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Original Article

Dentistry Section

Assessment of White Spot Lesions and In-Vivo Evaluation of the Effect of CPP-ACP on White Spot Lesions in Permanent Molars of Children

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ABSTRACT

Introduction: As hindrance of remineralisation process occurs during orthodontic therapy resulting in decalcification of enamel because number of plaque retention sites increases due to banding and bonding of appliances to teeth.

Aim: The present analytic study was undertaken to assess the occurrence of white spot lesions in permanent molars of children with and without orthodontic therapy and to evaluate the effect of Casein PhosphoPeptide-Amorphous Calcium Phosphate (CPP-ACP) on white spot lesions in post-orthodontic patients in a given period of time.

Materials and Methods: The study comprised of examination of 679 first permanent molars which were examined to assess the occurrence of smooth surface white spot lesions in children of 8 to 16 years age group. Group I comprised subjects without any orthodontic treatment and Group II comprised of subjects who had undergone orthodontic therapy. The sample size was calculated using the epi-info6 computer package. Treatment group included 20 post-orthodontic patients examined with at least one white spot lesion within the enamel who received remineralizing cream (GC Tooth Mousse, Recaldent, GC Corporation.) i.e., CPP–ACP cream two times a day for 12 consecutive weeks. Computerized image analysis method was taken to evaluate white spot lesions. These frequency and percentages were compared with chi-square test. For comparison of numeric data, paired t-test was used.

Results: Of the total 278 (49.6%) first permanent molars showed occurrence of smooth surface white spot lesions out of 560 in Group I and 107 (89.9%) first permanent molars showed presence of white spot lesions out of 119 debanded first permanent molars of children examined in Group II. CPP-ACP therapy group showed reduction in severity of codes which was found to be highly significant after 12 weeks and eight weeks on gingival-third, p-value (<0.001) and significant after eight weeks and four weeks on middle-third according to ICDAS II criteria and computerized image analysis.

Conclusion: CPP-ACP therapy minimum for 12 weeks is highly recommended as post-orthodontic treatment need in management of smooth surface white spot lesions on teeth undergoing fixed orthodontic therapy according to the present study.

Keywords: Casein phosphopeptide-amorphous calcium phosphate, Decalcification, Incipient caries lesions, Orthodontics

INTRODUCTION

White spot lesion is defined as sub-surface enamel porosity from carious demineralization that presents itself as milky-white opacity when located on smooth surfaces. White spot lesions have been termed as the most important iatrogenic effects of orthodontic fixed appliance therapy [1]. The number of plaque retention sites increases because of banding and bonding of orthodontic appliances to teeth, due to which oral hygiene becomes more difficult to carry out. The reduced pH of plaque surrounding orthodontic bands causes hindrance in the remineralization process of enamel and thus decalcification takes place [2].

Diagnosis and clinical assessment of white spot lesions is a great challenge for researchers. In view of lack of consistency among the contemporary criteria systems, International Caries Detection and Assessment System II (ICDAS II) criteria was developed by an International team of caries researchers to integrate several new criteria systems into one standard system for caries detection and assessment [3]. It determines the potential histological depth as well as surface changes of carious lesions by observing surface characteristics [4]. A newer software method, computerized image analysis is used to assess the clinical effect of therapeutic agents used in treatment of white spot lesions [5]. Assessment of enamel demineralization from coloured images appeared to be more reproducible than direct clinical observation with only the naked eye.

The overall prevalence of white spot lesions amongst orthodontic patients has been reported between 2% and 96% [6]. In view of the specific position occupied by the orthodontic band on the crown of each tooth, following orthodontic treatment, the distribution of lesions on individual teeth is related to the characteristic band position for each tooth. The increase is greatest on the cervical and middle thirds of the vestibular surfaces of these teeth [7]. However, reports of in-vivo studies of intervention to treat white spot lesion in post orthodontic patients are limited [8]. Potential of CPP-ACP to prevent enamel demineralization and promote lesion remineralization has been demonstrated in animal caries models, in vitro models, human in situ models and clinical trials [9]. CPP-ACP is a delivering system that allows freely available calcium and phosphate ions to attach to enamel and reform into calcium phosphate crystals [10]. CPP-ACP nano complexes stabilize calcium and phosphate ions, prevent their transformation into crystalline phases and maintain a highly supersaturated solution [9].

AIM

The present study was undertaken to study the occurrence of white spot lesions in permanent molars of children with and without orthodontic therapy and to assess an in vivo effect of CPP-ACP on white spot lesions in post-orthodontic patients in a given period of time. Main purpose of this study is to apply knowledge

in prevention of occurrence of disease during orthodontic therapy in children.

MATERIALS AND METHODS

The present analytic and prospective study was carried out in the Department of Pediatrics and Preventive Dentistry in coordination with Department of Orthodontics, M. M College of Dental Sciences and Research, Mullana, Ambala, Haryana, India. The study comprised of examination of 679 first permanent molars in children of 8 to 16 years age group and treatment of 20 patients out of all the post-orthodontic patients examined with at least one white spot lesion within the enamel. The study was conducted in two parts. In first part, analysis was carried out by examination of total 679 first permanent molars were divided into two groups. Group I comprised of 560 first permanent molars that were examined to assess the occurrence of smooth surface white spot lesions in children who had not undergone any orthodontic treatment. Group II comprised of examination of 119 first permanent molars for the occurrence of the smooth surface white spot lesions in first permanent molars in children who had undergone orthodontic therapy i.e., postorthodontically after debanding. All these samples were finalized as per the guidelines of the research protocol of a study. The sample size was calculated using the epi-info 6 computer package by the statistician. Number of first permanent molars taken for prevalence determination of white spot lesions due to unknown etiology, in normal population was taken five times than special group with known aetiology i.e., orthodontic therapy. This study was approved by ethical committee of the institute and written informed consent was obtained from children and their guardians. The buccal and palatal/lingual surfaces of the first permanent molars in each quadrant were examined for the presence of white spot lesions according to ICDAS II criteria [3]. Codes for detection and classification of carious lesions on the smooth surfaces according to ICDAS II criteria are as follows:

Code 0: Sound tooth surface

Code 1: First visual change in enamel

Code 2: Distinct visual change in enamel when viewed wet

Code 3: Localized enamel breakdown due to caries with no visible dentin

Code 4: Underlying dark shadow from dentin with or without localized enamel breakdown

For inter-examiner and intra-examiner bias, calibration test was conducted before the start of the study for satisfactory conformity. Consequently the risk of methodological misjudgement was considered to be minimal. For the selection of subjects in Group I, two nearby schools were selected by random sampling for examination of study in the required age group i.e., 8-16 years. Permission letter was taken from respective school authorities. Age was confirmed by birth certificate and information provided by school records. It was carried out in the school playgrounds as provided by the school authorities. The children were made comfortable and seated in upright chair and examined under natural light. Prior to examination, a cheek retractor was inserted into the mouth and surfaces of teeth were wiped with cotton and dried with chip blower. A ball end explorer was used to aid in examination. Prior to examination, ultrasonic scaling of the teeth was done to remove any plaque and debris and all tooth surfaces were polished with pumice and prophylaxis cup. Ultrasonic scaling was done in the college mobile dental van equipped with ultrasonic facility. For the enrolment of subjects in Group II, 8 to 16 years old children who had been treated earlier with orthodontic fixed appliances in the orthodontic department of the institute were recalled in the department for follow-up. A total of 119 first permanent molars were examined in this part of study. The children were made comfortable and seated in upright chair and examined under natural light and same procedure was followed in order to detect white spot lesions using ICDAS II criteria for severity of lesion. After the clinical examination, photographs of first permanent molars were made using digital camera. For the second part of the study, 20 post-orthodontic patients who were ready to participate in the study and had white lesion adjacent to the gingiva or middle-third of the tooth regarding the position and with lesion of 2mm^2 or larger regarding the size were selected for treatment group. Both the subjects and their parents received verbal and written information and signed a consent protocol. The selected volunteers received remineralizing cream (GC Tooth Mousse, Recaldent, GC Corporation), in this treatment part of study. After daily oral hygiene procedures using dentifrices and soft texture toothbrush, patients were instructed to locally apply a standardised amount of CPP–ACP cream two times a day for 12 consecutive weeks. Patients were instructed to make the diet chart and fill the given calendar form. Pre-treatment and post-treatment records of buccal and palatal/lingual surfaces of all these included permanent first molars in 20 volunteers were taken for the presence of white spot lesions. Identification number was given to every selected patient to prepare data for statistical evaluation and they were examined every four, eight and twelve weeks according to ICDAS II criteria. ICDAS II criteria codes were given on each particular visit to every observed lesion. Atleast one most severe white spot lesion on gingival surface was chosen for follow up. Effect of remineralizing cream was seen for increase or decrease in codes after four, eight and twelve weeks. If a lesion ceased to fulfil the lesion criteria, it was scored 0 (sound enamel).

Digital photographs of the permanent first molars were taken using Sony cyber-shot DSC-H55 14.1 mega pixels camera with 25mm wide-angle G lens having 10X optical zoom. Images were captured using camera and saved in JPEG format. Then they were opened in Adobe Photoshop version 7.0 and image edge enhancement was done by filtering the images and using brush strokes with accented edges. Then these images were stored as new JPEG file format. The images were carefully examined and white spot lesions were identified and traced using the free hand tool and computer mouse. Clinical assessment of change in white spot lesions was done by comparing pre therapy and post therapy record [Table/Fig-1,2].

The transitions for each white spot lesion between baseline and follow-up assessments were scored according to a transition

[Table/Fig-1a-c]: Representative photographs showing decrease in codes of ICDAS II criteria after treatment with CPP-ACP cream. (a) Ist permanent molar with accented edges showing code 2 ICDAS II criteria. (b) Ist permanent molar with accented edges showing code 2 ICDAS II criteria. (c) After intervention period of 12 weeks, showing code 0 of ICDAS II criteria.

[Table/Fig-2a-c]: Permanent I[st] molars showing code 1 of ICDAS II criteria, after intervention period of 12 weeks.
Permanent I[st] molars showing marking of white spot lesion.
Permanent I[st] molars with accented edges showing code 2 of ICDAS II criteria.
Representative photographs showing decrease in codes of ICDAS II criteria after treatment with CPP-ACP cream.

matrix. Lesion regression was accommodated, in that (<) baseline was given when transitions to less severe grades occurred. (>) baseline represented lesion progression, and stable (S) scores represented stable lesions that did not progress or regress. For numeric data, mean and standard deviation were calculated. For categorical data, frequency and percentage were calculated. These frequency and percentages were compared with chi-square test. For comparison of numeric data, paired t-test was used.

RESULTS

Results found that 278 (49.6%) first permanent molars, 132 maxillary (47.48%) and 146 mandibular (52.52%) showed occurrence of smooth surface white spot lesions out of 560 first permanent molars of children without orthodontic therapy examined in the age group of 8-16 years with International Caries Detection and Assessment System II. Total 107 (89.9%) first permanent molars, 52 (48.6%) maxillary and 55 (51.40 %) mandibular first permanent molars showed presence of white spot lesions out of 119 debanded first permanent molars of children examined in the age group of 8-16 years in post-orthodontic patients [Table/Fig-3]. To compare and evaluate the differences in occurrence and distribution of white spot lesions with and without orthodontic treatment groups, chi-square test was applied; it was found that the difference was highly significant with p-value < 0.0001 with chi-square value of 64.84% [Table/Fig-3].

Number of white spot lesions	Group I Permanent molars without orthodontic treatment.		Group II Permanent molars with orthodontic treatment.		p-value
	Frequency	Percent	Frequency	Percent	
Absent	282	50.3%	12	10.1%	<0.0001**
Present	278	49.6%	107	89.9%	
Total	560	100.0%	119	100.0%	
Maxillary first permanent molars	132	47.48	52	48.60%	0.251
Mandibular first permanent molars	146	52.52	55	51.40%	

[Table/Fig-3]: Frequency of white spot lesions in permanent molars with and without orthodontic treatment
(* Statistically significant, ** Statistically highly significant using Chi-square test).

In subjects without orthodontic therapy, white spot lesions were most prevalent on gingival third of buccal surface {code 1 (18.9%), code 2(14.3%) followed by middle-third [code 2(4.6 %), code 1(0.7%), code 3 (0.4 %) followed by on occlusal-third [code 0(100%)]}.

In case of subjects with debanded first permanent molars, white spot lesions were most prevalent on gingival third of buccal surface {[code 2(49.6 %), code 1(9.2%)] followed by middle-third [code 2 (19.3 %), code 4 (12.6%)] followed by on occlusal-third [code 4 (0.8%)]}. The occurrence of white spot lesions were found highly significant for gingival-third of buccal and palatal /lingual surface and also for middle-third of buccal and palatal /lingual surface of debanded first permanent molars when comparison of tooth divisions of buccal and palatal/lingual surfaces of first permanent molars of post-orthodontic group was done. The difference in occurrence of white spot lesions was not found significant between maxillary and mandibular first permanent molars in children with and without orthodontic therapy.

[Table/Fig-4] shows percentage of regression of white spot lesions in post-orthodontic treatment group. After four weeks, 17 (27.9%) teeth showed regression whereas 44 teeth (72.1%) showed no regression; after eight weeks, 40 (65.6%) teeth showed regression whereas 21 teeth (34.4%) showed no regression and after 12 weeks, 45 (73.8%) teeth showed regression whereas 21 teeth (34.4%) showed no regression on treatment with GC Tooth Mousse in post-orthodontic treatment group.

[Table/Fig-5] depicts the comparison of tooth divisions of buccal surfaces at time interval of four weeks, eight weeks and twelve

Regression (Y/N)	After 4 weeks		After 8 weeks		After 12 weeks	
	Frequency	Percent	Frequency	Percent	Frequency	Percent
Yes	17	27.9	40	65.6	45	73.8
No	44	72.1	21	34.4	21	34.4
Total	61	100.0	61	100.0	61	100.0

[Table/Fig-4]: Percentage of regression of white spot lesions after 4, 8 and 12 weeks in post-orthodontic treatment group.

	Paired Differences					T	Df	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
BG After debanding - BG At 4 weeks	0.164	0.610	0.078	0.008	0.320	2.097	60	0.040*
BG After debanding - BG At 8 weeks	0.656	0.814	0.104	0.447	0.864	6.291	60	<0.001**
BG After debanding - BG At 12 weeks	1.131	1.087	0.139	0.853	1.410	8.124	60	<0.001**
BM After debanding - BM At 4 weeks	0.230	0.883	0.113	0.003	0.456	2.030	60	0.047*
BM After debanding - BM At 8 weeks	0.410	1.116	0.143	0.124	0.696	2.868	60	0.006**
BO After debanding - BO At 12 weeks	.066	0.512	0.066	-0.066	0.197	1.000	60	0.321

[Table/Fig-5]: Mean differences between tooth divisions of buccal surfaces of debanded first permanent molars at different time intervals of post-orthodontic treatment group (n=20).
(* Statistically significant, ** Statistically highly significant using paired t-test)
BG= gingival-third of buccal surface, BM= middle-third of buccal surface, BO= occlusal-third of buccal surface.

weeks. Statistical paired t-test with t-value <0.001 shows that results were highly significant for gingival-third of buccal surface at debanding and after eight weeks and also after twelve weeks. Results were significant for gingival-third and middle-third of buccal surface after debanding and after four weeks. Results were highly significant for middle-third of buccal surface after debanding and after eight weeks.

[Table/Fig-6] depicts the comparison of tooth divisions of palatal/lingual surfaces at time interval of four weeks, eight weeks and twelve weeks. Statistical paired t-test with t-value <0.001 shows that results were highly significant for gingival-third of palatal/lingual surface at debanding and after eight weeks and results were also highly significant for gingival-third of palatal/lingual surface at debanding and after twelve weeks. Results were significant for middle-third of palatal/lingual surface after debanding and after four weeks. Results were highly significant for middle-third of palatal/lingual surface at debanding and after eight weeks. Results were also highly significant for middle-third of palatal/lingual surface at debanding and after twelve weeks.

	Paired Differences					T	df	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
P/L G After debanding - P/L G At 4 weeks	0.033	0.547	0.070	-0.107	0.173	0.468	60	0.641
P/L G After debanding - P/L G At 8 weeks	0.475	0.673	0.086	0.303	0.648	5.513	60	<0.001**
P/L G After debanding - P/L G At 12 weeks	0.689	0.904	0.116	0.457	0.920	5.946	60	<0.001**
P/L M After debanding - P/L M At 4 weeks	0.197	0.601	0.077	0.043	0.351	2.558	60	0.013*
P/L M After debanding - P/L M At 8 weeks	0.230	0.589	0.075	0.079	0.380	3.045	60	0.003**
P/L M After debanding - P/L M At 12 weeks	0.295	0.691	0.089	0.118	0.472	3.333	60	0.001**

[Table/Fig-6]: Mean differences between tooth divisions on palatal/lingual surfaces of debanded first permanent molars at different time intervals of post-orthodontic treatment group.
(* Statistically significant, ** Statistically highly significant using paired t-test) P/L G= gingival-third of palatal/lingual surface, P/L M= middle-third of palatal/lingual surface.

DISCUSSION

Demineralization is an initial phase of dental caries that initiates when plaque is allowed to remain on the tooth surface for a significant duration of time. In orthodontic patients, demineralization usually takes place in the form of white or brown spots on the enamel around the brackets and can lead to cavitation [11].

The present study was performed to determine the occurrence, severity and distribution of white spot lesions in permanent molars of children who have undergone orthodontic treatment and children without orthodontic therapy. There was variation in the sample size as only a small proportion of children undergo orthodontic treatment. Thus, it is difficult to find such a large sample of children who underwent orthodontic therapy. Further the present study was an analytic study and not a comparative study; hence there was a variation in Group I and Group II. Moreover, Sagarika N et al., conducted a comparative study among a test group that comprised of 90 subjects who underwent orthodontic treatment for a period of 12-15 months and control group that consisted of 90 subjects who were in need of orthodontic treatment and found a high prevalence rate of 75.6% in test group as compared to 15.6% in control group in Indian patients which reported to be about five times more prevalent in orthodontic patient as compared to normal population [12].

Children of age group 8-16 years were included in our study as in this age group children are in mixed dentition or late mixed dentition period and literature shows that fixed orthodontic therapy is usually carried out in this period. Space management is done in mixed dentition period and late mixed dentition period and therapeutic management is done after eruption of all permanent molars. First permanent molars were selected as the study teeth as these teeth are usually banded and bonded during fixed orthodontic therapy.

Diagnosis of white spot lesions in first permanent molars was carried out by using International Caries Detection and Assessment System Criteria for smooth surfaces i.e., ICDAS II criteria. This method was chosen as this system measures different stages of the caries process. It was developed from the systematic reviews of literature on the clinical caries detection system and other sources and thus imparts a new technique for the measurement of dental caries [13,14]. In present study the distribution of the ICDAS codes was difficult to explain. The code 2 (enamel caries detected on a wet tooth surface) might be easier to detect than the code 1 (enamel caries visible only when tooth surface is dry) and code 3 (minor cavitation on enamel only).

The present study found difference in percentage of occurrence of smooth surface white spot lesions in permanent molars with and without orthodontic banding and thus bonding results in definite high susceptibility of patients with fixed orthodontic therapy to white spot lesions. This shows that white spot lesions can occur on any tooth surface in the oral cavity where the microbial biofilm is allowed to develop and remain for a period of time i.e. banding and bonding being definite modifying factors impacting their development. The occurrence of white spot lesions in children without orthodontic treatment was 49.6% and in children with post-orthodontic white spot lesions in first permanent molars was found out to be 89.9%. Mahmoud E et al., reported that before orthodontic treatment, 32.3% of the patients had white spot lesions (WSL) and after orthodontic treatment, 73.5% of the patients presented WSL and thus reported incidence of WSL during multibracket treatment was 60.9% of the patients [15] . Sandvik et al., reported 11% and 50%, Lovrov S et al., reported 15.5% and 95.3% prevalence of white spot lesions without treatment and after orthodontic treatment respectively [16,17].

The high occurrence of white spot lesions in post-orthodontic group in present study may be due to the younger age group involved or may be due to method of examination involved. Since younger children are not able to maintain their oral hygiene so chances of plaque accumulation are high and so is the prevalence of white spot lesions. The data of present study depicts that maximum number of white spot lesions were present on gingival third followed by middle third on both buccal and palatal surfaces of first permanent molars of post-orthodontic group. This illustrates that surface enamel is changed in almost every single patient receiving fixed orthodontic treatment. The surfaces are therefore at great risk to develop visible WSLs during treatment if the patient does not comply with a caries-preventive program.

Enamel remineralization has been a topic of interest for researchers for about 100 years, and the literature recommends that the non-invasive management of early caries lesions by remineralization process offers better method for the clinical management of the disease. For the treatment part of the present study, CPP-ACP was chosen as the medicament. It has been GRAS-affirmed (Generally Recognized as Safe) by the Food and Drug Administration of the

United States of America and other regulatory bodies around the world and can be incorporated into oral care products and foods. Another advantage of this therapy is that these products are ingestible. In contrast, topical fluoride therapy poses a risk if the patient ingests a significant amount of fluoride [18].

CPP-ACP has been shown to localize and stabilize calcium and phosphate ions at the tooth surface in a bioavailable form that can promote remineralization of enamel subsurface lesions in situ, restoring the white opaque appearance of the lesions to translucency, even in the presence of fluoride. In the presence of fluoride, CPP-ACP has been shown to promote the formation of fluorapatite deep in the subsurface lesion [19].

Pukallus M et al., investigated the effect of casein phosphopeptide-amorphous calcium phosphate cream to reduce mutans streptococci and revealed reduction in the bacterial count [20]. In the present study, the treatment for 12 weeks with GC Tooth Mousse (GC Corporation) resulted in significant improvements in change of visual scores compared to baseline. Regression of white spot lesions was found out to be 27.9% after four weeks, 65.6% after eight weeks and 73.8% after twelve weeks. This data is in accordance with a study conducted by Bailey et al., who found regression of 72% after intervention period of 12 weeks [21]. Similarly Andersson A et al., found regression in 55% in randomized controlled trial in 26 subjects [10]. Uysal et al., found that there was reduced demineralization when ACP was incorporated in the orthodontic composite [8]. Mohanty P et al., carried out an in-vitro study to assess the effects of Novamin® on enamel demineralization around the orthodontic brackets and revealed that manual brushing with Novamin® containing remineralization toothpaste showed significant remineralizing potential in inhibition of artificial enamel sub-surface lesion around bracket after 10 days of remineralization phase [22]. Similarly, Jayarajan J et al., carried out an in vitro study to find out the remineralizing efficacy of CPP-ACP and casein phosphopeptide amorphous calcium phosphate fluoride (CPP-ACPF) and revealed that both had showed significantly higher amount of remineralization than artificial saliva [23]. Various similar studies are compared in [Table/Fig-7].

Thus all these clinical trials also retrieved clinical evidence that daily applications of the remineralizing cream could reverse the severity and visual appearance of WSL more effectively in postorthodontic patients.

Author (Year)	Type of study	Remineralizing agent	Results
Shetty S et al., (2014) [24]	In-vitro	Casein PhosphoPeptide Amorphous Calcium Phosphate (CPP-ACP) [GC Tooth Mousse], Casein PhosphoPeptide Amorphous Calcium Phosphate with Fluoride [CPP-ACPF] [GC Tooth Mousse Plus], Sodium Fluoride [Phos-Flur].	Enamel remineralization.
Mohanty P et al., (2014) [22]	In-vitro	Novamin® (Calcium Sodium Phosphosilicate).	Significant remineralizing potential in inhibition of artificial enamel sub-surface lesion around bracket after 10 days of remineralization phase.
Mehta R et al., (2013) [25]	In-vitro	Casein PhosphoPeptide-Amorphous Calcium Phosphate (CPP-ACP), Casein Phospho Peptide-Amorphous Calcium Fluoride Phosphate (CPP-ACFP).	Remineralization of the artificial enamel white spot lesion.
Patil N et al., (2013) [26]	In-vitro	Casein PhosphoPeptide-Amorphous Calcium Phosphate (CPP-ACP), Casein PhosphoPeptide-Amorphous Calcium Phosphate Fluoride (CPP-ACPF), and Tricalcium Phosphate Fluoride (TCP-F).	Remineralization.
Jayarajan J et al., (2011) [23]	In-vitro	Casein PhosphoPeptide-Amorphous Calcium Phosphate (CPP-ACP) and Casein PhosphoPeptideAmorphous Calcium Phosphate Fluoride (CPP-ACPF).	Remineralization.
Uysal T et al., (2010) [8]	In-vivo	Amorphous Calcium Phosphate-containing orthodontic composite for bonding orthodontic brackets.	Successfully inhibited demineralization.
Poggio C et al., (2009) [27]	In-vitro	CPP–ACP paste (Tooth Mousse).	Protective effect on enamel demineralization.

[Table/Fig-7]: Comparison of various similar studies

LIMITATION

Limitation of the present study is that subjects in 8-16 years age group were evaluated; further studies with patients undergoing orthodontic therapy in various age groups should be compared to draw further conclusions. However further studies are needed to investigate and clarify the clinical role of the calcium-based remineralization systems and regarding the application of CPP-ACP in post-orthodontic patients of different age groups in different population groups.

Present study prepared the data on occurrence of white spot lesions on different locations of tooth surfaces in children with and without orthodontic therapy and excellently proved the success of CPP-ACP cream in treatment of post-orthodontic white spot lesions in age group of 8 to 16 years old children. Hence CPP-ACP therapy for minimum of twelve weeks is highly recommended to implicate clinically as post-orthodontic treatment need in management of smooth surface white spot lesions on teeth undergoing fixed orthodontic therapy according to the present study.

CONCLUSION

The present study concludes that marked difference in occurrence of smooth surface white spot lesions in permanent molars with and without orthodontic therapy showed definite high susceptibility of patients with fixed orthodontic therapy to white spot lesions. CPP-ACP therapy minimum for twelve weeks is highly recommended as post-orthodontic treatment need in management of smooth surface white spot lesions on teeth undergoing fixed orthodontic therapy according to the present study.

REFERENCES

[1] Khan M, Fida M. White spot lesions in orthodontic patients part I: frequency and pattern of distribution. *Journal of Khyber College of Dentistry*. 2010;1(1):20-24.
[2] Bishara SE and Ostby AW. White spot lesions: formation, prevention and treatment. *Semin Orthod*. 2008; 14(3):174-82.
[3] International Caries Detection and Assessment System Coordinating Committee. Available at: https://www.icdas.org.
[4] Mizrahi E. Surface distribution of enamel opacities following orthodontic treatment. *Am J Ortho Dentofac Orthop*. 1983; 84:323–31.
[5] Ekstrand KR, Ricketts DN and Kidd EA. Reproducibility and accuracy of three methods for assessment of demineralization depth on the occlusal surface: an in vitro examination. *Caries Res*. 1997; 31(3):224-31.
[6] Kanthathas K, Willmot DR and Benson PE. Differentiation of developmental and post-orthodontic white lesions using image analysis. *Euro J Orthod*. 2005; 27:167-72.
[7] Benson PE, Parkin N, Millett DT, Dyer FE, Vine S, Shah A. Fluorides for the prevention of white spots on teeth during fixed brace treatment. *Cochrane Database Syst Rev*. 2004; 3:CD003809.
[8] Uysal T, Amasyali M, Ozcan S, Koyuturk AE, Akyol M, Sagdic D. In vivo effects of amorphous calcium phosphate-containing orthodontic composite on enamel demineralization around orthodontic brackets. *Aust Dent J*. 2010; 55: 285-91.
[9] Willmot DR. White lesions after orthodontic treatment: does low fluoride make a difference? *J Orthod*. 2004;31(3):235-42.
[10] Andersson A, Skold-Larsson K, Hallgren A, Petersson LG, Twetman S. Effect of a dental cream containing amorphous calcium phosphate complexes on white spot lesion regression assessed by laser fluorescence. *Oral Health Prev Dent*. 2007; 5:229-33.
[11] Al Maaitah EF, Adeyemi AA, Higham SM, Pender N, Harrison JE. Factors affecting demineralization during orthodontic treatment: a post-hoc analysis of RCT recruits. *AJODO*. 2011;139(2):181-91.
[12] Sagarika N, Suchindran S, Loganathan S, Gopikrishna V. Prevalence of white spot lesion in a section of Indian population undergoing fixed orthodontic treatment: an in vivo assessment using the visual International Caries Detection and Assessment System II criteria. *Journal of Conservative Dentistry*. 2012; 15(2):104-08.

[13] Chang HS, Walsh LJ, Freer TJ. Enamel demineralization during orthodontic treatment, aetiology and prevention. *Aust Dent J*. 1997; 42(5):322-27.
[14] Chesters RK, Pitts NB, Matuliene G, Kvedariene A, Huntington E, Bendinskaite R et al. An abbreviated caries clinical trial design validated over 24 months. *J Dent Res*. 2002; 81: 637-40.
[15] Mahmoud E, Niko B and Ruf S. White-spot lesions during multibracket appliance treatment: a challenge for clinical excellence. *Am J Orthod Dentofac Orthop*. 2011; 140: e17-24.
[16] Sandvik K, Hadler-Olsen S, El-Agroudi M, Ogaard B. Caries and white spot lesions in orthodontically treated adolescents-a prospective study. *Eur J Orthod*. 2006; 28:e258.
[17] Lovrov S, Hertrich K, Hirschfelder U. Enamel demineralization during fixed orthodontic treatment—incidence and correlation to various oral-hygiene parameters. *J Orofac Orthop*. 2007; 68:353-63.
[18] Hawkins R, Locker D, Noble J, Kay EJ. Prevention, part 7: professionally applied topical fluorides for caries prevention. *Br Dent J*. 2003; 195(6):313-17.
[19] Clark, Sarah Elizabeth. Remineralization effectiveness of MI paste plus - a clinical pilot study. MS (Master of Science) thesis, University of Iowa, 2011. http://ir.uiowa.edu/etd/939.
[20] Pukallus ML, Plonka KA, Holcombe TF, Barnett AG, Walsh LJ, Seow WK. A randomized controlled trial of a 10 Percent CPP-ACP cream to reduce mutans Sstreptococci colonization. *Pediatric Dentistry*. 2013; 35(7):550-55.
[21] Bailey DL, Adams GG, Tsao CE, Hyslop A, Escobar K, Manton DJ et al. Regression of post-orthodontic lesions by a remineralizing cream. *J Dent Res*. 2009; 88(12):1148-153.
[22] Mohanty P, Padmanabhan S, Chitharanjan AB. An in vitro evaluation of remineralization potential of novamin on artificial enamel sub-surface lesions around orthodontic brackets using energy dispersive X-ray analysis (Edx). *Journal of Clinical and Diagnostic Research*. 2014; 8(11):88-91.
[23] Jayarajan J, Janardhanam P, Jayakumar PD. Efficacy of CPP-ACP and CPP-ACPF on enamel remineralization - an in vitro study using scanning electron microscope and DIAGNOdent®. *Indian J Dent Res*. 2011; 22:77-82.
[24] Shetty S, Hegde MN, Bopanna TP. Enamel remineralization assessment after treatment with three different remineralizing agents using surface microhardness: an in vitro study. *J Conserv Dent*. 2014; 17(1):49–52.
[25] Mehta R, Nandlal B, Prashanth S. Comparative evaluation of remineralization potential of casein phosphopeptide amorphous calcium phosphate and casein phosphopeptide amorphous calcium phosphate fluoride on artificial enamel white spot lesion: an in vitro light fluorescence study. *Indian Journal of Dental Research*. 2013; 24(6): 681- 89.
[26] Patil N, Choudhari S, Kulkarni S, Joshi SR. Comparative evaluation of remineralizing potential of three agents on artificially demineralized human enamel: an in vitro study. *JCD*. 2013;16(2):116-20.
[27] Poggio C, Lombardini M, Dagna A, Chiesa M, Bianchi S. Protective effect on enamel demineralization of a CPP–ACP paste: an AFM in vitro study. *Journal of Dentistry* 2009; 37:949-54.

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