

Comparative Evaluation Of Retention And Microleakage In Acid-Etch And Self-Etch Pit And Fissure Sealants In Primary Teeth Post Radiotherapy: An In-Vitro Study

Dr. Dzuthohulu Thirah,¹ Dr. Sonvita Debnath^{2*}, Dr. Shabana Praveen Alam³, Dr. Ankita Singh⁴, Dr Kedoneikho⁵, Pranay Kumar Singh⁶

¹Urban Primary Health Centre, National Health Mission

^{2*}Assistant Professor, Department of Pediatric and Preventive Dentistry, Manav Rachna Dental College, Manav Rachna International Institute of Research and Studies, Faridabad, Haryana, India (Corresponding Author)

Email ID: sonvitad7@gmail.com

³ Clinician, Plush Dental

⁴Post Graduate Second year, Vyas Dental College and Hospital, Jodhpur, Rajasthan

⁵Post Graduate Second Year, Department of Oral Pathology and Microbiology, Jaipur Dental College, Jaipur, Rajasthan

⁶Under Graduate Intern, Manav Rachna Dental College, Manav Rachna International Institute of Research and Studies, Faridabad, Haryana, India

ABSTRACT

Background: The effect of radiotherapy on dental hard tissues increases their susceptibility to demineralization. In addition, irradiation can interfere with the stability of both the enamel and the dentin, compromising the bonding of sealants and leading to microleakage.

Aim: To evaluate and compare the retention and microleakage in two types of pit and fissure sealants of primary teeth post-radiotherapy.

Materials and method: 60 primary molars were allocated into four groups. In the designated 3 teeth, irradiation was carried out, and the sealants were placed as follows: GROUP 1 (n = 15) = Embrace™ Wetbond™. GROUP 2 (n = 15) = Prevent Seal. Another 30 teeth, which were taken as a control, were not irradiated prior to the placement of the sealant in the following: GROUP 3(n = 15) = Embrace™ Wetbond™. GROUP 4(n = 15) = Prevent Seal. The specimens were subjected to a fractionated radiation dose of up to 60 Gy, and the sealants were assessed for retention and microleakage.

Results: When comparing the two sealants, a significant difference ($P < 0.05$) was found between them, favouring the Self-etch sealant Prevent seal than acid-etch sealant Embrace™ Wetbond™. In retention stability, a higher proportion has been shown in Embrace™ Wetbond™ without irradiation (66.7%) than with irradiation (26.7%). And in Microleakage scores, prevent seal without irradiation (86.6%) revealed a more significant percentage than with irradiation (73.3%).

Conclusion: The self-etch sealant Prevent seal showed higher retention stability and less microleakage than the acid-etch sealant Embrace™ Wetbond™.

Key-word: Radiotherapy, Pit and fissure sealants, Microleakage, Bonding, primary teeth

INTRODUCTION

The statistics shows that over 2,15,000 cancer cases in children under the age of 15 are found each year, with 80,000 cancer fatalities reported worldwide¹. According to the National Cancer Institute (NCI), in a population of 0–14 years, the most prevalent malignancies reported were brain tumors (17%) and leukaemia (47%)². During head and neck radiotherapy, radiation typically causes direct damage to the teeth, resulting in dental hard tissue damage and changes in the mechano morphological properties of enamel^{4,5}. These mechano morphological properties seen in the demineralization behaviour of irradiated enamel damage the enamel, resulting in progressive tooth decay, with reports of a reduction in enamel microhardness due to decarboxylation of the hard tissue, inducing micro fracture in the hydroxyapatite minerals, affecting the mineral content, composition, structure, and mechanical properties⁶.

In numerous in-vitro studies on radiation caries, areas of demineralization in enamel with characteristics of diffused brown spots on the non-cavitated smooth surface enamel were discovered, and concluded that in irradiated patients, the brown spots are distinctive signs and should be taken into consideration and treated as early caries^{7,8}. Therefore, applying sealants is one of the non-invasive treatments for incipient caries that is clinically advised. Non-cavitated carious lesions are the earliest stages of caries lesion development, causing discoloration of teeth, loss of lusture, and changes in surface structure, due to demineralization before macroscopic breakdown takes place⁹. According to various studies, applying sealants to

non-cavitated lesions can stop or reduce the progression of caries. The existing data and the AAPD recommendations support sealing early caries that are non-cavitated on the occlusal surface in children and young adults¹⁰. Studies reported that, children receiving radiation or chemotherapy may need to consistently take highly cariogenic dietary supplements or take drugs high in sugar to maintain their weight, which could lead to more active caries. Additionally, parents and other caregivers may be too preoccupied with a child's medical condition to pay attention to the oral hygiene aspect^{11,12}. Pits and fissures are also more susceptible to caries because of how difficult they are to clean and how well they retain plaque. Also, primary dentition can be at risk for caries based on fissure anatomy or patient risk factors. Therefore, sealants are recommended for primary teeth with discoloured and deep retentive pits and fissures that show evidence of decalcification, as they are the main methods of preventing dental caries, or they can be used as a secondary method of halting the progression of the disease and, as a result, would benefit from sealant administration^{13,14}. Microleakage has been a common problem encountered with pit and fissure sealants. It gives way to the movement of bacteria or fluids between the surface of the cavity and the restorative material. In addition, the stability of both enamel and dentin after irradiation is interrupted, affecting the bond strength and compromising the success of the material¹⁵. Hence, there is a need to compare microleakage in teeth that are irradiated with non-irradiated teeth. Since there is a paucity of literature on the *in vitro* retention and microleakage of sealants after irradiation, the current *in vitro* study is being undertaken to compare the retention of acid-etch sealant and self-etching sealant, in primary molars after irradiation. Two sealants, Embrace™ Wetbond™ for acid etch sealant and Prevent Seal for self-etching sealant were used, as studies have demonstrated their clinical success. Embrace™ Wetbond™ is a resin-based, moisture-tolerant sealant that can bond to a relatively wet tooth surface, and Prevent Seal is effective in bonding the sealant in a quick and simple application. In the assessment study, the microleakage and retention of two materials used as pit- and fissure sealants were evaluated.

METHODS

Following approval from the ethical committee, the study was carried out in the Department of Pedodontics and Preventive Dentistry, Inderprastha Dental College and Hospital, Sahibabad, Ghaziabad. A sample size of 60 was determined using G power (Version 3.0). No teeth were extracted for the purpose of this study, and only those primary molar teeth that were already extracted by the clinicians were procured for the study. Inclusion criteria for the extracted teeth were: caries-free primary molars; teeth with deep retentive pit and fissure sealant; teeth with no fracture lines; hypoplasia or any developmental anomalies; and teeth with numerous appearances of calcification. Exclusion criteria for the extracted teeth were: a soft base of the cavity on probing; the presence of a cracked line; a wide self-cleansable pit and fissure; teeth with cavitation or carious dentin.

In the set of 30 teeth, irradiation was carried out, and the sealants were placed as per the following:

Group 1 (n = 15) = acid-etch sealant, Embrace™ Wetbond™. Group 2 (n = 15) = self-etch sealant, Prevent Seal. 30 teeth, which were taken as a control, were not irradiated prior to the placement of the sealant as per the following: Group 3 (n = 15) = acid-etch sealant, Embrace™ Wetbond™. and Group 4 (n = 15) = self-etch sealant, Prevent Seal.

30 teeth in groups 1 and 2 were subjected to radiotherapy at a fractionated dose of two Gy over five consecutive days for six weeks, with a final dose of 60 Gy prior to sealant placement. The occlusal fissures of the specimens were then prepared by using a fissurotomy bur prior to the sealant placement, followed by a thorough rinsing with water to remove any remnants, and air-dried. Self-etch sealant (prevent seal) was directly applied to the cleaned pit and fissure without etching according to the manufacturer's instructions. In the case of acid-etch sealant (Embrace™ wetbond), the occlusal surface was dried and etched with 37% phosphoric acid and rinsed thoroughly, followed by the application of pit and fissure sealant, which was cured for 20 seconds. Samples were made to undergo thermocycling in a water bath (according to the International Organization for Standardization, 2003)¹⁶. The test comprises 500 cycles in a water bath between 5°C and 55°C starting after 20 hours to 24 hours storage in the water at 37°C.

For the retention assessment of sealants, clinical evaluation was done by both visual and tactile examination using Simenson criteria¹⁷ as: completely present (C), partially present (P), completely missing (M) and decayed (D). The degrees of microleakage were scored using the criteria given by Övrebö and Raadal¹⁸: score 0: no dye penetration; score 1: dye penetration restricted to the outer half of the sealant; score 2: dye penetration to the inner half of the sealant; and score 3: dye penetration into the underlying fissure.

Statistical analysis was performed using MS Excel and SPSS 27.0. The proportions (%) were calculated for each clinical parameter, and the statistical test of significance was used. Nominal categorical data between the groups was compared using the chi-square goodness-of-fit test.

RESULT

The distribution of retention stability of two sealants is shown in Table (1-2). Embrace™ Wetbond™ without irradiation had a higher proportion (66.7%) than with irradiation (26.7%) (P = 0.023). In comparison to Prevent Seal with irradiation (80.0%), Prevent Seal without irradiation exhibited a higher percentage (86.7%) (P = 0.025). The comparison of retention stability between two sealant groups with and without irradiation is shown in Tables (3-4). Two sealants were statistically significant from one another with irradiation (P = 0.012) and without irradiation (P = 0.024).

The distribution of microleakage scores for both sealant materials with and without irradiation is shown in Tables (5-6). The two sealant materials showed statistically significant differences. Without irradiation, the Microleakage score-0 of

Embrace™ Wetbond™ showed a higher percentage (46.7%) than with irradiation (20.0%) (P=0.018). Without irradiation, the Prevent Seal's microleakage score-0, revealed a larger percentage (86.6%) than they did with irradiation (73.3%) (P = 0.039). Table (7-8) compares the microleakage scores of two sealant materials, Embrace™ Wetbond™ and Prevent Seal, with and without irradiation. The difference between the two sealants with and without irradiation was statistically significant (P=0.032).

Table 1: Retention capability of Embrace™ Wetbond™ with and without irradiation

Retention	With irradiation		Without irradiation		Chi square	p-value
	N	%	N	%		
C	4	26.7	10	66.7	14.828	0.023
P	9	60.0	4	26.6		
M	2	13.3	1	6.7		
D	0	0	0	0		

Table 2: Retention capability of Prevent Seal with and without irradiation

Retention	With irradiation		Without irradiation		Chi square	p-value
	N	%	N	%		
C	12	80.0	13	86.7	4.040	0.025
P	2	13.3	2	13.3		
M	1	6.7	0	0		
D	0	0	0	0		

Table 3: Retention capability of two sealant groups with irradiation

Retention	Embrace wetbond		Prevent Seal		Chi square	p-value
	N	%	N	%		
C	4	26.7	12	80.0	8.788	0.012
P	9	60.0	2	13.3		
M	2	13.3	1	6.7		
D	0	0	0	0		

Table 4: Retention capability of two sealant groups without irradiation

Retention	Embrace wetbond		Prevent Seal		Chi square	p-value
	N	%	N	%		
C	10	66.7	13	86.7	8.848	0.024
P	4	26.6	2	13.3		
M	1	6.7	0	0		
D	0	0	0	0		

Table 5: Microleakage scores of Embrace™ Wetbond™ with and without irradiation

Score	With irradiation		Without irradiation		Chi square	p-value
	N	%	N	%		
0	3	20.0	7	46.7	4.400	0.018
1	8	53.3	3	20.0		
2	2	13.3	1	6.7		
3	2	13.3	4	26.7		

Table 6: Microleakage scores of Prevent Seal with and without irradiation

Score	With irradiation		Without irradiation		Chi square	p-value
	N	%	N	%		
0	11	73.3	13	86.7	3.581	0.039
1	2	13.3	2	13.3		
2	1	6.7	0	0		
3	1	6.7	0	0		

Table 7: Microleakage scores of two sealant groups with irradiation

Score	Embrace wetbond		Prevent Seal		Chi square	p-value
	N	%	N	%		
0	3	20.0	11	73.3	8.838	0.032
1	8	53.3	2	13.3		
2	2	13.3	1	6.7		
3	2	13.3	1	6.7		

Table 8: Microleakage scores of two sealant groups without irradiation

Score	Embrace wetbond		Prevent Seal		Chi square	p-value
	N	%	N	%		
0	7	46.7	13	86.7	7.000	0.032
1	3	20.0	2	13.3		
2	1	6.7	0	0		
3	4	26.7	0	0		

DISCUSSION

It was reported that there is minimal tooth damage below 30 Gy (salivary gland threshold), but a critical threshold of ≥ 60 Gy may be related to the direct effect of radiation on the tooth structure. These findings suggested that care should be taken during the treatment planning process to limit the tooth dose to less than 60 Gy¹⁹. In accordance with this report and the study done by (Mellara TS et al.,2020)²⁰, the same RT regimen was adopted in this investigation, which consists of cumulative fractionated daily sessions of 2 Gy across the weekdays to a final dose of 60 Gy. The fractionated RT protocol was followed to adhere to the 5R's (repair, redistribution, reoxygenation, regeneration, and radiosensitivity)²¹.

(Naves et al.,2012) investigated the bond strengths of adhesives to restored enamel and dentin before and after radiotherapy, and they discovered that when the two dental substrates were restored after radiotherapy, both substrates experienced effects that compromised their bonding capacities and potentially harmed the formation of the hybrid layer²². This finding is comparable with the current investigation, though the materials used were different, which found that sealants without irradiation had a higher proportion of retention stability and less microleakage than sealants after irradiation. As a result, it may be proposed that sealants procedures be performed prior to irradiation in patients with head and neck cancer. Furthermore, because radiotherapy effects on the dentition are cumulative and dosage dependent²³, sealants applied during the radiotherapy time frame may have provided better clinical service than restorations placed after radiotherapy. During eating, drinking, and breathing, all dental components in the mouth cavity are constantly subjected to heat and pH changes. Therefore, samples were made to undergo thermocycling in a water bath to simulate the oral environment and like studies done by (Garrocho-Rangel A et al.,2015)²⁴ to be most clinically relevant. In this investigation, 1% Methylene blue dye was employed since it is easily detectable under visible light, is soluble in water, and diffuses freely, similar to (Panse AM et al.,2018)²⁵ studies. In this work, the samples were sectioned buccolingually using a water-cooled diamond disc, and inspected under a stereomicroscope, as (Garg D et al.,2019)²⁶ did. The sealant adheres to the enamel by micromechanical interlocking with the microporosities of the enamel. This property allows better bonding to enamel than to dentin²⁷. A good sealant material is biocompatible, has good retention, and is resistance to abrasion and wear. Sealant bonding to enamel is critical because microleakage at the tooth-material contact can result in treatment

failure²⁸. In this study, both the sealants revealed some dye penetration. This finding is consistent with the findings of (Theodoridou-Pahini et al.,1996) and (Do Rego et al.,1999), who concluded that microleakage is to be expected in all restorative materials. This could be related to the fact that the coefficient of thermal expansion of sealants is substantially greater than the coefficient of thermal expansion of teeth²⁹. Etch-and-rinse and self-etch are two different processes for sealants. Researchers investigated self-etch sealants for an efficient sealing process and came to different conclusions when compared to the traditional etch and rinse method. Some studies reported that self-etch sealants were found to be a good alternative to the traditional acid-etch technique, especially for children with behavioural issues and serious tooth decay, as well as for individuals who have nausea, reflux, or who are uncooperative as the washing and acid etching processes are omitted^{30,31}. In addition, the self-etching technology cuts down on application time because it eliminates the need for a separate etching process. It is less method sensitive and improves clinical performance^{32,33}. Its reduced incidence of post-operative sensitivity compared with the etch-and-rinse technique is another factor that is therapeutically significant. The results of the current study, showed that the self-etch sealant, produced better results when compared to the acid-etch sealant. This supports the findings of a systematic review suggesting that self-etch adhesives should be used to restore the teeth of head and neck cancer patients either before or after radiation³⁴. The acid in acid-etch sealants induces microporosities to improve bondi g agent penetration, and as previously proven, radiation causes the enamel to become more porous. The use of acid on the damaged enamel after radiotherapy may produce excessive conditioning³⁵. The results of our study were favourable for making use of self-etch Prevent Seal after irradiation, besides considering the compliance and limited treatment time available for a pediatric patient. Features like 21-MPa retention with the enamel, release of fluoride, greater flow and simplified application have the potential to make any clinician choose a self-etch sealant over the acid-etch sealant³⁶. Presumably, this is the first study that has evaluated acid-etch and self-etch sealants after RT in extracted primary teeth. Only a few studies have evaluated the effects of RT on different restorations. Future research is advised to assess the long-term effectiveness of sealants after radiation under in vivo circumstances. Furthermore, it is impossible to directly compare the results due to the lack of research on primary teeth exposed to radiation.

CONCLUSION

The retention stability and microleakage of acid-etch and self-etch without irradiation exhibited a higher percentage when compared with irradiation. Hence, sealant procedures may be suggested prior to irradiation in patients with head and neck cancer. And sealants applied during the radiotherapy time frame may have provided better clinical service than restorations placed after radiotherapy completion. Overall comparison between acid-etch and self-etch sealant as a measure to determine its effectiveness post radiotherapy, revealed that self-etch sealant showed a reliable and predictably good performance. The effectiveness of acid-etch sealant was less favourable.

REFERENCES

1. World Health Organization. International childhood cancer day: Much remains to be done to fight childhood cancer. WHO Press Release No. 241. Lyon, France: World Health Organization; 2016.
2. National Cancer Institute, Control and Prevention Center. Cancer Registry. Vėžys Lietuvoje 2012 metais. Vilnius; 2015.
3. Walker MP, Wichman B, Cheng AL, Coster J, Williams KB. Impact of Radiotherapy Dose on Dentition Breakdown in Head and Neck Cancer Patients. *Pract Radiat Oncol*. 2011;1:142-8.
4. Lieshout HF and Bots CP. The effect of radiotherapy on dental hard tissue.A systematic review. *Clin Oral Investig*. 2014;18(1):17-24.
5. Seyedmahmoud R et al. Oral cancer radiotherapy affects enamel microhardness and associated indentation pattern morphology. *Clin Oral Investig*. 2018 ;22(4):1795-1803.
6. Reed R, Xu C, Liu Y, Gorski JP, Wang Y, Walker MP. Radiotherapy effect on nano-mechanical properties and chemical composition of enamel and dentine. *Arch Oral Biol*. 2015;60(5):690-7
7. Silva AR, Alves FA, Antunes A, Goes MF, Lopes MA. Patterns of demineralization and dentin reactions in radiation-related caries. *Caries Res*. 2009;43(1):43-9.
8. Jongebloed WL, Gravenmade EJ, Retief DH. Radiation caries. A review and SEM study. *Am J Dent*. 1988 ;1(4):139-46.
9. Young D.A, Novy B.B., Zeller G.G., Hale R, HartT.C, Truelove E.L. The American Dental Association Caries Classification System for clinical practice: A report of the American Dental Association Council on Scientific Affairs. *J. Am. Dent. Assoc*. 2015;146, 79–86.
10. American Academy of Pediatric Dentistry. Evidence-based Clinical Practice Guideline for the Use of Pit-and-Fissure Sealants. *Pediatr. Dent*. 2016;38, 263–279.
11. Lieshout HF, Bots CP. The effect of radiotherapy on dental hard tissue--a systematic review. *Clin Oral Investig*. 2014 ;18(1):17-24.
12. Ritwik P. Dental Care for Patients With Childhood Cancers. *Ochsner J*. 2018;18(4):351-7.
13. Kitchens, D.H. The economics of pit and fissure sealants in preventive dentistry: A review. *J. Contemp. Dent. Pract*. 2005;6:95–103
14. Feigal, R.J. The use of pit and fissure sealants. *Pediatr. Dent*. 2002; 24:415–422

15. Kidd EA. Microleakage: a review. *J Dent.* 1976;4(5):199-206.
16. Garrocho-Rangel A, Lozano-Vázquez C, Butrón-Tellez-Girón C, Escobar-García D, Ruíz-Rodríguez S, Pozos-Guillén A. In vitro assessment of retention and microleakage in pit and fissure sealants following enamel pre-etching with sodium hypochlorite deproteinisation. *Eur J Paediatr Dent.* 2015 Sep;16(3):212-6.
17. Simonsen RJ. Retention and effectiveness of dental sealant after 15 years. *J Am Dent Assoc.* 1991;122(10):34.
18. Ovrebo RC, Raadal M. Microleakage in fissures sealed with resin or glass ionomer cement. *Scand J Dent Res.* 1990 Feb;98(1):66-9
19. Walker MP et al. Impact of Radiotherapy Dose on Dentition Breakdown in Head and Neck Cancer Patients. *Pract Radiat Oncol.* 2011;1(3):142-8.
20. Mellara TS, Paula-Silva FWG, Arid J, et al. Radiotherapy Impairs Adhesive Bonding in Primary Teeth: An In Vitro Study. *Journal of Dentistry for Children (Chicago, Ill.).* 2020;87(2):69-76.
21. Prabhakar AR, Murthy SA, Sugandhan S. Comparative evaluation of the length of resin tags, viscosity and microleakage of pit and fissure sealants—an in vitro scanning electron microscope study. *Contemp. Clin. Dent.* 2011;2(4):324
22. Naves LZ, Novais VR, Armstrong SR, Correr-Sobrinho L, Soares CJ. Effect of gamma radiation on bonding to human enamel and dentin. *Support Care Cancer.* 2012;20(11):2873-8.
23. Kielbassa Harrington K et al. Molecular biology for the radiation oncologist: The 5Rs of radiobiology meet the hallmarks of cancer. *Clin Oncol* 2007;19(8):561-71.
24. Alani AH, Toh CG. Detection of microleakage around dental restorations: a review. *Oper Dent.* 1997;22(4):173-85.
25. Panse AM, Nair MC, Patil AS, Bahutule SR. Comparison of microleakage, bond strength, and fracture strength of no etch no bond novel flowable composite as a pit and fissure sealant in comparison to the conventional sealants: An In vitro Study. *Int. J. Pedod. Rehabil.* 2018;3(1):28.
26. Garg D, Mahabala K, Lewis A, Natarajan S, Nayak A, Rao A. Comparative evaluation of sealing ability, penetration and adaptation of a self etching pit and fissure sealant- stereomicroscopic and scanning electron microscopic analyses. *J Clin Exp Dent.* 2019;11(6):e547-e552.
27. Waggoner WF, Siegal M. Pit and fissure sealant application: Updating the technique. *J Am Dent Assoc.* 1996;127:351–61.
28. Pérez-Lajarin L, Cortés-Lillo O, García-Ballesta C, CózarBraz Hidalgo A .Marginal microleakage of teo fissure sealants: acomparative study. *J Dent Child* 2003;70(1):24
29. Garrocho-Rangel A et al. In vitro assessment of retention and microleakage in pit and fissure sealants following enamel pre-etching with sodium hypochlorite deproteinisation. *Eur J Paediatr Dent.* 2015;16(3):212-6.
30. Burbridge L, Nugent Z, Deery C. A randomized controlled trial of the effectiveness of a one-step conditioning agent in sealant placement: 6-month results. *Int J Paediatr Dent.* 2006;16:424–30.
31. Ram D, Mamber E, Fuks AB. Clinical performance of a non-rinse conditioning sealant in three paediatric dental practices: A retrospective study. *Int J Paediatr Dent.* 2005;15:61–6.
32. Peumans M, Kanumilli P, De Munck J, Van Landuyt K, Lambrechts P, Van Meerbeek B. Clinical effectiveness of contemporary adhesives: A systematic review of current clinical trials. *Dent Mater* 2005;21(9):864-81.
33. Peumans M, De Munck J, Van Landuyt KL, Poitevin A, Lambrechts P, Van Meerbeek B. Eight-year clinical evaluation of a 2-step self-etch adhesive with and without selective enamel etching. *Dent Mater* 2010;26(12):1176-84.
34. Madrid Troconis CC, Santos-Silva AR, Brandao TB, Lopes MA, de Goes MF. Impact of head and neck radiotherapy on the mechanical behavior of composite resins and adhesive systems: A systematic review. *Dent Mater* 2017;33(11):1229-43
35. Goncalves LM et al. Radiation therapy alters microhardness and micro structure of enamel and dentin of permanent human teeth. *J Dent* 2014;42(8):986-92.
36. Nahvi A, Razavian A, Abedi H, Charati JY. A comparison of microleakage in self-etch fissure sealants and conventional fissure sealants with total-etch or self-etch adhesive systems. *Eur J Dent.* 2018;12:242-6.