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Influence of Staining Solution and Bleaching on Color Stability of Resin Used for Caries Infiltration

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Clinical Relevance

A bleaching treatment of resin-infiltrated enamel lesions may improve appearance after staining.

SUMMARY

Objective: The objective of this study was to evaluate the color stability of Icon-infiltrated white spot lesions after staining and the bleaching effect on the infiltrated and stained surfaces.

Methods and Materials: Enamel-dentin specimens (N=30, 5 × 5 × 3 mm, 1-mm enamel + 2-mm dentin thickness) were prepared from bovine incisors and randomly allocated into three groups (n=10): control, demineralized, and infiltrated. Artificial enamel subsurface lesions were created using 50 mL of 0.05 M acetate buffer solution. Specimens were produced by Icon application in enamel caries-like lesions, according to the manufacturer's instruction. Baseline color readings were assessed using a spectrophotometer, and CIE L*a*b* measurements of each specimen were performed using a white background. To simulate extrinsic dietary staining, specimens were placed into a 4-mL coffee infusion, three times daily for 15 minutes, for 14 days. After the staining procedure, color measurements were performed again. Then, bleaching procedures were performed using 16% carbamide peroxide gel for four hours daily for 21 days, and a final color assessment was performed. To compare the baseline and final measurements, *t*-test was used (α =0.05). The statistical comparison between the groups was performed using the one-way analysis of variance and Tukey tests (α =0.05).

Results: Coffee staining provided a significant reduction of L* values and an increase of a* and b* in all groups (control, decayed, and infiltrated). The bleaching procedure provided

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DOI: 10.2341/14-290-L

a significant increase in L* and decrease of a* and b* values in all groups. There was no significant difference in ΔE values between decayed and infiltrated groups before bleaching, and after bleaching, the infiltrated group showed the lowest ΔE values.

Conclusion: It can be concluded that enamel infiltrated with Icon presents significant alteration of color after staining when compared with sound enamel. However, if there is discoloration of the infiltrant, the bleaching treatment can be used successfully.

INTRODUCTION

Orthodontic treatment with fixed appliances increases the risk of developing demineralized white spot lesions. The main reason for the development of caries is plaque stagnation around braces, mostly underneath the arch wires and between the bracket and gingival margin.[1] An established active white spot lesion has a chalky, opaque appearance, since the light is scattered mainly within the lesion body. Scattering is caused in interfaces between substances with different refractive indexes. Untreated white spot lesions may result in progression to cavities and severe esthetic problems.[2] In addition, sometimes during the remineralization of these white spot lesions, stains get incorporated into the lesion, leading to the creation of brown spots and thus increasing the esthetic problem. Hence, the treatment of these white spot lesions should aim both to prevent the caries progression and to improve esthetics by decreasing opacity and staining.[3]

Lately, a new approach has been launched to arrest noncavitated enamel lesions. After erosion of the pseudo-intact surface layer with hydrochloric acid, low-viscosity resins (Icon) penetrate within the lesion.[4] Thus, the porosities of carious lesions can be occluded, and the diffusion of acids and minerals is reduced. In this way, lesion progression is hampered, and caries progression is slowed down or even arrested.[4] A study conducted by Paris and Meyer-Lueckel[5] demonstrated that the infiltration technique might be an alternative to microabrasion and restorative treatment, particularly for white spot lesions of esthetically relevant teeth, as in cases of white spot lesions in facial surfaces.

According to the manufacturer, Icon is a TEGDMA-based resin matrix. Some authors have suggested that the addition of TEGDMA to a restorative material could increase water sorption,[6,7] decrease general mechanical properties,[8] and hinder color stability.[9] The discoloration of restorative materials might be attributable to water sorption and the hydrophilicity of the matrix resin. If the resin material can absorb water, it can also absorb other fluids, resulting in the alteration of color.[10] This process may cause plasticization and softening of the resin matrix and reduction of color stability. Discoloration of tooth-colored resin-based materials may be caused by intrinsic and extrinsic factors. Extrinsic factors such as adsorption of stains may also cause discoloration.[11] In this way, surfaces containing the infiltrant and submitted to colored solutions can be stained, since their composition contains a greater amount of TEGDMA. For infiltrated lesions (white spot lesions treated with Icon), this could be an esthetic problem.

To solve this problem caused by staining solutions, some recent studies reported polishing of the infiltrated lesions and concluded that polishing increases their resistance to staining challenges.[12,13] However, it seems clinically impossible to remove only the infiltrant because it penetrates the enamel. Over time, the infiltrant could stain again, and the polishing procedures would need to be performed again, resulting in greater wear and excessive loss of enamel.

Some physical and mechanical properties of infiltrants have already been studied,[14] but few studies have been published regarding their esthetic behavior.[13,15] Rey and others[15] showed that Icon showed a higher staining susceptibility compared with adhesive systems. Thus, it is necessary to develop a method to improve Icon appearance after staining. Borges and others[13] suggest repolishing the infiltrated lesions to minimize the staining effect. However, this polishing alternative may result in unnecessary enamel wear caused by abrasives.

A very common procedure is to reach esthetic conditions using bleaching. Bleaching methods have been developed, and peroxide compounds at different concentrations are used for tooth-whitening procedures. Contemporary bleaching agents are typically either hydrogen peroxide or carbamide peroxide. The procedure may be performed at a dental office or by applying the agent by the patient in gel form within the confines of a custom tray.[16]

As a highly hydrophilic material, the infiltrant can be stained, but its behavior in bleaching procedures is unknown. No study has assessed the staining behavior of infiltrated lesions after staining and bleaching. It is questionable whether bleaching treatment is effective after infiltrant staining. Thus,

the aims of this study were 1) to evaluate the color stability of white spot lesions with Icon after staining and 2) to evaluate the bleaching effect on these infiltrated and stained surfaces. The hypotheses tested were 1) coffee staining causes a color change in the infiltrated enamel and 2) a bleaching procedure provides color recovery for the infiltrated enamel.

METHODS AND MATERIALS

Specimen Preparation

Enamel-dentin specimens (N=30, 5×5×3 mm, 1-mm enamel + 2-mm dentin thickness) were prepared from bovine incisors. Enamel surfaces were polished (silicon carbide paper 1200, 2400, 4000), and dentin surfaces were covered with two layers of acid-resistant nail varnish (Colorama, São Paulo, Brazil), leaving a 4×4 mm^2 exposed area. All specimens presented the same enamel thickness, and using a simple drawing, they were randomly allocated into three groups (n=10): control, demineralized, or infiltrated.

Artificial enamel subsurface lesions on the unprotected areas were created by storing specimens in 50 mL of 0.05 M acetate buffer solution, considering a ratio of 2.0 mL/mm^2 of exposed enamel, pH 5.0, at 50% hydroxyapatite saturation, for 10 hours at 37°C.[17] Caries-like lesions were etched with 37% phosphoric acid gel Scotch Bond Etchant (3M ESPE, St Paul, MN, USA) for 60 seconds,[18] washed with water spray, and dried for 15 seconds. Then, the specimens were dried by immersion in 100% ethanol. Icon (DMG, Hamburg, Germany) was applied to the etched enamel surface using a microbrush, resting for 3 minutes, according to the manufacturer's instructions. Specimens were light cured for 40 seconds using Free Light 2 (3M/ESPE) with 1000 mW/cm^2 irradiance, measured with a power meter (Ophir Optronics Inc, Danvers, MA, USA). The infiltrant was applied for a second time, resting for one minute, and then light cured for 40 seconds. All specimens were immersed in artificial saliva for 24 hours.

Color Assessment

An initial color reading of the specimen (baseline) was performed using a spectrophotometer (CM-700d, Konica Minolta, Tokyo, Japan) in reflectance mode. The samples were positioned in a sample carrier in a light cabin (GTI Mini Matcher MM1e, GTI Graphic Technology Inc, Newburgh, NY, USA) to standardize the ambient light during the measurement process, and then the samples were subjected to a reading with the spectrophotometer. The CIE L*a*b* measurements of each specimen were performed using a white background. In the color space, L* indicates lightness (L+ = lightness and L– = darkness), the a* coordinate represents the red/green range (a*+ = redness and a*– = greenness), and the b* coordinate represents the yellow/blue range (b*+ = yellowness and b*– = blueness). The values of the coordinates a* and b*, when approaching zero, indicate neutral colors (white and gray) and an increase in magnitude for more saturated or intense colors. The L*a*b* color space includes all perceivable colors and is based on a cube root transformation of the color data. This detailed analysis through the study of the coordinates L* a* and b* separately helps to better understand which one was more responsible for the total color change (ΔE).

The total color change (ΔE) was calculated according to the following formula[19]: $\Delta E = [(L_1 - L_0)^2 + (a_1 - a_0)^2 + (b_1 - b_0)^2]^{1/2}$, using On Color QC Lite software (Konica Minolta) to generate spectral measurements as a function of wavelength for data processing and analysis.

Staining Procedure

To simulate extrinsic dietary staining, specimens were placed into 4 mL of coffee infusion (Tradition Nescafé, Nestlé, Araras, SP, Brazil) of 8 g (powder) to 100 mL boiled water. After preparation, the specimens were immediately placed in contact with the coffee. This procedure was repeated for 15 minutes three times daily for 14 days. After the staining procedure, color measurements were performed again.

Bleaching Procedure

For the bleaching procedure, specimens were fixed in a device, and approximately 1 mm of the bleaching agent was applied to the exposed enamel surface. The 16% carbamide peroxide gel (Whiteness Perfect, FGM, Santa Catarina, Brazil) was left in contact with the enamel surfaces for four hours daily for 21 days.

The samples were maintained in relative humidity until the final color assessment (after 21 days), which was performed following the procedure of the initial color assessment. The L*, a*, b* and ΔE parameters were subjected to statistical analysis. To compare the baseline and final measurements, a *t*-test was used ($p<0.05$). The statistical comparison between groups was performed using one-way

Table 1: *Means and Standard Deviations of L*, a*, and b* Parameters Observed for the Groups Considering Time and Treatment for Each Group (No Bleaching and Bleaching) Individually*[a]

	Color Parameter	Time	Control	Decayed	Icon Infiltrated
No bleaching	L	Baseline	88.20 (1.37) Ab	93.10 (1.53) Aa	87.60 (1.14) Ab
		After staining	75.91 (3.41) Ba	60.50 (3.71) Bb	62.54 (1.80) Bb
	a	Baseline	−0.09 (0.33) Bb	0.26 (0.14) Bb	0.96 (0.43) Ba
		After staining	3.42 (0.91) Ab	3.08 (0.69) Ab	8.45 (0.64) Aa
	b	Baseline	11.78 (1.77) Ba	5.07 (0.68) Bb	9.33 (1.13) Bc
		After staining	14.08 (1.27) Aa	10.41 (1.23) Aa	18.79 (1.50) Aa
Bleaching	L	Baseline	75.90 (3.41) Ba	60.49 (3.71) Bb	62.17 (1.86) Bb
		After bleaching	87.19 (2.12) Ab	91.65 (2.92) Aa	88.27 (1.30) Ab
	a	Baseline	3.42 (0.91) Ab	3.07 (0.69) Ab	8.34 (0.57) Aa
		After bleaching	0.40 (0.66) Bb	1.19 (0.79) Ba	0.62 (0.34) Bb
	b	Baseline	14.07 (1.27) Aa	10.41 (1.23) Aa	18.64 (1.28) Aa
		After bleaching	11.54 (2.72) Ba	2.48(2.33) Bb	10.18 (1.63) Ba

[a] *Capital letters indicate comparison between the baseline and final measurement of the column. Lowercase letters demonstrate comparison between groups in rows. Different letters indicate statistically significant differences (*$p<0.05$*).*

analysis of variance and Tukey tests ($p<0.05$). Statistical analyses were performed by Assistat software (Campina Grande, Brazil).

RESULTS

Comparison of Values Between Baseline and After Staining

Table 1 shows the L*, a*, and b* mean values of the baseline and after the staining procedures, before and after bleaching, and Table 2 shows the ΔE mean values for all groups.

Coffee staining provided a significant reduction of L* values for all groups (control, decayed, and infiltrated). After coffee staining, the control group presented the highest L* values compared with the other groups. There was a significant increase for the a* and b* values for all groups ($p<0.05$). Infiltrated enamel showed, at baseline, significantly higher a* and b* values than decayed and sound enamel. Despite a significant baseline difference among all groups concerning the a* and b* values, all groups showed significantly higher a* and b* values after coffee staining, but they did not show any differences among each other after staining.

The bleaching procedure provided a significant increase in the L* values for all groups (control, decayed, and infiltrated). After bleaching, decayed enamel presented the highest L* values compared with the other groups, but there was no significant difference between the control and infiltrated groups. There was a significant decrease for the a* and b* values for all groups ($p<0.05$). After bleaching, decayed enamel showed significantly higher a* values than infiltrated and sound enamel, and there was no significant difference between infiltrated and control groups for the b* values. The lowest b* values were exhibited by the decayed group.

The lowest ΔE values were observed in the control group ($p>0.05$) when considering baseline × staining. However, after bleaching, the stained groups showed the highest ΔE for the decayed group compared with the group infiltrated with Icon. There was no significant difference in the ΔE values between the decayed and infiltrated groups ($p<0.05$) before bleaching, indicating that the Icon-infiltrated group underwent a significant color change similar to the decayed group. However, after bleaching, the infiltrated group showed the lowest ΔE values.

DISCUSSION

Color changes in direct restorative materials, and more specifically in resin materials, have a direct influence on esthetics and therefore on the clinical longevity of a restoration. Enamel infiltrated by no-filler low-viscosity resin may become an esthetic

Table 2: *ΔE Values Observed by All Groups in All Experimental Periods*[a]

Group	ΔE	
	Baseline × Staining	Staining × Bleaching
Control	13.1206 b	3.7379 ab
Decayed	33.1940 a	5.6464 a
Infiltrated	41.1049 a	2.2845 b

[a] *Different letters indicate statistically significant differences (*$p<0.05$*) for independent comparisons considering baseline × staining and staining × bleaching individually.*

problem over time because, like any resin material, it can be subject to alteration of color. The extent of discoloration varies according to the habits of the patient, such as oral hygiene and diet.[20]

Coffee was chosen as a dye-testing substance in this study because it is frequently consumed. Coffee exhibits a strong potential for staining both tooth structure and resin materials.[21] The compatibility between the brown dye from coffee and the resin polymer chain has been suggested to facilitate the adsorption and penetration of the dye in the resin.[21]

This study was performed using bovine teeth instead of human teeth because Attia and others[22] found that bovine and human enamel substrates behave similarly in terms of staining and bleaching effect. Moreover, the composition, density, and microhardness of bovine substrate are very similar to those of human enamel.[23,24]

Many methods are currently used to assess tooth color. Since spectrophotometers allow an objective color assessment and provide precise quantitative data,[25] this was the method used in the present study. The color alteration measurements were evaluated using reflectance measurements with the CIE Lab Color coordinate system. According to Dietschi and others,[26] when the three coordinates of color dimensions are analyzed separately, the L* values, which represent the lightness of the object, appear to be the most relevant parameter for comparisons under experimental conditions. Also, the ΔE values were analyzed because they indicate the magnitude of the color change at two different moments.[12]

The first hypothesis tested in this study was supported, as it was verified that after staining there was a significant L* value decrease for all groups. According to Joiner,[19] the L* value is a measurement of the lightness of an object and is quantified on a scale such that a perfect black has an L* value of zero and a perfect reflecting diffuser has an L* value of 100. The L* values decreased, probably because of the incorporation of the dye present in coffee into the infiltrant. Moreover, the sound enamel presented the highest L* values, which were significantly different from those of decayed enamel infiltrated with Icon. This means that infiltrated enamel is able to incorporate more dye. Clinically, this could be an esthetic problem. In addition, a* and b* coordinates increased after the staining procedures. The values of the a* and b* coordinates approached zero, indicating neutral colors (white and gray) and an increase in magnitude for more saturated or intense colors.[19]

These alterations can be explained by the composition of Icon. Its matrix is TEGDMA based, with no filler particles.[4] Some authors have suggested that TEGDMA is a monomer that presents high water sorption and has a hydrophilic behavior compared with other monomers.[4,7] Thus, it can be assumed that Icon would easily absorb dyes that are present in beverages and food. The a* and b* coordinate changes drove the alterations of the ΔE values, as sound enamel exhibited the lowest ΔE values, and enamel infiltrated with Icon presented almost three times higher ΔE values.

The second hypothesis was also supported, as there were significant alterations of the color coordinates and the ΔE values. After staining, all groups submitted to the bleaching treatment showed increased L* values, indicating higher lightness because of the application of carbamide peroxide. Moreover, decreased a* and b* values indicate that there was dye neutralization. Joiner[19] assumed that, as the values of the a* and b* coordinates approach zero, they indicate neutral colors. The carbamide peroxide action is due to its own breakdown into hydrogen peroxide and urea. Urea further breaks down into ammonia and carbon dioxide, which accounts for the elevation of the intraoral pH. Hydrogen peroxide breaks down into water, oxygen, and free radicals, which result in oxidation of the pigments in teeth.[27] The ΔE values showed that enamel infiltrated with Icon provided similar alteration of color after bleaching when compared with sound enamel. These results indicate that it is possible to increase the brightness of the enamel infiltrated with Icon, making the use of polishing procedures unnecessary.

Borges and others[13] showed that demineralized enamel treated with resin infiltration showed significant staining when exposed to coffee and wine, consistent with the results presented in this study. They suggested that the repolishing of the specimens could minimize the staining effect. However, polishing procedures may remove unnecessary enamel structure, causing iatrogenic damage to the enamel. This study suggests a more conservative approach: the bleaching procedure. A bleaching procedure does not promote the removal of enamel, and according to the results presented in this study, after carbamide peroxide bleaching, the ΔE values of the demineralized enamel treated with resin infiltration were statistically similar to those of the control group. According to the CIE/Lab units, a ΔE of less than 1 is excellent, and a ΔE value less than 3.3 is considered clinically insignificant.[28]

The effects of staining and bleaching on composite resins have been reported. Villalta and others[29] observed that composite resins were affected by staining solutions, such as wine and coffee, after bleaching. After bleaching, discoloration was removed completely from the composite resins, probably because of the superficial cleansing of the specimens, which may explain the results of this study regarding resin infiltration.

CONCLUSION

Based on the results obtained on this study, it can be concluded that enamel infiltrated with Icon presents significant color alteration after staining when compared with sound enamel. Therefore, patients should avoid the consumption of colored beverages and foods to increase the longevity of the resin infiltration in esthetically important areas. However, if the discoloration of the infiltrant occurs, bleaching treatment can be used successfully. *In vivo* studies should be performed to assess more accurately the staining behavior of Icon.

Acknowledgment

The authors wish to thank DMG (Brazil) for providing Icon kits for this research.

Regulatory Statement

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of UNICAMP.

Conflict of Interest

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

(Accepted 27 May 2015)

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