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Effect of 10% and 15% Carbamide Peroxide on Fracture Toughness of Human Dentin *In Situ*

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Clinical Relevance

There was no significant decrease in mean dentin fracture toughness after 10% and 15% carbamide peroxide bleaching *in situ*. This provides some reassurance that dentin is not overtly weakened by the bleaching protocol used in this study.

SUMMARY

Purpose: **Although damage to the structural integrity of the tooth is not usually considered a significant problem associated with tooth bleaching, there have been some reported negative effects of bleaching on dental hard tissues *in vitro*. More studies are needed to determine whether the observed *in vitro* effects have practical clinical implications regarding tooth structural durability.**

Objectives: **This *in situ* study evaluated the effect of 10% and 15% carbamide peroxide (CP) dental bleach, applied using conventional whitening trays by participants at home, on the fracture toughness of dentin.**

Methods: **Ninety-one adult volunteers were recruited (n ≈ 30/group). Compact fracture toughness specimens (approximately 4.5 × 4.6 × 1.7 mm) were prepared from the coronal dentin of recently extracted human molars and gamma-radiated. One specimen was fitted into a prepared slot, adjacent to a maxillary premolar, within a custom-made bleaching tray that was made for each adult participant. The participants were instructed to wear the tray containing the dentin specimen with placebo, 10% CP, or 15% CP treatment gel overnight for 14 nights and to store it in artificial saliva when not in use. Pre-bleach and post-bleach tooth color and tooth sensitivity were also evaluated using ranked shade tab values and visual analogue scales (VASs), respectively.**

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Within 24–48 hours after the last bleach session, the dentin specimens were tested for fracture toughness using tensile loading at 10 mm/min. Analysis of variance, Kruskal-Wallis, χ^2, Tukey's, and Mann-Whitney U tests were used for statistical analysis. The level of significance was set at $p<0.05$ for all tests, except for the Mann-Whitney U tests, which used a Bonferroni correction for post hoc analyses of the nonparametric data ($p<0.017$).

Results: The placebo, 10% CP, and 15% CP groups contained 30, 31, and 30 participants, respectively. Mean fracture toughness (± standard deviation) for the placebo, 10% CP, and 15% CP groups were 2.3 ± 0.9, 2.2 ± 0.7, and 2.0 ± 0.5 MPa*m$^{1/2}$ respectively. There were no significant differences in mean fracture toughness results among the groups ($p=0.241$).

The tooth sensitivity VAS scores indicated a significantly greater incidence ($p=0.000$) and degree of tooth sensitivity ($p=0.049$ for VAS change and $p=0.003$ for max VAS) in the bleach groups than in the placebo group. The color change results showed generally greater color change in the bleach groups than in the placebo group ($p=0.008$ for shade guide determination and $p=0.000$ for colorimeter determination).

Conclusions: There were no significant differences in *in situ* dentin fracture toughness results among the groups. The results of this study provide some reassurance that dentin is not overtly weakened by the bleaching protocol used in this study. However, the lack of a statistically significant difference cannot be used to state that there is no effect of bleach on dentin fracture toughness.

INTRODUCTION

Tooth bleaching is a popular procedure that can be prone to overuse in an attempt to achieve a whiter tooth color, either by using a bleach concentration that is too high or by bleaching for a prolonged period of time. However, it is not known whether the active ingredients in tooth bleaching materials (hydrogen peroxide or carbamide peroxide [CP]) damage the structural integrity of the tooth. Thus, identifying the short- and long-term effects of tooth bleaching is a targeted research priority on the American Dental Association Research Agenda.[1] Although many studies have investigated the effects of tooth bleaching on enamel and dentin surface properties, such as hardness,[2–4] surface morphology,[5–7] bonding,[3,4,8–10] surface demineralization,[11,12] and abrasion/erosion,[13] relatively fewer studies have investigated the effects of tooth bleaching on enamel and dentin mechanical properties, such as strength or fracture toughness.[14,15]

It has been reported that the flexural strength and modulus of bovine dentin decreased after an *in vitro* direct daily application of carbamide peroxide.[16] Significant reductions in tensile and shear strengths of dentin were reported after an *in vitro* direct intracoronal bleach application of 30% hydrogen peroxide.[17] It has been shown in another *in vitro* study that the fracture toughness of dentin was significantly reduced by the indirect (through intact enamel) application of peroxide bleaching agents (this represents a simulation of clinical bleaching of teeth with full enamel coverage) and by a direct bleach application method to dentin, and that fracture toughness was further reduced with a longer application time period (8 weeks vs 2 weeks) and a higher (16% vs 10%) bleach concentration.[18] The clinical relevance of these *in vitro* studies is uncertain. The fracture resistance of endodontically treated teeth was reported to decrease after an internal and external bleaching procedure.[19] However, the observed decrease in tooth fracture resistance was attributed more to a bleach-induced decrease in bond strength rather than a bleach-induced effect on the tooth structure itself. In the clinical setting, where the estimated number of tooth bleachings performed on or by patients ranges in the millions, there have been no published reports of tooth fractures attributable to tooth bleaching procedures. *In situ* or *in vivo* studies are needed to determine whether the observed *in vitro* effects have practical clinical implications regarding tooth structural durability.

The objective of this study was to determine the effect of dental bleaches, applied in a conventional manner by patients, on the fracture toughness of dentin placed *in situ*. If a decrease in *in situ* dentin fracture toughness or surface hardness is found, the results of this study would provide valuable confirmation to the *in vitro* literature that suggests that tooth weakening may occur as a result of direct bleach treatment. The null hypothesis for this study was that bleaching has no effect on the fracture toughness of dentin *in situ*.

Tooth sensitivity and color changes were also evaluated in this study to confirm the bleaching effect for each patient.

MATERIALS AND METHODS

Ethics approval for the collection of teeth and for this *in situ* study was obtained (University of Toronto Office of Research Ethics Protocol #21379 and #24941, respectively).

Human molars (extracted within 3 months of the experiment and stored in 1% chloramine solution) were collected to provide the dentin for testing. One dentin specimen was obtained from each tooth. Compact tension test specimens (Figure 1), based on an American Society for Testing and Materials standard specimen[20] and described previously,[18,21] were prepared from the dentin below the occlusal enamel initially using a water-cooled, low-speed diamond saw (Buehler Ltd, Lake Bluff, IL, USA), keeping the location and orientation of the dentin standardized. High-speed dental instrumentation was then used to form the rectangular block-shaped specimen with approximate dimensions 4.5×4.6×1.7 mm. A central notch was made with a 0.28 mm thick diamond disc (ThinFlex, Premier Products Co, Plymouth Meeting, PA, USA) and sharpened with a razor blade to act as a stress concentrator. A tungsten carbide drill bit (LA ¼ round, catalog 00400327, Brasseler, Savannah, GA, USA) was used to drill two cylindrical holes, approximately 0.8 mm in diameter, in each specimen to provide means of attachment for mounting. A micrometer (Digimatic Caliper, Mitutoyo Corporation, Kanagawa, Japan) was used to measure specimen dimensions (a, B, W, H, and N) to the nearest 0.01 mm. Equipment, instruments, and preparation materials were disinfected, sterilized, and/or disposed for each individual specimen's preparation following standard universal precautions infection-control procedures. Finally, the dentin specimens were stored in artificial saliva[22] in individual containers and sterilized using gamma-irradiation at 2.5 MRad for 1500 minutes (cobalt source [Co-60] from GammaCell-220, Atomic Energy of Canada Limited, Kanata, Canada).

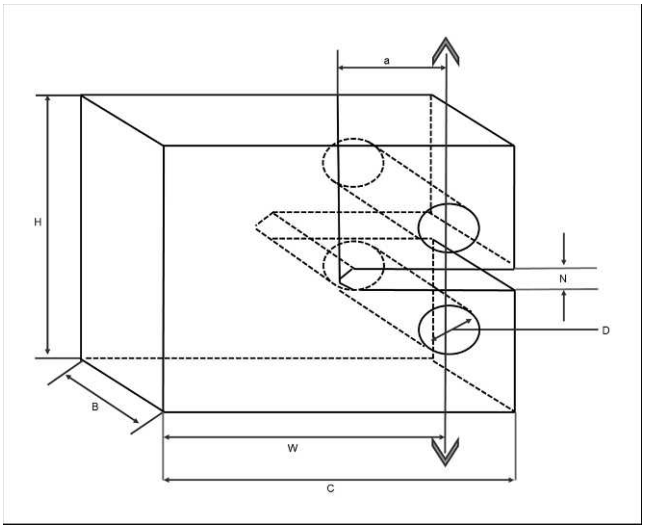

Figure 1. *Diagram of fracture toughness specimen. Total width (C) ≈ 4.6 mm. Net width (W) ≈ 3.75 mm. Height (H) ≈ 4.5 mm. Thickness (B) ≈ 1.7 mm. a/W ratio ≈ 0.45–0.55. Notch width (N) < 0.65W. Effective notch length (a) = 0.25W – 0.4W. Hole diameter (D) ≈ 0.8 mm. B/W ratio = 0.25–1.25. Large arrowheads indicate direction of tensile loading.*

The selected sample size (n=30/group) was based on a sample-size calculation of 27 using a Type 1 error alpha = 0.05, Type 2 error beta = 0.2, a high standard deviation (0.8 $MPa*m^{1/2}$) value compared with those generally obtained in previous *in vitro* dentin fracture toughness testing done by the author to reflect the higher variance seen *in situ* compared with *in vitro*, and a smallest difference of clinical interest value = 0.7 $MPa*m^{1/2}$. The criteria for acceptance into the study were as follows: adults (≥18 years old), willingness to participate in the study, a noncontributory medical history (not pregnant, not lactating), and a noncontributory dental history (ie, no xerostomia, no untreated carious lesions, not presently undergoing orthodontic treatment). Compensation of $20 was provided for each participant in the study. Participants who had recently (within 1 year) bleached their teeth were not included in the study. The one-year exclusion period was based on general current recommendations for frequency of tooth bleaching (based on need for re-bleaching after color regression).

The bleach materials included 10% or 15% CP (Opalescence or Opalescence 15% PF, Ultradent Products Inc, South Jordan, UT, USA). A placebo gel (Ultradent Products), without the active CP, was used as the control. The investigators gave participants the opportunity to join the placebo, 10% CP, or 15% CP groups, and recruitment continued until the approximate selected sample size was reached for each group.

Custom-made bleaching trays were constructed to deliver the bleach to the participant's teeth and to hold the dentin specimen. LC Block Out Resin (Ultradent Products) was applied to the facial surfaces of the incisors and premolars on the dental stone model to an approximate thickness of 0.5 mm. A composite resin block with the approximate dimensions of the dentin fracture toughness specimen was bonded onto the buccal surface of a maxillary premolar on each dental stone model to create a space for the actual dentin specimen in the bleaching tray. Vinyl tray material (Sof-Tray Regu-

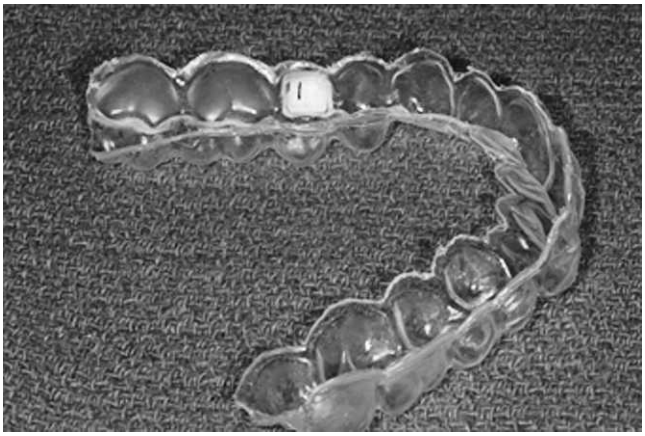

Figure 2. *The dentin fracture toughness specimen is shown sutured in place within the custom-made bleach tray.*

lar 0.35", Ultradent Products) was then vacuum-formed to the stone model and trimmed along the gingival margins. A randomly selected dentin fracture toughness specimen was inserted into the tray in the space created by the composite resin block and sutured to the tray (4-0 silk black-braided, Ethicon Inc, Somerville, NJ, USA) (Figure 2).

All participants received a demonstration and instructions concerning the proper use of the bleaching agents. To standardize participants' oral hygiene regimens, they were asked to not use dentifrices that contained whitening agents. Participants were instructed to wear the bleaching tray containing the dentin specimen overnight for 14 nights to simulate a typical at-home bleaching regimen. At the end of each daily bleach treatment, the participants rinsed the tray and dentin specimen with tap water to remove all external traces of bleach and stored the tray and dentin specimen in artificial saliva until the next bleach treatment. If the participants experienced tooth sensitivity, they were advised that they could choose to skip one or two days of bleaching. Participants were also advised that they were free to stop participating at any time during the study. Participants were asked to make a daily record of the following on a provided log form: (1) the number of hours of bleaching done and (2) the degree of tooth sensitivity experienced as shown on a 10-mm visual analogue scale (VAS) (Table 1).

Tooth sensitivity was also tested by before-bleaching and after-bleaching cold testing. The cold testing involved placing 0.3 mL of 7°C glycerine gel to the middle of the facial surface of the lateral incisor for 3 seconds. At the end of 3 seconds, the gel was wiped off, and the patient was asked to score the lateral incisor tooth sensitivity on a VAS. The daily tooth sensitivity and the cold test VASs were measured to the nearest millimeter to determine the change in tooth sensitivity score for the cold test after the study compared with before the study (VAS change = VAS score for cold test after the study – VAS score for cold test before the study), the maximum tooth sensitivity score during the study period (max VAS), and the percentage of participants who experienced no sensitivity (where VAS=0 for each treatment day).

Pre- and post-bleach tooth shades for one central incisor were recorded by visual matching and by using an Easy Shade colorimeter (Vident, Brea, CA, USA). Shade selection was carried out under similar clinical conditions for each participant in a neutral-colored room. The visual evaluation was made by

Table 1. *Form Participants Used to Log Daily Records of Number of Hours of Tray Wear and Degree of Tooth Sensitivity Based on a 10 mm Visual Analogue Scale (VAS)*

Date	Hours of Tray Wear	Tooth Sensitivity after indicated hours of tray wear: Please mark an X on the line to represent the maximum degree of tooth sensitivity experienced during the day. "A" represents no sensitivity and "B" represents extreme sensitivity.
		A tooth sensitivity B

Table 2. *Rank Scores Assigned to Each Shade Tab According to Value*

	Darkest----------																			Lightest
	Classic Shade Guide[1]																Bleach Shade Guide[2]			
Shade Tab	C4	A4	C3	B4	A3.5	B3	D3	A3	D4	C2	C1	A2	D2	B2	A1	B1	040	030	020	010
Rank	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20

[1] Vita Classic Shade Guide (Vident, Brea).
[2] *Bleach Shade Guide (Ivoclar Vivadent).*

comparing the shade tabs (Vita Classic shade guide, Vident, and Bleach Shade Guide, Ivoclar Vivadent, Amherst, NY, USA) with the middle third of the selected upper central incisor by the same investigator before and after treatment. There was no attempt to use or calibrate more than one investigator for pre- and post-bleach shade tab color assessment. A custom-made self-cure resin jig was used to ensure standardized positioning of the Easy Shade probe. The 16 shade tabs from the Vita Classic guide and 4 shade tabs from the Bleach Shade guide tabs were ranked according to value from 20 (highest value = 010) to 1 (lowest value = C4) (Table 2). The colorimeter shade results were limited to the 16 Vita Classic shade tabs only. Color change was determined by subtracting the pre-bleach shade from the post-bleach shade for each participant. Pre- and post-bleach digital photographs of the anterior teeth (in a fixed position) were also taken under standardized lighting conditions in a neutral-color room.

Within 24 hours after the last bleaching session, the dentin fracture toughness specimens and log forms were returned to the investigator, and the post-bleach tooth color and sensitivity assessments were done. Within 24 to 48 hours after the last bleaching session, the dentin specimens were mounted on an Instron universal testing machine (Model 4301, Instron Corp, Canton, MA, USA) for fracture toughness testing using a custom-designed mounting jig. Tensile loading was applied at a rate of 10 mm/minute until specimen fracture. The force recorded at fracture was used to calculate fracture toughness, K_{1C}.

The patient age, number of nights and hours of bleach or placebo treatment, fracture toughness results, and tooth sensitivity scores (VAS change and max VAS) were analyzed using analysis of variance ($p<0.05$). The Kruskal-Wallis ($p<0.05$) test was performed to analyze the color data, and χ^2 ($p<0.05$) were conducted to compare the gender and incidence of no tooth sensitivity (VAS = 0). Tukey's test ($p<0.05$) and Mann-Whitney U test with a Bonferroni correction ($p<0.017$) were used for post hoc analyses of the parametric and nonparametric data, respectively.

RESULTS

There were 30, 31, and 30 participants in the placebo, 10% CP, and 15% CP groups, respectively. The gender, mean age, and mean number of nights and hours of bleach treatment for each group are shown in Table 3. There were no significant differences among the groups for gender ($p=0.519$),

Table 3. *Gender and Age Distribution of Groups and Number of Nights and Hours of Placebo or Bleach Application (mean ± standard deviation)*[a]

	No.	Men	Women	Age, years	No. of Nights	No. of Hours
Placebo	30	14	16	29.4 ± 8.0^{y}	13.8 ± 0.7^{y}	85 ± 22^{y}
10% CP	31	10	21	27.2 ± 6.4^{y}	13.7 ± 1.8^{y}	$91 \pm 19^{y,z}$
15% CP	30	12	18	26.3 ± 6.0^{y}	14.1 ± 0.7^{y}	101 ± 17^{z}

[a] *Results denoted with the same superscript letters indicate no significant difference ($p>0.05$).*

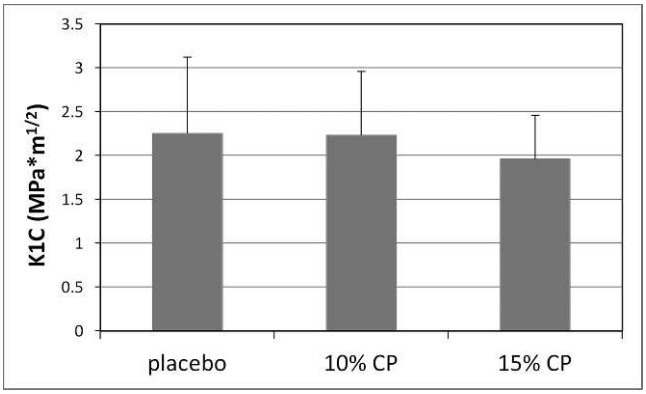

Figure 3. *Mean fracture toughness (K_{1C}) (± standard deviation) for the placebo, 10% CP, and 15% CP groups. There were no significant differences in fracture toughness results among the groups (*p*=0.241).*

age (p=0.216), and nights bleached (p=0.381). There was a significant difference in the number of hours bleached (p=0.007); the 15% CP bleach group wore the tray for a significantly greater number of hours than the placebo group.

The K_{1C} results are shown in Figure 3. Mean (± standard deviation) fracture toughness results for the placebo, 10% CP, and 15% CP groups were 2.3 ± 0.9, 2.2 ± 0.7, and 2.0 ± 0.5 MPa*$m^{1/2}$, respectively. There were no significant differences in fracture toughness results among the groups (p=0.241).

Tooth sensitivity data from the VAS scores are tabulated in Table 4. Compared with the placebo group, one or both of the bleach groups reported a significantly greater increase in the tooth sensitivity score for the cold test after the study compared with before the study (VAS change, p=0.049), a higher degree of maximum tooth sensitivity during the study period (max VAS, p=0.003), and a smaller percentage of sensitivity-free patients (VAS = 0, p=0.000).

Table 4. *Tooth Sensitivity Results*[a]

Group	VAS Change[b]	max VAS[c]	% VAS = 0[d]
Placebo	$0 + 8^{y}$	12 + 23	75
10% CP	$2 + 13^{y,z}$	$36 + 32^{y}$	17^{y}
15% CP	$7 + 13^{z}$	$31 + 28^{y}$	10^{y}

[a] *Results denoted with the same superscript letters indicate no significant difference (p>0.05 for VAS change and max VAS; p>0.017 for % VAS).*
[b] *Difference (mean + standard deviation) in VAS scores, in millimeters, for the cold test after the study compared with before the study (VAS change = VAS score for cold test after the study – VAS score for cold test before the study).*
[c] *Maximum tooth sensitivity VAS score (mean ± standard deviation) in millimeters, reported during the study period (max VAS).*
[d] *Percentage of participants who experienced no sensitivity (VAS = 0 for each treatment day).*

Tooth (central incisor) color data are shown in Table 5. There was no significant difference in initial (pre-bleach) tooth color among the groups when determined visually using the shade guides (p=0.175) or the Easy Shade colorimeter (p=0.182). There were significant differences in color change results when determined visually using the shade guides (p=0.008) or the Easy Shade colorimeter (p=0.000); color changes were generally greater in the bleach groups than in the placebo group. Although the color of the dentin specimens was not specifically measured, whitening of the dentin specimens in the bleach groups was evident.

DISCUSSION

This study investigated the effects of a placebo, 10% CP, and 15% CP treatment on dentin fracture toughness *in situ* and assessed tooth color change and tooth sensitivity *in vivo*. In contrast to the number of *in vitro* studies that have assessed the effect of tooth bleaching on enamel and dentin, the number of *in situ* studies is relatively small. A few *in situ* studies have measured the effects of tooth bleaching on enamel microhardness and have reported no significant differences.[23–26] The effect of tooth bleaching on the fracture toughness of dentin has not been studied *in situ*.

The participants and investigators were not blinded to the treatment used in this study. The lack of blinding was recognized as a potential source of bias, especially during shade assessment by the investigator and during recording of tooth sensitivity by the participant. However, it was decided to give participants the opportunity to choose their treatment group. This precluded the possibility of patient blinding. Furthermore, we considered that blinding throughout the experimental period would have been difficult because teeth generally do become noticeably whiter and more sensitive in the bleach groups. The main objective of this study was fracture toughness assessment, and the lack of blinding was not expected to have a significant effect on the eventual fracture toughness tests, in which the results are not subjective.

Fracture toughness is an intrinsic material property that measures the fracture resistance of the material. A small reduction in the fracture resistance of the tooth could have a great impact over the

Table 5. *Tooth (Central Incisor) Color Data*[a]

Group	Pre-bleach Shade Rank (Visual)[b]	Color Change (Visual)[c]	Pre-bleach Shade Rank (Easy Shade)[b]	Color Change (Easy Shade)[c]
Placebo	14.7 ± 2.4^{y}	1.1 ± 2.0^{y}	12.5 ± 4.2^{y}	0.3 ± 0.9^{y}
10% CP	15.0 ± 3.2^{y}	2.8 ± 2.8^{z}	13.6 ± 3.5^{y}	$1.3 \pm 1.3^{y,z}$
15% CP	13.9 ± 2.8^{y}	3.2 ± 2.3^{z}	13.9 ± 2.3^{y}	1.7 ± 2.0^{z}

[a] *Results denoted with the same superscript letters indicate no significant difference ($p>0.017$).*
[b] *Pre-bleach shade ranks (mean ± standard deviation) as determined by the operator (visual) or the Easy Shade colorimeter (Vident) using ranked shade tabs (Vita Classic Shade Guide,Vident, Brea or Bleach Shade Guide, Ivoclar Vivadent). A higher shade rank represents a lighter shade.*
[c] *Color change results (mean ± standard deviation) were determined by subtracting the pre-bleach shade rank from the post-bleach shade rank for each participant.*

lifetime of the tooth as a result of fatigue and crack propagation. Therefore, it is important to characterize any changes to the structural integrity of dentin that could occur as a result of bleaching treatment. The previously reported *in vitro* reductions in dentin fracture toughness observed as a result of bleach treatment[18] were not confirmed by this *in situ* study. Factors that may have contributed to the different *in vitro* and *in situ* dentin fracture toughness results include saliva and bleach considerations.

The *in vitro* specimens were stored in 37°C artificial saliva during the entire study period,[18] while the *in situ* specimens were exposed to human saliva during bleach treatment and stored in room-temperature artificial saliva when not undergoing bleach treatment. Human saliva has a buffering action because of the bicarbonate and phosphate systems, and it contains inorganic electrolytes, such as calcium phosphorus and fluoride, as well as enzymes and bacteria. Smidt and others[27] stated that the buffering capacity and remineralization potential of saliva *in vivo* could overcome the detrimental effects of bleach on enamel microhardness and surface morphology observed *in vitro*. In an *in situ* study of enamel microhardness, it was concluded that saliva led to mineral reposition on bleached enamel surfaces and reestablishment of hardness values similar to those of non-bleached specimens.[24] It is possible that human saliva played a role in this *in situ* study by preventing or reversing a potential reduction in dentin fracture toughness caused by bleach application.

Although there would have been significant variation in the amount of bleach applied to the bleaching tray because of participant variability, overall the *in situ* dentin specimens were probably exposed to a smaller amount of bleach than the *in vitro* specimens. The *in vitro* dentin specimens were exposed to bleach on all of its surfaces.[18] The *in situ* dentin specimens in our study were exposed to direct bleach application primarily on one surface only. The bleach on the *in situ* dentin specimens was also subject to washout, salivary dilution, and salivary antioxidants. The reduced amount of bleach on the *in situ* dentin specimens may partially explain why there was no significant difference in dentin fracture toughness among the groups in this study. However, there was sufficient bleach in the tray to cause a bleach effect, as evidenced by the color change and tooth sensitivity results.

Dentin is chromatic and enamel is not. Dentin is therefore the target tissue for bleaching. Studies of direct applications of bleach to dentin are relevant because dentin can be exposed to direct bleach application in clinical situations. It is impossible to avoid direct contact of bleach to dentin when there is exposed dentin during a typical home bleaching treatment using a tray or strips. Two common clinical situations in which dentin is exposed are occlusal attrition or root recession. In those situations, one or two surfaces of dentin would be exposed to direct bleach application. This *in situ* study mimicked these clinical situations better than the previous *in vitro* study[18] by limiting the direct application of bleach primarily to one dentin surface.

As expected, the bleach groups showed significant increases in tooth sensitivity results and color change results compared with the placebo group, and there was a trend for greater color change and tooth sensitivity with the greater bleach concentration. The standard deviations for the tooth sensitivity and color change results were high, suggesting a wide range of potential bleach treatment results. The lack of significant difference in color change (Easy Shade) between the placebo and 10% CP group

was probably because of the high standard deviation in the color change results.

The number of total bleaching hours for the 15% CP group was significantly greater than that for the placebo group. It is likely that the patients in the 15% CP group were more motivated to wear the bleaching tray for longer periods of time because they perceived that their teeth were becoming whiter. The greater number of bleach hours, in addition to the higher bleach concentration, for the 15% CP group may have further contributed to these participants' increased tooth sensitivity and color change results compared to the placebo group.

The design of this *in situ* study is more clinically relevant than the design of *in vitro* studies. The results of this study therefore could provide some reassurance that dentin is not overtly weakened by the bleaching protocol used in this study. However, the lack of a statistically significant difference cannot be used to state that there is no effect of bleach on dentin fracture toughness (this would risk committing a Type 2 error). An inadequate sample size or study design may have contributed to the lack of significant findings. The *in situ* specimens were subject to greater variability than the *in vitro* specimens in the form of different tray-wear patterns by the different participants, fluctuating intraoral temperatures, and varying intraoral conditions. An *in vivo* situation would add even more variability. Vital teeth would have an outward movement of fluid through dentinal tubules, which would tend to expel and buffer the applied bleach. Further studies with a greater sample size or different study design are needed to find more evidence to accept or reject the null hypothesis.

Although there was no statistical difference, the results of this study did show a slight trend for reduced dentin fracture toughness with the higher bleach concentration, which is in accordance with previously reported *in vitro* results.[18] Tooth bleaching materials are available over-the-counter and patients may overuse these products in an attempt to further whiten their teeth. Clinically, it is quite common for a patient to repeat the bleaching procedure several times for several weeks or to use higher bleach concentrations in order to achieve a satisfactory lightening of tooth color. It is not known whether repeated bleaching or use of a higher bleach concentration would cause a significant weakening of the dentin. The results of this study cannot be extrapolated to bleach concentrations or application times higher or longer than those used in this study. Until the specific effects of tooth bleach on dentin are clarified, it remains prudent to keep bleaching concentrations and times to a minimum and to avoid direct application of bleach to areas of exposed dentin, such as in gingival recession or occlusal attrition cases, whenever possible.

CONCLUSION

1. Mean fracture toughness (± standard deviation) for the placebo, 10% CP, and 15% CP groups were 2.3 ± 0.9, 2.2 ± 0.7, and 2.0 ± 0.5 MPa*$m^{1/2}$, respectively. There were no significant differences in *in situ* dentin fracture toughness results among the groups (p=0.241). The results of this study therefore could provide some reassurance that dentin is not overtly weakened by the bleaching protocol used in this study. However, the lack of a statistically significant difference cannot be used to state that there is no effect of bleach on dentin fracture toughness.
2. Compared with the placebo group, the bleach groups reported a significantly greater increase in the tooth sensitivity score for the cold test after the study compared with before the study (p=0.049), a higher degree of maximum tooth sensitivity during the study period (p=0.003), and a fewer number of sensitivity-free days (p=0.000).
3. There was a significant difference in color-change results among the groups when measured by visual shade-matching (p=0.008) and by the Easy Shade spectrophotometer (p=0.000).

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Conflict of Interest Declaration

The authors 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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REFERENCES

1. American Dental Association (2010) ADA Research Agenda. Research of importance to the practicing dentist, 2010–2011. Retrieved online January 5, 2011 from: http://www.ada.org/sections/about/pdfs/doc_research__10.pdf
2. Zantner C, Beheim-Schwarzbach N, Neuman K, & Kielbassa AM (2007) Surface microhardness of enamel after different home bleaching procedures *Dental Materials* **23(2)** 243-250.

3. Bitter NC, & Sanders JL (1993) The effect of four bleaching agents on the enamel surface: a scanning electron microscopic study *Quintessence International* **24(11)** 817-824.
4. de Freitas PM, Basting RT, Rodrigues Jr AL, & Serra MS (2002) Effects of two 10% peroxide carbamide bleaching agents on dentin microhardness at different time intervals *Quintessence International* **33(5)** 370-375.
5. Bitter NC (1998) A scanning electron microscope study of the long-term effect of bleaching agents on the enamel surface in vivo *General Dentistry* **46(1)** 84-88.
6. Hosoya N, Honda K, Iino F, & Arai T (2003) Changes in enamel surface roughness and adhesion of *Streptococcus mutans* to enamel after vital bleaching *Journal of Dentistry* **31(8)** 543-548.
7. Lopes GC, Bonissoni L, Baratieri LN, Vieira LCC, & Monteiro SJr. (2002) Effect of bleaching agents on the hardness and morphology of enamel *Journal of Esthetic and Restorative Dentistry* **14(1)** 24-30.
8. Titley K, Torneck C, & Ruse N (1988) Adhesion of composite resin to bleached and unbleached bovine enamel *Journal of Dental Research* **67(12)** 1523-1528.
9. Titley KC, Torneck CD, & Ruse ND (1992) The effect of carbamide-peroxide gel on the shear bond strength of a microfil resin to bovine enamel *Journal of Dental Research* **71(1)** 20-24.
10. Torneck CD, Titley KC, Smith DC, & Abdifar A (1990) Adhesion of light-cured resin to bleached and unbleached bovine dentin *Endodontics & Dental Traumatology* **6(3)** 97-103.
11. Al-Salehi SK, Wood DJ, & Hatton PV (2007) The effect of 24 h non-stop hydrogen peroxide concentration on bovine enamel and dentine mineral content and microhardness *Journal of Dentistry* **35(11)** 845-850.
12. Efeoglu N, Wood DJ, & Efeoglu C (2007) Thirty-five percent carbamide peroxide application cause in vitro demineralization of enamel *Dental Materials* **23(7)** 900-904.
13. Sulieman M, Addy M, Macdonald E, & Rees J (2004) A safety study in vitro for the effects of an in-office bleaching system on the integrity of enamel and dentine. *Journal of Dentistry* **32(7)** 581-590.
14. Ghavamnasiri M, Abedini S, & Mehdizadeh Tazangi A (2007) Effect of different time periods of vital bleaching on flexural strength of the bovine enamel and dentin complex *Journal of Contemporary Dental Practice* **8(3)** 21-28.
15. Seghi RR, & Denry I (1992) Effects of external bleaching on indentation and abrasion characteristics of human enamel in vitro *Journal of Dental Research* **71(6)** 1340-1344.
16. Tam LE, Lim M, & Khanna S (2005) Effect of direct peroxide bleach application to bovine dentin on flexural strength and modulus in vitro *Journal of Dentistry* **33(6)** 451-458.
17. Chng HK, Palamara JEA, & Messer HH (2002) Effect of hydrogen peroxide and sodium perborate on biomechanical properties of human dentin *Journal of Endodontics* **28(2)** 62-67.
18. Tam LE, & Noroozi A (2007) Effect of direct and indirect bleaching on dentin fracture toughness *Journal of Dental Research* **86(12)** 1193-1197.
19. Khouroushi M, Feiz A, & Khodamoradi R (2010) Fracture resistance of endodontically-treated teeth: effect of combination bleaching and an antioxidant *Operative Dentistry* **35(5)** 530-537.
20. Annual book of ASTM standards (2001) ASTM E 399–90 (Reapproved 1997). Metals-mechanical testing; elevated and low-temperature tests; metallurgy *West Conshohocken: American Society for Testing Materials* **03.01** 465-468.
21. El Mowafy OM, & Watts DC (1986) Fracture toughness of human dentin *Journal of Dental Research* **65(5)** 677-681.
22. Söderholm KJM, Mukerjhee R, & Longmate J (1996) Filler leachability of composites stored in distilled water or artificial saliva *Journal of Dental Research* **75(9)** 1692-1699.
23. Araujo FO, Baratieri LN, & Araujo E (2010) In situ study of in-office bleaching procedures using light sources on human enamel microhardness *Operative Dentistry* **35(2)** 139-146.
24. Justino L, Tames D, & Demarco E (2004) In situ and in vitro effects of bleaching with carbamide peroxide on human enamel *Operative Dentistry* **29(2)** 214-225.
25. Rodrigues JA, Marchi GM, Ambrosano GMB, Heymann HO, & Pimenta LA (2005) Microhardness evaluation of in situ vital bleaching on human dental enamel using a novel study design *Dental Materials* **21**(11) 1059-1067.
26. Araujo EM, Baratieri LN, Vieira LC, & Ritter AV (2003) In situ effect of 10% carbamide peroxide on microhardness of human enamel: function of time *Journal of Esthetic and Restorative Dentistry* **15(3)** 166-174.
27. Smidt A, Fueurstein O, & Topei M (2011) Mechanical, morphologic and chemical effects of carbamide peroxide bleaching agents on human enamel in situ *Quintessence International* **42(5)** 407-412.