Effect of Antioxidant on Coronal Seal of Dentin Following Sodium Hypochlorite and Hydrogen-Peroxide Irrigation

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ABSTRACT

Aim: Comparative analysis of the coronal microleakage of a self-etching priming dentin adhesive with the use of antioxidant (Sodium Ascorbate (SA) and herbal quercetin) after Sodium hypochloride (NaOCl) and hydrogen-peroxide (H_2O_2) irrigation.

Materials and Methods: Seventy single root canals were enlarged and seven different treatment protocols were applied throughout the root canal treatment. The groups 1, 2 and 3 were irrigated with NaOCl. Group 1 was used as the negative control with only NaOCl irrigation whereas in group2, Sodium Ascorbate was applied as an additional irrigation agent following NaOCl.In group 3,herbal antioxidant Quercetin(Q) was applied as an additional irrigation agent following NaOCl.Hydrogen-peroxide irrigation was used in groups 4, 5 and 6, the procedural steps were similar to those of groups 1, 2 and 3: H_2O_2 in group 4, Sodium Ascorbate application in group 5,and herbal antioxidant Quercetin group 6.Group 7 was positive control with saline irrigation alone. All root were obturated with A-H Plus root canal sealer and protaper F_4 gutta-percha cones immediately after irrigation. A self-etching priming adhesive plus ceram-x mono composite resin was applied after the endodontic treatment. The microleakage of dentin margins was determined using dye-penetration test under microscope.

Statistical Analysis:-In this study,Kruskal-Wallis test,Mann-Whitney U and Wilcoxon W tests were used.

Results: Kruskal-Wallis test with P=0.000 indicated that a significant difference exists between all the seven groups. NaOCl, H_2O_2 groups demonstrated higher leakage scores than the controls ($P<0.005$).Leakage pattern of groups 2, 3, 5 and 6 did not differ significantly from the control ($P>0.05$).

Conclusion: After control group 7, group 2 showed significantly less amount of dye penetration as compared with the other groups.

Keywords: Antioxidant, coronal seal, micro leakage

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INTRODUCTION

Endodontic treatment is essentially performed to prevent and control infection of pulpal & periapical tissues. The outcome of endodontic therapy depends on the method and quality of instrumentation, irrigation, disinfection, three dimensional obturation of root and good intracoronal restoration. Technical quality of the coronal restoration may be significantly more important than that of the endodontic treatment itself for apical periodontal health (1).

Sodium hypochlorite and hydrogen peroxide both antibacterial irrigating solution have been extensively used as separate irrigating solution or combination with each other,due to their debridement, deproteinization and antimicrobial properties (2).

Dentin bond strengths of resinous restorative materials to dentin are adversely influenced by use of Sodium hypochlorite and hydrogen peroxide (3-5)

Lai et al (6) suggested that the decreased bond strength observed in Sodium hypochlorite and hydrogen peroxide treated dentin could be reversed by use of antioxidant Sodium Ascorbate.

The other herbal antioxidant used in the study was Quercetin which belongs to a group of plant pigments called flavonoids that give many fruits, flowers, and vegetables their color. Flavonoids, such as Quercetin, are antioxidants -- they scavenge damaging particles in the body known as free radicals.

The most commonly used method for the evaluation of coronal microleakage is the linear measurement of tracer penetration e.g. dyes-different concentrations of aqueous solutions of methylene blue or India ink (7) dyes or bacteria (8) along the root filling.

The objective of this study was to find out microleakage of dentin margins by using antioxidant [Sodium Ascorbate & Quercetin] as final irrigant used after Sodium hypochlorite or hydrogen peroxide irrigation with a dye penetration test under stereomicroscope.

MATERIAL AND METHODS

Seventy freshly extracted, intact, non-carious human single-rooted teeth were selected for the study. The crowns were removed at CEJ with a carborundum disk in the presence of water. A #10 K-file was introduced into the root canal to establish the patency of the canal. The working length was determined by passing a #15 k-file into the canal until the tip was just visible at the apical foramen and then reducing 1mm from this length. This was kept as actual working length .Chemo-mechanical preparation was done with rotary Protaper [Dentsply Maillefer Switzerland] till F_4.

Distribution of Samples:-

Seventy teeth were assigned to 7 groups of 10 teeth each on the basis of irrigant used for canal preparation,

In group 1:- Root canals were irrigated with 5% Sodium hypochlorite.

In group 2:- Root canals were irrigated with 5% Sodium hypochlorite followed by 10% Sodium-Ascorbate .

In group 3:- Root canals were irrigated with 5% Sodium hypochlorite followed by Quercetin.

In group 4:- Root canals were irrigated with 3% Hydrogen peroxide.

In group 5:- Root canals were irrigated with 3% Hydrogen peroxide followed by 10% Sodium-Ascorbate.

In group 6:- Root canals were irrigated with 3% Hydrogen peroxide followed by Quercetin.

In group 7:- Root canals were irrigated with normal saline.

Group 1 [Sodium hypochlorite] and group 4 [hydrogen peroxide] were separated as negative controls, whereas, group 7 was the positive control [saline]. During the instrumentation, each canal of groups 1-3 was irrigated with 5% Sodium hypochlorite, whereas in groups 4-6, 3% hydrogen peroxide irrigation was applied to root canals. The same irrigation procedure was applied to all groups: 2 ml of irrigant was used after each change of endodontic instrument and 5 ml for final irrigation. The time of exposure to irrigants was estimated to 30 minutes per tooth. Following the final irrigation, all roots were flushed with 2 ml de-ionized water. Then, 10 ml of 10% Sodium Ascorbate was applied to the root canals as an added irrigation solution with flow-rate of 1 ml/minute in groups 2 and 5. Groups 3 and 6 were applied with 10 ml quercetin to root canal as an added irrigation solution with flow-rate of 1 ml/minute

The specimens irrigated with sterile saline without any combination were used as group 7 [positive control].

All roots were obturated with A-H Plus root canal sealer [Dentsply, India] and protaper F_4 gutta-percha cones [Dentsply Maillefer, India]. Excess gutta percha was sealed off with a hot instrument 3 mm below the decoronated root surface.

The root canals were sealed with a self-etching priming adhesive system [Xeno-V , Dentsply India]. A layer of bonding resin was applied with a brush, spread gently with air and cured for 10 seconds using an Light Cure Unit [LED, Guilin Woodpecker Medical Instrument Co. Ltd.] After dentin bonding application, stainless steel tofflemire matrices were custom-fitted to the prepared teeth. In order to ensure uniform thickness of the coronal restoration, a scribed line was placed on the inner surface of the matrix at a depth of 2 mm from the root surface. Resin composite [Ceram-x mono] was placed incrementally, and each increment was cured for 40 seconds. After removal of the tofflemire matrix, an additional curing time of 40 seconds was applied.

The specimens were then stored in a humidor at 37°C for 24 hours. The specimens were subjected to 500 thermal cycles between 5°C and 55°C water baths with a dwell time of 30 second. Thereafter, in all 7-groups three layers of nail polish was applied to the tooth surface, except for the coronal 2 mm, which remained exposed, in such a way that dye could penetrate the canal only via coronal region. Each group was separately immersed in 2% methylene blue and stored at 37°C for 48 hrs, after which the teeth were thoroughly washed in running water. The mesial and distal surface of all the teeth were grooved with the use of a tapered fissure carbide bur in a high speed hand piece without water spray nearly to the depth of the canal. The tooth was split longitudinally. Both halves of the split tooth were examined by stereomicroscope [OLYMPUS SZX, Japan].The observation was done after illumination with Nikon 100 W optical fiber and under maximum magnification of the microscope [4 X] and image was captured.

The scoring criteria is as follows: [Fig-1]

0-No evidence of dye penetration at the tooth /restoration interface [Fig-2]

1-Slight, just horizontal dye penetration [Fig-3]

2-Moderate, vertical Penetration less than half of radicular composite restoration [Fig-4]

3-Extensive, vertical Penetration greater than half but less than total of radicular composite restoration [Fig-5]

4-Gross, Vertical Penetration extending beyond the radicular composite restoration [Fig-6]

RESULT

In this study, the kruskal-wallis test as shown in table 1, with P=0.000, indicated that a significant difference exists between the all seven groups.

Table 1, in group 1 maximum number of teeth showed score 4 that is maximum penetration of dye. In group 7 maximum number of teeth showed score 0 that is minimum penetration of dye.

According to Mann-Whitney U test and Wilcoxon W test, as shown, the difference between micro leakage score of negative control groups [Groups 1 & 4,in which NaOCl and H_2O_2 were used alone, respectively] was not significant [P>0.05].

The use of an Sodium-Ascorbate antioxidant after NaOCl [groups 2] and H_2O_2 [group 5] irrigation significantly reduced the microleakage compared to their negative controls [P<0.05], such as herbal Quercetin after NaOCl [group 3] and H_2O_2 [group 6] irrigations significantly reduced the micro leakage compared to their negative controls [P<0.05].

However, in this study, micro leakage caused by the use of both antioxidants was not statistically different from those of the positive control [saline solution] group for both irrigation solution [P>0.05].

DISCUSSION

The success of endodontic treatment depends on adequate coronal restoration (9). Coronal micro leakage as a source of contamination has been cited as a significant cause of endodontic failure. Ideally, the coronal restoration should provide a hermetic seal to prevent ingress into the access cavity, since gutta-percha and sealer provide minimal resistance to bacterial micro leakage (10, 11).

Irrigation of root canal system is one of the most important steps in root canal treatment. NaOCl & H_2O_2 Both are widely used as an irrigation solution.

In this study, the deterioration effect of both irrigant on the micro leakage of dentin margin was also observed because both irrigants are a potent biological oxidant[12]. Nikaido et al (13) and Sasafuchi et al (14) reported that irrigation of the root canals of bovine incisors with NaOCl and H_2O_2 adversely affected the bond strength of several dentin bonding systems to coronal superficial dentin. Morris et al (15) also found that irrigation of the root canals with NaOCl produced a significantly large reduction in bond strength of dentin bonding resin to the canal dentin. Yiu et al (16) found that the micro tensile bond strengths of bonding resin to dentin were significantly reduced after NaOCl treatment.

However, these chemical agents differ from each other because of their mechanisms of action on dentin. Daumer et al (17) reported that pyridinoline cross-links that occur in collagen Type-1 and 2 were found to be disrupted by NaOCl, but not by H_2O_2. The formation of chloramines and protein-derived radical intermediates were also observed solely in the NaOCl treatment. Lai et al stated that the reduction in tesile bond strength of NaOCl treated dentin was connected with the presence of these reactive residual free-radicals. However, they suggested that residual solution in the collagen matrix and dentin tubules could be responsible for the reduction of bond strength in H_2O_2 treated dentin. Liberation of oxygen could either interfere with resin infiltration (18) or inhibit polymerization of resin (19) thus affecting bond strength.

In this study, in order to prevent micro leakage after endodontic treatment, antioxidant solution was used as final irrigation solution, following which the obturation was done. According Rose et al, ascorbic acid and its salt are well known antioxidants and are capable of reducing various oxidative compounds, especially free radicals. In this study Sodium Ascorbate increased the sealing ability of bonding system by neutralizing and reversing the

oxidizing effect of NaOCl and H_2O_2, when it was used as an additional and final irrigation agent. Ahmed et al found (20) Quercetin to possess good anti-oxidant property to heal aphthous ulcer. The use of Quercetin reduced the micro leakage induced by NaOCl and H_2O_2 irrigations in this study.

Microleakage score of groups 3 and 6, in which Quercetin was used as an additional irrigation solution, were different from negative controls [group 1 and 4] but not from that of positive control [group 7].

2% methylene blue dye was used in this study, it has a low molecular weight, and hence it has a deeper penetration as compared to other dyes.

CONCLUSION

The present study concludes that NaOCl and H_2O_2 adversely affected the adhesiveness of resin composite to tooth structures. The neutralization of adverse effects of NaOCl and H_2O_2, and subsequent reduction of micro leakage was seen by using antioxidant [Sodium Ascorbate and Quercetine] as an additional irrigation solution.

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Table 1: Dye penetration scores for Microleakage & Kruskal-Wallis test in all groups.

Groups	Irrigation solution	0	1	2	3	4	Mean	df	P
1	NaOCl	0	0	1	1	8	56.10		
2	NaOCl+SA	1	5	2	1	1	26.25		
3	NaOCl+Q	0	4	2	3	1	32.95		
4	H_2O_2	0	0	2	1	7	53.25	6	0.000
5	H_2O_2 +SA	2	3	3	1	1	26.50		
6	H_2O_2 +Q	1	1	4	3	1	34.65		
7	Saline	6	1	1	1	1	18.80		

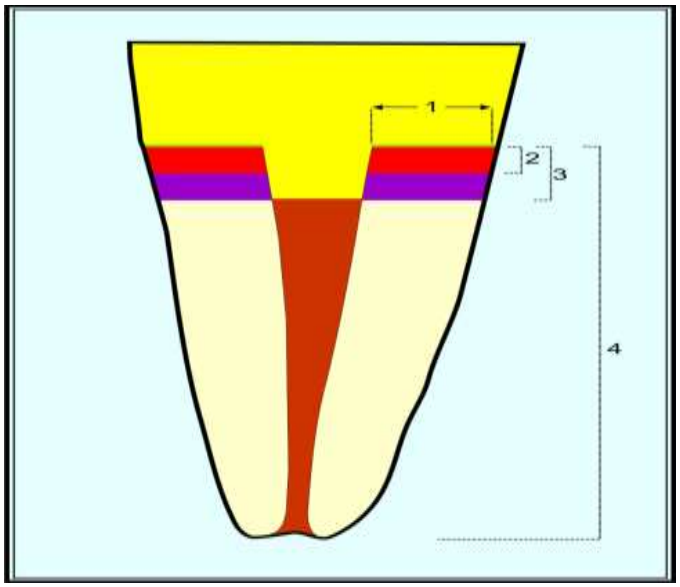

Figure 1: Scoring Criteria related to 0 to 4.

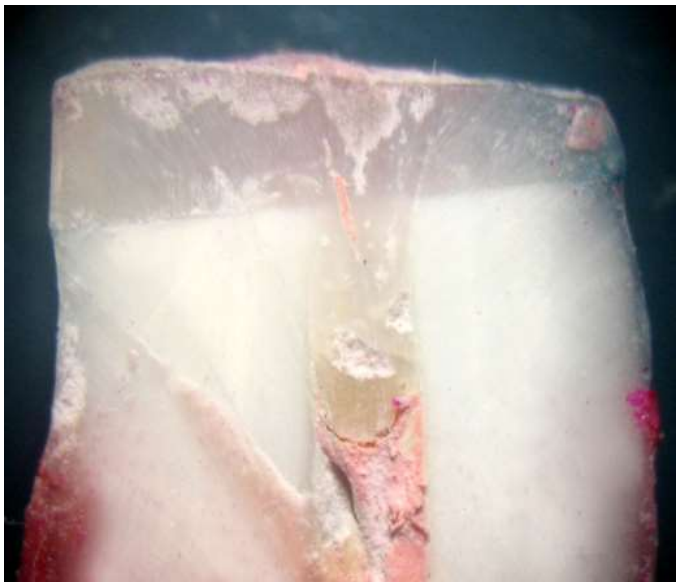

Fig 2: Score 0 No evidence of dye penetration at the tooth /restoration interface

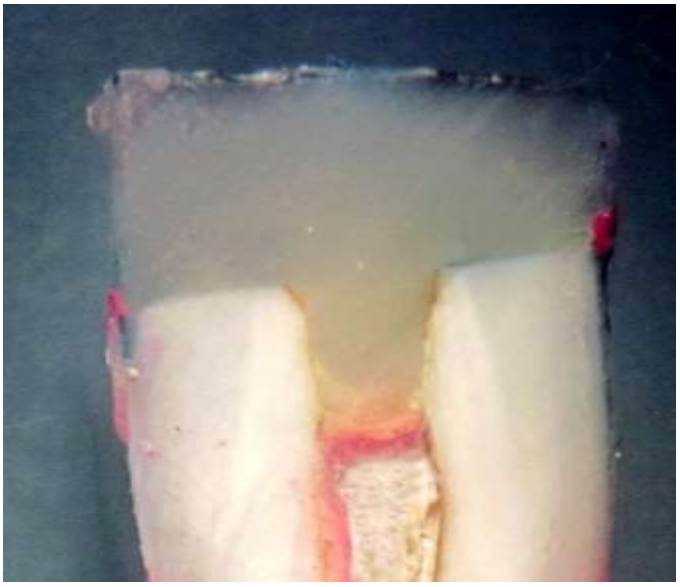

Fig 3: Score 1 Slight, Just horizontal dye penetration

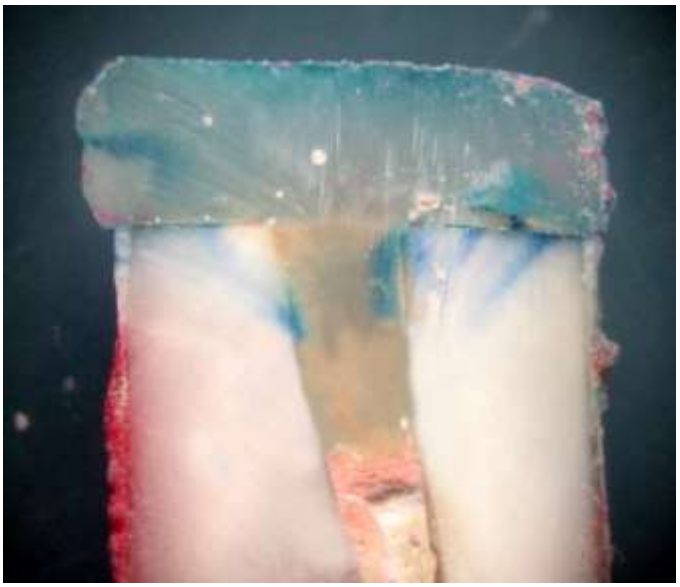

Fig 4: Score 2 Moderate, Vertical Penetration less than half of radicular composite restoration

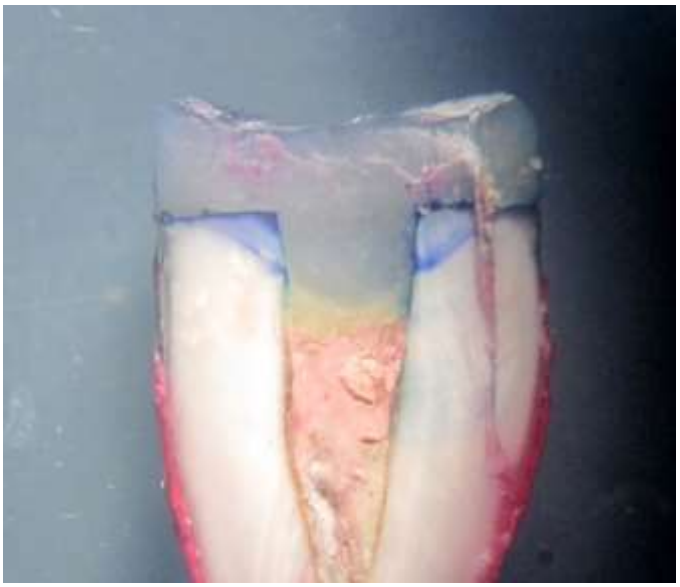

Fig 5: Score 3 Extensive, Vertical Penetration greater than half but less than total of radicular composite restoration.

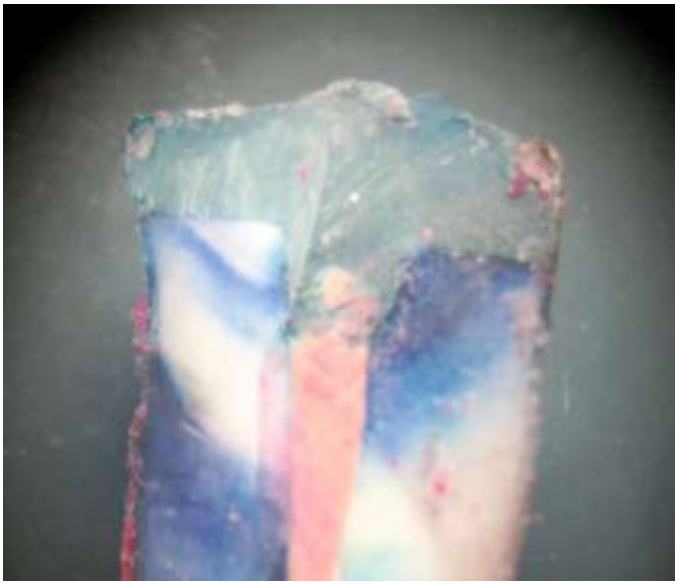

Fig 6:- Score 4 Gross, Vertical Penetration extending beyond the radicular composite restoration.