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ORIGINAL ARTICLE

Effect of Toothpaste Fluoride Concentration on Color Stability of Resin Composites: An *In Vitro* Study

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ABSTRACT
Objective: To investigate the effects of two different dentifrice fluoride concentrations on the color stability of the composite. **Material and Methods:** Twenty-seven specimens (2×4×5 mm) each of microfilled (Gradia, GC, Japan) and nanohybrid (Grandio, VOCO, Germany) composites were prepared. The specimens were randomly divided into six groups (control, Fluoflor caries protection toothpaste with 1450ppm Fluoride (EXW, France), and Fluoflor kids toothpaste with 500ppm Fluoride (EXW, France) (n = 9). The specimens were immersed in a mixture of artificial saliva and toothpaste in a ratio of 1:3 and applied for 60 seconds every 12 hours for 42 days. The control samples were incubated in artificial saliva at 37°C. Primary and secondary color measurements were performed using color parameters (L*a*b) with a spectrophotoshade (MHT Optic Research AG, Niederhasli, Switzerland). Data were analyzed using a two-way analysis of variance at a significance level of 0.05. **Results:** According to the two-way ANOVA analysis, there was no significant difference in color change between the composites and no difference in the level of discoloration between different fluoride concentrations($p>0.05$). Also, None of the dentifrices caused clinically significant color changes($\Delta E<3.3$). **Conclusion:** No clinically unacceptable color changes were observed in the microfilled and nanofilled composites with different concentrations of fluoride toothpaste.

Keywords: Color; Composite Resins; Fluorides; Toothpastes.

Introduction

The demand for esthetic composite materials has increased as more and more attention is paid to cosmetic dentistry to achieve a bright white smile [1]. The efficiency of esthetic restorative materials depends on their resistance to degradation and longevity in the oral cavity [2]. Endogenous and exogenous factors affect the color harmony and stability of materials. The exogenous factors that cause discoloration include poor oral hygiene, diet, smoking, and adsorption or absorption of dye-containing solutions by the resin matrix. Endogenous factors that cause changes include the resin matrix, matrix/filler particle interface, resin matrix, photoinitiator system, light curing device used for polymerization, and irradiation time [1-4].

The most common and efficient method of oral hygiene in most countries is brushing the teeth with toothpaste. Fluoridated dentifrices are responsible for reducing the prevalence of caries observed in industrialized countries in recent decades [5]. The caries-preventive effect depends on the solubility of the fluoride-containing compound, which causes the fluoride to adhere to the tooth surface. *In vitro* experiments indicate that inorganic and organic fluorides in toothpaste systems significantly stimulate fluoride uptake on the tooth surface, leading to remineralization [6]. In addition, the amount of toothpaste and the extent of tooth brushing greatly affected the uptake and remineralization of enamel fluoride [7].

Different types of commercial toothpaste are available on the market, which could contain various fluoride content, such as sodium fluoride, sodium monofluorophosphate, and amine fluoride [5]. In addition, toothpastes contain different fluoride concentrations. Most people use dentifrice with a fluoride concentration of 1000-1100 ppm. Lower concentrations are used for children because of the risk of fluorosis. Higher fluoride concentrations (1500 ppm) are prescribed for children and adults with a higher caries risk or living in a non-fluoridated area. The results of the studies show that the higher the fluoride concentration, the higher the enamel fluoride uptake (EFU) [5,8].

While some studies have shown no difference in color change with the use of mouth rinses or fluoride solutions, some studies have shown that they can change the color of composites. There is controversy in various studies regarding the color change [9-11]. The fluoride ions of sodium fluoride present in topical fluoride solutions may dissolve the surface layer of dental composites, causing discoloration by increasing the roughness of the surface. Long-term use also increases discoloration. Due to the daily use of toothpaste, this may happen when fluoridated toothpaste is used [11-13]. In addition, one study showed that fluoride toothpaste could cause a significant clinical color change in a subgroup of the composite resin [14]. Also, in another study, the color change of the composite was clinically unacceptable [15].

While most studies have focused on the effect of different fluoride compounds on the color change of composites [16], no article was found that closely examined the color change of composites with the change in fluoride concentration. However, very few studies have investigated the effects of fluoridated dentifrices on the color stability of composite materials, which are limited to whitening toothpaste or charcoal-based products [17-20]. Therefore, the present study aimed to investigate the effects of different concentrations of fluoridated toothpaste on the color stability of two different resin composites. The null hypothesis tested in this study was that fluoridated toothpaste did not affect the color stability of composite resins.

Material and Methods

Study Design and Sample

In this in vitro study the minimum sample size required for each of the six study groups was 9 samples, according to Lepri et al. [2]. using the one-way ANOVA power analysis feature of PASS 11 software (power 0.95, $\alpha = 0.05$, $\beta = 0.15$).

Materials

The restorative materials and different dentifrices used in the present study are listed in Table 1.

Table 1. The Resin composites and dentifrices were used for this study.

Material	Type	Matrix	Composition (Filler Type)	Manufacturer
Grandio	Nanohybrid	Bis-GMA, TEGDMA	Microfiller 1µm (Glass-ceramic), Nanofiller 20-60 nm spherical (Si02)	Voco, Cuxhaven, Germany
Gradia	Microfilled hybrid	UDMA	Silica, Pre-polymerized filler, Flouro-aluminosilicate glass	GC Dental Products Corp. 2-285 Torllmastsu-Cho, Kasugai, Aichi, Japan
Fluoflor Caries Protection Toothpaste			Aqua, Sorbitol, Hydrated Silica, Peg-8, Sodium Lauryl Sulfate, Cellulose Gum, Sodium Monofluorophosphate (1450 ppm F), Titanium Dioxide/Ci Sodium 77891, Aroma, Limonene, Pvp, Sodium Saccharin, Methylparaben, Calcium Glycerophosphate, Hydrated Allantoin, Calcium Silica Ci 74160, 2-Bromo-2 Nitropropane-1,3-Diol.	EXW France
Kids Fluoflor Toothpaste			Sorbitol, Aqua, Hydrated Silica, Glycerin, Peg-32, Sodium Lauryl Sulfate, Cellulose Gum, Aroma, Monofluorophosphate (500ppm of Fluorine), Sodium Saccharin, P34 Gluconate, Sodium Methylparaben Propylparaben, Ci 12490	EXW France

Laboratorial Procedures

Twenty-seven specimens of two resin composites listed in Table 1 were prepared and converted into rectangular-shaped samples using the A2 shade of each resin composite and a stainless-steel mold (2×4×5mm). The specimens were inserted into the mold over glass slides and were covered with a celluloid strip (Polydentia, Swiss product). The material was polymerized on both sides using an LED (Guilin Woodpecker Medical Instrument Co., Ltd, Guilin, China) device for 20 seconds. A radiometer was used to check the intensity of visible light at 1000 mW/cm^2. The upper surfaces of the specimens were polished and struck ten times with fine and superfine soflex discs. Superfine soflex discs are equivalent to 2500-grit silicon carbide papers [18]. After each polishing, a new disk was used. Only one operator did the polishing process to control the variability, and the amount of brushing time was determined by conducting a pilot study. The samples were incubated in artificial saliva at 37°C (1±°C) for 24 hours [21].

The initial color values of each specimen (L*, a*, b*) were determined using a spectrophotoshade (MHT Optic Research AG, Niederhasli, Switzerland) against a white background. The colorimeter was calibrated before every measurement session according to the manufacturer's instructions using the existing white calibration standard [21].

Subsequently, the specimens were randomly divided into six groups (n = 9) (Table 2), including Fluoflor caries protection toothpaste with 1450 ppm fluoride, Fluoflor kids toothpaste with 500 ppm fluoride, and a control group (artificial saliva). The samples were immersed in a mixture of artificial saliva and toothpaste in a ratio of 1:3. This mixture was applied for 60 seconds every 12 hours for 42 days for the second color measurement. During this time, the samples of the control group were incubated in artificial saliva at 37 °C. The specimens were subject to the daily cycle for 42 days [22]. The same spectrophotoshade was used for the second color measurement.

Table 2. Division of the prepared specimens alongside a selected toothpaste into different groups.

Specimens	Materials/Toothpastes, Control
G_{NC}	Nanohybrid/control(artificial saliva)
G_{NF}	Nanohybrid/ Fluoflor caries protection toothpaste with 1450 ppm fluoride
G_{NK}	Nanohybrid/ Fluoflor kids' toothpaste with 500 ppm fluoride
G_{MC}	Microfilled/control(artificial saliva)
G_{MF}	Microfilled/ Fluoflor caries protection toothpaste with 1450 ppm fluoride
G_{MK}	Microfilled/ Fluoflor kids' toothpaste with 500 ppm fluoride

The color change of the specimens was determined by the color difference formula (ΔE), which is the difference between the initial and final values. The three-dimensional color space representing color perception (three axes of L*, a*, and b*) has been proposed by the International Commission on Illumination (CIE) [23].

Color difference (ΔE) was calculated for each specimen according to the following equation: $\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]1/2$ where L* represents lightness, a* green-red (-a=green; +a=red) and b* blue-yellow (-b=blue;+b=yellow) [19,24]. The ΔE values >3.3 are visually perceptible and clinically unacceptable [15,25].

Data Analysis

Statistical analysis was done using two-way analysis of variance (ANOVA). P-values <0.05 were considered significant.

Results

The mean and standard deviation of ΔL*, Δa*, Δb*, and ΔE* before and after the exposure to fluoride-containing toothpaste are presented in Table 3. According to the two-way ANOVA analysis, there is no interaction between the composite and the type of fluoride toothpaste (p=0.223). The type of composite does not affect the color change (ΔE) of the composite (p=0.763), and the relationship between the color change and the type of fluoridated toothpaste is also not significant (p=0.518). None of the specimens showed color change with values above the clinically acceptable (ΔE >3.3).

Table 3. ΔL*, Δa*, Δb*, and ΔE* mean and standard deviation, before and after exposure within each specimen.

Specimen	ΔE* (Mean±SD)	Δa*	Δb*	ΔL*
G_{NC}	0.31±0.14	0.07±0.17	0.11±0.23	0.05±0.15
G_{NF}	0.17±0.1	-0.02±0.13	-0.02±0.13	0.00±0.08
G_{NK}	0.24±0.14	0.01±0.1	0.03±0.21	-0.02±0.17
G_{MC}	0.20±0.14	0.02±0.12	-0.01±0.19	-0.01±0.12
G_{MF}	0.24±0.24	0.03±0.17	0.06±028	0.01±0.1
G_{MK}	0.21±0.2	0.05±0.15	0.08±0.2	0.03±0.11

The N_1 group showed the maximum, and the N_2 group showed the minimum color change in the Grandio direct nanohybrid composite. As for Gradia's direct microfilled composite, the M_2 group showed the maximum, and the M_1 group showed the minimum color change. However, there was no significant difference between the two composites.

Moreover, all the specimens experienced positive Δa changes (a redshift) except N_2. Δb of all groups was positive (a yellow shift) except for the N_2 and M_1 groups. All groups had positive changes in ΔL except N_3 and M_1 groups, indicating that the brightness level of most specimens increased (Figure 1).

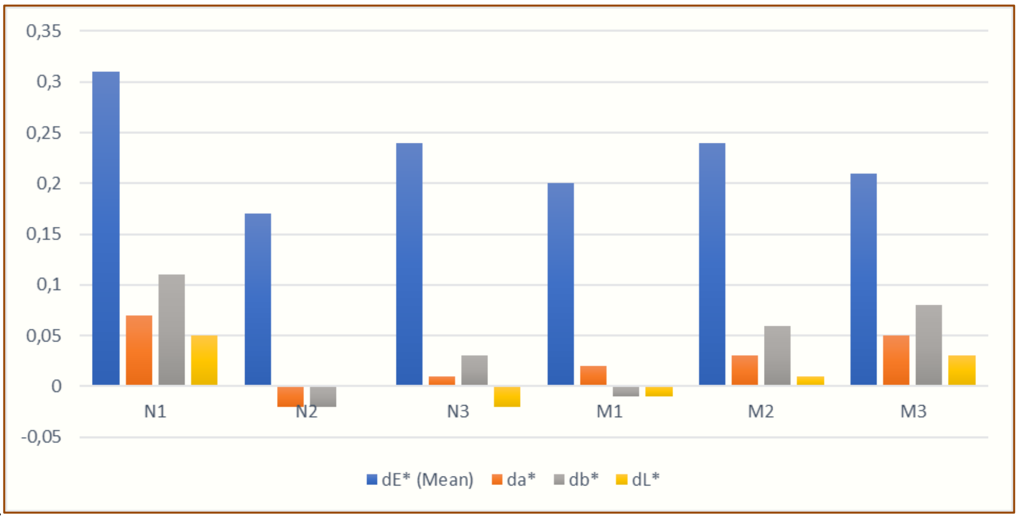

Figure 1. Average ΔE*, Δa*, Δb*, and ΔL* parameters.

Discussion

Composite materials are a significant group of restorative materials widely used in dentistry. The significant use of composite materials prompts various studies to develop their physical and chemical properties. One of the most critical factors for any restorative material is its longevity, which depends on its color, surface roughness, and microhardness [17,18]. Restorative materials require more attention due to their stay in the oral cavity and constant exposure to foods, beverages, mouth rinses, and toothpaste, as these materials can change the color of composites. The color stability of composite restorations is influenced by several internal and external factors [1,2].

This study investigated the effect of fluoridated toothpaste on the color stability of composite restorations. As the results of our research, the null hypothesis was accepted due to not obtaining significant differences between the color changes of restorative materials after being immersed in different dentifrices. Accordingly, none of the dentifrices caused clinically substantial color changes in the composite materials. There was no significant difference in color change between the microfilled and nanohybrid composites. Furthermore, there was no difference in the color change of composite restorations between different concentrations of fluoride toothpaste.

The two most common methods of measuring tooth color are visual examination, the usual method used in dentistry, and computer analysis of digital photographs. Other research methods for examining tooth color include spectrophotometry, spectroradiometry, tristimulus colorimetry, and digital color analysis. Using a spectrophotometer can improve inherent objective problems and standardization and is accurate and reliable for the quantitative measurement of tooth color [26]. Color perception varies in different individuals and even in the same individual at different times. The characteristics of the human eye, lighting conditions, translucency, opacity, and light scattering can affect color perception [27]. In addition, $\Delta E < 1$ is not detectable by the human eye; experts can detect ΔE between 1 and 3.3, and $\Delta E > 3.3$ can be detected by laypeople. $\Delta E > 3.3$ is not clinically acceptable [25,28].

Some studies investigating whitening or charcoal toothpaste used conventional toothpaste containing 1450-1550 ppm fluoride as a control group [15,18-20,24,29]. The composite discoloration caused by fluoride toothpaste in these studies was consistent with the results of the present study [18-20,24,29]. However, Pintado-Palomino et al. [15] reported inconsistent results in a double-masked randomized controlled trial. Although the discoloration caused by fluoride toothpaste was not statistically significant in composites, the color change was clinically unacceptable, as the ΔE value was reported to be 4.4. This color alteration is attributed to other ingredients in the toothpaste and the reduction in tooth staining. Accordingly, it is impossible to compare these studies and our study [15] accurately. In addition, the examination method differed in these studies, and the specimens were brushed [15,18-20,24,29]. The frequency and technique of toothbrushing may influence the discoloration of restorative materials [15]. Nonetheless, in a study by Torso et al. [20], the discoloration caused by fluoride-containing toothpaste was clinically acceptable in the control group regardless of the number of toothbrushing cycles and the abrasive effect. In the present study, we omitted toothbrushing to limit the investigation of the impacts of fluoride-containing materials on composite color change.

Different restorative materials were used in previous studies. Researchers have shown that Compomer discoloration is clinically acceptable when fluoride toothpaste is used [16,18]. On the other hand, the color change caused by fluoridated toothpaste is not clinically acceptable, with a mean ΔE value of 5.14 for cention N (a subgroup of composite) [14]. Nonetheless, the materials used in the present study were composites, so an exact comparison was impossible.

Color changes in composites are due to adhesion failure at the matrix-filler interface, water absorption, surface roughness, nutrition, and oral hygiene. The chemical structure and size/type of filler particles may also influence the color change of composites [17,30,31]. The resin matrix, the main component of the composite, plays an essential role in color stability, and different pH values and alcohol concentrations in the solution may affect discoloration [32]. In addition, the penetration of dyes in the surface of the resin composite exposed to the oral environment and their physicochemical properties are the factors that cause negative color changes in the resin composite, which were not considered in our study [3,30,33]. However, the analysis of Mundim et al. [11] has shown that fluoride solutions change the surface roughness of the composite below the critical limit.

Water sorption is a major factor initiating degradation. The hydrophilic resin matrix affects the water sorption of the resin composite through its polymer network; as a result, it causes a whiter and more opaque color change [30,34]. Some studies concluded that TEGDMA, as part of the matrix in resin materials, has a significant hydrophilic capacity and increases the sensitivity of Bis-GMA to water absorption compared to DMA (UDMA) so that UDMA provides better color stability than Bis-GMA [3]. The higher the degree of water absorption in the resin composite, the more the color stability reduction as the free volume of the formed polymer increases. Subsequently, more space is created to diffuse water molecules in the polymer structure [33]. This phenomenon is called plasticization of the composite, which softens the polymer matrix and leads to more color changes in the resin composite [25,34]. Therefore, GRADIA, in the presence of UDMA in the matrix, is expected to be more resistant to discoloration than GRANDIO with Bis-GMA and TEGDMA in the matrix. In addition, studies have shown that composites with large filler particles are more prone to water absorption and discoloration [30]. De Moraes Rego Roselino et al. [24] reported that the color change induced by fluoridated toothpaste was more significant in the z250 composite (microhybrid) with larger filler particles compared to the z350 composite (nanofill). However, the present study showed no difference in color change between the GRADIA and GRANDIO composites.

This finding could be related to the fact that the toothpaste used in the present studies contained no potential causes of composite discoloration, such as chlorhexidine, low pH, and alcohol [32]. It could also be related to the short study period, lack of toothbrushing, and the resulting roughness. In clinical practice, fluoride toothpaste's effects on composite materials' color stability depend on many factors, such as saliva and salivary pellicles, foods, beverages, and oral hygiene habits that cannot be simulated in vitro. Therefore, further *in vivo* studies are required. It is suggested to use other materials, such as fluoride alternatives, or compare with other artificial stainings, such as tea and chlorhexidine, to study the discoloration of different composites in future studies.

Conclusion

Within the limitations of this *in vitro* study, the selected fluoridated toothpaste containing different fluoride concentrations did not have any critical effects on the color stability of the two resin composites, i.e., there was no significant difference in the color change between the different compositions used. However, the result of a laboratory study may differ from clinical conditions.

Authors' Contributions

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EA	https://orcid.org/0000-0002-3428-1045	Conceptualization, Methodology and Writing - Review and Editing.
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All authors declare that they contributed to a critical review of intellectual content and approval of the final version to be published.

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Conflict of Interest

The authors declare no conflicts of interest.

Data Availability

The data used to support the findings of this study can be made available upon request to the corresponding author.

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