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Research article

Marginal integrity of aesthetic restorations following intracoronal bleaching with sweet potato extract as an additive: An SEM study

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ABSTRACT

The aim of this *in vitro* study was to evaluate the marginal integrity of an alkasite restoration in comparison to that of a conventional composite resin restoration following intra-coronal bleaching with 30% hydrogen peroxide (HP) containing sweet potato extract (SPE) as an additive. Access cavities were prepared in 60 extracted human incisors. The teeth were decoronated 2mm below the CEJ and the pulp chambers were sealed cervically. The samples were divided into two groups (n = 30) based on the type of restorative material - group I: Hybrid composite resin and group II: Alkasite restorative material. Both the groups were divided into three subgroups (n = 10) based on the intra-coronal bleaching agent used namely, subgroup A: no bleaching (NB); subgroup B: 30% hydrogen peroxide (HP) and subgroup C: 30% hydrogen peroxide containing SPE (HSP). The tooth-restorative interface was observed under a scanning electron microscope (SEM) to determine the marginal integrity. The results were tabulated and statistically analyzed using one-way ANOVA. Specimens bleached with HP alone showed higher marginal gaps, irrespective of the restorative material used. Subgroups A and C showed lesser marginal gaps under both the restorative materials. An improved marginal integrity was seen with the alkasite material. It can be concluded that the addition of SPE to HP improves the marginal integrity of the coronal restorative material placed immediately post-bleaching. The new alkasite material holds promise as a permanent coronal seal in cases of intra-coronal bleaching.

1. Introduction

With increasing insistence on conserving the integrity of remaining tooth structure, it is the responsibility of the clinician to contain restorations only to the extent of lost tooth structure (Polesel, 2014). Intracoronal bleaching of discoloured intact pulpless anterior teeth with 3%–30% hydrogen peroxide (HP) is considered an ideal choice to refurbish esthetics as well as to preserve the structural integrity of the tooth. It indeed reduces the need for invasive procedures like veneers and crowns (Klaric et al., 2015).

During bleaching, hydrogen peroxide (HP) ionizes to release free radicals and nascent oxygen, which break down the larger chromogenic pigments to smaller less intensive ones thereby lightening the color of the tooth (Bernardon et al., 2010). However, the clinician faces multiple challenges when composite restorations are to be done immediately following bleaching (Teixeira et al., 2003; Jacob and Dhanya Kumar, 2007; de Oliveira et al., 2011). HP causes deleterious effects on the enamel by oxidizing the organic and inorganic components of the latter. Studies have shown that HP causes micro- to nano-morphological changes on the enamel surface, depending upon its concentration and exposure time, resulting in cracks and craters. In addition to this, the residual peroxide radicals interfere with the polymerization of composite resin, as well as reduce the penetrating ability of resin adhesives into enamel which is essential for micromechanical retention (Elfallah et al., 2015; Klaric et al., 2015). The marginal integrity of composite restorations is also compromised when they are used immediately as permanent restorations in access cavities of teeth subjected to intracoronal bleaching. Loss of marginal integrity opens up a microscopic gap between the tooth and the restoration, leading to a cascade of events that ultimately result in failure of the restoration (Teixeira et al., 2003; Jacob and Dhanya Kumar, 2007; de Oliveira et al., 2011).

The poor bonding characteristics of composite to bleached teeth can be countered by either postponing restorative work by two to three weeks or using antioxidants such as sodium ascorbate, proanthocyanidin or green tea extract for immediate bonding, as suggested by various authors. These antioxidants can scavenge the free radicals and reverse the bond strength of resin adhesives allowing immediate restoration (Vidhya et al., 2011; Sharafeddin et al., 2016). However, this requires an

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additional step, implying that these agents cannot prevent the initial deleterious effects on enamel and involves extended overall clinical time (Al-Hassani and Al-Shamma, 2018).

In the quest for aids to reduce the harmful effects of HP, studies have been focused on plant tubers like Ipomoea batatas L (sweet potato) which contains a diversity of antioxidant molecules like catalase, superoxide dismutase and anthocyanins with high radical scavenging activity (Zhao et al., 2013; Sun et al., 2018; Li et al., 2019). Gopinath et al. (2013) have shown that sweet potato extract (SPE) when added to HP resulted in enhanced bleaching effects of the latter, besides decreasing its harmful effects on enamel. Also, Nair et al. (2016) reported that SPE improved the reduced bond strength associated with immediate bonding of resin composites to bleached enamel. Whether the use of SPE-incorporated HP as a bleaching agent will avert the loss of marginal integrity associated with the immediate bonding of composite restorations in teeth subjected to intracoronal bleaching is not yet explored.

Recently, an improved marginal seal has been demonstrated with the introduction of a new alkasite restorative material over conventional packable composite (George and Bhandary, 2018). Whether this alkasite material will perform better than conventional composites when placed in such a challenging clinical situation is not yet known. A careful review of literature revealed that marginal integrity after application of SPE-incorporated HP followed by restoration with alkasite or composite restorative material has not yet been evaluated. Hence, the aim of this *in vitro* study was to evaluate and compare the marginal integrity of alkasite and conventional composite restorations following intracoronal bleaching using 30% HP with SPE as an additive.

2. Materials and methods

2.1. Preparation of sweet potato extract (SPE)

The methodology was adopted from Gopinath et al. (2013). 200g of purple flesh sweet potato were cut into smaller pieces and blended using a blender. The filtrate was taken and centrifuged at 2000 rpm for 20 min at 4 °C. The centrifuged clear liquid was refrigerated at 4 °C until use.

2.2. Preparation of experimental bleaching solution (HSP)

1 mL of SPE added to 28 mL of phosphate buffered saline (PBS, Thermo fisher scientific India Pvt. Ltd., Mumbai, India) and 1mL of 30% HP (Thermo fisher scientific India Pvt. Ltd., Mumbai, India) formed the experimental bleaching solution (HSP).

2.3. In vitro study

2.3.1. Preparation of specimens

60 extracted upper incisors were cleaned of tissues and debris and stored in 0.1% thymol (Thermo fisher scientific India Pvt. Ltd., Mumbai, India). Access opening was done and enlarged upto size 3 gates-glidden drill (MANI, Inc. Tochigi, Japan). The teeth were sectioned 2mm below the CEJ and the cervical end (2mm) was sealed with glass ionomer cement (Vivaglass CEM PL, Ivoclar Vivadent, Liechtenstein). The specimens were randomly divided into two groups ($n = 30$) depending upon the permanent coronal seal; namely group I: Hybrid composite resin (C) (TE Econom Plus, Ivoclar Vivadent, Liechtenstein) and group II: Alkasite restorative material (A) (Cention N, Ivoclar Vivadent, Liechstenstein).

2.3.2. Intracoronal bleaching procedure

Each of these groups were further divided into three subgroups based on the intracoronal bleaching agent used: subgroup A: no bleaching; subgroup B: bleaching with 30% HP - a pellet of cotton dipped in 30% HP was placed inside the pulp chamber and sealed temporarily with Cavit (3M ESPE, St. Paul, MN, USA) and; subgroup C: bleaching with HSP - the same protocol as in subgroup B was followed. All the specimens were stored in distilled water until the treatment period of 4 weeks, with the solution being changed every week. At the end of 4 weeks, the pulp chambers were cleaned and restored with the respective materials, manipulated according to manufacturers' instructions. The restorations were finished and polished using Sof-Lex finishing and polishing system (3M ESPE, St. Paul, MN, USA).

2.3.3. SEM analysis

The palatal surface of the specimens were then sputter coated with gold and viewed using a scanning electron microscope (SEM Phenom Pro, Einthovan, Netherlands) under 1000x magnification. The tooth-restorative interface was focussed and the marginal gap was measured using an inbuilt tool.

2.4. Statistical analysis

Statistical analysis was done using SPSS software version 22 by applying paramteric one-way ANOVA. Multiple comparison was done using Tukey's HSD post hoc test. The significance was set at $p < 0.05$.

3. Results

The representative SEM micrographs of all the groups are shown in Figure 1. The descriptive statistics of the mean marginal gap of all the groups is given in Table 1 and their multiple comparisons are given in Table 2. Samples bleached with HP (IB and IIB) showed the highest gap in both the groups ($p < 0.05$). The mean marginal gap of unbleached samples (IA and IIA) were significantly lesser than the HP bleached samples (IB and IIB) in both the groups ($p < 0.05$). Samples bleached with HSP (IC and IIC) showed significantly lesser gaps compared to HP bleached samples ($p < 0.05$). No significant difference was seen between the unbleached samples and HSP bleached samples ($p > 0.05$). No significant difference was seen in intergroup comparison ($p > 0.05$).

4. Discussion

Marginal integrity of a restorative material with enamel is an important criterion in determining the long term success of a restoration (Rakhshan, 2015). Researchers have pointed out that a marginal gap as low as 50 μm can result in adherence of *S. mutans,* resulting in secondary caries (Diercke et al., 2009). Marginal integrity can be studied by various methods including dye leakage studies, resin replica technique, as well as direct visualization of specimen using stereo- or scanning electron microscope (Rakhshan, 2015). SEM was chosen in this study as it allowed clear visualization and accurate measurement of marginal gaps. This resulted in a quantifiable data that could be statistically compared.

Bleaching with only 30% HP showed the widest marginal gap values with both the restorative materials. This can be attributed to the presence of free radicals from HP, which not only act on the chromogenic pigments, but also attack the enamel surface causing surface alterations. This results in loss of prismatic substance and an over-etched appearance (Abouassi et al., 2011; Xu et al., 2011; Gopinath et al., 2013; Alqahtani, 2014). Moreover, the retained peroxide layer on the subsurface of enamel interferes with the polymerization of composite resins and ultimately impedes its bonding to enamel (Vidhya et al., 2011; Elfallah et al., 2015; Klaric et al., 2015).

Samples bleached with HSP showed minimal or no marginal gap which was comparable to that of the unbleached controls. This can be ascribed to the presence of enzyme catalase and other antioxidants in SPE (Bovell-Benjamin, 2007; Kabel, 2014). Catalase catalyses the decomposition of hydrogen peroxide to water and oxygen with the formation of hydrogen peroxide - catalase complex. This complex provides an alternate pathway with lower activation energy (Kabel, 2014). Activation energy is the energy required to convert a stable molecule into a reactive molecule. Thus, the activation energy of HP is lowered from 76 kJ/mol to less than 8 kJ/mol (Atkins and de Paula, 2010). One molecule of catalase can decompose 40 million molecules of HP (Rhodes, 2011; Milgrom,

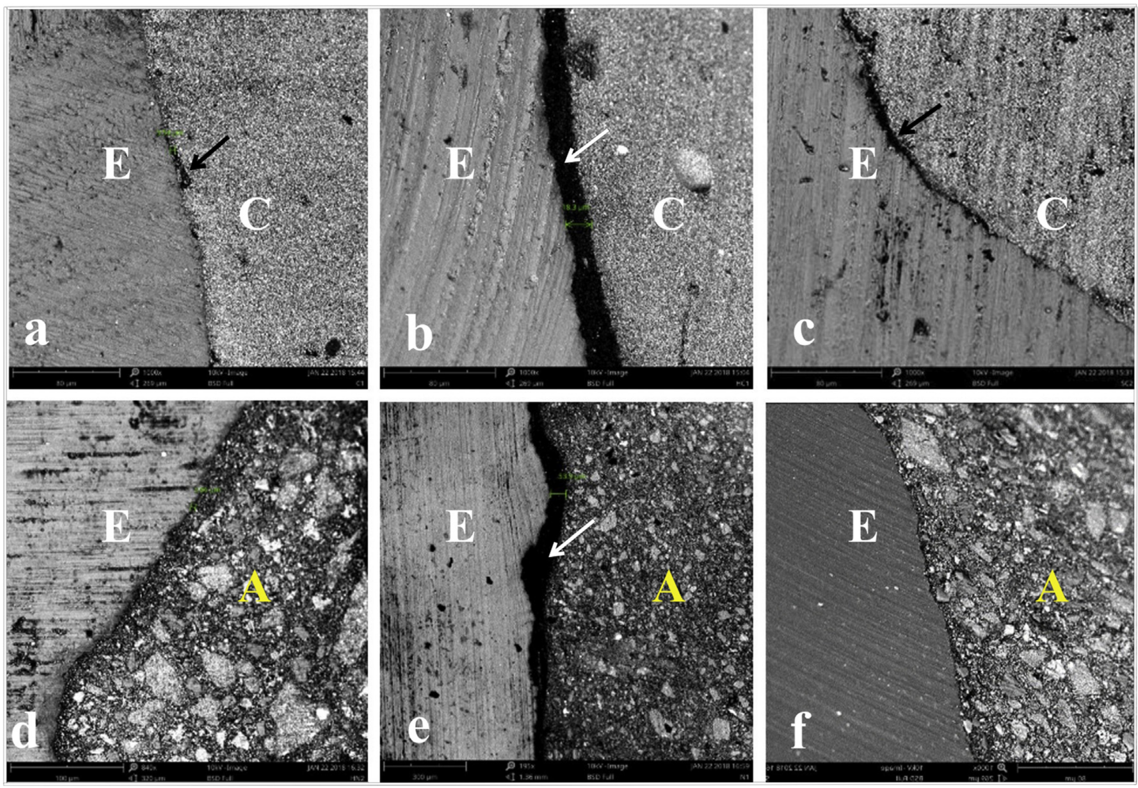

Figure 1. Representative SEM micrographs showing wider marginal gap (white arrows) at the tooth (E)-restorative (C-Composite, A-Alkasite) interface in samples bleached with 30% hydrogen peroxide (b,e) and lesser marginal gap (black arrows) in samples bleached with 30% hydrogen peroxide containing sweet potato extract (c,f) and unbleached controls (a,d).

2016). As a result, there is an outburst of free radicals within a short period of time, ultimately minimizing the contact time of HP with the enamel surface and its consequent effects (Gopinath et al., 2013).

Antioxidants such as flavonoids, superoxide dismutase, tocopherol, anthocyanin and beta carotene present in sweet potato possess high radical scavenging activity (Zhao et al., 2013; Sun et al., 2018; Li et al., 2019). Thus, the excess free radicals will be expunged by these antioxidants. The results of the present study correlates well with these findings as there were minimal surface changes with little or no marginal gap in the groups treated with HSP. Thus, bleaching with HSP did not affect the marginal integrity of both composite and alkasite restorations. The more purple the sweet potato flesh, more are the anthocyanins present in it (Xu et al., 2015). Hence a purple flesh one was chosen for this study.

Comparing the alkasite restorative material with conventional packable composite, there seems to be a superior marginal seal with no marginal gap in samples restored with Cention N. This can be related to the lower polymerization shrinkage, increased wettability and low water sorption exhibited by this new material (George and Bhandary, 2018; Liu et al., 2003; Ilie, 2018; Mazumdar et al., 2019). The presence of less hydrophilic monomer urethane dimethyl methacrylate (UDMA) and the absence of hydrophilic hydroxyethyl methacrylate (HEMA) are responsible for its low water sorption (Akin et al., 2015; Ilie, 2018). The polyethylene glycol 400 dimethacrylate (PEG 400 DMA) is a hydrophilic monomer which augments the wettability of the material and enables it to flow on the tooth substrate sustaining an intact seal (Liu et al., 2003; Ilie, 2018). It contains a patented isofiller partially functionalized by silanes, which act as shrinkage stress reliever during polymerization. This isofiller owing to its low modulus of elasticity (MOE-10 GPa) acts like a spring amongst the standardized glass fillers with a higher MOE (71 GPa), resulting in low volumetric shrinkage

Table 1. Descriptive statistics (mean and standard deviation, S.D) showing the mean marginal gap of all the groups.

Groups	Subgroups	Mean (in μm)	S.D	Standard error	95% Confidence interval for mean		Minimum	Maximum
					Lower bound	Upper bound		
Group I: Hybrid composite resin	**Subgroup A:** No bleaching (NB)	3.7650	2.89613	1.44806	-0.8434	8.3734	0.00	6.98
	Subgroup B: Bleaching with 30% hydrogen peroxide (HP)	26.7500	5.24754	2.62377	18.4000	35.1000	21.20	33.00
	Subgroup C: Bleaching with 30% hydrogen peroxide containing SPE* (HSP)	4.2725	2.90169	1.45085	-0.3447	8.8897	0.00	6.48
Group II: Alkasite restorative material	**Subgroup A:** No bleaching (NB)	1.6550	1.93228	0.96614	-1.4197	4.7297	0.00	3.66
	Subgroup B: Bleaching with 30% hydrogen peroxide (HP)	27.4600	23.4101	11.70505	-9.7907	64.7107	6.15	50.00
	Subgroup C: Bleaching with 30% hydrogen peroxide containing SPE (HSP)	0.1000	0.20000	0.10000	-.2182	0.4182	0.00	0.40
Total		10.6671	14.85691	3.03265	4.3936	16.9406	0.00	50.00

* SPE – Sweet potato extract.

Table 2. Multiple comparisons of the mean marginal gap of all the groups.

(I) Groups	(J) Groups	Mean difference (I-J)	Standard error	Significance	95% Confidence interval	
					Lower bound	Upper bound
Composite-NB	Composite-HP	-22.98500*	7.04835	0.042	-45.3849	-0.5851
	Composite-HSP	-0.50750	7.04835	1.000	-22.9074	21.8924
	Alkasite-NB	2.11000	7.04835	1.000	-20.2899	24.5099
	Alkasite-HP	-23.69500*	7.04835	0.035	-46.0949	-1.2951
	Alkasite-HSP	3.66500	7.04835	0.995	-18.7349	26.0649
Composite-HP	Composite-NB	22.98500*	7.04835	0.042	0.5851	45.3849
	Composite-HSP	22.47750*	7.04835	0.049	0.0776	44.8774
	Alkasite-NB	25.09500*	7.04835	0.023	2.6951	47.4949
	Alkasite-HP	-0.71000	7.04835	1.000	-23.1099	21.6899
	Alkasite-HSP	26.65000*	7.04835	0.015	4.2501	49.0499
Composite-HSP	Composite-NB	0.50750	7.04835	1.000	-21.8924	22.9074
	Composite-HP	-22.47750*	7.04835	0.049	-44.8774	-0.0776
	Alkasite-NB	2.61750	7.04835	0.999	-19.7824	25.0174
	Alkasite-HP	-23.18750*	7.04835	0.040	-45.5874	-0.7876
	Alkasite-HSP	4.17250	7.04835	0.990	-18.2274	26.5724
Alkasite-NB	Composite-NB	-2.11000	7.04835	1.000	-24.5099	20.2899
	Composite-HP	-25.09500*	7.04835	0.023	-47.4949	-2.6951
	Composite-HSP	-2.61750	7.04835	0.999	-25.0174	19.7824
	Alkasite-HP	-25.80500*	7.04835	0.019	-48.2049	-3.4051
	Alkasite-HSP	1.55500	7.04835	1.000	-20.8449	23.9549
Alkasite-HP	Composite-NB	23.69500*	7.04835	0.035	1.2951	46.0949
	Composite-HP	0.71000	7.04835	1.000	-21.6899	23.1099
	Composite-HSP	23.18750*	7.04835	0.040	0.7876	45.5874
	Alkasite-NB	25.80500*	7.04835	0.019	3.4051	48.2049
	Alkasite-HSP	27.36000*	7.04835	0.012	4.9601	49.7599
Alkasite-HSP	Composite-NB	-3.66500	7.04835	0.995	-26.0649	18.7349
	Composite-HP	-26.65000*	7.04835	0.015	-49.0499	-4.2501
	Composite-HSP	-4.17250	7.04835	0.990	-26.5724	18.2274
	Alkasite-NB	-1.55500	7.04835	1.000	-23.9549	20.8449
	Alkasite-HP	-27.36000*	7.04835	0.012	-49.7599	-4.9601

NB-No bleaching, HP-Bleaching with 30% hydrogen peroxide, HSP- Bleaching with 30% hydrogen peroxide containing sweet potato extract.
* The mean difference is significant at the 0.05 level ($p < 0.05$).

during polymerization (Chole et al., 2018; George and Bhandary, 2018).

The results of the study show that single step bleaching using HP with SPE as an additive has the potential to nullify the deleterious effects of HP on enamel and scavenge the free radicals resulting in a good marginal seal of the ensuing restoration. This is the first study to report the effect of HP/SPE combination on the marginal integrity of adhesive permanent restorations following intracoronal bleaching. The results of this study are clinically viable. The implication of the study is that the risk of microleakage is increased when resin restorations are done immediately after bleaching. Addition of SPE to the bleaching agent is capable of averting this risk, enabling immediate and durable bonding procedures. With adequate literature available on the advantages of the alkasite material, Cention N could be considered as a potential permanent restorative material in such challenging clinical situations.

In vitro studies offer insights into the performance of a material or a technique under controlled experimental situations, which are otherwise difficult to achieve clinically. The limitations of this study include exclusion of other confounding factors present in the oral cavity such as saliva, which could influence the results of the study. Hence, future studies should assess the role of such factors on the marginal integrity of resin restorations. The effective use of this combination in various other concentrations also needs to be studied. Long term clinical studies are also required to reinforce and validate the results of this *in vitro* study.

5. Conclusion

Within the limitations of this *in vitro* study, it can be concluded that intracoronal bleaching of intact pulpless teeth using 30% HP containing SPE as an additive followed by coronal seal with composite or alkasite material paves the way for an immediate bonding procedure without any deleterious alterations in enamel, allowing for an intact marginal seal.

Declarations

Author contribution statement

Gurucharan Ishwarya: Conceived and designed the experiments; Performed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Sampath Vidhya & Sekar Mahalaxmi: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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