The effect of carbamide peroxide treatment on metal ion release from dental amalgam

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**Abstract**

Objectives. There is concern that hydrogen peroxide generated by tooth bleaching agents may cause enhanced metal ion release (including mercury) from dental amalgam following contact. The aim of this in vitro study was therefore to investigate the effect of a carbamide peroxide (CP) based tooth bleaching gel on metal ion release from dental amalgam.

Methods. Dental amalgam discs were prepared according to the manufacturers’ instructions. These were treated with either a 10% carbamide peroxide (CP) gel or a 0% CP gel for 24 h. Discs were carefully wiped with cotton wool before immersion in distilled water (20 ml) for 24 h at 37 °C. Following immersion, water samples were taken for metal ion release determination (Ag, Cu, Hg and Sn) using inductively coupled plasma mass spectrometry methods. The specimens were further evaluated for surface changes using scanning electron microscopy (SEM) and Talysurf surface roughness measurements.

Results. The differences in concentration of metal ions released after treatment with the 10% CP gel and a placebo gel treatment were not statistically significant (\(p>0.05\)). For example, mercury release following treatment with the 10% CP gel and the 0% CP gel was found to be 1.17(0.5) and 0.57(0.1) \(\mu g\) cm\(^{-2}\), respectively. Roughness measurements for samples treated with the 10% CP gel and 0% CP gel were 2.23(0.47) and 1.74(0.16) \(\mu m\), respectively, again showing no significant difference between groups (\(p>0.05\)). SEM images of the amalgam surfaces showed no apparent differences between treatments.

Significance. Treatment with a 10% CP gel did not significantly enhance subsequent metal ion release from dental amalgams compared to a control gel, contradicting previously published studies.

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1. Introduction

In recent years there has been an increased demand for tooth bleaching in order to improve the whiteness and perceived aesthetic appearance of tooth tissue [1–4]. This process commonly uses hydrogen peroxide, either directly or via its generation in a carbamide peroxide (CP) gel. The effects of peroxide on enamel and dentin have been extensively studied and there are numerous studies that report peroxide-containing products do not adversely affect enamel and dentin [5–10]. In contrast, there have been studies that provided evidence that high concentrations of peroxide could alter the chemical and
morphological structures of tooth tissues [11–14]. While work is ongoing to determine the optimum, safe and effective concentration of peroxide for tooth whitening procedures, it is important to also consider other potential interactions that may occur in the oral cavity.

One potential interaction is that between peroxide and dental materials. Various reports have described the effects of bleaching agents on dental materials including glass-ionomer cements, ceramics and gold [8,15–18]. These reports generally concluded that there was little evidence for the bleaching systems causing significant changes to the materials. In the case of dental amalgam, however, several in vitro studies have reported a significant increase in mercury release as a result of treatment with peroxides compared to control treatments [19–21]. Robertello et al. [19] compared the effects of three peroxide-containing commercial tooth whitening products and saline control on a zinc-free, palladium-enriched high copper amalgam. After 80 h of bleaching, 0.98 mg m\(^{−1}\) of mercury was detected for one of the products. Hummert et al. [20] studied the effects of two tooth whitening products and saline on mercury release from four different amalgams. After 8 h treatment, the level of mercury released was between 109 and 158 ng ml\(^{−1}\) for the tooth whitening products and 5 ng ml\(^{−1}\) for saline. Rotstein et al. [21] studied the effects of 10% carabamide peroxide and phosphate buffer (both at pH 6.5) on four different amalgams. After 48 h, the level of mercury released was reported to be in the range 23–161 μg ml\(^{−1}\).

Consideration of published studies shows that a wide variety of methods have been used to model the effects of bleaching agents on tooth tissue and dental materials. This lack of standardisation is reflected by the data generated, and this in turn is the most likely explanation for the very different conclusions reached by authors. In addition to this broad criticism, it was also noted that relatively little attention has been directed at understanding the mechanisms responsible for metal ion release from dental amalgam following exposure to peroxides. The aim of this study was therefore to investigate metal ion release from amalgam discs which were prepared and finished to simulate clinical preparation.

2. Materials and methods

2.1. Materials

The amalgam selected for this study was Sybraloy® (Kerr UK Ltd., Peterborough, Lot 71062). This is a typical restorative material based on a high copper, unicompositional spheroidal alloy. The composition (% w/w) of this alloy is reported as 41.8 Ag, 29.3 Sn, 28.2 Cu and 0.03 Zn. It is mixed at an alloy to mercury ratio of 1–0.92 (w/w). The Kerr data sheet gives the final mercury concentration as 45%, while the US material safety data sheet gives 44.5% (w/w). Discs (10 mm diameter × 2 mm thickness) were prepared according to the manufacturer’s instructions and aged for 7 days at 37°C in air. They were then polished using standard dental equipment (silicone polishers) and left overnight in air at 37°C. The disc specimens were treated with either a 10% carabamide peroxide (CP) gel, the 0% CP control gel (2.0 g), a control gel based on lutrol polymer containing no peroxide (2.0 g) or a carbonated beverage (Sprite Light, The Coca Cola Co., Uxbridge, UK, containing citric and carbonic acids, pH 2.84): (20 ml) for 24 h (n = 5). After treatment the specimens were wiped clean with cotton wool and placed in distilled water (20 ml) for 24 h at 37°C.

2.2. Scanning electron microscope

The amalgam samples were studied using SEM to evaluate any changes in surface morphology or features. SEM photomicrographs were taken of the surface of amalgam discs following the experimental treatments.

2.3. Ion release

Following immersion of the discs in the water, samples (2× 10 ml) were taken for analysis of ion release by inductively coupled plasma-mass spectrometry (ICP-MS – Agilent 4500). All ion release samples were acidified with 200 μl of nitric acid (for Ag determination) or hydrochloric acid (for all other ions).

2.4. Surface roughness

Following ion release experiments, one disc from each series was also taken for SEM, along with an untreated control in order to determine any changes in surface morphology or features. The surface roughness of the discs was determined using a Talysurf (Mitutoyo Corporation, Kawasaki, Japan), which was calibrated by setting the appropriate zero reading prior to roughness measurement of the discs. The roughness of the uppermost surface was then measured by moving the stylus across its diameter. This procedure was repeated eight times for each disc and the results averaged.

2.5. Statistical analysis

The data were analysed using one-way ANOVA followed by Bonferroni post hoc analyses.

3. Results

3.1. Metal ion release

The ion release data for mercury, silver, tin and copper are shown in Figs. 1–4, respectively. Treatment with Sprite Light did not result in significant release of mercury or silver, but gave the highest release of copper in this study. There were no significant differences in metal ion release between 10% CP and 0% CP gel (p > 0.05).

3.2. Scanning electron microscopy

The surface of a typical amalgam disc under SEM, before and after treatment with the 10% carbamide peroxide gel, is shown in Fig. 5(a+b respectively). All of the micrographs showed the typical structure of the set amalgam where the spheroidal nature of the alloy was obvious, with no major differences in the surface before and after treatment.
Fig. 1 – Mercury ion release (µg cm⁻²) from amalgam discs following treatment with experimental and control materials. Results are the arithmetic mean of one analysis of each of five samples (error bars show standard error of mean).

3.3. Surface roughness

The average roughness for each specimen after treatment is shown in Table 1. The difference in roughness values for the discs treated with the 10% CP gel and the 0% CP gel was not of statistical significance (p > 0.05).

3.4. Statistical analysis

The data were analysed using one-way ANOVA followed by Bonferroni post hoc analyses.

4. Discussion

Metal ions, including mercury, were detected in water samples following all of the treatments including controls (Figs. 1–4).

The greatest mercury release followed treatment with the 10% CP gel. However, this was not significantly greater than the 0% CP gel treated group. This observation suggested that the physical removal of gel from the surface of the amalgam disc itself contributed to subsequent metal ion release. Treatment with Sprite-Light® (pH 2.84) resulted in relatively low levels of mercury release compared to 10% CP gel, although copper ion release was elevated. Sprite-Light® was selected as an additional control solution because it has been commonly employed in dental studies [22] and has been compared to tooth bleaching agents [10,23]. Tin ion release was consistently higher than silver, and of a similar order to that detected for mercury. However, it was noted that tin release was higher than mercury following treatment with Sprite Light. The ion release profiles for acidic Sprite-Light® and the gel treatments were consistent with the operation of different mechanisms. The carbonated beverage acted on the amalgam by acidic

<table>
<thead>
<tr>
<th>Sample</th>
<th>Lutrol Gel</th>
<th>Sprite</th>
<th>10% CP</th>
<th>0% CP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Roughness (µm)</td>
<td>1.23 (0.12)</td>
<td>1.91 (1.23)</td>
<td>2.23 (0.47)</td>
<td>1.74 (0.16)</td>
</tr>
</tbody>
</table>

Surface roughness (µm) determination of discs exposed to different treatments.
attack, while the active gel oxidised the surface of the material. This did not however account for the increased ion release following placebo gel treatment.

SEM did not show any differences between samples. All of the micrographs showed the structure of the set amalgam, and the spherical nature of the alloy was apparent. Differences in ion release mechanisms were not detected using SEM, even though ion release profiles suggested differences between acidic and oxidative systems. These observations were supported by the study of surface roughness of treated materials. Statistical analysis of surface roughness results showed no significant difference ($p > 0.05$) between the surface of amalgam discs treated with 0% and 10% CP gel. Similar results were reported by Potocnik et al. [23] on the effects of 10% CP on human enamel as substrate.

It is difficult to make direct comparisons between the data from this study and published figures, due to limitations of either method or presentation of data in these papers. Ion release should be expressed in a standard form, ideally as an amount released per unit surface area of specimen. Surprisingly few authors have elected to use this approach to date. Rotstein et al. [21] reported a concentration of between 23 and 161$\mu$g$\text{mL}^{-1}$ of mercury released from their samples after 48 h of bleaching with 10% CP. Their amalgam samples had dimensions of 10 mm $\times$ 5 mm $\times$ 3 mm giving a total surface area of 1.9 cm$^2$ for each sample. Their data may be recalculated to give between 60 and 424$\mu$g cm$^{-2}$ of mercury release directly into CP solutions. This data suggests very high metal ion release, almost certainly due to the use of unpolished specimens along with relatively aggressive test procedures. Hummert et al. [20] used cylindrical amalgam specimens (4 mm $\times$ 8 mm diameter). Their published mercury release values can be recalculated in terms of specimen surface area and are found to be in the range 1.42–2.04$\mu$g cm$^{-2}$ for the bleaching agents and 0.07$\mu$g cm$^{-2}$ for saline. These values are more similar to the data reported in this study. Recalculation of Robertello et al. [19] data was not possible due to the lack of experimental details given in their paper.

The release of mercury from restorations is time dependent and proportional to the surface area of the restoration [24]. Mackert and Berglund [25] have reported the rate of unstimulated mercury release from amalgam to be on average 0.4$\mu$g per amalgam surface per day, calculated from six different in vivo studies. Assuming an average amalgam surface in vivo is 5 mm $\times$ 5 mm, the Mackert and Berglund figure would give a mercury release value from one amalgam of 1.6$\mu$g cm$^{-2}$ per day. Comparing this value with the values of the current study shows that the bleaching gel system has a similar level of mercury release, thus indicating that bleaching does not unduly accelerate subsequent mercury release. Assuming a surface area of 1 cm$^2$ is equivalent to four mercury amalgam...
surfaces in vivo, the mercury release value from the current study of approximately 1.0 ng cm$^{-2}$ in 24 h would mean that four bleached amalgam surfaces would subsequently release only 1.0 ng of mercury into the oral cavity. This is well within the World Health Organisation’s maximum acceptable daily intake (ADI) for mercury of 40 ng [26]. Indeed, to exceed the ADI for mercury, a patient would require mercury to be released from approximately 160 amalgam restorations.

In considering the quantity of product applied in these experiments, the amount can be considered to be in excess of in vivo applications since there was no elution of the gel from the amalgam surface. This is in contrast to the in vivo situation where it is known that peroxide levels within bleaching products are depleted during use [27]. Thus the experimental design in the current studies was in excess of what is anticipated under normal use, and can be used as further confirmation of the safety of these types of products. It is therefore highly unlikely that, assuming manufacturers’ instructions are followed, metal ion release from amalgam following contact with tooth bleaching gels containing up to 10% CP constitutes a health hazard.

In the current study, no preliminary data were available for power calculations. Based on the current data and using Altman’s [28] method of power calculations, a sample of 30 discs would be required to yield an 80% power to find a difference in the magnitude observed for mercury ion release in these data at an alpha of 0.05. In future studies, it may be possible to reduce the sample size by increasing the effect of the bleach (for example, by possibly using longer and more exaggerated exposure conditions) and/or by reducing possible variability of the sample preparation.

5. Conclusions

Metal ions, including mercury, silver, copper and tin, were released from amalgam following all treatments. Mercury release from the amalgam samples postbleaching was approximately 1 ng cm$^{-2}$ over a 24 h period. While metal ion release was found to be above that associated with control treatment (0% CP gel) this was not statistically significant and the levels of metal ion release were not considered sufficiently high to represent a health hazard. It was also concluded that the mechanism for ion release was different for acidic carbonated drinks and tooth bleaching gels based on a carbamide peroxide system.

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Fig. 5 – Scanning electron photomicrograph showing typical amalgam surface before (A) and after (B) treatment with a 10% carbamide peroxide gel.
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