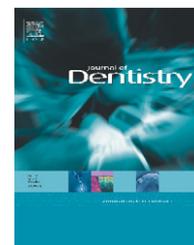


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Review

Review of the effects of peroxide on enamel and dentine properties

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ABSTRACT

Objectives: To review the available literature investigating the effects of peroxide-based products and solutions on enamel and dentine properties.

Sources: All original scientific full papers listed in ISI Web of Science and Medline were included in this review using the search terms peroxide AND (enamel OR dentin*) up to the end of 2006.

Conclusions: The majority of studies indicate that peroxide containing products and solutions have no significant deleterious effects on enamel and dentine surface morphology and chemistry, surface microhardness, subsurface enamel and dentine microhardness or ultra-structure. In addition, *in vitro* studies indicate that they have no significant clinically relevant effects on subsequent enamel and dentine loss caused by acidic erosive challenges, toothpaste abrasion or caries lesion formation. The contrasting studies that do show an effect on some of the above properties, in general, have some limitations in the *in vitro* methods used which do not accurately reflect the *in vivo* situation or use products/solutions that have a particularly low pH where acidic erosive processes are likely to dominate and explain the observed changes in enamel and dentine.

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1. Introduction

There are a number of methods and approaches that have been described in the literature for the bleaching of vital teeth. For examples, methods utilising different bleach agents, concentrations, times of application, product format, application mode and light activation.¹ Contemporary bleaching agents are typically either hydrogen peroxide (HP) or carbamide peroxide (CP). HP is capable of oxidizing a wide range of coloured organic and inorganic compounds, causing decolourisation and hence bleaching of the substrate. CP is a chemical adduct of urea and HP, which upon dissolving in water or saliva disassociates back into HP and urea. Thus, CP

can be considered as a precursor of the active bleach species HP.

Three fundamental vital tooth bleaching approaches exist, namely, dentist-supervised nightguard bleaching, in-office or power bleaching and over the counter (OTC) bleaching products. Nightguard bleaching typically uses a relatively low level of whitening agent applied to the teeth via a custom fabricated mouth guard and is worn at night for at least 2 weeks. In-office bleaching generally uses relatively high levels of whitening agents, for examples 25–35% HP or 35% CP containing products, for shorter time periods. The whitening gel is applied to the teeth after protection of the soft tissues and the peroxide may be further activated by heat or light. OTC

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products typically contain low levels of whitening agent (e.g. 3–6% hydrogen peroxide) that are self-applied to the teeth via gum shields, strips or paint-on product formats and typically require twice daily application for up to 2 weeks.¹

The purpose of the current review is to summarise the available literature concerning the effects of peroxide containing bleaching agents on enamel and dentine. All English language original scientific full papers listed in ISI Web of Science and Medline were included in this review using the search terms peroxide AND (enamel OR dentin*) up to the end of 2006. A total of 88 papers were included and 278 papers excluded in this review. Papers that were relevant to the dental material science field were excluded since this topic has recently been extensively reviewed.² In addition, the following were not included in the current review: abstracts; peroxide delivered via toothpaste formats; papers where the level of peroxide was not stated; formulations directed towards intra-coronal bleaching methods, and papers not relevant to the current review.

The influence of hydrogen peroxide (HP) and carbamide peroxide (CP) treatments on enamel and dentine properties have been extensively investigated in the literature, primarily using *in vitro* studies and the levels of HP and CP used ranged from 5.3 to 38% and 10 to 37%, respectively. The current review has collated these studies into investigations on the surface morphology and chemistry, surface microhardness, subsurface properties such as microhardness and ultrastructure, and the subsequent effects of acid challenges and abrasion on enamel and dentine, and these are summarised in Table 1.

2. Enamel and dentine surface morphology and chemistry

Scanning electron microscopy (SEM) is a rapid and convenient method for qualitatively analysing the surface morphology of enamel and dentine specimens following bleaching. Another relevant technique is profilometry which typically uses either a stylus probe run across the surface of a specimen or optical scanning methods to determine the surface profile of the

specimen. By measuring pre- and post-treatment profiles, it is possible to quantitatively determine changes in surface roughness and loss of surface material from the specimen.

For the studies that used these techniques in the current review, the majority showed no significant changes in enamel surface morphology following bleaching, even with one of the highest concentrations of 35% HP^{3,4} or 35% CP.⁵ Similarly, the lower levels of 6.5% HP^{6,7} and 6.0% HP,^{7–9} 25% CP,¹⁰ 15% CP,¹¹ and 10% CP^{6,9,11–19} were also shown to have no significant effects on enamel surface morphology following simulated product usages. In addition, enamel treated with 10% CP for 250 h simulated treatment showed no significant differences in surface morphology from control surfaces.^{20,21}

Using a replica technique on enamel surfaces treated *in vivo*, Leonard et al.²² confirmed previous laboratory observations in that nightguard vital bleaching did not alter the surface texture of enamel after 14 treatment days with 10% CP or after a 6 months follow up examination. Using similar methods, Turkun et al.²³ showed a slight increase in enamel porosity immediately following 14 treatment days with 10% CP, but was reversed within 3 months following treatment.

This is contrasted with *in vitro* studies^{13,24–35} that observed some changes in enamel morphology following treatment with HP or CP, which Yeh et al.³¹ described as mild surface pitting at localized areas, Ben-Amar et al.²⁶ as slight, Hegedus et al.³⁴ as mild and Akal et al.²⁷ considered as negligible when compared to dental prophylaxis procedures. In addition, Spalding et al.²⁸ observed minor changes after 35% HP (20 min) followed by 10% CP (12 h/day for 1 week) treatments, but considered these changes to be within the normal variations existing in natural teeth and concluded that the bleaching procedures were safe for enamel.

The differences between the positive and null effects on enamel are most likely due to either differences in the *in vitro* protocols used and this is reflected in their differences with respect to replicating the *in vivo* environment and/or the products used had a particularly low pH. For example, Ben-Amar et al.²⁶ and Yeh et al.³¹ stored their samples between bleaching sessions in distilled water, or other studies^{33–35} that air-dried the specimens between bleaching sessions, thus

Table 1 – Summary of references relating to the effects of peroxide on enamel and dentine

Measurement	No significant changes observed		Changes observed	
	Whitening measured	Whitening not measured	Whitening measured	Whitening not measured
Enamel surface morphology	6, 7, 8, 14, 20, 21, 23	3, 4, 5, 9, 10, 11, 12, 13, 15, 16, 17, 18, 19, 22, 28	33	13, 24, 25, 26, 27, 29, 30, 31, 32, 35, 36, 37
Dentine surface morphology	7, 8	4, 11, 46		13, 40
Enamel chemistry	39	10, 38, 40, 41, 42, 43, 44, 45		
Dentine chemistry	7			35
Enamel surface microhardness	6, 7, 8, 50, 59, 63	4, 10, 16, 17, 24, 27, 41, 49, 51, 52, 53, 54, 55, 56, 57, 58, 60, 61, 62		29, 64, 66, 67, 68, 69, 70, 71
Dentine surface microhardness	7, 8, 77	4, 57, 60, 61, 70, 73, 74, 75, 76, 77		61, 67, 78, 79
Enamel subsurface properties	6, 7, 50, 81, 82, 85	49, 80, 83		52, 84, 86, 87, 88, 89, 90
Dentine subsurface properties	7, 50, 81, 82, 85	83, 84		
Acid and abrasion effects on enamel	91	4, 54, 92, 93, 94		
Acid and abrasion effects on dentine	91	4		

negating any opportunity for remineralisation from salivary factors. Other studies^{29,30,32} used artificial saliva consisting only of inorganic calcium and phosphate components, and were devoid of any organic components which could have the potential of forming a protective salivary pellicle. In the case of three studies^{7,8,17} which showed no effect of bleaching products on enamel surface morphology, human whole saliva was used as a key part of replicating the *in vivo* situation. Indeed, Justino et al.¹⁷ demonstrated that any adverse effects evident for *in vitro* bleached and stored in water enamel specimens were not seen for similarly treated specimens placed on an intra-oral device and worn in the mouth and thus exposed to the remineralisation and protective benefits of saliva.

The effect of low pH of some of the peroxide treatments is exemplified by Shannon et al.²⁴ who found that the most severe changes in enamel topography occurred with products of lower pH (4.3 and 4.9). A study by Zalkind et al.¹³ showed that a commercial solution of 30% HP gave morphological changes in enamel after 7 days continuous exposure. Although no pH of the 30% HP solution is quoted, the authors describe the solution as “highly acidic”. Similarly, McGuckin et al.³⁶ found enamel surface morphology changes following treatment with 30% HP with a pH of 3.0. In addition, studies by Bitter³⁷ and Bitter and Sanders²⁵ investigated some early bleaching products that used acidic pre-rinses containing citric acid or acetic acid prior to the application of a peroxide gel and it is likely that these pre-rinses caused the enamel surface changes as observed by SEM. Indeed, these authors discuss that “the type of acid and its pH most likely have a significant bearing on the result”. Thus, it is most likely that studies which use products or solutions with relatively low pH are probably describing primarily demineralisation effects that are caused by acidic erosion processes rather than effects by peroxide per se.

In terms of changes in human enamel surface chemical composition, no differences were found between enamel treated with 30% HP (120 h treatment), 10% CP (6 h/day for 14 days) or water controls as measured by Raman spectroscopy.^{38,39} Similar results were obtained for electron spectroscopy for chemical analysis (ESCA) techniques used on human enamel treated with either 10% CP (continuous for 7 days),⁴⁰ 10% CP, 7% HP or 12% HP (7 h/day for 14 days),⁴¹ 25% CP (42 × 30 min)¹⁰ or 35% HP for 1 h⁴² and infra-red analysis and X-ray diffraction studies on human enamel treated with 10% CP and 16% CP (8 h/day for 6 weeks).⁴³ Teeth exposed to 10% CP solution for 6 h lost small amounts of surface calcium as determined by atomic absorption spectroscopy. However, this was considered not clinically significant since it was comparable to the calcium loss due to a cola beverage during a 2.5 min exposure.⁴⁴ A similar result and conclusion was drawn by Lee et al.⁴⁵ who observed slight changes in bovine enamel calcium–phosphate ratios following exposure to 30% HP solution for 120 h.

The majority of studies that investigated the surface morphology of dentine using SEM found no significant changes following bleaching.^{4,7,8,11,46} However, Zalkind et al.¹³ found that dentine treated with 30% HP or 10% CP solutions (7 days continuous) showed morphological surface changes of dentine. From the same research group, it was also

found that the calcium:phosphate ratio of dentine was modified following 7 days of continuous treatment with 30% HP or 10% CP solutions compared to saline control.⁴⁰ However, the authors describe the solutions as “highly acidic” so again it is likely the observed effects are due to primarily erosive processes. The chemical composition of dentine has been assessed by micro-Raman spectroscopy, a non-destructive technique that allows the chemical groups in a material to be evaluated, e.g. phosphate, carbonate, amide, etc. Using this technique, it was found that bleached dentine specimens had principally the same Raman spectra as unbleached controls.⁷

Overall, the majority of studies indicate that HP and CP containing products have no significant deleterious effects on enamel and dentine surface morphology and chemistry, even if one of the highest concentrations of HP or CP are used. The few contrasting studies that do show an effect, in general, have some limitations in the *in vitro* methodologies used which do not reflect the *in vivo* situation accurately or use products/solutions that are highly acidic.

3. Enamel and dentine surface microhardness

Surface microhardness (SMH) measurement is a simple method for determining the mechanical properties of enamel and dentine surfaces and it is related to a loss or gain of mineral of the dental structure. It has been previously shown to be a suitable method for determining small changes in SMH of enamel and dentine following erosive challenges from acids and acidic beverages.^{47,48}

In the context of the current review, SMH was the most frequently used technique for evaluating the effects of peroxide and bleaching products on enamel and dentine and the majority of studies demonstrated that peroxide and bleaching products had no significant effects on SMH. For example, Sulieman et al.⁴ showed that a 35% HP treatment for 30 min on human enamel showed no significant reduction of SMH. Cycling experiments on enamel with 6–9.5% HP where treatments were 30 min, twice/day for 14 days simulated use,⁴⁹ 6% and 6.5% HP for 30 min twice/day for 28 days,⁷ 5.3% and 6.5% HP for 2 h twice/day for a total of 14 h or 70 h,^{6,50} 12% HP for 7 h/day for 14 days,⁴¹ 6% HP for 20 min, twice/day for 14 days⁸ all showed no reduction in SMH. A similar conclusion was obtained for other simulated use cycling experiments when 10% CP,^{6,16,17,24,41,50–58} 12% CP,²⁷ 15% CP,⁵⁷ 20% CP,^{6,50} 25% CP¹⁰ and 37% CP⁵⁹ products or solutions were evaluated.

Lewinstein et al.⁶⁰ observed a slight reduction in enamel SMH following 35% HP or 35% CP treatments on human enamel which was completely reversed when treated with a 0.05% fluoride solution. Similarly, Basting et al.⁶¹ also observed a slight reduction in SMH of human enamel following 8 h/day for 42 days of 10% CP treatments which recovered to above baseline following 7 days in a remineralisation solution. In an *in situ* type study, Rodrigues et al.⁶² noted a slight reduction in SMH following in office 37% CP treatment (30 min × 2 for 3 days) plus at home use of 10% CP (6 h for 21 days). However, this was not significantly different from an equivalent series of placebo treatments and the authors considered the observed SMH reductions as clinically insignificant. In another *in situ* study, human enamel specimens were treated with a 10% CP

gel for either 1 or 7 h/day for a total of 21 days and in both cases neither treatment regime caused a significant affect on enamel SMH.⁶³

A reduction in enamel SMH was observed by Lewinstein et al.⁶⁴ following a 15 min treatment with 30% HP. However, in reviewing this study Kellehar and Roe⁶⁵ considered its conclusions flawed because it used HP with a pH of 3.0, which would in effect erode the enamel surface and produce the observed surface softening. Similarly, Smidt et al.⁶⁶ observed a slight reduction in enamel SMH following treatment with three different 10% CP (6 h/day for 16 days) products, with pH values in the range 4.3–5.5. In addition, no mention is made in the methods used as to what the specimens were stored in between and after bleach treatments so it is unclear if the specimens had opportunity to undergo any remineralisation processes.

A reduction in enamel SMH was also observed by Hairul Nizam et al.⁶⁷ following 24 h treatment with 30% HP solution. However, the pH of this solution was not stated and is likely to be of an acidic nature since commercial HP solutions are highly acidic to maintain long-term stability. In addition, the method used was nanoindentation which will only sample microhardness changes in the top 200 nm, so there is unlikely to be any clinical significance for these changes.

Other studies have observed a reduction in enamel SMH following bleaching with up to 35% HP or 35% CP.^{29,68–71} This conflicting data may again be due to differences of the *in vitro* methods used. For examples, these studies exposed specimens between bleaching treatments to either water or artificial saliva containing no organic components which could have formed a protective pellicle layer, there were no additional fluoride treatments to aid remineralisation and the study by Attin et al.⁷⁰ used bovine enamel which is known to have a threefold faster rate of lesion progression compared to human enamel.⁷²

For dentine, no significant changes in SMH were reported in studies involving 35% HP for 30 min,⁴ up to 35% HP (10 min) or 35% CP (2 × 1 h),⁷⁰ 10% CP and 15% CP treatments for up to 28 h,⁵⁷ or in cycling experiments using, 6% HP or 6.5% HP treatments for 30 min twice/day for 28 days⁷ or 6% HP for 20 min twice/day for 14 days.⁸ Further, in an *in situ* model there were found no reductions in SMH for sound and demineralised dentine following 10% CP treatments for 8 h/day for 3 weeks.⁷³

A transitory decrease in dentine SMH has been observed in some studies but recovered following a remineralisation period^{74–76} or 0.05% fluoride solution treatment.⁶⁰ Arcari et al.⁷⁷ reported small reductions in dentine SMH (5.4%), however, they concluded that this value probably has no clinical significance. A significant reduction in dentine SMH was observed for one 10% CP product, however, in the same study another 10% CP product only gave minor changes in SMH which recovered after a remineralisation period.⁶¹

Pecora et al.⁷⁸ observed a significant reduction in human dentine SMH following a 72 h treatment with 30% HP. Similarly, a nanoindentation study by Chng et al.⁷⁹ observed decreases in dentine hardness and Young's modulus following 24 h exposure to 30% HP. However, the pH of the 30% HP solutions in these two studies is stated as 2.0 and 2.05, respectively, and this particularly low pH is the likely primary cause of the observed changes. The nanoindentation study by

Hairul Nizam et al.⁶⁷ also showed a reduction in dentine SMH, but the limitations of this study have already been discussed.

Overall, the majority of studies indicate that HP and CP containing products have no significant deleterious effects on enamel and dentine SMH. The few contrasting studies that do show an effect, in general, again have some limitations in the *in vitro* methodologies used which do not reflect the *in vivo* situation accurately or used products/solutions which were highly acidic. Further, some studies that demonstrated a transitory reduction in SMH showed recovery to baseline values following a remineralisation period.

4. Subsurface enamel and dentine

Since peroxide will diffuse through enamel towards the enamel–dentine junction, some studies have investigated the effects of bleach agents on subsurface enamel and dentine. This effect can be investigated by bleaching whole teeth or fragments and then cutting and polishing the specimens to reveal the internal subsurface enamel and dentine areas, followed by microhardness measurements.

Using the above approach, Potocnik et al.⁸⁰ found no reduction in enamel subsurface microhardness following a 336 h treatment with 10% CP and Teixeira et al.⁴⁹ found no reductions in enamel subsurface microhardness following treatments with 6–9.5% HP (30 min × 2/day) or 10% CP (6 h/day) for 14 days in total. Similar results were found for both subsurface enamel and dentine when evaluating 6% HP following 20 min × 2/day for 14 days⁸¹; 5.3% HP following 14 and 70 h total bleaching time,⁸² and 10% CP following 1 h × 2/day for 14 days.⁸³ In contrast, the study by Attin et al.⁸⁴ showed some reduction in subsurface enamel but not subsurface dentine following bleaching protocols with up to 35% HP or 35% CP. Again this contrast may be due to differences in the methodology as previously described and discussed by the authors themselves. Indeed, Attin et al.⁸⁴ point out that the hardness reduction was “confined to superficial layers” and advise on the use of fluoride applications to minimize the observed effects.

An alternative approach to investigating the effects of bleaching on subsurface enamel, dentine and the enamel–dentine junction is to use confocal laser scanning microscopy which enables the ultrastructure of subsurface enamel and dentine to be investigated in a non-destructive way. Studies on bleached tooth specimens have demonstrated no changes in enamel and dentine ultrastructure following simulated bleaching protocols.^{7,50,82,85} In contrast, Efeoglu et al.⁸⁶ found significant demineralisation in the uppermost 50 μm of human enamel specimens following 10% CP treatments of 8 h/day for 15 days using microcomputerised tomography, another non-destructive method. However, the authors state that “positive controls to examine the clinical significance of the results of our study are needed” and so the clinical relevance of this result needs verification. Further, Bizhang et al.⁸⁷ evaluated the mineral loss of bovine enamel with microradiography following treatments of 10% CP (8 h/day, 2 weeks), 5.3% HP (1 h/day, 2 weeks) or a cola beverage (1 h/day, 2 weeks) and found median lesion depths of only 4.85, 1.65 and 13.70 μm, respectively, and an overall significantly greater

mineral loss for the cola beverage. In addition, in the latter study it was shown that the post-treatment of specimens with fluoride helped to further prevent mineral loss of bleached bovine enamel surfaces.

The tooth fracture susceptibility *in vitro* has been investigated by Seghi and Denry⁵² and it was demonstrated that 10% CP bleaching for 12 h gave a 30% decrease in tooth fracture toughness. This is in contrast to a study where enamel specimens were bleached with 6.5% HP or 20% CP (2 h/treatment) for a total of 70 h where no significant reduction in fracture toughness was observed compared to control treatments.⁶ These differences may be explained by the differences in the methods used, particularly with respect to the extra precautions taken in the latter study to rehydrate specimens prior to the analyses, where specimens were exposed to human whole saliva between bleaching treatments and after the final treatment for at least 40 h as opposed to phosphate buffer saline for an undisclosed period of time in the former study, thus better simulating *in vivo* conditions.

The ultimate tensile strength of subsurface enamel following treatment with up to 35% HP and 37% CP^{88,89} and 10–20% CP⁹⁰ has been shown to be reduced compared to non-bleached controls. The limitations of this method have been discussed by Cavalli et al.⁹⁰ and include that the “microtensile testing method creates four ground enamel surfaces that are all within 0.35 mm of the center of the specimen. This can be used to accelerate or exaggerate the potential damage to dental hard tissues by dental products or procedures”. Further, bleaching agents are applied to the outer surfaces of intact, hypermineralised enamel and not to a ground and highly polished subsurface enamel. In addition, the tensile forces used in these studies are not readily extrapolated to the clinical situation. Thus, although a possible mechanical reduction in enamel was observed the clinical relevance of the test method has to be questioned. Indeed, no clinical reports about fractures or cracks in tooth structures following bleaching have been presented in the dental literature.⁸⁸

Overall, the majority of studies indicate that HP and CP containing products have no significant deleterious effects on subsurface enamel and dentine microhardness or ultrastructure.

5. Effects of acid challenges and abrasion on bleached enamel/dentine

Suliman et al.⁴ found that pre-bleaching human enamel and dentine with 35% HP for 30 min had no subsequent deleterious effect on enamel and dentine loss caused by citric acid erosive challenges or brushing with toothpaste, as measured by profilometry. Similarly, Burgmaier et al.⁵⁴ evaluated the effect of an erosive challenge (1% citric acid, 20 min) on bleached (10% CP, 4 × 8 h) and unbleached enamel specimens and found no significant difference in SMH between bleached and unbleached. Further, bleaching human enamel and dentine with 10–22% CP for 2 h × 20 treatments did not increase their susceptibility to acid erosion or caries lesion formation as measured by quantitative light-induced fluorescence and transverse microradiography.⁹¹ This latter result of no increase in caries susceptibility of enamel after bleaching

was confirmed by Al-Qunaian⁹² in an *in vitro* microbial caries model following treatments with either 10% CP, 20% CP plus fluoride or 35% HP and by Kraigher et al.⁹³ in rat molars *in vivo* following treatment with 10% CP (1 h/day for 14 days).

The study by Wiegand et al.⁹⁴ showed that bleaching with 35% HP or 38% HP (15 min × 2/day for 4 days) or 35% CP (1 h/day for 4 days) gave no significant increase in enamel wear caused by brushing with a toothpaste. In the same study, they did show a significant increase in enamel wear following treatment protocols with 5.3% HP, 10% CP and 15% CP. However, this increase in abrasion was of the order of 0.1 μm, a level of abrasion the authors conclude as “clinically less relevant” than other wear processes that could occur in the mouth.

Overall, literature studies indicate that HP and CP containing products have no significant clinically relevant effects on subsequent enamel and dentine loss caused by acidic erosive challenges, toothpaste abrasion or caries lesion formation.

6. Conclusions

In conclusion, the vast majority of studies indicate that HP and CP containing products have no significant deleterious effects on enamel and dentine surface morphology and chemistry, SMH, subsurface enamel and dentine microhardness or ultrastructure, even if one of the highest concentrations of HP or CP are used. In addition, *in vitro* studies indicate that HP and CP containing products have no significant clinically relevant effects on subsequent enamel and dentine loss caused by acidic erosive challenges, toothpaste abrasion or caries lesion formation. The few contrasting studies that do show an effect on some of the above properties, in general, have some limitations in the *in vitro* methodologies used which do not accurately reflect the *in vivo* situation or used HP/CP products or solutions of a particularly low pH where erosive processes are likely to dominate and explain the observed changes in enamel and dentine specimens. It is also interesting to note that the confirmation of tooth whitening actually occurring has largely not been assessed in many of the hard tissue safety studies described in the literature (Table 1).

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