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# Assessment of white spot lesions around Orthodontic brackets using different Bonding Agents: an SEM study

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#### **ABSTRACT**

#### **Introduction:**

White spot lesions (WSLs) around orthodontic brackets are a significant concern due to their impact on aesthetics and oral health. This study delves into the factors contributing to WSL formation, exploring their genesis, pathophysiology, and clinical implications. Various factors, such as orthodontic attachment design and archwire ligation, influence WSL development. Remedial approaches and adjunctive treatments aim to prevent or mitigate demineralization during orthodontic intervention.

**Methods**: This study examined WSLs in 10 patients aged 14 to 23 undergoing orthodontic treatment with different bonding composites (AEGIS ORTHO and TRANSBOND XT). Samples were evaluated using scanning electron microscopy (SEM) and image analysis software to quantify WSL severity and lesion depth. Statistical analysis compared the outcomes between the two groups.

**Results**:Both groups experienced increased WSL severity after debonding, with Group A (TRANSBOND XT) showing higher demineralization. Within Group A, demineralization increased significantly after debonding, while Group B (AEGIS ORTHO) remained relatively stable. The net mean change in WSL severity showed 23.0% higher demineralization in Group A. Group B had significantly higher WSL severity and significantly lower lesion depth compared to Group A.

**Discussion**: The study highlights the role of bonding agents in WSL formation and their impact on demineralization. It aligns with previous research on bacterial acids, plaque dynamics, bracket design, and innovative materials like "smart composites."

Conclusion: This study underscores the significance of bonding materials in orthodontic treatments and their effects on WSLs. Aegis orthodontic bonding composite with Amorphous Calcium Phosphate (AEGIS ORTHO) exhibited better outcomes in terms of both WSL severity and lesion depth compared to TRANSBOND XT. Further research can build upon these insights to enhance bonding practices and improve the quality of orthodontic interventions.

*Keywords:* White Spot Lesions (WSLs), Transbond XT, Aegis Ortho, Scanning Electron Microscopy (SEM), Amorphous Calcium Phosphate (ACP), Casein Phosphopeptidase (CPP)..

#### 1. INTRODUCTION

It is imperative to explore in-depth the intricate underpinnings of white spot lesions (WSLs) around orthodontic brackets, a topic of profound significance. A mechanical force coupled to a biochemical dynamic creates a microcosm between the orthodontic apparatus and dental enamel, resulting in the appearance of these demineralised areas. <sup>[1,2]</sup> It requires a meticulous dissection of multifaceted factors that orchestrate the formation of such lesions in order to understand their genesis, pathophysiology, and clinical implications.

It is believed that the dissolution of enamel is caused by acids produced by bacteria within plaque which cause the enamel to dissolve. Most cases of demineralisation appear in the subsurface area of the enamel before four weeks, but it has been known to occur earlier. There is a strong correlation between the continued retention of bacteria in the form of plaque after the fixed orthodontic appliance is placed, and this increases the risk of caries developing. Plaque retention may be influenced by the surface characteristics, design, and design of both orthodontic attachments as well as composites. Additionally, it is of importance to note that the method of ligation of the archwire plays a significant role in developing WSLs. [5]

The manifestation of these WSLs ranges from 2-96% on the tooth surface [6,7] and this elicits a convergence of financial, emotional, and biological perturbations for patients, thereby imparting a distinctive clinical quandary to orthodontic practitioners. Despite the widespread acceptance of the remedial approach in dentistry to WSLs with the objective of attaining a beautiful enamel surface characterised by aesthetic appeal, there is still an enigmatic paradox which eludes resolution. As a result, it becomes essential to employ adjunctive agents as auxiliary treatments, which are aimed at preventing and/or ameliorating the processes of demineralisation that may occur during or after orthodontic intervention.

In the past decade, the works of Schumacher et al <sup>[8]</sup> have illuminated the restorative materials endowed with biological activity, showcasing their potential to incite tooth structure repair by releasing bioactive substances like calcium and phosphorous, effective in combatting caries. These innovative composites, coined as "smart composites," incorporate fillers such as amorphous calcium phosphate (ACP), which is thoughtfully encapsulated within a polymer binder to serve as a bioactive agent. Thorough investigations encompassing laboratory, animal, and human in situ studies have corroborated the anti-cariogenic prowess of casein phosphopeptidase amorphous calcium phosphate (CPP-ACP). The released calcium and phosphate ions from ACP materials have been demonstrated to precipitate onto dental structures, forming an apatite mineral akin to the hydroxyl-apatite inherent in tooth enamel, as evidenced by its striking similarity.<sup>[9]</sup>

WSLs are characterised by a variety of macroscopic and microscopic methodologies which significantly contribute to the identification and quantification of WSLs, along with the assessment of the extent to which demineralisation has occurred. In addition to clinical and photographic examinations, optical methods that don't use fluorescent dye, and optical fluorescent approaches that do use fluorescent dye are also included among these techniques. Moreover, microscopic analyses are integral to this endeavour, with a particular emphasis placed upon backscattered scanning electron microscopy (SEM) being one of the most efficient methods. As the name implies, backscattered SEM is based on the idea of high-energy electron beams scattering and reflecting when they interact with treated tooth samples. Underlying changes in the material composition under the surface can be explained by means of this intricate interaction, which provides profound insights into the underlying processes.

Its prowess in producing mineral density images is demonstrated by the fact that it is capable of capturing both naturally occurring WSLs as well as demineralised enamel lesions that are intentionally induced. With the use of this technology, nuances within enamel structural integrity can be discerned more clearly.

The primary objective of this microscopic investigation was to leverage the potential of backscattered scanning electron microscopy for a comprehensive assessment of the extent and depth of WSLs development during orthodontic treatment. This approach aimed to elucidate the demineralization process associated with orthodontic interventions. Additionally, this study sought to evaluate the impact of different bonding composites, specifically Aegis Orthodontic Bonding Light Cure Composite with Amorphous Calcium Phosphate (AEGIS ORTHO) and Transbond XT (3M UNITEK), on the severity and depth of WSLs around orthodontic brackets. The research also considered the broader implications of composite selection on overall oral health outcomes for patients, both during and after orthodontic treatment.

Our investigation suggests that smart composites like Aegis Ortho have the potential to effectively mitigate WSLs. However, it's important to note that this material is relatively novel in the market, necessitating further research to validate its efficacy in orthodontic patients. Furthermore, future clinical studies will be essential to determine if similar results can be replicated in diverse and complex oral environments.

### 2. MATERIALS AND METHODS:

# **Study Participants:**

The study involved 10 patients aged between 14 and 23 years who were undergoing orthodontic treatment that included the

extraction of all four first permanent premolars. There were two groups of samples: Control (Group A) and Study (Group B). Study design was a randomised cross-arch design.

#### **Bonding Composites:**

Two different bonding composites were used in this study. Group A received Transbond XT (3M UNITEK) for bracket bonding, while Group B received Aegis Orthodontic Bonding Light Cure Composite with Amorphous Calcium Phosphate (AEGIS ORTHO, The Harry J. Bosworth® Company, USA).

# **Bracket Bonding and Premolar Extraction:**

The brackets were attached to the premolars using the respective bonding composites, and light curing was performed to secure them in place. Premolar extractions were scheduled for 16 weeks after the bracket bonding. Upon extraction, the brackets were removed, leaving behind the composite material on the teeth surfaces. The extracted teeth were stored in artificial saliva to preserve their condition for Microscopic examination.

# Microscopic Analysis:

Microscopic analysis of demineralisation was carried out at Birbal Sahni Institute of Paleobotany, Lucknow. The teeth were divided into buccolingual halves, and sections of the bracket-bonded areas were cut for analysis. A palatal furrow was created on the sectioned teeth for identification purposes. The samples were subjected to microscopic analysis using scanning electron microscopy (SEM) at 25 kV. Aluminized stubs were used to mount the samples, and gold and palladium sputtering was performed to coat the tooth surfaces with a 7.5 nm layer before SEM analysis.

### **Quantitative Analysis:**

To quantify the extent of demineralization, image analysis software (Image J version 1.33u for Windows XP, US National Institutes of Health, Bethesda, MD) was used. The SEM images of the control and experimental groups were imported into the software and converted into 8-bit grayscale images. The software was configured to measure the depth of the white spot lesions (WSLs). The outline of the WSLs was traced using the freehand preselection tool.

# **Statistical Analysis:**

Data were summarized as Mean ±SD. Student's t-test and Mann-Whitney U test were used for comparisons between the two independent groups, depending on the data distribution. Kruskal-Wallis one-way analysis of variance (ANOVA) was used to assess inter-observer variability. The significance of mean differences between groups was determined by Dunn's test. Pearson correlation analysis was employed to evaluate inter-observer variability. A p-value less than 0.05 was considered statistically significant. All statistical analyses were conducted using STATISTICA software (Windows version 6.0).

### 3. RESULTS:

Different bonding agents, Transbond XT and AEGIS ORTHO light-cure composites, were evaluated for their impact on white spot lesions (WSL) in orthodontic patients. Ten subjects were cross-sectionally randomized into two groups: Group A received treatment with Transbond XT, while Group B received treatment with AEGIS ORTHO. A total of 20 samples were collected from each subject, consisting of teeth on both the upper and lower sides. The primary outcomes of interest were WSL severity (assessed via SEM examination) and white spot lesion severity.

## A. Basic Characteristics:

The age range of all subjects was 14 to 23 years, with a mean age of  $18.30 \pm 2.98$  years and a median of 18.0 years. Among the subjects, there was an equal distribution of 5 males (50.0%) and 5 females (50.0%).

#### **B. Primary Outcome Measures:**

#### I. WSL Severity:

Table 1 presents a comparison of WSL severity scores in Group A and Group B before and after bonding. In both groups, WSL severity increased after debonding, indicating demineralization.

Table 1: WSL severity (Mean  $\pm$  SD) of two groups at before bonding and after debonding

Groups	Before bonding	After debonding	Change	W	p
	(n=20)	(n=20)	(After-Before)	value	value
Group A	$0.80 \pm 0.77$	$1.40 \pm 0.68$	$0.60 \pm 0.50$	78.00	0.001
Group B	$0.80 \pm 0.77$	$1.00 \pm 0.65$	$0.20 \pm 0.41$	10.00	0.125
U value	200.00	136.00	120.00	-	-

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P value 0.989 0.082 0.028	
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When analyzing the mean WSL severity within each group (Table 1 and Fig. 1), the Wilcoxon test revealed a significant increase in WSL severity after debonding compared to before bonding in Group A  $(0.80 \pm 0.77 \text{ vs. } 1.40 \pm 0.68, \text{W} = 78.00, \text{p} = 0.001)$ . In contrast, Group B did not show a significant increase in WSL severity after debonding compared to before bonding  $(0.80 \pm 0.77 \text{ vs. } 1.00 \pm 0.65, \text{W} = 10.00, \text{p} = 0.125)$ .

Comparing the mean WSL severity between the two groups (Tables 1 and 2) (Figures 1 and 2), the Mann-Whitney test revealed similar WSL severity at both time periods (before bonding:  $0.80 \pm 0.77$  vs.  $0.80 \pm 0.77$ , U = 200.00, p = 1.000; after debonding:  $1.40 \pm 0.68$  vs.  $1.00 \pm 0.65$ , U = 136.00, p = 0.082). However, when examining the net mean change (after debonding minus before debonding) in WSL severity for both groups (Table 1 and Fig. 3), the Mann-Whitney test revealed a significantly different result, with a 23.0% higher degree of demineralization in Group A compared to Group B (0.60  $\pm$  0.50 vs.  $0.20 \pm 0.41$ , U = 120.00, p = 0.028).

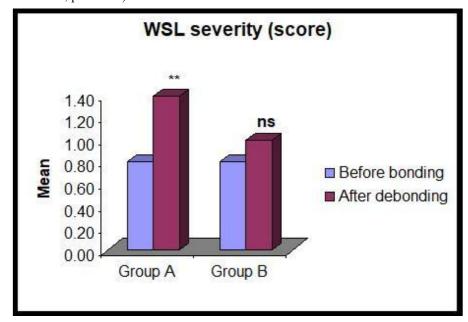


Fig 1- Mean WSL severity within the two groups.

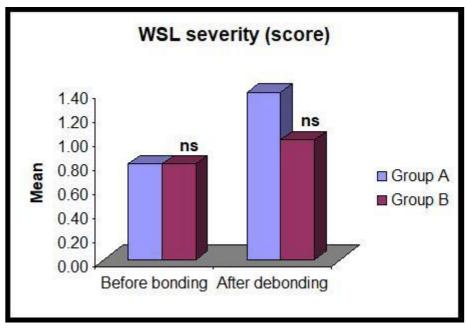


Fig 2- Mean WSL severity between the two groups.

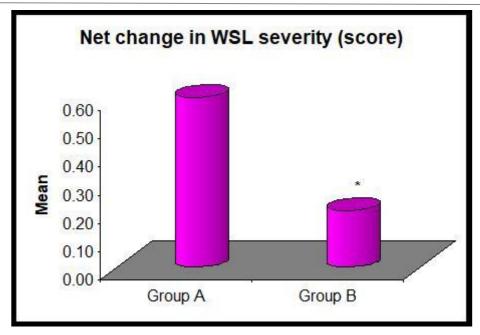


Fig 3- Net mean change (after debonding – before debonding)

Table 2: Frequency distribution of WSL severity of two groups

WSL severity	Group A (n=20)	Group B (n=20)	χ² value (DF=1)	p value
No WSL	8 (40.0%)	16 (80.0%)	6.67	0.010
Slight	12 (60.0%)	4 (20.0%)		
Severe	0 (0.0%)	0 (0.0%)		

The net mean change scores for WSL severity (score) were further categorized as "No WSL" (0), "Slight" (1), and "Severe" (2), and the results are summarized in Table 2 and presented in graphical form in Fig. 4. At the final evaluation, 40.0% of the subjects in Group A had "No WSL" (no severity), and 60.0% had "Slight WSL." In contrast, in Group B, 80.0% had "No WSL," and 20.0% had "Slight WSL." Notably, there were no cases of "Severe WSL" in either group. A  $\chi 2$  test revealed a significant difference between the groups, with 40.0% more instances of "No WSL" in Group B compared to Group A (p = 0.010).

## II. White Spot Lesion Depth (mm):

White spot lesion depth data were available for four subjects in each group and are summarized in Table 3 and shown graphically in Fig. 5. The white spot lesion depth in Group A ranged from 0.024 mm to 0.042 mm, with a mean depth of  $0.033 \pm 0.006$  mm. In contrast, Group B had a depth range of 0.004 mm to 0.005 mm, with a mean depth of  $0.011 \pm 0.004$  mm. Comparing the mean white spot lesion depth between the two groups, a t-test revealed a significant difference, with Group B having a 65.7% shallower white spot lesion depth compared to Group A  $(0.033 \pm 0.006$  mm vs.  $0.011 \pm 0.004$  mm, t = 8.58, p < 0.001).

Table 3: White spot lesion depth (Mean  $\pm$  SD) of two groups

Group A (n=8)	Group B (n=8)	t value (DF=6)	p value
$0.033 \pm 0.006$	$0.011 \pm 0.004$	8.58	0.014

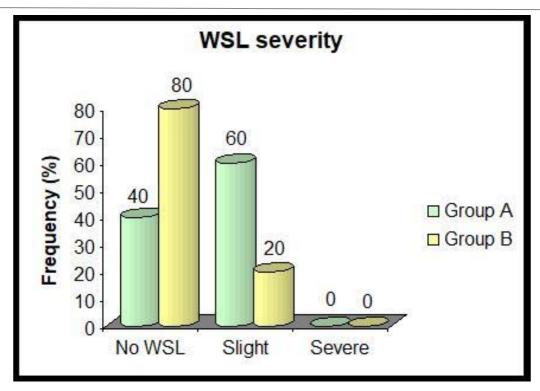


Fig 4- Frequency distribution of WSL severity of two groups.

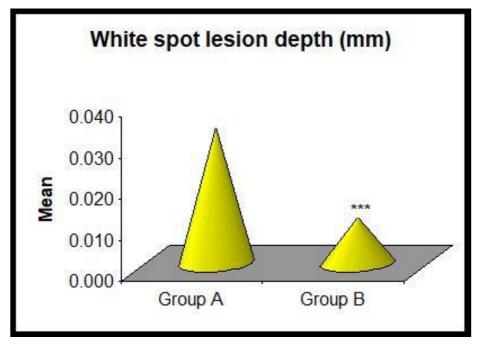


Fig 5- Mean white spot lesion depth of two groups.

#### 4. DISCUSSION:

WSLs adjacent to orthodontic brackets are a multifactorial problem that warrants a thorough evaluation and deliberation when it comes to preventing them. [9]

There is an intrinsic link between WSLs and dental plaque dynamics, which is a complex biofilm consortium harbouring a wide variety of microorganisms. This milieu is characterised by acidogenic bacteria, such as Streptococcus mutans, which cause pH reduction and demineralisation of enamel by catalysing the fermentation of dietary carbohydrates. In orthodontic brackets, retentive niches encourage biofilm growth, resulting in localised plaque burdens and acidogenic potential when biofilm accumulates.<sup>[10]</sup>

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When left untreated, WSLs can cause structural damage and cavitation. Additionally, microbial colonies can re-establish themselves on the compromised enamel surface. It compromises orthodontic therapy's intended cosmetic enhancement because these opalescent lesions contrast starkly with the enamel surrounding them.

A pivotal element of preventive dentistry is Casein phosphopeptide-ACP (CPP-ACP), which has exhibited topical anticariogenic effects due to its ability to stabilise calcium and phosphate in an amorphous state. The inclusion of CPP-ACP into dental plaque results in a significant escalation in the levels of plaque calcium and phosphate ions. This mechanism proves highly efficacious in mitigating enamel demineralisation, as an inverse relationship appears to exist between levels of plaque calcium and phosphate and the incidence of caries. Subsequently, the localised application of CPP-ACP serves to equilibrate free calcium and phosphate ions present within the plaque fluid, thereby sustaining a state of ACP over-saturation concerning enamel mineral. As a consequence, this approach effectively restrains enamel demineralisation while concurrently augmenting the remineralisation process. [11]

The findings of the current study echo those of a study conducted by Featherstone and colleagues (2003) [12], in which the crucial role of bacterial acids in the demineralisation of enamel was highlighted. According to their research, biofilms may have a high acidogenic potential, which can affect enamel integrity by causing acidification. This is in line with the current study's focus on the interaction between orthodontic appliances, plaque dynamics, and enamel dissolution as part of the current study.

A study published by Tufekci et al. (2015) [13] has also placed emphasis on the significance of orthodontic bracket design in retaining plaque and the development of wind-shield lesions in relation to plaque retention. Their research revealed that the bracket design had a significant impact on plaque accumulation and, in turn, on the risk of demineralisation for the patient. This echoes the current study's emphasis on the surface characteristics and design of orthodontic attachments as contributing factors to WSL formation.

The idea of "smart composites" based on amorphous calcium phosphate (ACP) aligns well with the research conducted by Meyer et al. (2017) [14]. ACP-containing materials were evaluated for their remineralisation potential and it was demonstrated that they could enhance enamel remineralisation by enhancing the remineralisation of enamel. This supports the current study's approach of using ACP-based bonding agents to mitigate WSLs.

Among the various methods of assessing WSLs, backscattered scanning electron microscopy, or SEM, has been used extensively in this study that is highly related to the work that was conducted by Tufekci et al. (2013) [15]. After the enamel had been debonded, they used SEM in order to analyse the surface characteristics of the enamel. Similar to the present study, quantitative measurements of demineralisation were carried out using SEM to provide comparable data on the extent of demineralisation.

Paris et al. (2017) [16] investigated the impact of different ligation methods on the development of a WSL, emphasising the role that archwire ligation plays in the formation of a lesion. Accordingly, the current study's assertion that the method of ligation of the archwire plays a significant role in the development of WSL is corroborated by these empirical data.

In future research, Wang et al. (2018) suggests incorporating antimicrobial agents into orthodontic materials <sup>[17]</sup>. Using quaternary ammonium compounds in orthodontic adhesive resins may inhibit biofilm formation and dental caries.

The current study aimed to compare white spot lesions (WSL) around orthodontic brackets using different bonding agents. The participants were split into Group A (Transbond XT) and Group B (AEGIS ORTHO). Both groups experienced increased WSL severity after debonding, with higher demineralisation in Group A. Within Group A, there was a significant increase in WSL severity after debonding, while Group B showed no significant change. Comparing mean WSL severity between groups showed no significant difference before and after bonding.

However, analysing the net mean change in WSL severity revealed 23.0% higher demineralisation in Group A compared to Group B. Group A had 40.0% with no severity and 60.0% with slight severity, while Group B had 80.0% with no severity and 20.0% with slight severity. The  $\chi^2$  test showed significantly higher no WSL cases in Group B compared to Group A.

In terms of white spot lesion depth, Group B had significantly lower lesion depth compared to Group A, with a 65.7% reduction.

The study concludes that Group B (AEGIS ORTHO) showed better outcomes in terms of both WSL severity and lesion depth compared to Group A (Transbond XT).

#### 5. CONCLUSION:

In conclusion, this study examined the impact of different bonding composites on WSLs in orthodontic patients. By utilizing scanning electron microscopy and image analysis software, the extent and depth of WSLs were quantitatively assessed. The findings revealed that Aegis Orthodontic Bonding Light Cure Composite with Amorphous Calcium Phosphate (AEGIS ORTHO) showed better outcomes compared to Transbond XT in terms of WSL severity and lesion depth. These results underscore the significance of bonding materials in orthodontic treatments and their potential effects on tooth health during

and after orthodontic interventions. Further research can build upon these insights to enhance bonding practices and improve the overall quality of orthodontic care.

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