"EFFECT OF REMINERALISATION AND RESIN INFILTRATION ON THE SURFACE HARDNESS AND EVALUATION OF MICROSHEAR BOND STRENGTH USING COMPOSITE RESIN ON THE REMINERALISED DENTIN : AN IN-VITRO STUDY "

DISSERTATION

Submitted to

BABU BANARASI DAS UNIVERSITY,

LUCKNOW, UTTAR PRADESH

In the partial fulfilment of the requirements for the degree

of

MASTER OF DENTAL SURGERY

In

CONSERVATIVE DENTISTRY & ENDODONTICS

By

Dr. RECHU RAJU

Under the guidance of

Dr. TANU TEWARI

Reader

Department of Conservative Dentistry & Endodontics

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES, LUCKNOW

(Faculty of Babu Banarasi Das University)

BATCH: 2020-2023 Enrollment No.: 12003220320

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DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled **" EFFECT OF REMINERALISATION AND RESIN INFILTRATION ON THE SURFACE HARDNESS AND EVALUATION OF MICROSHEAR BOND STRENGTH USING COMPOSITE RESIN ON THE REMINERALISED DENTIN : AN IN-VITRO STUDY "** is a bonafide and genuine research work carried out by me under the guidance of **Dr. TANU TEWARI,** Reader, and **Dr. PRAGYAN PALIWAL ,** Senior Lecturer as Co-Guide in Department of Conservative Dentistry & Endodontics, Babu Banarasi Das College of Dental Sciences, Babu Banarasi Das University, Lucknow, Uttar Pradesh.

Date: 14/02/2023

Place: Lucknow

Dr. RECHU RAJU

CERTIFICATE BY THE GUIDE

This is to certify that the dissertation entitled **"EFFECT OF REMINERALISATION AND RESIN INFILTRATION ON THE SURFACE HARDNESS AND EVALUATION OF MICROSHEAR BOND STRENGTH USING COMPOSITE RESIN ON THE REMINERALISED DENTIN : AN IN-VITRO STUDY "** is a bonafide work done by **Dr. RECHU RAJU**, under my direct supervision and guidance in partial fulfilment of the requirement for the degree of MDS in Department of Conservative Dentistry & Endodontics.

Date: 14/02/2023

Dr. TANU TEWARI
Reader
Department of Conservative
Dentistry & Endodontics.
BBDCODS, BBDU
Lucknow

CERTIFICATE BY THE CO-GUIDE

This is to certify that the dissertation entitled **" EFFECT OF REMINERALISATION AND RESIN INFILTRATION ON THE SURFACE HARDNESS AND EVALUATION OF MICROSHEAR BOND STRENGTH USING COMPOSITE RESIN ON THE REMINERALISED DENTIN : AN IN-VITRO STUDY "** is a bonafide work done by **Dr. Rechu Raju** under the supervision of **Dr. Pragyan Paliwal** as Co-Guide, Senior Lecturer, Department of Conservative Dentistry & Endodontics., Babu Banarasi Das College Of Dental Sciences, Babu Banarasi Das University, Lucknow, Uttar Pradesh.

Pragyan Paliwal

Dr. PRAGYAN PALIWAL
Senior Lecturer
Department of Conservative
Dentistry & Endodontics
BBDCODS, BBDU
Lucknow

ENDORSEMENT BY THE HOD / HEAD OF THE INSTITUTION

This is to certify that the dissertation entitled **" EFFECT OF REMINERALISATION AND RESIN INFILTRATION ON THE SURFACE HARDNESS AND EVALUATION OF MICROSHEAR BOND STRENGTH USING COMPOSITE RESIN ON THE REMINERALISED DENTIN : AN IN-VITRO STUDY "** is a bonafide work done by **Dr. Rechu Raju** under the supervision of **Dr Tanu Tewari**, Reader, Department of Conservative Dentistry & Endodontics , Babu Banarasi Das College Of Dental Sciences, Babu Banarasi Das University, Lucknow, Uttar Pradesh.

Dr. PRAVEEN SINGH SAMANT
Professor & Head
Department of Conservative Dentistry
& Endodontics
BBDCODS, BBDU
Lucknow

Dr. PUNEET AHUJA
Principal
BBDCODS, BBDU
Lucknow

PRINCIPAL
Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)
BBD City, Faizabad Road, Lucknow-226028

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Place: Lucknow.

Dr. RECHU RAJU

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TO GOD ALONE BE THE GLORY

Dr. Rechu Raju

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ABSTRACT

ABSTRACT

Non-Carious Cervical Lesions (NCCL) are characterized by loss of hard dental tissue near the CEJ. Demineralization occuring in NCCLs and long term remineralizing treatment alter the structure of dentin. Thus, it is a unique challenge for the clinician to successfully restore such demineralized and partially remineralized NCCLs..

The purpose of this study was to assess the effect of surface micro hardness of demineralised dentin after remineralising with MI Paste Plus, Remin Pro and Icon Infiltrant and to evaluate the microshear bond strength (μSBS) of composite to remineralised dentin.A total of 50 extracted premolar teeth were taken for this study. All the teeth samples were decoronated and sectioned into buccal & lingual halves making the total sample size to be 100.Then the Dentin was exposed at the cervical aspect. 80 tooth samples were demineralised using citric acid solution and divided into 5 different groups according to the remineralising agents used, so that each group contained 20 samples. Then 10 samples from each group were subjected to analyse their surface hardness after 15 days using Vickers hardness tester. The remaining 10 samples from each group were bonded with composite using Etch & Rinse technique & μSBS of the bonded samples were evaluated using Universal testing machine.

Sample size estimation was done by using GPower software (version 3.0). Sample size was estimated for ANOVA. The significance was set at $p < 0.05$. Failure modes were analyzed using chi-square test.

The results showed that Remineralization and Resin Infiltration efficiently increased the surface hardness of demineralised dentin. Also Resin infiltration groups showed significant higher bond strength values to composite than the Positive control (Sound dentin) group, Negative control (demineralised dentin) group & remineralised dentin group. The present In-vitro study indicates that composite forms a weaker bond with remineralized dentin due to the interference of acid resistant hypermineralized zone

INTRODUCTION

INTRODUCTION

Dental caries is caused by the interaction of microbial factors, diet, host factors, and time. When there is an imbalance in the de/remineralization process on the tooth surface, demineralization takes over and the first carious lesions appear (1). The canges caused by initial demineralisation are mineral loss from the body of the lesion, an increase in intercrystalline spaces, and a decrease in the microhardness of the subsurface, while the surface remains relatively highly mineralized (2,3). The pores in the enamel give it its distinctive whitish appearance and help acids and dissolved minerals to diffuse through the enamel leading to White spot lesions(4,5).

White spot lesions (WSLs) are the first clinical sign of dental demineralization. The progression of the lesion can be prevented during this phase by shifting the reactions toward remineralization. However, if demineralization continues, it leads to the formation of dental caries (6). Non-invasive remineralization of lesions may simply prevent the progression of incipient carious lesions, eliminating the need for invasive and costly restorative procedures (7). Hence, currently a lot of emphasis is laid on caries prevention and minially invasive dentistry (8).

Initial enamel carious lesions can regress or even disappear with remineralization treatment (1,2,3). Non-cavitated caries lesions can be repaired by saliva, if there is control of diet and plaque. Additionally, treatment with fluoride is an alternative noninvasive method used for remineralizing carious processes, as the fluoride improves the acid resistance of enamel and interferes with bacterial metabolism and enzymatic processes. Thus, less invasive treatments have long been adopted to control the progression of initial enamel carious lesions (9,10).

A breakthrough in medicine and preventive dentistry has extended the retention period of dentition in the oral cavity. This has also steadily increased the prevalence of non-carious cervical lesions (NCCLs). Loss of hard tissue at the cementoenamel junction (CEJ) is a condition commonly encountered in clinical practice. The prevalence rate of cervical lesions makes about 18% of the permanent teeth defects. NCCLs are caused by tooth structure loss caused by a variety of etiologies such as chemical agents, occlusal stress, mechanical factors,

or a combination of all of these. They are the result of increased speed and level of demineralization, which becomes dominant over time (11). Erosion, abrasion, and abfraction occur as a result of the interaction of several such factors (12,13) .

Dentinal hypersensitivity, plaque retention, caries incidence, structural integrity loss, and pulp vitality all complicate NCCL treatment. Non-carious cervical lesions present in a variety of forms. They vary from shallow grooves to broad dished-out lesions to large wedge-shaped defects.

The loss of cervical tooth structure is compensated by restoring the defect to meet aesthetic and functional demands. NCCLs are restored with glass ionomer cement (GIC) or composite resins. Composites are usually preferred when it comes to aesthetics (14). However, the durability of composite restoration is determined by the substrate to which it is bonded (15). Demineralization in NCCLs and long-term remineralizing treatment change the structure of dentin. Thus, it is a unique challenge for the clinician to successfully restore such demineralized and partially remineralized NCCLs (16).

Remineralization is defined as the process whereby calcium and phosphate ions are supplied from a source external to the tooth to promote ion deposition into the crystal voids formed in the demineralized enamel to produce net mineral gain (17). The availability of calcium and phosphate ions is the limiting factor for net remineralization to occur (18) . Many commercial products, such as fluoride compounds, have been introduced to date for remineralization of White Spot Lesions (WSLs). Despite the fact that the efficacy of fluoride for remineralization of WSLs has previously been established (19) , recent studies have called into question the use of fluoride for treatment of these lesions (20) . Fluoride uptake by the superficial layer of WSLs may reduce calcium and phosphate uptake, resulting in inadequate remineralization of deeper layers . Furthermore, applying fluoride alone does not improve the appearance of these lesions (21). As a result, several other compounds have been introduced for the remineralization of early caries.

Casein phosphopeptide amorphous calcium phosphate fluoride (CPP-ACPF), marketed as MI-Paste Plus, contains calcium, phosphate and fluoride stabilized by casein phosphopeptide. Dental remineralization technology has undergone advancements with combinations such as fluoride, hydroxyapatite (HA), and xylitol, ingredients used in Remin Pro (VOCO GmbH, Cuxhaven, Germany), which is believed to be associated with calcium in aqueous solution to

inhibit the dissolution of calcium and/or phosphate ions from enamel and to act as a carrier of calcium required for enamel remineralization. HA is a major mineral component found in the dental matrix and is what gives teeth its rigidity.

The resin infiltration technique is a minimally invasive alternative treatment for non-cavitated carious lesions. The goal of low viscosity light-activated resin infiltration is to seal the carious lesion microporosities and form a diffusion barrier within the lesion, preventing acids from penetrating and depositing material on the enamel surface (4). Several studies have demonstrated the efficacy of this technique (4,22,23). The resin matrix has the ability to strengthen the enamel structure by increasing surface microhardness and preventing enamel surface breakdown (9,10)

Nowadays, the request for esthetic restorations has been dramatically increased. Various adhesive systems are available to attach the restorations to dental tissue. The efficiency and quality of an adhesive systems is important in producing a stable bond between the composite and dental structure. Shear bond strength (SBS) is an important factor for measuring the efficiency and quality of adhesive systems. Inadequate SBS may cause early failure of restoration in the face of minimum masticatory forces . However, the literature regarding the bonding of composites to partially remineralized NCCLs is sparse (16).

Finding the best protocol for remineralization of incipient enamel lesions is a crucial step toward achieving preventive dentistry goals with minimal invasion. Thus the purpose of this study was to evaluate and compare the effects of MI-Paste Plus, Remin Pro, and ICON on the surface hardness of incipient enamel lesions and to evaluate the microshear bond strength (μSBS) of composite resin bonded to demineralized dentin which had been remineralized with MI-Paste Plus, Remin Pro, and ICON resin Infiltrant. The null hypothesis for this study stated that the three materials would not differ significantly in their effect on the surface hardness and microshear bond strength on remineralised dentin

AIM & OBJECTIVES

AIM OF THE STUDY:

The aim of the study was to assess the effect of surface micro hardness of demineralised dentin after remineralising with MI Paste Plus, Remin Pro and Icon Infiltrant and to evaluate the microshear bond strength (μSBS) of composite to remineralised dentin.

OBJECTIVES

1. To assess the effect of surface micro hardness of demineralised dentin after remineralising with MI Paste Plus using Vickers microhardness test.

2. To assess the effect of surface micro hardness of demineralised dentin after remineralising with Remin Pro using Vickers microhardness test.

3. To assess the effect of surface micro hardness of demineralised dentin after Resin Infiltration with Icon Infiltrant using Vickers microhardness test.

4. To compare and evaluate the microshear bond strength (μSBS) of the composite resin to dentin remineralised using MI Paste under the Universal Testing Machine.

5. To compare and evaluate the microshear bond strength (μSBS) of the composite resin to dentin remineralised using Remin Pro under the Universal Testing Machine

6. To compare and evaluate the microshear bond strength (μSBS) of the composite resin to dentin after Icon resin Infiltration under the Universal Testing Machine

7. To evaluate and compare the effect of the tested remineralising agents amongst each other.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

1) **H. Sano, T. Takatsu, B. Ciucchi et al in 1995** conducted an in-vitro study to compare the Tensile Properties of Resin-infiltrated Demineralized Human Dentin. The result obtained was that some adhesive resins, after infiltrating demineralized dentin, can restore and even exceed the ultimate tensile strength of mineralized dentin. They concluded that All Bond 2 resin infiltrated demineralised dentin has higher ultimate tensile strength than Scotchbond Multi-Purpose, Clearfil Photobond, Clearfil Liner Bond and Superbond C&B resin infiltrated demineralised dentin

2) **Franklin R. Tay, Share-Moon Kwong, Anut Itthagarun et al in 2000** conducted an invitro study to compare the Bonding of a Self-Etching Primer to Noncarious Cervicai Sclerotic Dentin: Interfacial Ultrastructure and Microtensiie Bond Strength Evaiuation. They concluded that overall bond strength of compsite to natural sclerotic dentin was about 20% lower than sound cervical dentin.

3) **Anderson T. Hara, Celso S. Queiroz et al , in 2004** conducted an invitro study to compare the influence of the mineral content and morphological pattern of artificial root caries lesion on composite resin bond strength. They found that Remineralised teeth samples had the least micro tensile bond strength values compared to the Demineralised and sound tooth samples. They concluded that Remineralised teeth samples had microhardness greater than demineralised teeth samples but less than Sound teeth Samples.

4) **Manuel Toledano, Raquel Osorio, Estrella Osorio, Carlo Prati, Ricardo M. Carvalho et al in 2004** conducted an invitro study to determine if superficial or deep dentine microhardness (MH) is affected by different chemical dentine pre-treatments performed for resin bonding. The results obtained was that Resin infiltration increased KHN but did not recover the initial micro-hardness values in both Superficial Dentin and Deep Dentin. Hence it was concluded that Treating dentine

with either H3PO4 or NaOCl caused marked reduction of its surface hardness and subsequent resin infiltration was not capable to restore it

5) **Yahya Orcun Zorbaa , Ali Erdemirb , Ertugrul Ercanc et al in 2010** conducted an invitro study to compare the effects of three different desensitizing agents on the shear bond strength of composite resin bonding agents. The Result showed that the bond strength to desensitized dentin was lower than that of sound dentin. Also that the use of different desensitizing agents does not affect the shear bond strength of various adhesive systems used to bond resin composite to dentin.

6) **Masatoshi Nakajima, Sitthikorn Kunawarote, Taweesak Prasansuttiporn et al in 2011**. In their study, Bonding to caries-affected dentin, they found that Self-etch systems have exhibited lower bond strengths to caries-affected dentin than normal dentin, and their hybrid layers in caries-affected dentin are also thicker than those of normal dentin but absolutely thinner than those of etch and rinse system. They concluded that when the adhesive interface of caries affected dentin is exposed to oral environment, the poor quality of the hybrid layer of caries-affected dentin would compromise the longevity of the composite restoration due to hydrolysis of the resin and collagen fibrils

7) **Nadia Malek Taher , Haifa Abdulrahman Alkhamis , Sarah Mesha'l Dowaidi et al in 2012** conducted an In vitro study to nvestigate the effect of a resin infiltrant on the surface microhardness and roughness of healthy enamel and to compare it with a fissure sealant. They concluded that enamel surfaces treated by infiltrant showed significantly higher VHN values than those treated with fissure sealant.

8) **Hossam Elzuhery, Ola Ibrahim Fahmy, Inas AbdElmoneum Elghandou et al in 2013** conducted an invitro study to compare the Bond strength and morphological interface of self-etching adhesives to demineralized and remineralized enamel. The found that the bond strength to enamel remineralized using Fluocal mouthwash or Tooth Mousse was higher than the normal and natural saliva-remineralized enamel. They concluded that etching improved the bond strengths of self-etching bonding agents especially if the enamel was remineralized

9) **Pui Ling Chay, David J. Manton & Joseph E.A. Palamara et al in 2013** conducted an invitro study to compare the effect of resin infiltration and oxidative pre-treatment on microshear bond strength of resin composite to hypomineralised enamel. They found that Increased bond strength of resin composite to hypomineralised enamel was obtained by pre-treatment of hypomineralised enamel specimens with 5.25% sodium hypochlorite with or without subsequent resin infiltration.

10) **S. Paris , F. Schwendicke , S. Seddig , W.-D. Mu¨ ller , C. Do¨ rfer , H. Meyer-Lueckel et al in 2013** conducted an Invitro study to evaluate the influence of infiltrant composition and application frequency on micro hardness and lesion progression after resin infiltration of artificial enamel lesions. They concluded that Resin infiltration significantly improves micro-hardness and demineralisation resistance of enamel lesions; these effects are significantly enhanced if resins are applied twice.

11) **Mahmoud Bahari, Siavash Savadi Oskoee, Soodabeh Kimyai et al in 2014** conducted an invitro study to compare the Effect of Casein Phosphopeptide-amorphous Calcium Phosphate Treatment on Microtensile Bond Strength to Carious Affected Dentin Using Two Adhesive Strategies. The results obtained showed Bond strength of Etch & Rinse adhesive systems to Normal Dentin was significantly higher than that of Self etch adhesive system. They concluded that the Bond strength of etch & rinse adhesive system to Remineralised dentin was lesser than that to Caries affected Dentin and Normal Dentin.

12) **Bruna Chrispim Dos REIS , Ana Júlia Faria De LACERDA & Taciana Marco Ferraz CANEPELE et al in 2016** conducted an invitro study to compare the bond strength of composite resin to demineralized enamel, exposed to remineralization and subjected to caries infiltration. They found that the Single Bond Universal (etch & rinse system) had statistically significant higher values of bond strength, superior when compared to Clearfil S3 Bond (self etch system). The surface infiltrated with an infiltrating resin (Icon) did not negatively affect the bond strength between resin composite and enamel. The demineralized and remineralized groups with sodium

fluoride and artificial saliva presented statistically lower results when compared to the other groups

13) **Elif Beril Gurdogan, Didem Ozdem_Ir-Ozenen, Nuket Sandalli et al in 2016** conducted an invitro study to evaluate the surface roughness via atomic force microscopy (AFM) as well as to evaluate the microhardness values of Icon in comparison with sound and demineralized enamel in a large subject. The result obtained was that the infiltrant IconVR had a significantly rougher surface than the control group. They concluded that microhardness of IconVR was found to be similar to healthy enamel and it was observed that Icon increased the microhardness of demineralized enamel

14) **Deepesh Prajapati, Rashmi Nayak, Deepika Pai, Nagraj Upadhya, Vipin K Bhaskar, Pujan Kamath et al in 2017** conducted an invitro study to evaluate the effectiveness of resin infiltration on artificial caries lesion by assessing the depth of resin penetration and the change in microhardness of lesion post infiltration. They found that the Penetration depth of the resin infiltrant was deep enough to render beneficial effects, while a significant increase in microhardness was observed post resin infiltration.

15) **Sidika Aynur Horuztepe, Meserret Baseren et al in 2017** conducted an Invitro study to investigate both the effect of resin infiltration on the color and microhardness of white spot lesions (WSLs) that were treated with bleaching and to analyse the penetration of the infiltrant.. The result obtained was that the resin infiltration enhanced the microhardness of the WSLs regardless of whether or not a bleaching treatment was used. Using a bleaching treatment before resin infiltration resulted in color alteration, and it was also found to negatively influence the penetration of the infiltrant

16) **Jyothi Mandava , Y. Shilpa Reddy , Sirisha Kantheti et al in 2017** conducted an invitro study to compare the Microhardness and Penetration of Artificial White Spot Lesions Treated with Resin or Colloidal Silica Infiltration. The result obtained was Resin infiltration group showed significantly greater increase in microhardness compared to colloidal silica infiltration. They concluded that the Resin infiltrates

performed better in regaining the baseline microhardness and penetrating deep into the porous white spot lesions, when compared to colloidal silica infiltrates

17) **Elangovan Sivapriya, Krishnamoorthy Sridevi, Ravishankar Periasam et al in 2017** condcted an invitro study to compare the Remineralization ability of sodium fluoride on the microhardness of enamel, dentin, and dentinoenamel junction. They concluded that the Long-term repeated application of sodium fluoride (226 ppm) can improve the microhardness of demineralized dental tissues on enamel, dentin, and DEJ-axial zone, except in the DEJ-cusp tip and DEJ-center of fissure.

18) **Sneha Jeetendra, Anithakumari Rangappa, Meena Naganathan et al in 2018** conducted an invitro study to compare the Influence of dentin desensitizers on the bond Strength of Self-etch Adhesive to Dentin. The result obtained was that Dentin pretreated with desensitizing agents has lower tensile bond strength with resin composites as compared with dentin that is untreated. They also concluded that hydroxyethyl methacrylate (HEMA)-containing desensitizing agents had better bond strength compared with the fluoridecontaining agents

19) **Buhler Borges Alessandra , Abu Hasna Amjad , Guedes Nogueira Matuda Amanda , Lopes Stephanie Ribeiro , Paula Valente Pinho Mafetano Ana et al (2019)** conducted an Invitro study to access the effect of Adhesive systems over bond strength of resin-infiltrated and de/remineralized enamel.. They found that the etch-and-rinse showed significantly higher values than self-etching and resin infiltration groups showed significantly higher bond strength values than the negative control (demineralised enamel), 0.05% fluoride solution and artificial saliva groups

20) **M. Chen, J.-Z. Li, Q.-L. Zuo, C. Liu, H. Jiang, M.-Q. Du et al in 2019** conducted an Invitro study to investigate characteristics associated with this infiltrant resin and to compare the Icon infiltrant with universal Filtek Z350 and flowable Filtek Z350 resins when exposed to artificial accelerated aging. The result obtained was that the ICON infiltrant displayed reduced microhardness when compared to universal Z350 and flowable Z350 under accelerated aging. They also concluded that e absorption peaks of the chemical bonds were significantly altered after the accelerated aging process

21) **LiLing Wu, Kun Geng and QingPing Gao et al in 2019** conducted an invitro study to compare Effects of different anti-caries agents on microhardness and superficial microstructure of irradiated permanent dentin. The result obtained was that the Infiltration resin, CPP-ACP, fluoride and their pairwise combination can effectively prevent radiation-dentindestruction. Among them, infiltration resin with CPP-ACP, infiltration resin with fluoride, CPP-ACP with fluoride, and infiltration resin have the most protective effects on irradiation-dentin-destructions

22) **Krithi Banka, Vidhya Sampath , Mahalaxmi Sekar et al (2020)** conducted an Invitro study to access the Microshear bond strength of composite resin to demineralized dentin after remineralization with sodium fluoride, CPP-ACP and NovaMin containing dentifrices.. They concluded that Composite forms a weaker bond to remineralized dentin. Self-etch bonding system is capable of achieving acceptable bond strength to dentin remineralized with NaF and NovaMin

23) **Alagha Ebaa et al (2020)** conducted an Invitro study to access the Effect of using Different Remineralizing Agents on Micro-shear Bond Strength of Nanohybrid Composite Resin. He concluded that Fluorohydroxyapatite and nanohydroxyapatite had a positive effect on micro-SBS to dentin, but sodium fluoride had a negative effect. Fluorohydroxyapatite had the highest bond strength mean values followed by nanohydroxyapatite, while the lowest value was the sodium fluoride group

24) **M. Rabee, K. Nomaan, A. Abdelhady et al in 2020** conducted an invitro study to compare the Effect Of Remineralizing Agents On Bond Strength Of Resin Composite To Dentin. The result showed that floro-hydroxyapatite group had the highest bond strength mean value followed by nanohydroxyapatite , while the lowest bond strength was in sodium fluoride group. They also concluded thatRemineralizing agents can be used successfully under resin composite restorative material.

25) **Marina Simunovic Anicic , Cecilia Goracci , Jelena Juloski et al in 2020** conducted an invitro study to compare the Influence of Resin Infiltration Pretreatment on Orthodontic Bonding to Demineralized Human Enamel. The result

obtained showed that the treatment with a resin infiltrant on Demineralised Enamel does not impair the shear bond strength when compared to Sound enamel, although it does produce a significantly higher strength when combined with Assure PLUS

26) **Deepti Rao M and R Nageswar Rao et al in 2020** conducted an invitro study to compare the Microtensile bond strength of Resin composite to Normal dentin and Caries - affected dentin with total etch and self-etch bonding adhesives before and after thermocycling. The results obtained was total-etch has a higher bond strength compared to self-etch. Bond strengths are higher to normal dentin than caries-affected dentin. Thermocycling lowers the bond strength values of all the groups.

27) **Behrouzi Parastou , Heshmat Haleh , Ganjkar Maryam Hoorizad, Farnaz Tabatabaei Seyedeh, Kharazifard Mohammad Javad et al (2020)** conducted an Invitro study to access the Effect of Two Methods of Remineralization and Resin Infiltration on Surface Hardness of Artificially Induced Enamel Lesions.. They concluded that MI-Paste Plus and Remin Pro can efficiently increase the hardness of incipient enamel lesions. but this increase was not significant in ICON group

28) **Sahiti J Sai, Krishna N Vamsee, Prasad S Datta, Kumar C Sunil, Kumar S Sunil, Babu K S Chandra et al (2020)** conducted an Invitro study to access the Comparative evaluation of enamel microhardness after using two different remineralizing agents on artificially demineralized human enamel. They concluded that both Remin Pro and Clinpro are equally effective as remineralizing agents. Although Remin Pro tended to yield a higher microhardness, no significant differences were observed between the two agents

29) **El Meligy Omar Abd El Sade, Alamoudi Najlaa Mohammed, Ibrahim Shimaa Tag Eldin , Felemban Osama Mahmood , Al-Tuwirqi Amani Ahmed et al (2020)** conducted an Invitro study to access the Effect of resin infiltration application on early proximal caries lesions.. They concluded that Resin Infiltration application on proximal incipient caries increased surface roughness and hardness significantly

30) **Himabindu Buddula[1], Naga Sribala Gantha, Manoj Kumar Mallela, Srinivas Nallanchakrava, Karunakar Parupalli, Sanjana Reddy Cheruku et al in 2020** conducted an in vitro study to to compare microhardness of initial enamel caries using two agents (Icon and CPP-ACP Plus). The result obtained was Resin infiltration technique showed enhancement in the microhardness of initial enamel caries lesions compared to CPP-ACP plus application

31) **Steffi N Dhillon , Anshula N Deshpande , Chirag Macwan , Kinjal S Patel , Yash S Shah , Aishwarya A Jain et al in 2020** conducted an Invitro study to compare the microhardness and enamel solubility (ES) of the treated surface enamel with resin infiltrant, fluoride varnish, and casein phosphopeptide-amorphous calcium phosphate. The results showed that all agents used in study remineralized initial carious lesion. It was also concluded that Fluoride varnish has the highest microhardness and showed least enamel solubility compared to other remineralizing agents.

32) **Hema Pulidindi, Jyothi Mandava, Roopesh Borugadda et al in 2021** conducted an invitro study to compare the Effect of remineralizing agents on resin-dentin bond durability of adhesive restorations. The result obtained was that immediate bond strength values were significantly high for the control group and low for the MI paste plus group

33) **Menna El Ghamrawy, Mohamed Nasser, Khaled Aly Nour et al in 2021** conducted an invitro study to compare the effect of silver diamine fluoride on bond strength of self-etch adhesives to demineralized dentin. The result obtained was the bond strength of resin composite to either sound or demineralized dentin is negatively affected by the application of SDF prior to the use of universal adhesives in self-etch mode

34) **Hend S. Ahmed, Nermeen Kamal Hamza and Hossam A. Alhussiny et al in 2021** conducted an invitro study to compare the effect of different surface pretreatment protocols on the micro-shear bond strength of resin composite to resin infiltrated demineralized enamel . The result obtained was highest μSBS mean values were in

sound enamel followed by Icon resin infiltrated demineralised enamel group,. While demineralised enamel group yielded lowest μSBS values. They concluded that surface pretreatment is mandatory to improve bonding to resin infiltrated enamel

35) **Nishita Rana, Namita Singh, Shaila, Abi. M. Thomas & Rajan Jairath et al in 2021** conducted an Invitro study to compare and evaluate the penetration depth and enamel microhardness of Resin Infiltrant, CPP-ACPF and Novamin on artificial demineralized enamel surface after orthodontic banding. The result obtained was Resin Infiltrant exhibited the highest significant reduction in demineralization and increased microhardness. The surface microhardness of enamel was in order Resin Infiltrant > Novamin > CPP-ACPF > Control. They concluded that Resin Infiltrant exhibited the highest potential to impede caries and constitutes a competent aerosol free micro-invasive strategy for combating non cavitated lesions approaching the outer layer dentine which are too advanced for remineralizing agents but do not necessarily require any drilling of tooth

36) **Ebaa Ibrahim Alagha1, Mustafa Ibrahim Alagha in 2021** conducted an invitro study to compare the impact of two resin infiltration systems on microhardness of demineralized enamel before and after an acidic challenge.The result obtained was that the Icon resin infiltration and single bond universal adhesive showed significantly higher mean microhardness than negative control, but significantly lower mean microhardness than positive control. After an acidic challenge, icon resin infiltration was more successful than single bond universal total-etch adhesive system in microhardness

37) **Mohamed A. Wakwak , Nabil Abd Alhameed Abd Alsalam Alaggan , Ahmed Safwat Morsy et al in 2021** conducted an invitro study to evaluate the effect of resin infiltration material (Icon) and remineralizing agents (n-HA, CPP-ACP) on micro hardness and surface roughness of enamel white spot lesion.The results of microhardness revealed that the nanohydroxyapatite (n-HA) was provided the highest microhardness mean value, followed by Icon, then CPP ACP while the control showed the lowest micro hardness mean value.The results of surface roughness revealed that the CPP ACP group was provided the highest surface

roughness mean value, followed by Control, then nanohydroxyapatite (n-HA) and Icon showed the lowest surface roughness.They concluded that the Resin infiltration material (Icon) had a positive effect on microhardness and surface roughness of enamel white spot lesion.

38) **Shirin P. Kshirsagar, Shalini Aggarwal, Pooja R. Gupta et al in 2022** conducted an invitro study to compare the bond strengths of affected dentin, using two different remineralizing solutions with or without lasers. They found that LASER Toothmin groups showed the highest increase (311.17%) in microhardness in HV (50 g load) as compared to other groups (GC-255.11%, LASER GC 255.42%, Toothmin 250.34%). Shear bond strength of the LASER-treated Toothmin was significantly higher compared to other groups ($P = 0.0021$). They concluded that Toothmin and GC Mousse with or without LASER were found to be effective in increasing mineralization. Bond strength of Toothmin with LASER treatment was higher compared to others

39) **Reem Majeed H.J. Al-Mamoori , Aseel Haidar M.J. Al Haidar et al in 2022 conducted an Invitro study to** evaluate the impact of resin infiltration and microabrasion in the microhardness of the artificial white spot lesions at various depths. The result obtained was Microhardness values decrease as the depth of the white spot lesion increase. There was an increase in the microhardness values after the treatment with the resin infiltration and the microabrasion. They concluded that the hardness values that found among the icon group were higher than that of the microabrasion group,

40) **Aranta Avinash Chindane, Anil T. Patil, B. Sandhyarani et al in 2022** conducted an invitro study to evaluate and compare the effectiveness of CPP-ACP fluoride (CPP-ACPF), resin infiltration, and colloidal silica infiltration on surface microhardness in artificial white spot lesions in primary incisors using the Vickers microhardness testing machine. They concluded that Enamel specimens treated with resin infiltration showed a high microhardness mean value compared to CPP-ACPF and colloidal silica infiltration.

MATERIALS & METHODS

MATERIALS AND METHODS

The present in-vitro study was conducted in the Department of Conservative Dentistry and Endodontics, Babu Banarasi Das College of Dental Sciences, Lucknow in collaboration with Praj Mettallurgical Laboratory, Pune.

A total of 50 human permanent maxillary & mandibular premolar teeth were collected following the inclusion and exclusion criteria. Teeth were cleaned using ultrasonic scalers for removing any tissue remnants, plaque and calculus on the roots.

The following inclusion & exclusion criteria were set to select the teeth:

INCLUSION CRITERIA

Sound tooth structure free from

- Cracks
- Restorations
- Stains
- White spot lesions
- Caries evident by naked eye .

Tooth had been extracted for Orthodontic or Periodontal reasons

EXCLUSION CRITERIA

Tooth with

- Enamel hypoplasia or any other developmental anomalies
- Teeth with any visible or detectable caries, cracks & restorations

SAMPLE SIZE ESTIMATION

Sample size estimation was done by using GPower software (version 3.0). Sample size was estimated for ANOVA: Fixed effects, main effects and interactions

A minimum total sample size of 96 teeth were found to be sufficient for an alpha of 0.05, power of 95
%, 2.4 as effect size (assessed from a similar study). The sample size was further rounded off to
100.(20 in each group)
F tests - ANCOVA: Fixed effects, main effects and interactions
Analysis: A priori: Compute required sample size
Input: Effect size f = 0.53
α err prob = 0.05
Power (1-β err prob) = 0.95
Numerator df = 10
Number of groups = 5
Number of covariates = 1
Output: Noncentrality parameter λ = 26.9664000
Critical F = 1.9375668
Denominator df = 90
Total sample size = 96
Actual power = 0.9502725

Table A : MATERIALS AND ARMAMENTARIUM USED FOR SAMPLE PREPARATION:

S. No.	Material & Armamentarium	Manufacturer
1	Ultrasonic Scaler with tips	Coltene, Switzerland
2	Straight hand piece	Marathon, Korea
3	Micro motor (Slow Speed)	NSK, Japan
4	Diamond disc & Mandrel	Horico, Germany
5	Silicon Carbide Sand Paper	Sandcastle, India
6	Cold Cure Acrylic Resin	Dpi, India
7	Dental Operating Microscope	Labomed Prima, USA

Table B : MATERIALS AND ARMAMENTARIUM USED FOR DEMINERALISING, RINSING AND STORAGE OF THE SAMPLES :

8	Citric Acid	Cero Care Products LLP, Mumbai, India
9	Wet Mouth	ICPA Health Products Ltd., India
10	Deionized Water	Charco Chemicals, Punjab, India
11	10% Formalin	Fizmerk India Chemicals, India

Table C : MATERIALS AND ARMAMENTARIUM USED FOR REMINERALISING THE SAMPLES :

12	MI Paste Plus	Gc Recadent Tooth Mousse Plus, Tokyo, Japan
13	Remin Pro	VOCO, Germany
14	Icon Smooth Surface	DMG, German

Table D : MATERIALS AND ARMAMENTARIUM USED FOR BONDING COMPOSITE :

15	Etchant Gel S	Coltene, Switzerland
16	One Coat Bond SL	Coltene, Switzerland
17	3M Filtek Z250 XT Composite	St. Paul, Minnesota
18	Light Cure Unit	Woodpecker, China

METHODOLOGY

SAMPLE COLLECTION

Fifty freshly extracted Premolar teeth were obtained from the Department of Oral and Maxillofacial Surgery, Babu Banarasi Das College Of Dental Sciences, Lucknow. Teeth were cleaned using ultrasonic scaler for removing any tissue remnants, plaque and calculus on the roots. Later teeth were examined clinically and radiographically to follow inclusion and exclusion criteria. The extracted teeth were then stored in 10% formalin until use.

SPECIMEN PREPARATION

The 50 crowns were separated from the roots by sectioning the teeth 1 mm apical to the cementoenamel junction using a diamond disc. The crown portions were sectioned longitudinally into buccal and lingual halves (n = 100). The sectioned halves were placed in silicon rectangular blocks filled with self-cure acrylic resin, until the resin set. Dentin was exposed on the cervical one-third of the buccal and lingual surfaces of the teeth. Both surfaces were wet polished using 600-grit silicon carbide paper to obtain a flat surface. They were then inspected under a dental operating microscope to ensure that no remnants of enamel were present.

The acrylic block samples were then allotted randomly to five groups (n = 20) according to the remineralizing agents used.

Table E : GROUPWISE DISTRIBUTION OF SAMPLES

GROUPS	NUMBER OF SAMPLES	REMINERALIZING AGENT USED
Group 1	20	Sound Dentin
Group 2	20	Demineralised Dentin
Group 3	20	Remin Pro
Group 4	20	MI-Paste Plus
Group 5	20	Icon Infiltrant

Samples in group 1 (sound dentin) did not receive any further treatment while those in the rest of the groups (Group 2-5) were immersed in 1% citric acid solution (pH 2.5) for 3 days. At the end of demineralization, the samples were washed with distilled water for 30 min. Samples in group 2 (demineralized dentin) did not receive any further treatment. The samples in groups 3–5 were subjected to remineralization treatment using Mi-Paste Plus, Remin Pro and Icon resin Infiltrant according to the manufacturer's instructions.

REMINERALISATION OF SAMPLES

In the group 3, Remin Pro (VOCO GmbH, Cuxhaven, Germany) were applied on the surface of tooth samples

Paste was made into slurry by dissolving dentifrice (5 g) in water (10 ml)

↓

Pastes was then applied on the exposed dentin in 1mm thickness and brushed with the help of an toothbrush for 2 min

↓

After 3 minutes, the samples were then rinsed with deionized water for 30 seconds and dried with gentle airflow for 5 seconds

This was repeated twice a day for 15 days.

All samples were stored in artificial saliva in between the experiment

.

In the group 4, Tooth Mousse Plus (MI Paste Plus; GC Corporation, Tokyo, Japan) were applied as explained in the group 3.

RESIN INFILTRATION TECHNIQUE

Finally, in the group 5, ICON (DMG, Hamburg, Germany) were used according to the manufacturer's instructions.

The ICON-Etch was applied on the surface of samples for 2 minutes

↓

Then rinsed with water for 30 seconds

↓

Surface was then dried with gentle air spray for 5 seconds

↓

ICON-Dry was applied on the surface of samples for 30 seconds

↓

Then again dried with gentle air flow for 5 seconds

↓

One layer of ICON-Infiltrant was then applied on the surface of samples and remained for 3 minutes

↓

Excess material were removed by a cotton roll and resin was light-cured for 40 seconds using a LED light-curing unit

↓

After that, another layer of ICON- Infiltrant was applied on the surface of samples and remained for 1 minute

↓

Afterwards, the resin was light-cured for 40 seconds

ANALYSIS OF THE SAMPLE'S

1) MICROHARDNESS EVALUATION

10 samples from each group were subjected to analyse their surface hardness after 15 days using Vickers hardness tester. In each sample, three indentations were made along tooth surface. At each measurement, indentation was made using a 50 g load perpendicular to the indentation surface with a dwell time of 10 seconds. The indentations were made at a distance of 1000 micrometer from each other. The hardness values for each specimen was obtained as the average of the results for the three indentations.
Raw data were obtained and sent for statistical analysis.

2) Micro Shear Bond Strength Evaluation

The Remaining 10 samples from each group were bonded with composite resin in 1 mm increments using Etch & Rinse technique and and light cured. Then the μSBS of the bonded samples were evaluated using Universal Testing Machine. Load application was done at the rate of 1 mm/min until the composite sheared off from the dentin surface. The load at failure divided by the surface area was taken as the μSBS value of the sample (in MPa). Raw data were obtained and sent for statistical analysis.

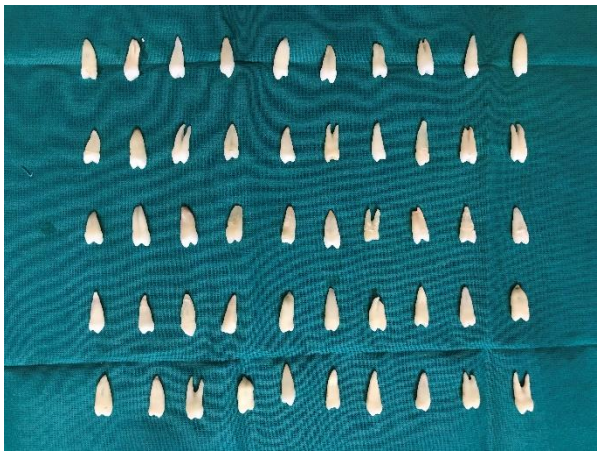


Fig 1 – Teeth Collected for study

Fig 2 – Tooth marked for sectioning

Fig 3 – Armamentarium for specimen preparation

Fig 4 – Sectioning of the teeth Samples

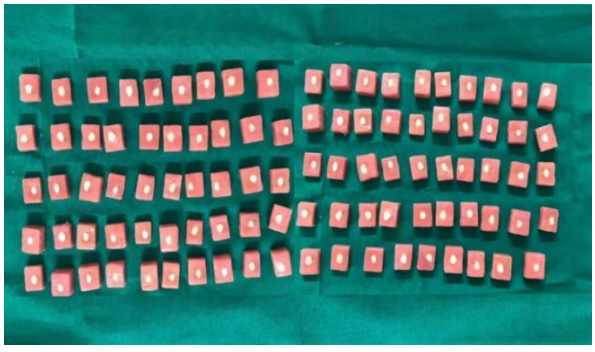


Fig 5 – Sectioned Teeth Samples mounted Samples in acrylic blocks

Fig 6 – Demineralisation of teeth using 1% Citric acid solution

Fig 7 – Deionized Water

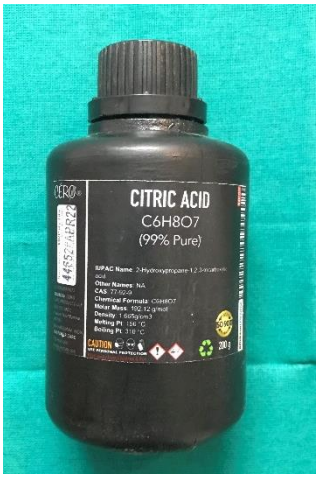


Fig 8 – Citric Acid

Fig 9 – Artificial Saliva

Fig 10 – Remin Pro

Fig 11 – MI Tooth Plus

Fig 12 – Icon Resin Infiltration Kit

Fig 13 – Bonding Agent

Fig 14 - Composite

Fig 15 - Armamentarium for Composite Restoration

Fig 16 – Application of Remin Pro

Fig 17 – Application of MI Paste Plus

Fig 18 – Application of Icon Etch

Fig 19- Application of Icon Dry

Fig 20 – Application of Icon Infiltrant

Fig 21 – Light Curing

Fig 22 – Application 37% Phosphoric Acid Etchant

Fig 23 – Application of Total Etch Adhesive

Fig 24 – Placement of Composite over teeth surface

Fig 25 – Light Curing of the Composite

Fig 26 - Vicker's Microhardness tester

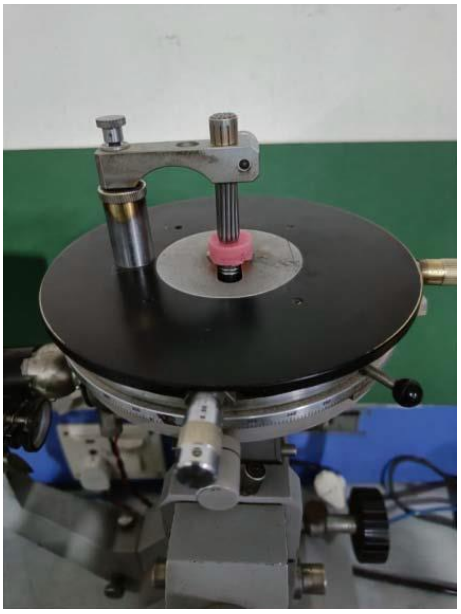


Fig 27 – Testing for Microhardness

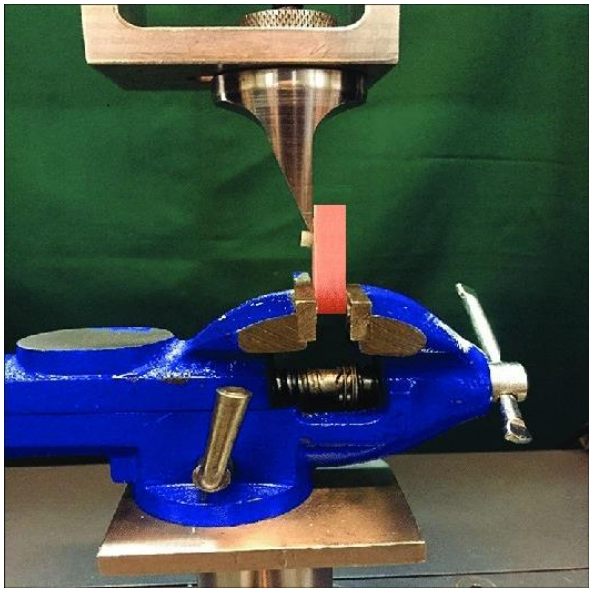


Fig 28 – Microshear Bond Strength Testing using Universal testing machine

OBSERVATION & RESULTS

OBSERVATION AND RESULTS

Table F: MEAN MICRO HARDNESS IN THE FIVE GROUPS

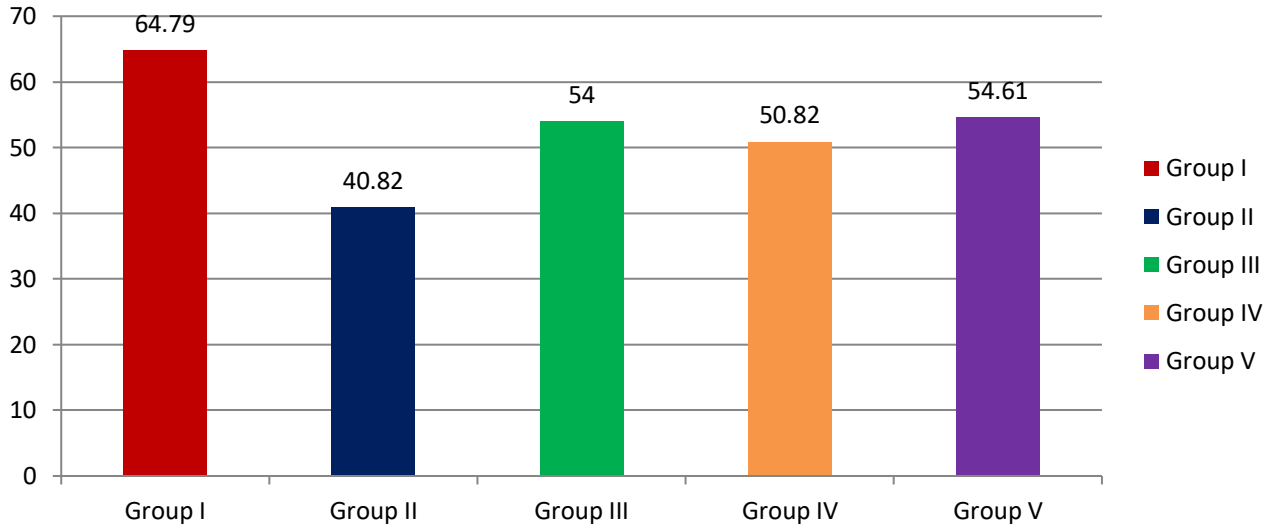
	Mean	Std. Deviation	Std. Error	Minimum	Maximum
Group I (Sound Dentin)	64.79	2.44	0.772	60.60	68.40
Group II (Demineralised Dentin)	40.82	2.98	0.942	36.30	45.60
Group III (Remin Pro)	54.00	2.88	0.912	49.40	58.40
Group IV (MI-Paste Plus)	50.82	3.02	0.957	46.80	55.60
Group V (Icon Infiltrant)	54.61	2.88	0.912	50.90	60.20

The mean micro hardness in the Group I was 64.79 (SD=2.44), in the Group II was 40.82 (SD=2.98). in the Group III was 54.00 (SD=2.88), in the Group IV was 50.82 (SD=3.02) and in the Group V was 54.61 (SD=2.88). The micro hardness was highest in the Group I, followed by Group V and least in the Group II

Table G: INTERGROUP COMPARISON OF MEAN MICROHARDNESS BETWEEN THE GROUPS

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2957.007	4	739.252	90.879	.000
Within Groups	366.050	45	8.134		
Total	3323.057	49			

The mean micro hardness in the Group I was 64.79 (SD=2.44), in the Group II was 40.82 (SD=2.98). in the Group III was 54.00 (SD=2.88), in the Group IV was 50.82 (SD=3.02) and in the Group V was 54.61 (SD=2.88). The micro hardness was highest in the Group I, followed by Group V and least in the Group II . The intergroup comparison between the five groups was done using the One Way ANOVA and difference between the five groups was found to be statistically significant with f value of 90.879 and p value of 0.001



Graph 1 : Mean Micro Shear Bond Strength In The Five Groups

Table H : <u>INTERGROUP COMPARISON OF MEAN MICROHARDNESS BETWEEN THE GROUPS</u>

	Mean	Std. Deviation	Std. Error	Minimum	Maximum	P value
Group I	64.79	2.44	0.772	60.60	68.40	0.001 (Sig)
Group II	40.82	2.98	0.942	36.30	45.60	
Group I	64.79	2.44	0.772	60.60	68.40	0.001 (Sig)
Group III	54.00	2.88	0.912	49.40	58.40	

Group I	64.79	2.44	0.772	60.60	68.40	0.001 (Sig)
Group IV	50.82	3.02	0.957	46.80	55.60	
Group I	64.79	2.44	0.772	60.60	68.40	0.001 (Sig)
Group V	54.61	2.88	0.912	50.90	60.20	
Group II	40.82	2.98	0.942	36.30	45.60	0.001 (Sig)
Group III	54.00	2.88	0.912	49.40	58.40	
Group II	40.82	2.98	0.942	36.30	45.60	0.001 (Sig)
Group IV	50.82	3.02	0.957	46.80	55.60	
Group II	40.82	2.98	0.942	36.30	45.60	0.001 (Sig)
Group V	54.61	2.88	0.912	50.90	60.20	
Group III	54.00	2.88	0.912	49.40	58.40	0.016 (Sig)
Group IV	50.82	3.02	0.957	46.80	55.60	
Group III	54.00	2.88	0.912	49.40	58.40	0.635 (Non-Sig)
Group V	54.61	2.88	0.912	50.90	60.20	
Group IV	50.82	3.02	0.957	46.80	55.60	0.005 (Sig)
Group V	54.61	2.88	0.912	50.90	60.20	

The mean micro hardness in the Group I was 64.79 (SD=2.44) and in the Group II was 40.82 (SD=2.98)). The intergroup comparison between the groups was done using the independent t test and difference between the five groups was found to be statistically significant with p value of 0.001

The mean micro hardness in the Group I was 64.79 (SD=2.44) and in the Group III was 54.00 (SD=2.88), The intergroup comparison between groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean micro hardness in the Group I was 64.79 (SD=2.44) and in the Group IV was 50.82 (SD=3.02) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean micro hardness in the Group I was 64.79 (SD=2.44) and in the Group V was 54.61 (SD=2.88).The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean micro hardness in the Group II was 40.82 (SD=2.98) and in the Group III was 54.00 (SD=2.88) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant p value of 0.001

The mean micro hardness in the Group II was 40.82 (SD=2.98) and in the Group IV was 50.82 (SD=3.02) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

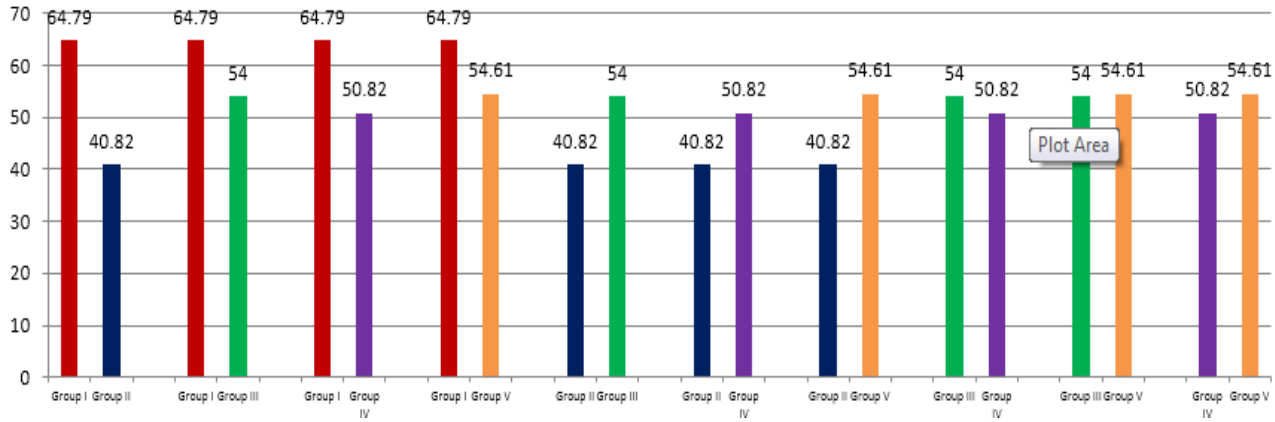
The mean micro hardness in the Group II was 40.82 (SD=2.98) and in the Group V was 54.61 (SD=2.88). The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean micro hardness in the Group III was 54.00 (SD=2.88), and in the Group IV was 50.82 (SD=3.02) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.016

The mean micro hardness in the in the Group III was 54.00 (SD=2.88) and in the Group V was 54.61 (SD=2.88). The intergroup comparison between the groups was done using the

independent t test and difference between the five groups was found to be **statistically non-significant with p value of 0.635**

The mean micro hardness in the in the Group IV was 50.82 (SD=3.02) and in the Group V was 54.61 (SD=2.88). The intergroup comparison between the five groups was done using the independent t test and difference between the five groups was found to be statistically significant with p value of 0.005



Graph 2 : Intergroup Comparison Of Mean Microhardness Between The Groups

Table I : MEAN MICRO SHEAR BOND STRENGTH IN THE FIVE GROUPS

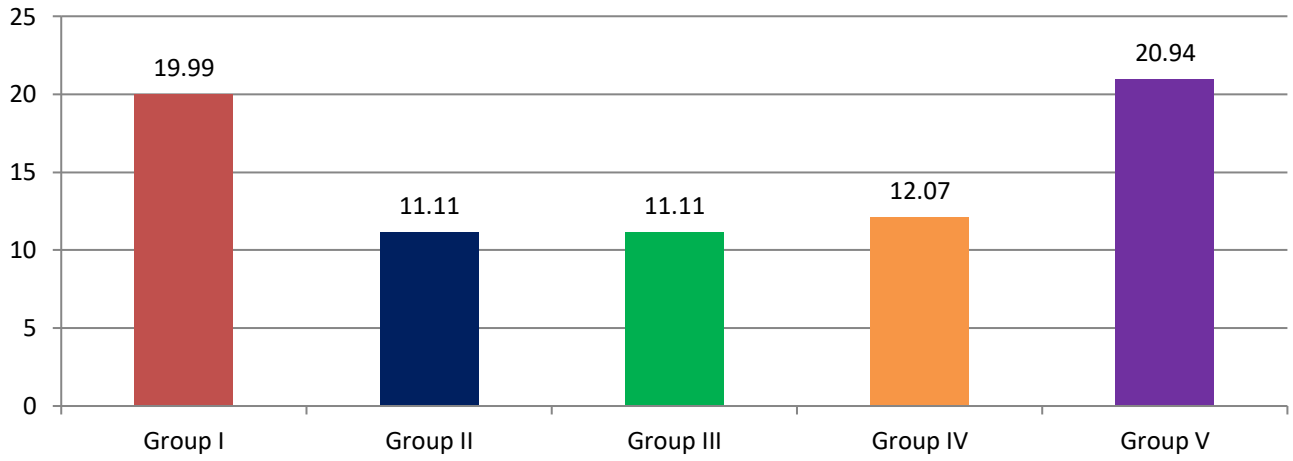
	Mean	Std. Deviation	Std. Error	Minimum	Maximum
Group I (Sound Dentin)	19.99	2.88	0.911	15.60	25.60
Group II (Demineralised Dentin)	11.11	1.81	0.574	8.40	14.70
Group III (Remin Pro)	11.11	2.35	0.744	7.40	14.20
Group IV (MI-Paste Plus)	12.07	1.72	0.544	9.40	14.60
Group V (Icon Infiltrant)	20.94	2.57	0.814	17.60	24.70

The mean shear bond strength in the Group I was 19.99 (SD=2.88), in the Group II was 11.11 (SD=1.81). in the Group III was 11.11 (SD=2.35), in the Group IV was 12.07 (SD=1.72) and in the Group V was 20.94 (SD=257). The bond strength was highest in the Group V, followed by Group I and least in the Group II and Group III

Table J : INTERGROUP COMPARISON OF MEAN SHEAR BONDS STRENGTH BETWEEN THE GROUPS

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	990.231	4	247.558	46.264	0.001
Within Groups	240.792	45	5.351		
Total	1231.023	49			

The mean shear bond strength in the Group I was 19.99 (SD=2.88), in the Group II was 11.11 (SD=1.81). in the Group III was 11.11 (SD=2.35), in the Group IV was 12.07 (SD=1.72) and in the Group V was 20.94 (SD=257). The intergroup comparison between the five groups was done using the One Way ANOVA and difference between the five groups was found to be statistically significant with f value of 46.264 and p value of 0.001



Graph 3 : Mean Micro Shear Bond Strength In The Five Groups

Table K : INTERGROUP COMPARISON OF MEAN SHEAR BONDS STRENGTH BETWEEN THE GROUPS

	Mean	Std. Deviation	Std. Error	Minimum	Maximum	P value
Group I	19.99	2.88	0.911	15.60	25.60	0.001 (Sig)
Group II	11.11	1.81	0.574	8.40	14.70	
Group I	19.99	2.88	0.911	15.60	25.60	0.001 (Sig)
Group III	11.11	2.35	0.744	7.40	14.20	
Group I	19.99	2.88	0.911	15.60	25.60	0.001 (Sig)
Group IV	12.07	1.72	0.544	9.40	14.60	
Group I	19.99	2.88	0.911	15.60	25.60	0.363 (Non-sig)
Group V	20.94	2.57	0.814	17.60	24.70	
Group II	11.11	1.81	0.574	8.40	14.70	1.000 (Non-Sig)
Group III	11.11	2.35	0.744	7.40	14.20	
Group II	11.11	1.81	0.574	8.40	14.70	0.358 (Non-Sig)
Group IV	12.07	1.72	0.544	9.40	14.60	
Group II	11.11	1.81	0.574	8.40	14.70	0.001 (Sig)
Group V	20.94	2.57	0.814	17.60	24.70	
Group III	11.11	2.35	0.744	7.40	14.20	

Group IV	12.07	1.72	0.544	9.40	14.60	0.358 (Non-Sig)
Group III	11.11	2.35	0.744	7.40	14.20	0.001 (Sig)
Group V	20.94	2.57	0.814	17.60	24.70	
Group IV	12.07	1.72	0.544	9.40	14.60	0.001 (Sig)
Group V	20.94	2.57	0.814	17.60	24.70	

The mean shear bond strength in the Group I was 19.99 (SD=2.88), in the Group II was 11.11 (SD=1.81). The intergroup comparison between the five groups was done using the independent t test and difference between the five groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in the Group I was 19.99 (SD=2.88) and in the Group III was 11.11 (SD=2.35), The intergroup comparison between the five groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in the Group I was 19.99 (SD=2.88) and in the Group IV was 12.07 (SD=1.72) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in the Group I was 19.99 (SD=2.88) and in the Group V was 20.94 (SD=257). The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in in the **Group II was 11.11 (SD=1.81) and in the Group III was 11.11 (SD=2.35)**The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically **non-significant p value of 1.000**

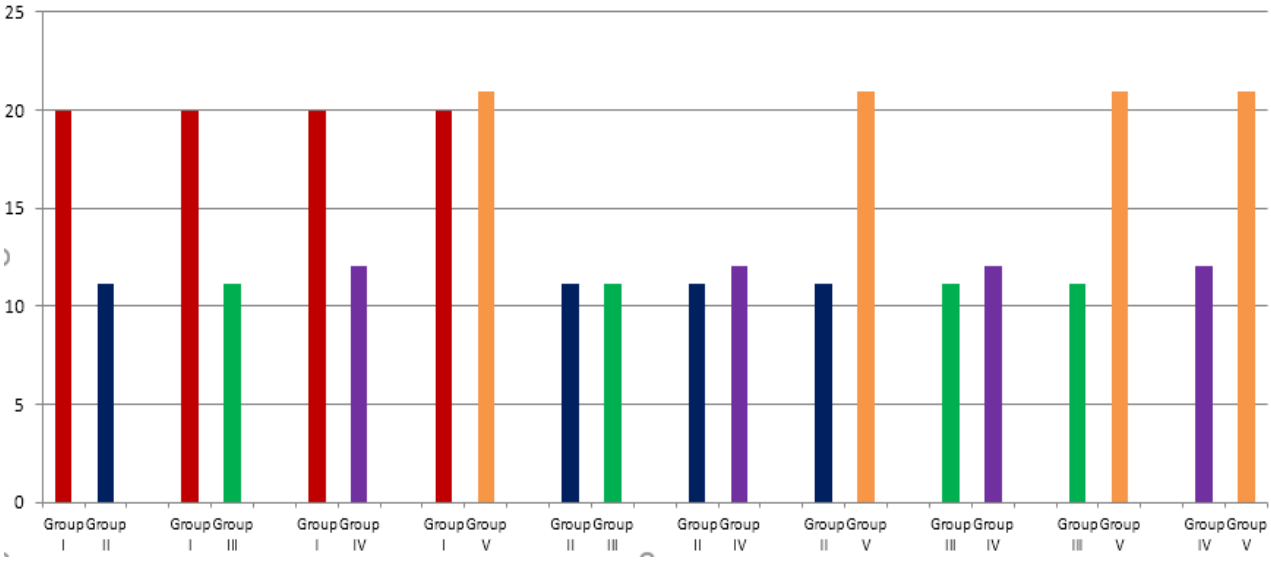
The mean shear bond strength in the in the **Group II was 11.11 (SD=1.81). and in the Group IV was 12.07 (SD=1.72)** The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be **statistically non-significant with p value of 0.358**

The mean shear bond strength in the in the Group II was 11.11 (SD=1.81) and in the Group V was 20.94 (SD=257). The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in the in the **Group III was 11.11 (SD=2.35) and in the Group IV was 12.07 (SD=1.72**) The intergroup comparison between the groups was done using the independent t test and difference between the groups was found to be statistically **non-significant with p value of 0.358**

The mean shear bond strength in the in the Group III was 11.11 (SD=2.35) and in the Group V was 20.94 (SD=257). The intergroup comparison between the groups was done using the independent t test and difference between the five groups was found to be statistically significant with p value of 0.001

The mean shear bond strength in the in the Group IV was 12.07 (SD=1.72) and in the Group V was 20.94 (SD=257). The intergroup comparison between the five groups was done using the independent t test and difference between the five groups was found to be statistically significant with p value of 0.001



Graph 4 : Intergroup Comparison Of Mean Shear Bonds Strength Between The Groups

DISCUSSION

Discussion

The present in-vitro study was conducted in the Department of Conservative Dentistry and Endodontics, Babu Banarasi Das College of Dental Sciences, Lucknow in collaboration with Praj Mettallurgical Lab, Pune.

The main objective of this study was to assess the effect of surface micro hardness of demineralised dentin after remineralising with MI Paste Plus, Remin Pro and Icon Infiltrant and to evaluate the microshear bond strength (μSBS) of composite to remineralised dentin. The results showed that MI paste plus, Remin Pro and Icon Infiltrant increased the surface microhardness of demineralised dentin but lower than the sound dentin. The result also showed that Icon Infiltrant has higher microshear bond strength than sound dentin and Remin Pro & MI paste plus showed significantly weaker bond strength than sound dentin.

The balance between demineralization and remineralization is largely mediated by the acidic microenvironment intraorally, produced by bacteria, and the buffering capacity of saliva in neutralizing acidic pH levels. A drop in pH of the oral cavity results in tooth demineralization. Conversely, a rise in pH results in the deposition of calcium, phosphate, and fluoride, hence a reversal of the demineralization process. To prevent demineralization effectively, early intervention is essential and possible through the application of commercially available preparations consisting of, for example, fluoride, calcium, phosphate-based systems, and calcium sodium phosphosilicate. (24)

Studies have shown that dentin remineralized at a much faster rate than enamel (25). Similar results were reported by Laheij *et al*. 2010 (26). who evaluated *in situ* remineralization of enamel and dentin. According to ten Cate in 2008 , proteins such as dentin phosphoprotein may play a role in enhanced mineralization of dentin.. Similarly van Strijp *et al*. observed full remineralization of dentinal lesions after using fluoride toothpastes and observed that remineralization conditions for dentin lesions may be favorable compared to enamel lesions. They suggested that the demineralized organic matrix of dentin may constitute a scaffold to enhance remineralization. (27,28,29)

The increasing demand for non-invasive therapeutic procedures for dental caries has encouraged researchers to assess and compare the efficacy of remineralizing agents available in the market (30). CPP-ACP is among the commonly used remineralizing agents and its efficacy has been widely investigated (31,32). MI Paste Plus is a type of CPP-ACPF available in the market that contains amorphous calcium and phosphate as well as 900-ppm fluoride, stabilized by casein phosphopeptide (33).

The remineralizing agent Remin Pro contains fluoride (1450 ppm), HA, and xylitol (antibacterial agent) and has been suggested that it can mitigate dental hypersensitivity, prevent demineralization, and enhance remineralization of subsurface lesions. HA provides remineralizing agents with the potential to repair microcracks in enamel and dentin, thereby improving the hardness of enamel and dentin by HA present in preparations like Remin Pro (24). According to Miake *et al*., xylitol can induce remineralization of deeper layers of demineralized enamel and dentin by facilitating calcium ion movement and accessibility (34).

Nowadays, Incipient decay can now also be managed with other conservative methods (35,36,37) such as resin infiltration (RI) which was developed in Germany to treat early enamel proximal caries and it is now commercially available in the name ICON. Its low viscosity that penetrates the porosities of the lesions, makes it superior to the former technique that depends on sealing of the occlusal lesions by sealing materials. (38,39) Resin infiltration has several benefits, like absence of tooth structure loss, allowing stability for white spot lesions, preventing caries progress, plugging of the micropore forms in the body of the lesion, delaying the necessity for a restoration, decreasing recurrent decay, absence of inflammation of the pulp and postoperative sensitivity, lowering the possibility of periodontitis and gingivitis and better aesthetic outcomes in covering demineralized enamel.(40,41)

The prevalence rate of cervical lesions makes about 18% of the permanent teeth defects. Premolars are the most commonly affected teeth. (42). Thus, the current study was conducted on Maxillary & Mandibular Premolar teeth. In the current study, dentin was exposed on the cervical aspect of the buccal and lingual surfaces of the teeth so that it closely relates to the condition of Non Caries Cervical Lesion's. (NCCL's) The demineralization-remineralization cycle was adopted from a previous study (43). This closely mimics an erosive intraoral environment, in which cycles of demineralization and remineralization occur.

In the present study, Filtek Z350, a methacrylate-based nanofill composite resin designed for use in anterior and posterior restorations was chosen. Qin et al. (2013), showed that Filtek Z350 can be used as a restorative material for an effective clinical performance in NCCL's (44).

In this study, Total etch (Etch & Rinse) adhesive system is used for bonding the composite to the dentin. Many studies has shown that the etch-and-rinse adhesive system presented superior μTBS value in comparison with the self-etching system (45) . The enamel etch-and-rinse method is based on selective demineralization of the hydroxyapatite crystals found in tooth enamel, resulting in a highly roughened surface with elevated energy. These features offer better wetting capacity of the resinous monomers that, when polymerized, result in prolongations named tags that 'anchor' the resin to the tooth (46,48). In dentistry, the substance most commonly used for this purpose is phosphoric acid at a concentration of 35-37%. Because of its high ionization potential, it results in a final pH of 0.6. As a consequence of the high availability of H+ ions, its application for short periods, such as the 15 seconds usually recommended, is capable of producing a suitable enamel etching pattern (47), resulting in exceptional micromechanical interlocking by the tags created (48).

Acidic adhesive agents, called self-etchants, were introduced with the objective of promoting simultaneous demineralization and impregnation of the substrate. The concentration of this adhesive solution in an aqueous solution and the number of ionizable radicals are lower in comparison with those in phosphoric acid, and consequently, the etching capacity of this adhesive system is more restricted. As a result, this one-step adhesive does not demineralize enamel to the same extent as phosphoric acid does, promoting a less microretentive surface and usually lower μTBS values (49). Moreover, according to Erickson, (2009) (50), phosphoric acid promotes a particular morphology of the resin-enamel that permits fairly extensive resin penetration, creating a three dimensional structure (i.e. scalloped), and the transition from resin to sound enamel is distributed over a variety of microns. This interface can be more resistant to crack propagation when compared with the relatively plainer interface promoted by self-etch adhesives

.Surface Microhardness (SMH) measurement is a useful technique that can be used to determine enamel and dentine surface hardness. SMH indentation provides a proportionately simple and abrupt method in demineralization and remineralization studies for materials with fine microstructure, non-homogenous, or prone to cracking such as dental enamel and dentin. Microhardness measurements can be evaluated by two different parameters, Knoop hardness number, and Vickers hardness number. VMHTs are used in most cases to assess the hardness of materials within the range of microhardness test loads (typically 1-1000 g). However, Knoop hardness test is often used when testing the hardness of thin layers such as ceramics or coatings to overcome the problem of cracking in brittle materials. The square shape of the residual indentation obtained by VMHT are easy and accurate to measure under a microscope. Minutest changes in square shape can be easily detected, whereas rhomboid shape indentations with opposing surfaces parallel to each other, obtained with Knoop hardness, make it difficult to detect errors (51).

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The Vickers microhardness test (VMHT) was used in this study to determine the microhardness of the tooth structure. VMHT uses a diamond indent area to impress a small area of the tooth surface with a predefined set load for a predetermined period of time. After microscopic examination of the indentation in relation to the used indentation load and the area of the remaining impression, the microhardness number is computed.. (52,53)

Microshear bond strength testing was chosen for testing the microshear bond strength in this study, as it is a distinctive test to assess the interfacial strength between the tooth and the polymeric composite restorative materials. The smaller bonded surface area would allow a more uniform stress distribution, which provides an accurate assessment when compared to the conventional shear test.(54)

The results of our study showed that Remin Pro to be more effective than MI Paste Plus in remineralization of demineralised dentin. The phosphopeptide stabilizes the calcium and phosphate ions and creates a super-saturated state of these ions adjacent to tooth structure. Remineralization of tooth structure occurs in presence of high levels of these ions and increased pH [44]. In fact, the density of hydroxyapatite crystals increases by penetration of calcium, phosphate and fluoride ions into these crystals (55). Leila *et al.* in their study (56) showed MI Paste Plus to be more effective than Remin Pro in remineralising the tooth structure, which was in contrast to our findings. Although the efficacy of high concentration

of calcium for dental remineralization has been previously confirmed, high concentration of calcium can cause its quick absorption by the superficial layers and thus, less remineralization occurs in deeper layers of the lesion (57). Casein phosphopeptide present in MI Paste Plus prevents its fast deposition and stabilizes the calcium and phosphate compounds [55]. According to Vyavhare *et al.* (58) MI Paste can be used as an adjunct to fluoride but due to lower remineralization level, it cannot be used as an alternative to fluoride. It should be noted that natural saliva and dental biofilm provide a suitable environment to preserve the afore-mentioned super-saturated state of ions in the oral cavity during treatment with CPP-ACP compounds (59). Considering the absence of these factors in vitro, the effect of these compounds can only be attributed to their application time on dental samples; this explains the difference in the results of in vivo and in vitro studies (60).

The efficacy of Remin Pro for remineralization of incipient enamel lesions has been previously confirmed (61). Kamath *et al.* (62) indicated that Remin Pro in-creases the hardness of bleached enamel. Shetty *et al.* (63) have also confirmed the efficacy of this material for increasing the micro hardness of incipient carious le-sions, which is in agreement with our findings. Remin Pro is a water-based paste made of hydroxyapatite, fluo-ride and xylitol . It seems that hydroxyapatite can fill the porosities of the incipient carious lesions. The fluoride in the composition of Remin Pro seals the tu-bules while xylitol exerts antibacterial effects. Thus, this compound can stop demineralization and induce remineralization of incipient enamel lesions (64). Our results are also in accordance with Thakur Sandeep *et al*. (65)and Jaya Shankara *et al*. (66). These studies were in accordance as the studies were similar to the present study on account of remineralization and the remineralizing agent Remin Pro had shown the better remineralization potential compared with other agents used in their studies.

In the present study, micro hardness values of deminerlised dentin following the use of ICON were higher than the other remineralising agents used in the study but the hardness value were lesser than the sound dentin. Resin infiltration of lesions is performed aiming to fill the enamel lesion porosities and increase enamel resistance to acid attacks. This can stop caries progres-sion and increase the strength of lesions (67). Arslan *et al.* (68) in their study showed that infiltration of incipient enamel lesions with ICON resin increased the micro hardness and

decreased bacterial accumulation. Prasada *et al.* (69) in their study indicated that use of ICON resin improved the appearance of WSLs.

ICON's 15% HCl gel erodes the surface layer more effectively than 37% phosphoric acid (H 3PO4) gel, and using the HCl conditioning for two minutes with ICON could have led to dee per resin penetration than using phosphoric acid gel for the etching process (70). The amount of tooth structure removed following the use of this etchant is around 40 μ; (45) while in micro-abrasion treatment of WSLs; the tooth structure is removed to 250-μ depth (71). Also, the use of ICON dry which contains 99% ethanol for 30 s before application of the infiltrant increases the penetration coefficient by reducing the viscosity and contact angle and improves infiltration depth. (72). In addition, higher ethanol content can inhibit polymerization of resin and thus reduce infiltration success. (73). ICON-Infiltrant is a methacry-late-based resin containing BISGMA and TEGDMA. Increased concentration of TEGDMA, HEMA and ethanol increases the resin penetration coefficient. Application of Resin Infiltrant was done twice in our study, which caused increase in microhardness of the lesions, compensated to polymerization shrinkage and filled porosities and crevices in the infiltrated lesion body, thereby increasing hardness and decreasing mineral loss when exposed to demineralization again. (74)

The results of this study also clearly show that bond strength values obtained in demineralized specimens and those treated with remineralizing agents were lesser than those obtained with normal dentin. In this study, the μSBS of composite to demineralized dentin was significantly lesser than to sound dentin. This could be attributed to the loss of minerals from the peritubular and intertubular dentin that compromises the bonding of adhesive monomers to demineralized dentin. The remaining unsupported collagen network serves as water filled channels. This hydrophilic substrate further lowers the bonding ability of adhesive resins. (75)

In this study, MI Paste Plus (CPP-ACPF) showed better bond strength than Remin Pro (Ha, F, Xylitol). Ishikawa et al. (76) showed that following CPP-ACP application, calcium phosphate (Ca–P) precipitation occurs on the superficial dentin and that the precipitates did not penetrate deep enough into the dentinal tubules in order to effectively occlude them. Also

this precipitation occurred so rapidly, that it prevented continuous Ca–P interaction in solution thus, resulting in precipitate formation only on the surface of dentin. The surface debris was sparse as the precipitates formed due to CPP-ACP were dissolved by the use of phosphoric acid as etchant. The low pH (< 1) of phosphoric acid could be attributed to this. Further, the rinsing step might have led to the washing away of the dissolved products, and the superficially deposited minerals, thus resulting in predominantly open dentinal tubules. This relatively precipitate free dentin surface could also be the reason behind the higher bond strength of this group, compared to Remin Pro.

The group treated with Remin Pro also showed the lowest μSBS values like the demineralised dentin. Saliva contains calcium (Ca) and phosphate (Pi) in supersaturated concentrations, and these ions are continually deposited or re-deposited on the tooth surface that has suffered loss of these ions (77). The fluoride ions present in the Remin Pro can also remain incorporated into remineralizing dentin, mainly in surface lesions, changing the carbonated apatite to a fluoroapatite-like form that is more acid tolerant and makes more acid resistant hard tissues (77,78,79). A previous study showed that both saliva exposure for eight weeks and daily sodium fluoride treatment resulted in increased surface microhardness of demineralized specimens(79). It is therefore hypothesized that the increased acid resistance of remineralized dentin impaired the conditioning effect of the etching system, thus promoting a less microretentive surface, and consequently, lower μSBS values

The groups infiltrated with the low-viscosity resin showed higher μSBS than the positive control group (sound enamel). The resin infiltrant contains monomers with high penetration coefficients and adequate hardening (80). The satisfactory μSBS values associated with infiltrated groups may be said to optimized due to the affinity between the monomers present in the infiltrant and the monomers of the adhesive systems. According to the results of the present study, resin infiltration resulted in increased bond strength, and therefore, restorative treatment can be indicated on tooth surfaces treated with resin infiltration, since it does not negatively interfere in the composite bond to enamel and dentin. Previous studies also showed that the application of an etch-and-rinse adhesive after resin infiltration did not alter enamel μTBS (81), or even increase the adhesion of a self-etching adhesive (80).

In order to infiltrate a carious lesion, resin infiltration requires the application of 15% hydrochloric acid to promote erosion of the surface layer and allow the resin to penetrate into

the porous spaces of the lesion body (81,82,83). An appropriate acid etching pattern enhances resin infiltration into the more porous lesion body structures, both in natural caries lesions (25) and also in artificial lesions (84,85) optimizing the μSBS to the substrate, as was observed in this study, in which the infiltrated groups reestablished the μSBS above the levels achieved in the sound enamel

.

This study did not reproduce the natural sclerotic dentin encountered in NCCLs, which is a difficult substrate to bond, when compared to normal cervical dentin. (49) The interaction of dentin and remineralising agents could be affected by numerous aspects like age of source of specimen, smear layer, dentinal tubule orientation, their density, branching, diameter and direction, and existing or absent peritubular dentin. Insufficient bonding could have also resulted from dehydration associated with the use of extracted teeth. Pulpal pressure, which was not simulated here, might also have significant consequence on bonding. Hence, further long-term clinical studies are mandatory to validate the results of this research

In this in vitro study, extracted human premolars were used and the procedure and time needed for the formation of lesions, the application of the treatment material and how the material works on the lesions may be different from what happens in vivo. The lesions in each tooth may also be different, i.e. the amount of demineralization in each tooth may vary depending on the amount of fluoride to which they were exposed before extraction. Other limitations unique to our research are that the pH cycling cannot fully replicate the oral environment. In about 3 days of exposure to demineralization acids, the lesions were formed, while oral cavity lesions may develop over a period of months or years..

Hence from this study, it can be concluded that ICON material performs well in increasing the hardness and the bond strength. The ICON material is used as an infiltrant and prevent further demineralization by formation of resin tags where the material penetrates deep into the tooth surface.The low-viscosity resin infiltration treatment did not affect dentin μSBS values while the demineralization and remineralization treatments reduced enamel μSBS values of the Composite to the tooth surface

CONCLUSION

CONCLUSION

Within the limitations of this *in vitro* study, the sound dentin shows greatest microhardness, yet along with ths, it has also been found that Remin Pro, MI Paste Plus and Icon Resin Infiltrant can efficiently increase the micro hardness of demineralised dentin. On compairing the remineralising agents, it can be stated that Icon resin infiltrant showed the highest microhardness value among the tested groups followed by Remin Pro and then MI paste plus.

Shear bond strength was found to be the highest for Icon Resin when compared with all other groups even sound dentin. The bond strength was significantly reduced in the Remin Pro, MI Paste Plus and demineralised dentin groups. MI paste plus showed slightly higher bond strength than Remin Pro and demineralised dentin.

Hence, it can be stated that bonding to remineralized dentin is compromised owing to the chemical and morphological changes that occur in this substrate. Icon material performed well in increasing the micro hardness and the bond strength. Hence it can be used as an infiltrant. The Icon material prevents further demineralization by formation of resin tags where the material penetrates into the enamel and dentin. Future long-term clinical trials should be conducted to determine the superiority of these agents in vital teeth.

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ANNEXURES

ANNEXURE 1

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES (FACULTY OF BBD UNIVERSITY), LUCKNOW

INSTITUTIONAL RESEARCH COMMITTEE APPROVAL

The project titled **"Effect of Remineralisation and Resin Infiltration on the Surface Hardness and Evaluation of Microshear Bond Strength using Composite Resin on the Remineralised Dentin: An In Vitro Study"** submitted by **Dr Rechu Raju** Post graduate student from the **Department of Conservative Dentistry and Endodontics** as part of MDS Curriculum for the academic year 2020-2023 with the accompanying proforma was reviewed by the Institutional Research Committee present on **11th October 2021** at BBDCODS.

The Committee has granted approval on the scientific content of the project. The proposal may now be reviewed by the Institutional Ethics Committee for granting ethical approval.

Prof. Vandana A Pant
Co-Chairperson

Prof. B. Rajkumar
Chairperson

ANNEXURE 2

Babu Banarasi Das University
Babu Banarasi Das College of Dental Sciences,
BBD City, Faizabad Road, Lucknow – 226028 (INDIA)

Dr. Lakshmi Bala
Professor and Head Biochemistry and
Member-Secretary, Institutional Ethics Committee

Communication of the Decision of the IXth Institutional Ethics Sub-Committee

IEC Code: 32 **BBDCODS/04/2022**

Title of the Project: Effect of remineralisation and resin infiltration on the surface hardness and evaluation of microshear bond strength using composite resin on the remineralised dentin: An in vitro study.

Principal Investigator: Dr Rechu Raju **Department:** Conservative Dentistry & Endodontics

Name and Address of the Institution: BBD College of Dental Sciences Lucknow.

Type of Submission: New, MDS Research

Dear Dr Rechu Raju,

The Institutional Ethics Sub-Committee meeting comprising following four members was held on 07th April, 2022.

1.	Dr. Lakshmi Bala Member Secretary	Prof. and Head, Department of Biochemistry, BBDCODS, Lucknow
2.	Dr. Amrit Tandan Member	Prof. & Head, Department of Prosthodontics and Crown & Bridge, BBDCODS, Lucknow
3.	Dr. Rana Pratap Maurya Member	Reader, Department of Orthodontics, BBDCODS, Lucknow
4.	Dr. Akanksha Bhatt Member	Reader, Department of Conservative Dentistry & Endodontics, BBDCODS, Lucknow

The committee reviewed and discussed your submitted documents of the current MDS Project Protocol in the meeting.
The comments were communicated to PI thereafter it was revised.

Decisions: The committee approved the above protocol from ethics point of view.

Forwarded by:

Lakshmi Bala

(Dr. Lakshmi Bala)
Member-Secretary
IEC

Member-Secretary
Institutional Ethic Committee
BBD College of Dental Sciences
BBD University
Faizabad Road, Lucknow-226028

(Dr. Puneet Ahuja)
Principal

PRINCIPAL BBDCODS
Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)
BBD City, Faizabad Road, Lucknow-226028

ANNEXURE 3

DENTAL MATERIAL TEST FACILITY
MDS Thesis in various fields
i.e. Orthodontic, Prosthodontics, Endodontics, Periodontics, Oral Surgery, etc.

PRAJ
Metallurgical Laboratory

B-38, Indira Shankar Nagari,
Paud Road, Near Kothrud PMT Depo,
Kothrud, Pune - 411 038.
Tel.: +91 - 20 - 2528 1584,
Cell : 96650 11314, 9158658634
E-mail : prajdental@gmail.com
Website: www.prajlab.com

Date: 9/11/2022

TO WHOMSOEVER IT MAY CONCERN

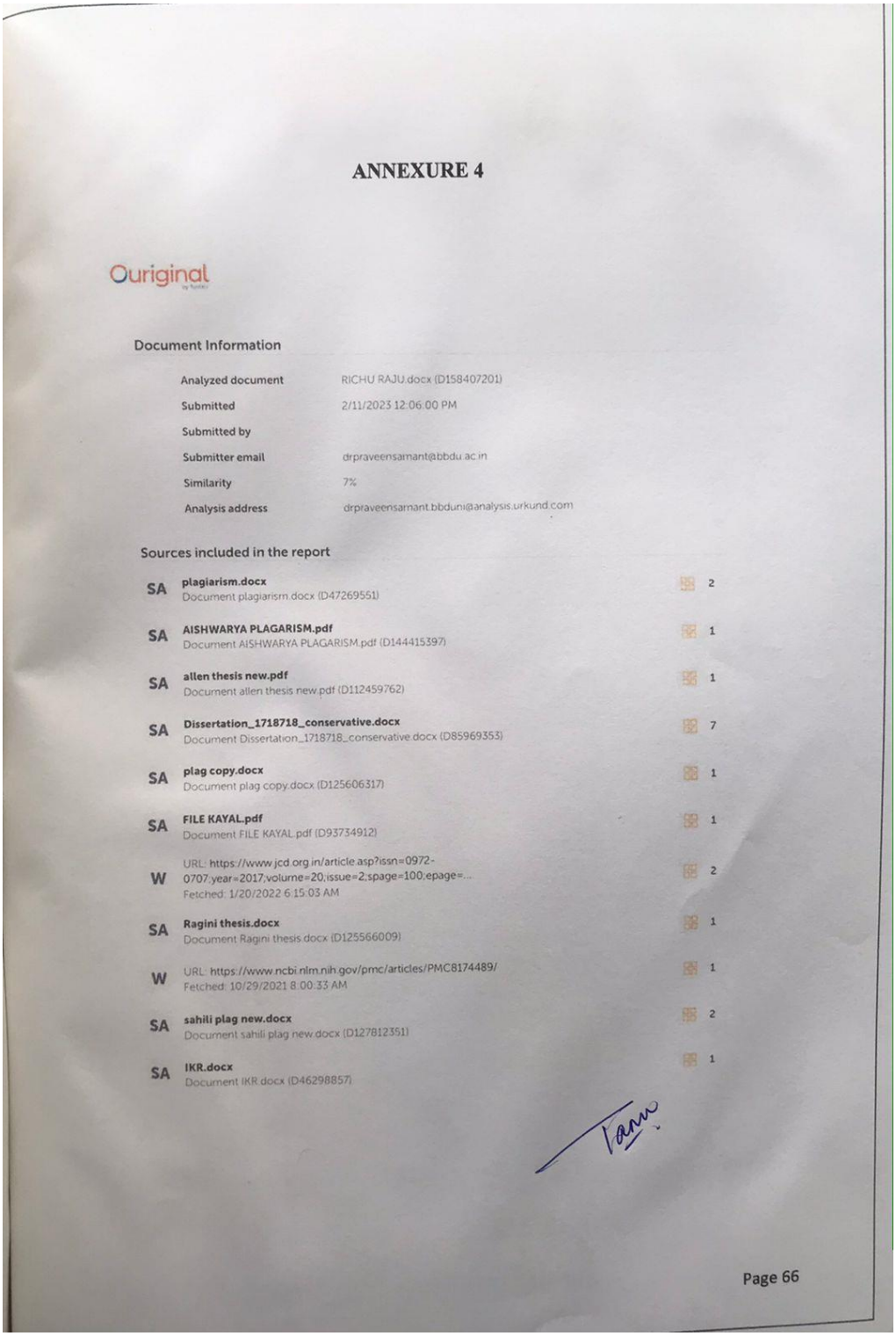
This is to certify that Dr. Rechu Raju , final year MDS , Babu Banarasi Das College of Dental Sciences , has done his Vickers Microhardness Testing & Micro Shear Bond Strength Testing of teeth samples from PRAJ Metallurical Laboratory , Kothrud , Pune

SACHIN
TEST ENGINEER

A.M.BHAGAT
PROPRIETOR

PRAJ METALLURGICAL LABORATORY ★ PUNE

ANNEXURE 4



ANNEXURE 4

Ouriginal

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Analysis address	drpraveensamant.bbduni@analysis.urkund.com

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