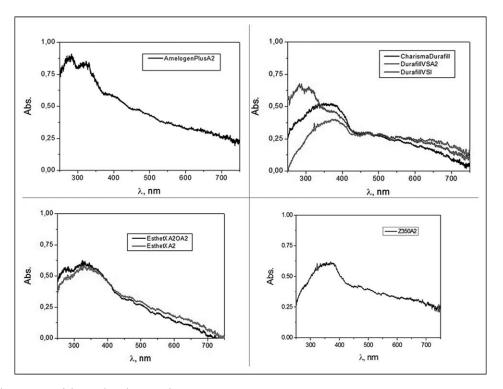


Figure 2. Absorption spectra of the analyzed composites.

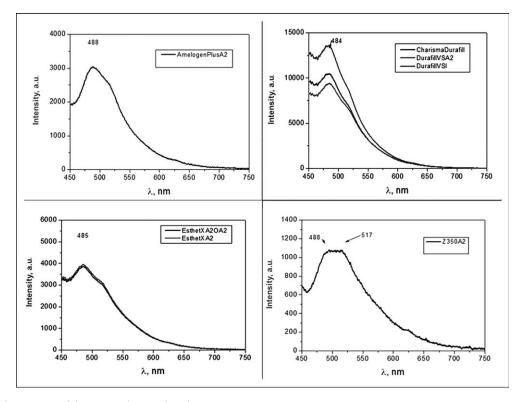


Figure 3. Emission spectra of the composites analyzed.

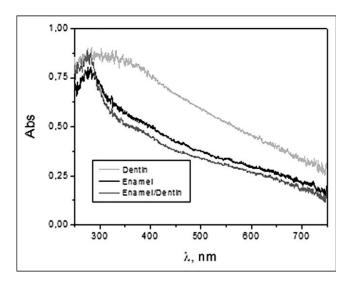


Figure 4. Absorption spectra of dental tissues.

488 nm, the beginning of a shoulder is observed at 520 nm, possibly due to its composition.

With regards to dental tissues, the light absorption spectrum was between 250 nm and 300 nm (Figure 4). Dentin presented the broadest and highest spectrum compared with tooth enamel. In the emission spectrum, the peaks were higher than 450 nm, as is shown in Figure 5, which means that the highest emission values (peak) are above 450 nm or are in the visible light spectrum area, more precisely, among the values close to 490 nm. This area (including the peak) is a result of the emission process (fluorescence) of the whole tooth due to the absorbed energy (during the light absorption process by the tooth or the resin). The absorption process implicates the transition of electrons (from the tooth or resin components) from the ground state to the excited state (higher energy level), while the emission process is implicated in the transition from the excited stated to the ground state.

Therefore, the absorption peaks of the composite resins are between 250 nm and 450 nm, and those of dental tissues are between 250 and 300 nm. All of the composites had maximum emissions close to 485 nm. There was a statistically significant difference between composites with regards to the fluorescence intensity, with the exception of the group Z-350. The translucent DVSI group exhibited the highest fluorescence intensity value (13539 AU), followed by ODVSI (10440 AU), DVS (10146.2 AU), ES (3946.2 AU), OES (3840.8 AU), A (3540.1 AU), and Z (1146.2 AU).

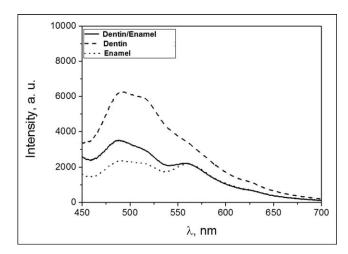


Figure 5. Emission spectra of enamel, dentin, and enamel/dentin.

All of the studied composite resins exhibited fluorescence intensities that differed statistically from those of the dental tissues (enamel=1380 AU; dentin=6262 AU; enamel/dentin=3251 AU). The Z group was the one that presented a fluorescence intensity that approximated dental enamel.

In this current study, the opaque composites did interfere in the final fluorescence analysis in mixed specimens, indicating that the subsuperficial layer may interfere in the fluorescence of composites.

DISCUSSION

The method used in this study is innovative because it is a direct fluorescence measurement method using an optical fiber. There are many reports on fluorescence measurement through lab spectrophotometers that are not suitable for clinical direct measurements. 1,8,19-21 Therefore, no studies were found in the literature about the direct measurement of fluorescence in dental tissues and composite resins samples. When the fluorescence intensity of composites and dental tissues is established, the direct method—using a spectrometer coupled with an optical fiber—will be an important clinical tool for selecting composites not only based on color shades but also on degree of fluorescence. Therefore, it will be possible to produce a new scale that involves color and fluorescence.

There is a wide variety of composite resins on the market that are true direct restoration systems presenting opaque composites for dentin and translucent composites for enamel, each with a different degree of translucence. However, fluorescence varies according to the resin brand and not according to the characteristics of its particles or opacity and trans-

lucence.¹³ The results of the present study are in disagreement with those of other studies because composites of the same brand but with distinct qualities also exhibited distinct fluorescence values.

In this current study, statistically different fluorescence intensity values were observed among the composite resins and between the dental tissues, probably because of the different compositions of each substrate. ^{1,13,15,22} For all of the composites, one may infer that the same chromophore is responsible for the phenomenon because of the similarities of the spectral profiles. However, the emission intensity might vary based on the composition of each of the composites (Filtek Z-350, 3M ESPE; Esthet-X, Dentsply; Durafill VS, Heraeus Kulzer; Amelogen Plus, Ultradent; Charisma Opal, Heraeus Kulzer). Nevertheless, manufacturers do not indicate which chemical substances are responsible for the fluorescence of their products, which encourages investigation into how the chemical composition may influence fluorescence.

A study Studies has shown lower emission of fluorescence from composite resins than from dental structure. 19 Those authors observed that dental tissues showed a greater intensity of excitation than did composite resins at a wavelength higher than 430 nm. Nevertheless, the present study showed that samples of translucent and opaque/translucent composites obtained higher fluorescence intensity values than did dental structure. Moreover, it could be perceived that, when analyzing the mixed opaque/translucent samples or translucent samples, the composites of the same commercial brand presented different fluorescence values, which occurs within natural teeth, for enamel and dentin values. This result is in disagreement with the study of Macedo and others, 13 in which the same brand of composites indicated for reproducing dentin, enamel, or the incisal edge presented equal fluorescence values.

Another relevant finding in the literature is that the fluorescence of composite resins does not follow the same model as that of dental tissues; that is to say that only the superficial layer of composite would be relevant in the fluorescence indices. ²³ In this study, composite resin for enamel samples did not obtain lower fluorescence intensities than the mixed opaque/translucent samples. The present study revealed the interference of the subsuperficial layer in the measurement of the fluorescence of composite resins, which contradicts other findings in the literature. Furthermore, other studies agree that

the application of sealants and accumulation of pigments may alter the fluorescence of a composite, both in the transmission of light on the surface of the material and in the absorption of the fluorescence emitted. 6,16,21,24

In some studies, the results indicated that there is a considerable variation in fluorescence between restorative materials and dental structure. Some authors found that the transmission of light was lower than that of dental tissues. Other authors found that the fluorescence intensity of the restorative material was higher, which compromised the quality of the restoration, affecting the esthetic success or failure of restorative treatment.

The behavior of the dental structure exposed to light has always been a complicating factor in adequate esthetic restorations, as the dental structure is polychromatic and exhibits different tonalities when exposed to different types of light. 11,25 In the current study, the dental structure could be analyzed in specimens of enamel/dentin, enamel, and dentin, which showed differences in their intensities, proving the polychromatic structure of dental tissues. Dentin exhibited greater fluorescence intensity than enamel because of a higher collagen content, which contains the amino acids responsible for fluorescence, such as tryptophan and hydroxypyridine.^{8,9} However, many studies did not include separate test specimens of enamel and dentin for their analyses, obtaining only the combined results of the fluorescence of the two tissues.

The results of this present study showed that the composite resin Filtek Z-350 (A2) most approximated the fluorescence intensity of tooth enamel. Whereas Esthet-X (A2), Amelogen Plus (A2), and Durafill VS (A2) obtained higher values than those of pure enamel, which would compromise the result of the restoration. However, these composite resins presented values closer to the values obtained with the enamel/dentin combination. These results differed from those of previous studies, in which no brand of composite resin for enamel or translucent composite had fluorescence intensities similar to that of enamel, and only Esthet-X OA2 had fluorescence similar to that of human dentin.⁸

All of the combinations of opaque, enamel, and translucent composite resins may vary in terms of fluorescence for different brands. This study demonstrated that different combinations of composite resins/shades interfere with the final fluorescence

result. Fluorescence varies from tooth to tooth and between dental tissues. Therefore, it is of great importance to develop a method capable of directly measuring tooth fluorescence in vivo for developing a future composite resins fluorescence guide. Further studies should establish the fluorescence of the available composite resins and their combinations so that this information can be used in clinical practice in the same way the shade guide is used.

CONCLUSIONS

According to the methodology used, it can be inferred that

- The null hypotheses were refuted, as there were significant differences in fluorescence among the analyzed composite resins and the dental tissues;
- The direct method of measuring fluorescence using a spectrophotometer is efficient, in addition to being a promising tool for selecting composite resins by fluorescence.

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Conflict of Interest

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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