Remineralization Capacity of Three Fissure Sealants with and without Gaseous Ozone on Non-Cavitated Incipient Pit and Fissure Caries

Unal M*/ Oztas N**

Objective: *The purpose of this study was to investigate the remineralization activation of the application of three fissure sealants (FSs), alone or with gaseous ozone (GO), on non-cavitated initial caries and evaluate the clinical success of FS.* ***Study design:*** *Sixty children who had DIAGNOdent scores between 10–30 on bilateral symmetric mandibular first permanent molars were included in study. In a split-mouth design, teeth were assigned to experimental (with GO) and control (without GO) groups. GO was applied to teeth on one side and then the same brand of randomly selected FSs was applied to the teeth on both sides. Children were divided into 3 groups based on type of FS (Group 1: Aegis {Bosworth Co, North Hamlin Avenue Skokie, Illinois, USA}, Group 2: Fuji Triage {GC, Tokyo, Japan}, Group 3: Helioseal {Ivoclar Vivadent, Liechtenstein, Germany}). All FSs were then examined for retention rates at 1, 3, 6, 9, and 12 months; at the end of 12 months, all FSs were removed with an air-abrasion device and DIAGNOdent scores noted to compare with the initial values.* ***Results:*** *The application of GO with either Fuji Triage or Aegis FS was effective on remineralization ($p<0.05$); however, the application of Helioseal FSs was not effective ($p>0.05$). The 1st and 12th months' full retention rates of Fuji Triage FSs was a significant difference ($p<0.05$) from other FSs (Aegis and Helioseal) ($p>0.05$).* ***Conclusions:*** *GO+Aegis FS showed the highest remineralization; and, at the end of 12 months, its clinical success was higher than other FSs.*

Key words: Ozone gaseous, remineralization, fissure sealant

INTRODUCTION

Dental caries are generally seen in molars and often found in school children all around the world.[1] This situation creates an important problem in crowded developing countries.[2] The rate of pit and fissure caries is almost 80–90% in permanent posterior teeth and 44% in primary teeth.[3] For the purpose of preventing formation of caries in pits and fissures of permanent molars in school children, effective programs are required.

*Murat UNAL DDS, PhD, Department of Pedodontics, Faculty of Dentistry, Cumhuriyet University, Sivas, Turkey.

**Nurhan Oztas DDS, PhD, Professor, Department of Pedodontics, Faculty of Dentistry, Gazi University, Ankara, Turkey.

Send all correspondence to:

Murat Unal
Department of Pedodontics, Faculty of Dentistry
Cumhuriyet University, Sivas, Turkey
Mobile : +90 506 2734583
Fax: +90 346 2191237
E-mail: gmuratunal@hotmail.com

Incipient enamel caries is difficult to detect and to diagnose. The DIAGNOdent (Kavo, Biberach, Germany), an instrument for caries detection, contains a laser diode (655 nm, modulated, 1 mW peak power) as an excitation light source and a photo diode combined with a long pass filter (transmission >680 nm) for use as a detector. The DIAGNOdent is an easy to use device for the noninvasive detection of hidden caries on occlusal surfaces. When researchers compared DIAGNOdent and visual and conventional radiography for occlusal caries, they found no differences between DIAGNOdent measurements and the other caries detection methods.[4,5] DIAGNOdent has been extensively tested in permanent teeth for occlusal caries detection both in vivo and in vitro and has been indicated as highly reliable.[6] As could be seen in a review[7], DIAGNOdent, when compared to visual examination for detecting occlusal dentinal caries, was more sensitive but less specific, while for enamel caries it tended to be less sensitive and more specific. It would appear wise at the current state of development to use early detection devices such as DIAGNOdent as an adjunct to clinical decisions made in a conventional manner. In the present study, we used the Clinical Severity Index (CSI) (Table 1) and visual scoring with a DIAGNOdent device to assess caries.

The occlusal surface of the first permanent molars is most vulnerable to dental caries.[8] Since these teeth have deep and narrow fissures, they are susceptible to plaque retention and caries; therefore, caries prevention could be achieved by using a preventive

treatment approach. One of the crucial procedures to prevent initial enamel caries is the use of fissure sealants (FSs). As barriers, they prevent the aggregation of oral bacteria and dietary carbohydrates in the pits and fissures and the development of acidic conditions that cause tooth caries.[8,9] In the present study, we used three different FSs (Aegis, Fuji Triage and Helioseal FSs) (Table 2). The composition of FSs is important to their success. Two main types of FSs were discovered to be present: glass ionomer (GI) or resin-based (RB) sealants

GI sealants are moisture friendly because they require the presence of a moist environment to develop and maintain their mechanical properties. Most importantly, the fluoride-release-potential glass ionomers provide cariostatic action by producing an acid-resistant surface of fluoride-modified Hydroxyapatite (HAP); this, in turn, promotes remineralization of enamel in teeth with incipient caries.[10,11]

RBSs are classified according to their filler, which features either fluoride or amorphous calcium-phosphate contents. Helioseal FS is an unfilled and non-fluoride-containing FS; on the other hand, Aegis FS is known as a calcium phosphate-containing FS. New FSs that can release calcium and phosphate due to the presence of amorphous calcium phosphate (ACP) (Aegis) have now been marketed. The premise behind this sealant type is that, during caries formation at or below pH 5.8, HAP is leached from the enamel surface.[12] At this low pH value, ACP is capable of breaking down and releasing saturating levels of Ca^{+2} and PO_4 ions.[12,13] These concentrations are conducive to the formation of HAP; which, in turn, can be used by the tooth for enamel remineralization.[14] ACP is not only considered an indication for HAP, but also exhibits anti-cariogenic properties with remineralization potential.[15] Alsaffar *et al*.[16] evaluated the effect of FSs (Delton{Conventional unfilled}, Aegis{ACP-containing}, UltraSeal XP plus and Clinpro {Fluoride-containing}, Fuji Triage {glass ionomer sealant}) in protecting adjacent enamel from acid demineralization. They concluded that RB FSs containing fluoride or ACP could provide a better preventive effect on demineralization of adjacent enamel than conventional non-fluoride FS.

One of the alternative management strategies used for non-invasive interventions in recent years involves the use of ozone in reference to initial pit and fissure caries.[17,18] The ozone gas quickly kills microorganisms by virtue of oxidative degradation of the unsaturated fatty acids of the cell walls.[19] The ozone is used in two ways, as a gas or diluted in water. The authors of some studies indicate that the ozone decomposes into a harmless, non-toxic, and environmentally safe oxygen molecule and indicate that its unique characteristics enable it to serve as a disinfectant.[20,21] An ozone-generating device, HealOzone (Kavo, Biberach, Germany), has been developed and studied in dentistry in recent years. This device applies gaseous ozone (2100±200 ppm at a flow rate of 615 ccs/min) in high concentrations to the tooth surface under controlled conditions.[22] It must generate the ozone onsite during treatment due to the substance's molecular instability. In addition, its oxidation potential is high. When the cell membranes of bacteria in the lesion come into contact with the ozone, this feature enables their destruction.

In the minimally invasive dentistry, air abrasion has developed a new concept for the preservation of a maximal sound tooth structure. Air abrasion devices can be used at higher pressures for removal of old amalgam restorations and fissure sealants for replacing them or for removal repair of composites, glass ionomers, and the other restorations. Additionally, appropriate operating distance for the air abrasion was 0.5 to 2 mm from the tooth surface. Further distances produce a more diffuse stream that results in a diminished cutting ability.[23] For this reason, in the present study, we removed FSs by using air-abrasion higher than 2 mm distance from the tooth without any damage to the tooth enamel.

In recent studies, in-vitro application of ozone has been indicated to be a useful prophylactic antimicrobial treatment prior to etching and placement of dental sealants, one with no negative interactions with the physical properties of enamel and adhesive restorations.[24,25] The purpose of this research was to evaluate the remineralizing capacity of FSs used with gaseous ozone and their clinical success for a period of 12 months.

MATERIALS AND METHOD

This study was conducted with the approval of the Ethics Committee of Cumhuriyet University, Faculty of Dentistry. Informed consent forms clearly specifying the purpose and scope of the study were read and signed by the parents of all individuals included in the study.

This study was conducted on 60 patients, consisting of 29 girls and 31 boys between the ages of 7 and 9 (average age of 7.6) who had applied to Cumhuriyet University, Faculty of Dentistry, Department of Pedodontics for treatment. In this study, the female-to-male ratio was 0.93.

This study included children with both mandibular-symmetric first molars fully erupted, and those on the occlusal level. Before the restorative treatments were initiated, detailed medical and dental medical histories of the patients and their parents were obtained. Children with no acute or chronic systemic diseases, non-cavitated on their symmetric first permanent molars, but having initial pit and fissure caries on enamel, participated in this clinical research. Initially, standard oral-hygiene training was provided to the children. They were asked to brush their teeth in the instructed way after breakfast and before going to sleep every night during the treatment and control period.

Research Protocol and Treatment Groups

After the teeth to be used in the study were selected, the dental surfaces were cleaned using an air-flow device (Prophyflex 3, Kavo®, Germany) and a strong saliva absorber, and were then isolated with sterile cotton rolls. Following the surface cleaning, the teeth were washed with an air-water spray and dried. The Clinical Severity Index (CSI) and visual scoring were assessed for two contra-lateral molars as baseline reference values through use of a score system also developed by Ekstrand *et al* (Table 1).[26] Measurements with the DIAGNOdent device were done after calibration of the device with a ceramic standard .[18]

Measurements were made twice for each tooth and the arithmetic mean of the highest two values digitally shown by the device were recorded as the DIAGNOdent value. In consequence of the double-blind measurements made by two independent researchers, teeth scoring between 10 and 30 were included in the study (Table 3). Sixty patients included in the study were randomly divided into 3 study groups of 20 subjects each; these 3 study groups were then divided into an experimental (ozone) and a control (no ozone) group.

In every group, the teeth were also divided into an experimental group and a control group based on the use of gaseous ozone.

Treatment of the teeth in the experimental (ozone) group

Ozone treatment was implemented via an ozone generator (HealOzone, Kavo®, Germany) to the teeth in the experimental group as follows:

- Grasping the tooth tightly, a single-use silicon head fitting the sterilizable ozone generator head was placed against the teeth to be treated.
- The ozone generator was calibrated to an ozone concentration of 2100 ppm and a 615 cm^3/min flow rate. Ozone was applied to the treatment area under control with a vacuum pump for 40 seconds.
- The remaining gaseous ozone was vacuumed back within the next 10 seconds by the generator and automatically broken up by the decomposition system.

Following ozone application, a randomly selected treatment protocol was applied to the teeth in the experimental group.

Treatment of the teeth in the control (no ozone) group

A randomly selected PFS treatment protocol was applied without gaseous ozone application to the teeth in the control group.

After the teeth in the experimental and control groups were determined, the teeth were divided into 3 groups in terms of the applied PFS type.

GROUP 1 (Aegis ACP)

Following the application of 35% phosphoric acid (Bosworth Acid Etch, Bosworth Co, North Hamlin Avenue Skokie, Illinois, USA) to the pits and fissures, acid was pushed deep inside the fissure via a probe and kept there for 30 seconds. After that, the applied acid etching was washed off by an air-water spray for 20 seconds. Then the isolation was performed via the cotton roll, at which point, the tooth was dried with the air spray for 10 seconds. PFS (Aegis ACP, Bosworth Co, Illusiana, USA) was applied to all pits and fissures by means of a single-use brush. For photopolymerization purposes, a halogen light source with a light output power calibrated to 400 mW/cm^2 (Hilux 250, Benlioglu Dental, Ankara) was applied for 40 seconds.

GROUP 2 (Fuji Triage)

A glass ionomer-based fissure sealant produced in capsules (Fuji Triage, GC, Tokyo, Japan) was mixed in an amalgamator for 10 seconds in accordance with the manufacturer's instructions. The capsule, which was inserted into the gun after isolation of the tooth via the cotton roll, was triggered and applied to pits and fissures. When the PFS started losing its bright appearance, the product's varnish was applied with a cotton pellet for moisture isolation.

GROUP 3 (Helioseal)

Following the application of 37% phosphoric acid (Eco-Etch, Ivoclar Vivadent, Liechtenstein, Germany) to the pits and fissures, Helioseal fissure sealant was pushed deep inside the fissure via a probe and kept there for 30 seconds. After that, the applied acid etching was washed off by the air-water spray for 20 seconds. After isolation via a cotton roll, the tooth was dried with the air spray for 10 seconds. PFS (Helioseal, Vivadent, Liechtenstein, Germany) was applied to all pits and fissures with the help of a single-use brush. By means of the halogen light source (Hilux 250, Benlioglu Dental, Ankara), polymerization of the FS was done for 40 seconds.

After the PFS applications, in the 1st, 3rd, 6th, 9th, and 12th months of the study, the retention values of the PFSs that had been applied to the teeth in the experimental and control groups were clinically assessed. In the clinical assessment of the PFSs, partial loss and full retention of PFSs were evaluated as successful, but total losses were evaluated as unsuccessful.[27] Unsuccessful PFSs were excluded from the study. Two clinicians, who were pre-calibrated on the assessment criteria, performed the clinical assessments individually during each control session. We reached a consensus with discussion if our results were not the same. For remineralization and demineralization assessment, PFSs in all groups were removed after 12 months with an air-abrasion device (Rondoflex-plus 360 Kavo®, Biberach, Germany) and DIAGNOdent measurements were conducted again.

Statistical analysis

The intra- and inter-examiner of DIAGNOdent values were assessed by using Cohen's Kappa statistic test. The difference between the initial and 12th month DIAGNOdent values was evaluated based on alpha level adjusted for the Wilcoxon test. In each group, the retention assessment of the fissure sealants (experimental and control teeth) were analyzed by X^2 and Cochran's Q tests. McNemar's test was used to compare whether there was any statistical difference between the examination months. SPSS for 14.0 Windows (SPSS Inc. Chicago, IL, USA) was used to conduct all statistical analyses. Our data were shown on the tables as arithmetic mean ± standard deviation, sample numbers, and percentage, and significance level was taken as 0.05.

RESULTS

The intra-observer agreement between the first and second observers was found to be 0.88 and 0.93. The inter-observer agreement was found as 0.90 for the Cohen's Kappa.

In the study, when the initial and 12th month DIAGNOdent values were compared for the teeth in all test groups, group 1 and group 2 were found to have statistically significant differences ($p<0.05$). In group 3, no statistically significant difference was found ($p>0.05$). However, the results showed that, when initial and 12th month DIAGNOdent values were compared, a decrease was seen in all three groups (with ozone), but the difference was statistically insignificant ($p>0.05$). In consequence of these assessments, the highest DIAGNOdent change value was determined as group 1. In the control group teeth, when the initial and 12th month DIAGNOdent values were statistically assessed, the differences was found to be statistically insignificant in all three groups ($p>0.05$) (Table 4). In groups 1, 2 and 3, when the full retention values of the experimental and control groups on the 1st, 3rd, 6th, 9th and 12th months of the study were compared, the difference between the groups was insignificant ($p>0.05$) (Table 5). Comparing the 1st and 12th months' full retention values of the experimental and control in group 2, the difference was significant ($p<0.05$); when comparing other months in multiple, the differences were found not to be statistically significant ($p>0.05$).

Table 1. Clinical severity index scores(CSİ)

Scores	Description
0	No or slight change in enamel translucency after prolonged air drying (>5 seconds)
***1**	Opacity (white) hardly visible on the wet surface, but distinctly visible after air drying (>5 seconds)
***1a**	Opacity (brown) hardly visible on the wet surface, but distinctly visible after air drying (>5 seconds)
***2**	Opacity (white) distinctly visible without air drying
***2a**	Opacity (brown) distinctly visible without air drying
3	Localised enamel breakdown in opaque or discoloured enamel and or greyish discolouration from the underlying dentine
4	Cavitation in opaque or discoloured enamel exposing the dentine beneath

*The teeth were included in the study.

Table 2. Materials used in the study

Materials	Composition	Manufacturer	Lot Number
AEGİS	UDMA, mono-and di methacrylates resins TLV-TWA: 15 mg ⁄m3 TWA for ACP N⁄A for resin	Bosworth®-Compa-ny(USA)	0608-398
FUJİ TRİAGE	Glassionomer, alimunofluorosilicate glass, polyacrylic acid, distilled water, polybase carboxylic acid	GC Coopera-tion, Japan	0905281
HELİOSEAL	Bis-GMA, TEGD-MA(>99wt%) Additional contents are stabilizers and catalysts(<1 wt%)	İvoclar Vivadent Ets., (Schaan, Liechtenstein)	L24213

Abbreviations: Bis-GMA = bisphenol-aglycidylmethacrylate; N⁄A = Not available; TEGDMA = triethylene glycol dimethacrylate;

TLV-TWA = Threshold Limit Value-Time-Weighted Average; UDMA = urethane dimethacrylate.

Table 3. Guidelines for the clinical use of the DIAGNOdent

Value	Action
0-13:	Usual preventive measures
14 - ~ 20:	Intensified preventive care is advised
21 - ~ 29:	Intensified preventive or operative care is advised depending on the patient's caries risk, the recall interval, etc.
≥ = ~ 30:	Operative care (and intensified preventive measures) are advised

Note that the cut-off value is arange (~).

DISCUSSION

A number of studies have emphasized the fact that the success of fissure sealants (FSs), which is one of the most important applications in preventive dentistry, is directly associated with the material's ability to remain on the tooth surface.[11,28] The retention of FSs prevents the establishment of bacterial biofilm in the pits and fissures. Studies have revealed that there is a strong relationship between the retention of FSs, especially those that are resin-based, and the prevention of caries formation.[29]

Studies comparing the clinical effectiveness of FSs have observed that, generally, the method of testing the same mouth environment (split-mouth) is used.[30,31] The studies assessing the retention of FSs have determined that more accurate results were obtained in the mandibular first molars since it is easier to observe and assess them than other teeth.[32] For the reasons described above, the clinical success of FSs was observed on the mandibular first molars in this study.

A majority of the studies assessing the clinical success of resin and glass ionomer- (GI)-based FSs have reported that the effectiveness of the retention of GI-based FSs was lower than the resin-based products.[33,34] However, some researchers have put forward the idea that, although the retention values of GI-based FSs are lower than those of the resin-based ones, their caries-preventing effects are similar to or more effective than others.[35,36] In the assessment of the results of this study, when the 1st- and 12th-month retention values were statistically compared, both experimental groups and control groups of the Fuji-Triage (GI-based FS) group were found to be have been treated less successfully than the other groups. The clinical success of Fuji Triage was found to be low; however, when its initial and 12th- month DIAGNOdent values were statistically compared, the remineralization values were found to be similar to the remineralization values of the other two resin-based FSs (Control groups).

In the literature, there have been no clinical studies involving the Aegis FS (Amorphous-calcium-phosphate-containing, resin-based FS) other than the below-mentioned in-vitro studies. In the study conducted by Mark et al.[37] using Aegis and two FSs (Admira Seal {Ormoser based FS}, Conseal F {Fluoride-containing resin-based FS}), the researchers compared two different bond systems (Clearfil S3 Bond and OptiBond Solo Plus) with acid etching. It was determined that microleakage scores in groups to which Aegis was applied with phosphoric acid and OptiBond Solo Plus were lower than those in all other groups. Selecman *et al*.,[38] on the other hand, assessed the effect of occlusal surface treatments performed with air-abrasion and pumice by using Aegis and four different FSs (Admira Seal, Conseal F, Fuji Triage, and Delton Opaque {unfilled and non-containing fluoride FSs}) on the microleakage. Following the pumice application, the lowest microleakage values were obtained with Aegis FS. We have come to the conclusion that the low microleakage values given by Aegis FS in-vitro studies, according to the results of the above-mentioned studies, may be a result of the high retention values of Aegis in our study and may positively affect its clinical success. When the full retention results of Aegis were compared with the other FSs, this research reported that it provided similar results with Helioseal (unfilled and non-containing fluoride FS), although it was superior to the Fuji Triage.

Ozone, which has come into use in dentistry in recent years, is not only a preventive application, but also a non-invasive approach

for the treatment of existing initial caries. Many *in vitro* studies have shown the effectiveness of ozone on the pathogenic microorganisms that cause caries.[22,39] Moreover, ozone has a caries-preventing effect by causing the oxidation of acidic structures of carbohydrates in the saliva and caries lesions. The main philosophy in reference to ozone treatment is to change the balance of demineralization and remineralization cycles in favor of remineralization. In addition, it has been reported that remineralized tooth tissue has a more resistant structure in terms of new acid attacks. [17,20,22] Yamayoshi [40] have revealed that ozone is a strong oxidizer for cell walls and cytoplasmic membranes of microorganisms. Some studies have evaluated the antibacterial effects of ozone on oral microorganisms according to results indicating that when it is used the number of planktonic microorganism cells in-vitro significantly decreased.[21,41] Baysan [42] determined that remineralization of primary root-caries lesions takes place within 10 or 20 seconds after ozonated water application. Furthermore, the results of Hodson *et al* [43] are in line with the results of Baysan *et al* [42] the ozone has an effect on the remineralization of initial fissure and root-caries lesions. Atabek [44] reported that gaseous ozone application to mandibular first molars with non-cavitated initial fissure caries affected remineralization positively.

In our study, participants with DIAGNOdent values between 10–30 on bilateral first-permanent mandibular-molar teeth and non-cavitated initial pit and fissure caries were chosen because the carious depth was limited to the enamel stage. Data regarding the effect of gaseous ozone before application of FSs is limited. Celiberti [24] reported that gaseous ozone may be applied as a prophylactic treatment prior to etching and placement of FSs. Recent studies have reported that ozone does not have an adverse effect on the sealing ability of FSs (ozone fs). Machesil *et al*[45] reported that gaseous ozone did not immediately affect the enamel-bond strength or microleakage scores of FSs. In addition, findings in previous studies show that gaseous ozone does not change enamel or dentin structure:[24,25,45] a 40-second ozone gas application did not have any effect on the physical properties of enamel. For this reason, we preferred ozone applications of 40 seconds on the experimental group teeth in our study. As a result, comparisons between the experimental and control groups had statistically insignificant results. These findings confirm the findings in the above-mentioned research.

Sılva et al.[15] revealed that amorphous calcium phosphate (Aegis) and fluoride (Fluorshield) containing FSs promote remineralization of artificially induced caries lesions on enamel surfaces. Coudhary

Table 4. The comparison of DIAGNOdent values

	Group 1		Group 2		Group 3	
DIAGNOdent						
Scores	**Ozone**	**No ozone**	**Ozone**	**No ozone**	**Ozone**	**No ozone**
İnitial Values	23.25(5.29)[a]	20,12(5,63)	23.07(4.58)[b]	20,42(5,57)	22.70(4.60)	19,40(4,87)
12.Month Values	21.95(4.49)[a]	19,60(5,45)	22.42(4.80)[b]	20,65(5,06)	22.22(4.38)	19,47(4,95)
Results	*p=0.014	p=0.087 *	p=0.021	p=0.379	p=0.570	p=0.665

*Same letters are significantly different (P<0.05)

Table 5. Successful and full retantion of fissure sealants

	1.Month		3.Month		6.Month		9.Month		12.Month		***Results***	
Groups	**Ozone**	**No ozone**	**Ozone**	**No ozone**	**Ozone**	No ozone	Ozone	No ozone	Ozone	No ozone	Fr (1. and 12.month comparasion)	
	Ss/Fr	Ss/Fr	Ss/Fr	Ss/Fr	Ss/Fr	Ss/Fr	**Ss/Fr**	**Ss/Fr**	**Ss/Fr**	**Ss/Fr**	**Ozone**	**No ozone**
Group 1 (n) (%)	20/20	20/20	20/20	20/20	20/20	20/20	20/20	20/20	20/18	20/19	**p=0.092**	**p=0.406**
	100/100	100/100	100/95	100/100	100/95	100/100	100/90	100/100	100/90	100/95		
Group 2 (n) (%)	20/17	20/18	19/16	19/16	19/15	18/15	19/14	18/14	19/13	18/13	***p=0.040**	***p=0.015**
	100/85	100/90	95/84.2	95/84.2	95/78.9	90/83.3	95/73.7	90/77.8	95/68.4	90/72.2		
Group 3 (n) (%)	20/19	20/20	20/19	19/18	20/19	19/18	20/19	19/17	20/19	19/17	**p=1.000**	**p=0.406**
	100/95	100/100	100/95	95/94.7	100/95	95/94.7	100/95	95/89.5	100/95	95/89.5		
Results	**x^2=0.019**		**x^2=0.018**		**x^2=0.018**		**x^2=0.073**		**x^2=0.128**			
	p=0.990		**p=0.991**		**p=0.991**		**p=0.964**		**p=0.938**			

*p<0.05 significant

Ss(Successfull sealants)=Full retention+partial retention, Fr=Full retention

*From repeated observations for each group by months, in group 2 for the Fr , for the experimental and control groups were statistically significant differences between 1st and 12th months (p<0.05).

et al.[46] demonstrated that amorphous calcium phosphate (Aegis) and fluoride containing FSs (Teethmate F1) have the potential to remineralize because of ACP molecules and fluoroapatite formation; in addition, these results were supported by scanning electron microscopy. When the results of our study were assessed, it was observed that the decrease in the DIAGNOdent values were mostly in the Aegis-Fuji Triage-Helioseal FS groups, respectively. Because of ACP in the Aegis FS material and the fluoride content in Fuji Triage, we believe they would be more effective in remineralization of teeth after gaseous ozone application. There was also a decrease in the DIAGNOdent values based on ozone application in the group using Helioseal FS with no remineralizing agent, but this difference was not found to be statistically significant.

CONCLUSIONS

Gaseous ozone application before applying fissure sealant did not affect the clinical success of the Aegis, Fuji Triage and Heliseal fissure sealants,

Gaseous ozone may be helpful together with fissure sealants with amorphous calcium phosphate or fluoride content for initial caries prevention,

Resin-based fissure sealants may be preferable to glass ionomer fissure sealants for higher retention rates.

REFERENCES

1.Petersen PE. Global policy for improvement of oral health in the 21st century--implications to oral health research of World Health Assembly 2007, World Health Organization. Community Dent Oral Epidemiol; 37: 1-8. 2009.

2.Liu BY, Lo EC, Chu CH, Lin HC. Randomized trial on fluorides and sealants for fissure caries prevention. J Dent Res; 91: 753-758. 2012.

3.Feigal RJ, Donly KJ. The use of pit and fissure sealants. Pediatr Dent; 28: 143-150. 2006.

4.Kavvadia K, Lagouvardos P. Clinical performance of a diode laser fluorescence device for the detection of occlusal caries in primary teeth. Int J Paediatr Dent; 18: 197-204. 2008.

5.Toraman Alkurt M, Peker I, Deniz Arisu H, Bala O, Altunkaynak B. In vivo comparison of laser fluorescence measurements with conventional methods for occlusal caries detection. Lasers Med Sci; 23: 307-312. 2008.

6.Lussi A, Hibst Paulus R. DIAGNOdent™: an optical method for caries detection. J Dent Res; 83 (Spec Iss C): C80-C83. 2004.

7.Bader JD, Shugars DA. A systematic review of the performance of a laser fluorescence device for detecting caries. J Am Dent Assoc; 135: 1413-1426. 2004.

8.Brodeur JM, Payette M, Galarneau C. Treatment cost savings with universal coverage of dental pit and fissure sealants in Quebec. Journal of Canadian Dental Association; 63: 625-632. 1997.

9.Simonsen RJ. Pit and fissure sealants.In:Clinical application of the acid etch technique. Chicago: Quintessence Publishing Co. Inc.,:19-42, 1978.

10.Antonson SA, Antonson DE, Brener S, Crutchfield J, Larumbe J, Michaud C, Yazici AR, Hardigan PC, Alempour S, Evans D, Ocanto R. Twenty-four month clinical evaluation of fissure sealants on partially erupted permanent first molars: glass ionomer versus resin-based sealant. J Am Dent Assoc;143:115-122. 2012.

11.Beauchamp J, Caufield PW, Crall JJ, Donly K, Feigal R, Gooch B, Ismail A, Kohn W, Siegal M, Simonsen R. Evidence-based clinical recommendations for the use of pit-and-fissure sealants: a report of the American Dental Council on Scientific Affairs. J Am Dent Assoc; 139: 257-268. 2008.

12.Foster JA, Berzins DW, Bradley TG. Bond strenght of an amorphous calcium phosphate-containing orthodontic adhesive. Angle Orthod; 78: 339-344. 2008.

13.Skrtic D, Antonucci JM, Eanes ED. Improved properties of amorphous calcium phosphate fillers in remineralizing resin composites. Dent Mater; 12: 295-301. 1996.

14.Skrtic D, Antonucci JM, McDonough WG, Liu DW. Effect of chemical structure and composition of the resin phase on mechanical strength and vinyl conversion of amorphous calcium phosphate-based composites. J Biomed Mater Res A.; 68: 763-772. 2004.

15.Silva KG, Pedrini D, Delbem AC, Ferreira L, Cannon M. In situ evaluation of the remineralizing capacity of pit and fissure sealants containing amorphous calcium phosphate and/or fluoride. Acta Odontol Scand.; 68: 11-18. 2010.

16.Alsaffar A, Tantbirojn D, Versluis A, Beiraghi S. Protective effect of pit and fissure sealants on demineralization of adjacent enamel. Pediatr Dent ; 33: 491-495. 2011.

17.Baysan A, Lynch E. Effect of ozone on the oral microbiota and clinical severity of primary root caries. Am J Dent; 17: 56–60. 2004.

18.Huth KC, Paschos E, Brand K, Hickel R. Effect of ozone on non-cavitated fissure carious lesions in permanent molars. A controlled prospective clinical study. Am J Dent; 18: 223–228. 2005.

19.Dähnhardt JE, Jaeggi T, Lussi A. Treating open carious lesions in anxious children with ozone. A prospective controlled clinical study. Am J Dent; 19: 267–270. 2006.

20.Baysan A, Beighton D. Assessment of the ozone-mediated killing of bacteria in infected dentine associated with non-cavitated occlusal carious lesions. Caries Res.; 41: 337–341. 2007.

21.Velano HE, do Nascimento LC, de Barros LM, Panzeri H. In vitro assessment of antibacterial activity of ozonized water against Staphylococcus aureus. Pesqui Odontol Bras; 15: 18–22. 2001.

22.Baysan A, Whiley RA, Lynch E. Antimicrobial effect of a novel ozone-generating device on micro-organisms associated with primary root carious lesions in vitro. Caries Res; 34: 498–501. 2000.

23.Hegde VS, Khatavkar RA. A new dimension to conservative dentistry: Air abrasion. J Conserv Dent. 2010; 13: 4-8.
24.Celiberti P, Pazera P, Lussi A. The impact of ozone treatment on enamel physical properties. Am J Dent; 19: 67-72. 2006.
25.Schmidlin PR, Zimmermann J, Bindl A. Effect of ozone on enamel and dentin bond strength. J Adhes Dent; 7: 29-32. 2005.
26.Ekstrand KR, Ricketts DNJ, Kidd EAM, Ovist V, Schou S. Detection, diagnosis, monitoring and logical treatment of occlusal caries in relation to lesion activity and severity: an in vivo examination with histological validation. Caries Res; 32: 247–254. 1998.
27.Feigal RJ, Hitt J, Splieth C. Retaining sealant on salivary contaminated enamel. J Am Dent Assoc; 124: 88-97. 1993.
28.Simonsen RJ. Retention and effectiveness of dental sealant after 15 years. J Am Dent Assoc; 122: 34-42. 1991.
29.Simonsen RJ, Neal RC. A review of the clinical application and performance of pit and fissure sealants. Aust Dent J; 56 Suppl 1: 45-58. 2011.
30.Subramaniam P, Konde S, Mandanna DK. Retention of a resin-based sealant and a glass ionomer used as a fissure sealant: a comparative clinical study. J Indian Soc Pedod Prev Dent; 26: 114-120. 2008.
31.Poulsen S, Laurberg L, Vaeth M, Jensen U, Haubek D. A field trial of resin-based and glass-ionomer fissure sealants: clinical and radiographic assessment of caries. Community Dent Oral Epidemiol; 34: 36-40. 2006.
32.Brooks JD, Mertz-Fairhurst EJ, Della-Giustina VE, Williams JE, Fairhurst CW. A comparative study of two pit and fissure sealants: three-year results in Augusta, Ga. J Am Dent Assoc; 99:42-46. 1979.
33.Taifour D, Frencken JE, van't Hof MA, Beiruti N, Truin GJ. Effects of glass ionomer sealants in newly erupted first molars after 5 years: a pilot study. Community Dent Oral Epidemiol; 31: 314-319. 2003.
34.Yip HK, Smales RJ. Glass ionomer cements used as fissure sealants with the atraumatic restorative treatment (ART) approach: review of literature. Int Dent J; 52: 67-70. 2002.
35.Beiruti N, Frencken JE, van't Hof MA, Taifour D, van Palenstein Helderman WH. Caries-preventive effect of a one-time application of composite resin and glass ionomer sealants after 5 years. Caries Res; 40: 52-59. 2006.
36.Williams B, Laxton L, Holt RD, Winter GB. Fissure sealants: a 4-year clinical trial comparing an experimental glass polyalkenoate cement with a bis glycidyl methacrylate resin used as fissure sealants. ; 10;180:104-108. 1996.
37.Marks D, Owens BM, Johnson WW. Effect of adhesive agent and fissure morphology on the in vitro microleakage and penetrability of pit and fissure sealants. Quintessence Int; 40: 763-772. 2009.
38.Selecman JB, Owens BM. Effect of preparation technique, fissure morphology and material characteristics on the in vitro margin permeability and penetrability of pit and fissure sealants. Pediatr Dent; 29: 308-314. 2007.
39.Oizumi M, Suzuki T, Uchida M, Furuya J, Okamoto Y. In vitro testing of a denture cleaning method using ozone. J Med Dent Sci; 45: 135-139. 1998.
40.Yamayoshi T, Tatsumi N. Microbicidal effects of ozone solution on methicillin-resistant Staphylococcus aureus. Drugs Exp Clin Res; 19: 59–64. 1993.
41.Cardoso MG, de Oliveira LD, Koga-Ito CY, Jorge AO. Effectiveness of ozonated water on Candida albicans, Enterococcus faecalis, and endotoxins in root canals. Oral Surg Oral Med Oral Pathol Oral Radiol Endod; 105: e85–91. 2008.
42.Baysan A, Lynch E. The use of ozone in dentistry and medicine. Part 2. Ozone and root caries. Prim Dent Care. ; 13: 37-41. 2006.
43.Hodson N, Dunne SM. Using ozone to treat dental caries. J Esthet Restor Dent; 19: 303–305. 2007.
44.Atabek D, Oztas N. Effectiveness of Ozone with or without the Additional Use of Remineralizing Solution on Non-Cavitated Fissure Carious Lesions in Permanent Molars. Eur J Dent; 5: 393-399. 2011.
45.Marchesi G, Petris LC, Navarra CO, Locatelli R, Di Lenarda R, Breschi L, Cadenaro M. Effect of ozone application on the immediate shear bond strength and microleakage of dental sealants. Pediatr Dent; 34: 284-288. 2012.
46.Choudhary P, Tandon S, Ganesh M, Mehra A. Evaluation of the remineralization potential of amorphous calcium phosphate and fluoride containing pit and fissure sealants using scanning electron microscopy. Indian J Dent Res.; 23:157-163. 2012.