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Comparison Of Three Fluoride Dentifrice Products In The Prevention Of White Spot Lesions In Orthodontic Treatment

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COMPARISON OF THREE FLUORIDE DENTIFRICE PRODUCTS IN THE PREVENTION OF WHITE SPOT LESIONS IN ORTHODONTIC TREATMENT

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ABSTRACT

Introduction:. During the course of orthodontic treatment, enamel decalcification is a common problem. Demineralized enamel, the precursor to caries formation, can be attributed to fixed orthodontic appliances and prolonged exposure to bacterial plaque. Progression to clinically detectable white spot lesions may occur as early as one month following the placement of orthodontic appliances. Such problems have influenced clinicians to search for a solution. Because fluoride treatment immediately upon debonding is not advocated, clinicians have proposed fluoride treatment and fluoride-releasing materials at the commencement of the therapy. The two new anti-cavity toothpastes, Clinpro^{TM} 5000 with 1.1% Sodium Fluoride and Clinpro^{TM} Tooth Crème with 0.21% Sodium Fluoride have been shown to be useful in reduction of white spot lesions. Both the Clinpro ^{TM} products (3M ESPE) contain fluoride as well as tri-calcium phosphate, which are components naturally found in saliva. The aim of this study is to determine if Clinpro^{TM} 5000, Clinpro^{TM} Tooth Crème, or MI-Paste Plus has an effect on the formation and resolution of white spot lesions for patients undergoing orthodontic treatment. **Methods:** Three prospective groups of 40 patients undertaking routine orthodontic treatment were evaluated (total recruitment of 120 subjects). Patients must have their permanent dentition, be 12 years or older, and have not used extensive fluoride regimes before. Patients were chosen if they appear that

they would be compliant as judged by the investigator. The selected product was brushed on the teeth by the patient for two minutes, twice daily for a total of 4 months. After brushing, the patients were instructed to expectorate only, not rinse, eat or drink anything for at least 30 minutes. The subjects were observed every 4 weeks. At each visit, 3 intraoral photos were taken (frontal and buccal views) and measurements based on the Enamel Decalcification Index (EDI) will be recorded. The Enamel Decalcification Index was used to determine the number of white spot lesions and caries risk detected in these photographic records. Also, a brushing diary was reviewed. A trained member of the 3M ESPE Clinical Research group did monitor the study by means of visits to the clinic to evaluate patient charts, study data, and study photographs. **Results:** Altogether, the results provided strong support with regard for Clinpro™ 5000 providing superior enamel protection against decalcification when compared to Clinpro™ Crème, and MI Paste. **Conclusions:** The use of Clinpro™ 5000, Clinpro™ Crème, and MI paste demonstrated less formation of white spot lesions when compared to data in the literature. Clinpro™ 5000 had a marginally better affect compared to the other two products.

Keywords: white spot lesion, demineralization, fluoride dentifrice, casein phosphopeptide amorphous calcium phosphate, tri-Calcium Phosphate, orthodontic treatment.

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LIST OF ABBREVIATIONS

WSL	white spot lesion
wt%	percentage by weight
CPP-ACP	casein phosphopeptide amorphous calcium phosphate complex
fTCP	functionalized β-tricalcium phosphate
TCP	tricalcium phosphate
ADA	American Dental Association
ACP	Amorphous Calcium Phosphate
NaF	Sodium Fluoride
ppm	part-per million
QLF-D	Quantitative light-induced fluorescence
EDI	Enamel Decalcification Index
SnF2	Stannous Fluoride

CHAPTER 1

INTRODUCTION

During the course of orthodontic treatment, the practitioner normally faces two common iatrogenic treatment side effects: root resorption and enamel decalcification, with the latter occurring at a much higher frequency. While the processes that lead to enamel demineralization are well understood, methods to diminish or perhaps eliminate degradation of enamel surfaces are being pursued. Several approaches have been formulated to "counteract" demineralization of tooth structure. One approach involves patient compliance and consists of in--depth oral hygiene instructions, in--office fluoride applications, and at--home fluoride rinses, gels, and varnishes. An alternative approach, which possesses potential benefit regardless of patient compliance, includes the use of fluoride--releasing agents, such as composites, glass ionomers, sealants, and elastomeric ties.

Enamel decalcification or white spot formation, is a phenomenon occurring primarily on smooth enamel surfaces of teeth, notably within the gingival third of the crown.[1] Demineralized enamel, the precursor to caries formation, can be attributed to debris trapped on fixed orthodontic appliances[2], and prolonged exposure to bacterial plaque[3]. Bacterial plaque promotes the accumulation of acidic byproducts and demineralization that leads to successive changes in the optical properties of subsurface demineralized enamel. Progression to clinically detectable white spot lesions may occur as early as one month following the placement of orthodontic appliances.[4-6]

Over the past thirty years, numerous studies have reported an increase in white spot lesions following orthodontic treatment.[6-9] While a large portion of the non-orthodontically--treated population experiences some form of decalcification, orthodontically treated patient populations have shown both an increase in new lesions and an increase in the severity of pre-existing enamel opacities.[4-8] Approximately 50 percent of all orthodontically treated patients develop white spot lesions in one or more teeth, compared with only 24 percent in those not undergoing orthodontic treatment. [6-9]

Appliance removal drastically reduces white spot lesion formation and many cariogenic factors, diligent oral hygiene efforts tend to inactivate incipient lesions which may undergo regression over time.[1, 10, 11] Complete elimination of lesions is unlikely due to the rapid remineralization of the enamel surface with high concentration fluorides, which restrict passage of ions into the deeper, more affected layers. Therefore, immediate application of high concentration of fluoride is not recommended. Reduction in the white spot lesion appearance occurs as a result of gradual remineralization of the tooth surface from the inside out. .[1]

Two new anti--cavity toothpastes, Clinpro™ 5000 with 1.1% Sodium Fluoride and Clinpro™ Tooth Crème with 0.21% Sodium Fluoride, are currently available and have been shown in some initial case reports to be useful in the reduction of white spot lesions. Both the Clinpro™ products are advanced formulas containing an innovative tri-calcium phosphate ingredient. These products are available exclusively from 3M ESPE. Clinpro™ contains fluoride as well as calcium and phosphate, which are components naturally found in saliva.

This proprietary formula successfully integrates these components, enhancing,

rather than compromising, the product's performance. During the manufacturing process, a protective barrier is created around the calcium allowing it to coexist with the fluoride ions. Think of this as a coated bubble that transports the Tri-Calcium Phosphate to the teeth. As the toothpaste comes in contact with saliva during brushing, the bubble coat breaks down and allows the calcium, phosphate and fluoride to be readily available to the tooth surface. The tooth naturally absorbs these components, preventing further progression of demineralization and promoting remineralization.

MI Paste Plus has been shown in recent reports to be useful in the reduction of white spots lesions.[12-14] There are no studies in the current literature in which these products are tested together during the orthodontic treatment.

Aim of Study

Determine if Clinpro™ 5000, Clinpro™ Tooth Crème or MI-Paste has an effect on the formation and resolution of white spot lesions for patients undergoing orthodontic treatment.

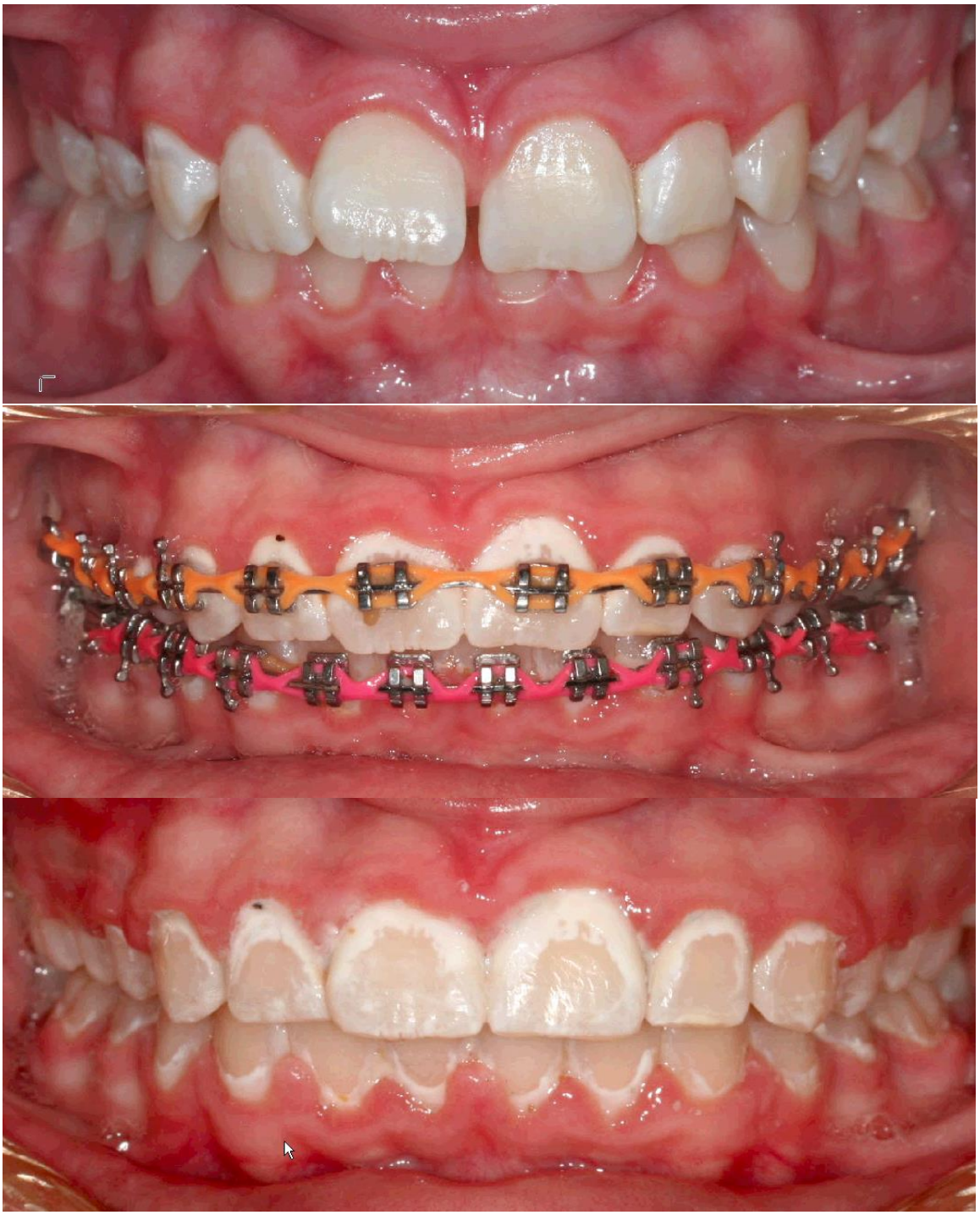

Figure 1. Clinical example of before orthodontic treatment (top) during orthodontic treatment (middle) and after orthodontic treatment (bottom) in a patient exhibiting severe enamel demineralization.

CHAPTER 2

LITERATURE REVIEW

Enamel demineralization has been reported as an unfortunate sequel of orthodontic treatment for nearly a century. The presence of numerous brackets, bonding material, arch wires, and ligatures make proper brushing and flossing difficult.[7] Ineffective removal of food debris and bacterial plaque can result in the buildup of *Streptococcus mutans* and *Lactobacilli spp*, which are sources of organic acid responsible for enamel demineralization. [15] These acids result in enamel caries formation (white spot lesions). Once fixed appliance therapy is concluded, these enamel lesions have the potential to remineralize overtime.

Prevalence of enamel demineralization in orthodontic patients

Numerous studies regarding enamel demineralization report varied frequencies within the general and orthodontic populations. The reported prevalence of white spots after fixed appliance treatment varies between 2 and 96 per cent.[8] These differences can be attributed to factors, such as diverse demographics, banding versus bonding, emphasis on oral hygiene, use of fluoridated water and other methods of fluoride delivery, and varying methods of analysis. However, several trends can be drawn from these studies. First, it is generally reported that white spot lesions can occur in as many as 50 percent of teeth with orthodontic appliances and in up to 50 percent of treated patients.[8] Second, white spot lesions are more pronounced at the gingival third of the crown of the tooth, where plaque accumulates. [16]Finally, bracketed orthodontic therapy tends to increase the prevalence of white spot lesions when compared to untreated control groups.[15]

Orthodontically treated patients show significant numbers of new areas of enamel opacities when compared to controls. Gorelick *et al.* reported 49.6 percent of de-bonded patients showed white spot formation on at least one tooth compared with only 24 percent of non-orthodontically-treated controls.[8] Similar to these findings, decalcifications in their control groups of orthodontic patients were found to be 58 percent. [17] Artun *et al.* concluded that with proper oral hygiene instructions, reinforcement, fluoride prescription usage, and removal of excess adhesive from multi-bonded appliances, there is no significant difference between treated and untreated individuals with respect to white spot lesion formation.[1]

There is certain diversity in locations for white spot lesions. The distribution of lesions is different between treated and control populations, with buccal and lingual surfaces being more susceptible in treated patients. Gorelick *et al.* reported control teeth most frequently affected by enamel demineralization are maxillary central and lateral incisors, followed closely by mandibular molars. However, during orthodontic treatment, maxillary lateral incisors tended to be most susceptible to decalcification, followed by mandibular canines and premolars.[8] Ogaard found that first molars showed the highest prevalence of decalcification in both control and treated groups. The small area of tooth structure located between the bracket and gingival margin is conducive to plaque retention and has decreased accessibility to oral hygiene measures, allowing for white spot formation. Interestingly, white spot formation does not tend to differ significantly with respect to banded versus bonded appliances. In addition, white spot formation with incisors and canines adjacent to bonded lingual retainers has not proven to be more susceptible.[15]

Etiology of enamel demineralization, the early caries process

[18] Dental caries begin when an acidic environment (low salivary and plaque pH, which favors cariogenic bacteria) is present. This unfavorable environment leads to the dissolution of the hydroxyapatite crystal structure of enamel. Following a period of acidic attack, the salivary and plaque pH stabilizes, and a period of remineralization occurs. These periods of demineralization and remineralization are part of a dynamic pattern of the oral environment and the caries process. [19] Presences of food debris, refined carbohydrate content of the diet, fluoride availability, mineral crystal composition, and salivary content are all factors that can influence the length of these acidic assaults (demineralization) and stabilization (remineralization) periods. [20]

Enamel solubility is affected by the composition, size, shape and packaging of the hydroxyapatite crystals making up the enamel.[21] Orthodontic treatment often begins during the mixed dentition period, when several teeth are erupting into the oral cavity. Soon after eruption, teeth will undergo post-eruptive maturation that changes the composition of the surface enamel.[22] Dental enamel is comprised chemically of approximately 96wt% of mineral, similar to hydroxyapatite, $Ca_{10}(P0_4)_6(OH)_2$. The chemical and structural nature of an apatite mineral are such that they allow many substitutions in its crystal lattice without losing the essentials of its identity. Carbonate can occupy the phosphate positions in the lattice, and both carbonate and fluoride may be substituted for hydroxyl ions. Approximately 2to5% of enamel mineral is carbonate. These substitutions change the dimensions of the unit cell[23-25] of the crystal, as well as the solubility product of the apatite.[26-28]

Maturation involves the loss of caries-susceptible carbonate-rich hydroxyapatite.

Carbonate is replaced by mineral phases that are less soluble and have higher fluoride content.[21] When present, fluoride ions may replace carbonate ions and become incorporated into the enamel hydroxyapatite structure to form fluoridated hydroxyapatite, a much more stable structure. More than 40 trace elements can be incorporated into the enamel and affect its solubility beneficially or adversely.[29] During the post-eruptive process, enamel is quite susceptible to carious attack.[22]

Enamel structure consists of long, thin hydroxyapatite crystals bundles together in rods or prisms. An organic matrix, comprising the prism sheath, surrounds these crystals. Larger and more uniform crystals that are closely packed demonstrate less enamel solubility. Water located between enamel crystals acts as a diffusion channel for acidic byproducts from bacteria to attack the enamel crystals. Therefore, tightly packed crystals offer more resistance to an acidic attack than loosely bound crystals.

Acids involved in the caries process are produced by plaque colonies located on either the dentition or orthodontic appliances.[30] Frequent consumption of fermentable refined carbohydrates favors the presence of cariogenic bacteria. These bacteria metabolize carbohydrates and generate organic acids as byproducts. Carbohydrates are broken down into sucrose, which aids in the synthesis of extracellular polysaccharides and favors accumulation of *Streptococci mutans and Lactobacilli* in dental plaque.[15]

The first stage of enamel demineralization involves surface softening, where preferential removal of inter-prismatic substance within the enamel surface occurs. Organic acids present in bacterial plaque create minute pores in the enamel surface that act as passageways for enamel dissolution. Larger quantities of acid produced by bacteria cause lowering of the pH, which in turn results in more rapid diffusion of acid into

enamel. These acids dissociate in the enamel and produce hydrogen ions, which solubilize the calcium phosphate mineral phases of the enamel. The second stage of demineralization typically dominates the first stage and can be seen as dissolution in the deeper part of the enamel. Free calcium and phosphate ions diffuse from sites deep within the enamel to the tooth surface. When this process continues for an extended period of time, frank cavitation will occur.[31]

Salivary composition and flow rate are also important host factors. Saliva provides mechanical cleansing properties to the tooth surface and dilutes and buffers acids. Moreover, saliva contains antimicrobial agents, and organic and inorganic components, which act to inhibit demineralization and assist remineralization.[19]

The white spot lesion is the first clinically observed manifestation of the caries process. It represents an area of demineralized enamel that may have lost up to 50 percent of its mineral content.[32] It appears histologically as an area of subsurface demineralization covered by a relatively intact enamel surface. A study using polarized light microscopy illustrate that the white spot lesion consists of four distinct histological zones. [33]

Placement of fixed orthodontic appliances increases plaque accumulation and initiates changes in plaque composition. [15] Up to 15 percent mineral loss can occur around orthodontic brackets in only one month.[4] Orthodontic therapy results in a continuous cariogenic challenge with an extremely rapid process of demineralization.

Moreover, there seems to be a difference in progression rate between traditional caries formation and white spot lesions induced by deficient oral hygiene combined with fixed orthodontic appliances. The latter has a rather superficial and more rapid character

and can become apparent within 1 month after placement of fixed appliances. The formation of a 'normal' caries lesion is usually a slower process, which takes at least 6 months. [34]

Nevertheless, upon completion of bracketed orthodontic therapy and appliance removal, further progression of the lesions may be prevented and potentially reversed if appropriate oral hygiene and fluoride treatment are initiated. [11, 34]

Remineralization of early caries lesions

Following loss of minerals during caries attacks, re-deposition of essential minerals can occur (remineralization). When salivary pH returns to a value greater than 5.5, soluble calcium, phosphate and fluoride derived from plaque, saliva and dissolved tooth mineral may be transformed into hydroxyapatite and fluoro-hydroxyapatite and re-precipitated into the demineralized enamel. Fluoride is redistributed as it is released from the advancing front of the white spot lesion and re-deposited as fluoride containing mineral in the surface zone and superficial body of the lesion. This process results in a less soluble, more acid resistant mineral with a lower critical pH. [29]

Following removal of orthodontic appliances, white spots lesions can remineralize, decrease in size and return to normal enamel luster.[11, 34] The remineralization process appears to follow an exponential pattern, whereby mineral recapture is accelerated in the first few months and then continues at a much lower rate.[5] Marcusson *et al.* found that lesion size decreased over a 2-year observation period.[10] Al-Khateeb *et al.* also reported a decrease in lesion size over a 1-year follow-up period. [11] Together remineralization and surface abrasion of tooth structure may result in clinical resolution of lesions.

Fluoride is known to increase the rate of remineralization, but complete repair is inhibited by precipitation of fluoride into the surface layer. Surface lesions remineralize more rapidly and more completely than subsurface lesions, and this process is accelerated by higher concentrations of fluoride. The presence of concentrated fluoride will arrest lesions and not allow for complete repair. This occurs because fluoro-hydroxyapatite is formed in the surface zone in the presence of calcium fluoride derived from high concentration fluoride rinses and gels. This fluoridated apatite is less soluble and may block diffusion of mineral into the deeper layers of the lesion.[5, 35-37] Therefore, during the repair of established lesions, slow-release of low fluoride concentration agents are advocated.[38, 39]

Role of fluoride in enamel demineralization and remineralization

Preventive fluoride therapy in addition to fluoridated dentifrice usage is imperative in contemporary orthodontic practices, and has proven to inhibit caries formation and decrease enamel demineralization. The presence of fluoride at the time of acidic attack may considerably slow rates of demineralization.[40] During both periods of demineralization and remineralization, enamel lesions preferentially absorb fluoride ions onto their partially demineralized hydroxyapatite crystals or redeposit fluoride as fluoridated hydroxyapatite. Fluoride's ability to prevent and arrest the caries process is dependent upon three mechanisms of action: 1) inhibiting demineralization when fluoride is present at the crystal surfaces during an acidic challenge, 2) inhibiting bacterial metabolism after diffusing into the bacteria as hydrogen fluoride when plaque is acidified, and 3) enhancing remineralization and forming a low-solubility fluoridated hydroxyapatite.

Not only can fluoride decrease demineralization, it can also remineralize existing early white spot lesions.[40] However, the ideal concentrations of fluoride should be considered. High levels of fluoride used to treat existing white spot lesions will only remineralize the surface layer of the lesion.[5, 35-37] This superficial layer might prevent calcium and phosphate from penetrating to the deeper layers of the enamel, thus inhibiting deeper remineralization and limiting the cosmetic improvement of the WSLs.[38, 39] Thus, low fluoride levels should be used in order to promote remineralization of the entire depth of the lesion. The goal in oral hygiene promotion should be frequent low concentrations fluoride exposure to prevent demineralization and to completely repair existing lesions.

Daily fluoride rinse

The use of fluoride dentifrice alone is insufficient to inhibit lesion development around orthodontic brackets. Most orthodontic patients cannot achieve basic tooth brushing that is meticulous enough to prevent enamel decalcification. Insufficient oral hygiene measures lead to a buildup of plaque with increased levels of *Streptococcus mutans* and *Lactobacilli spp*.[15] To counteract poor tooth brushing skills, many practitioners supplement their oral hygiene regimens with daily fluoride rinses and topical fluoride treatments. In-office topical fluoride treatments have been suggested to minimize the need for patient compliance.

Even with fluoridated toothpaste use two or three times per day, measurable demineralization occurs in 9 out of 10 patients in as little as one month. Clinical studies have demonstrated addition of daily sodium fluoride, stannous fluoride, or acidulated phosphate fluoride rinsing retard lesion development during orthodontic therapy

significantly. Geiger *et al.* reported a 21% reduction in the number of patients with white spot lesions while adhering to a daily fluoride rinse program.[41] Ogaard *et al.* reported fluoride rinsing reduced lesion depth by a factor of three and decreased mineral loss by 60%. The frequency of fluoride application, not the fluoride concentration, is most important in preventing lesions and limiting the extent of lesion formation. Fluoride application on a daily basis should provide enhanced benefit over less frequently used methods. Additionally, fluoride-rinsing programs augment the protection seen in children daily fluoride from their drinking water.

Fluoride gels and foams

Fluoridated gels and foams are commonly used both in offices at higher concentrations, and at home at lower concentrations. These are primarily used after nightly tooth brushing. The at-home gel is typically a 0.4%-1.1% solution of stannous fluoride (SnF2). Stratemann and Shannon reported brushing with SnF2 gel for 28 days more than doubled the fluoride concentration in the outer enamel surface. Also they found only 2 percent of patients using daily fluoride gel developed white spot lesions, while 58 percent of those without daily gel usage formed lesions. [17] Studies report that the use twice daily of SnF2 gel inhibited demineralization to a greater degree than once daily sodium fluoride (NaF) rinse.[17, 41] However, this difference may be explained by the fact that SnF2 gel was applied more frequently than the NaF rinse. Frequent applications of low concentrations of SnF2 have proven to reduce the solubility of tooth structure. Moreover, stannous fluoride displays an antibacterial effect that results in a significant reduction in plaque accumulation clinically. [17]

Studies have shown in-office application of acidulated phosphate fluoride (APF) and amine fluoride gels reduce the amount of visible decalcification in clinical settings. Dimitriadis and Sassouni reported a decrease in demineralization under orthodontic bands when topical APF gel was applied.[42] Garcia-Godoy *et al.* showed in-office application of 1.23% APF gel for 1 minute produced a 37 percent reduction in the depth of the body of the lesion. [35] In-office fluoride gels and foams delivered via foam trays at monthly visits, while beneficial, fall short of the protection offered to patients by topical fluoride delivered on a daily basis.

Fluoride varnish

To counteract lack of patient compliance with fluoride dentifrice and mouthwashes, clinicians can apply topical fluoride varnishes at regular intervals. Studies demonstrate a 35-50 percent reduction in enamel demineralization when fluoride varnish containing 5% sodium fluoride is applied following bracket cementation with composite resin.[43] The benefit of fluoride varnish use includes longer contact time with enamel due to enhanced adherence to tooth structure resulting in increased length of time available to incorporate fluoride into the enamel surface. This longer exposure period to fluoride increases the amount of fluoride retained in enamel, enhances the formation of fluoridated hydroxyapatite, and reduces the acid solubility of enamel.[44] Fluoride varnish may be necessary in patients with strong gag reflexes, where delivery of fluoride in trays is impossible. An additional benefit of fluoride varnish is that a prophylaxis to remove plaque is not required before application and varnish is not inactivated by dental plaque.[45] Disadvantages of fluoride varnish are temporary discoloration and increased appointment time per patient. Reapplication at regular intervals is necessary due to removal of the

varnish by mechanical brushing. Its use may be necessary in those high-risk patients unable to carry out proper oral hygiene. Though the majority of patients find the presence of varnish on their teeth acceptable, some patients dislike its presence as a thin film on their teeth or they find the taste of the varnish objectionable. [46] Also, some orthodontists are reluctant to use fluoride varnish due to the increased chair time required for varnish application.

Patient compliance during orthodontic treatment

With all the preventive protocols at the disposal of the orthodontist, the major problem seen is lack of patient compliance. There are numerous reports demonstrating the benefits of proper oral hygiene and fluoride treatment on inhibiting dental caries and re-mineralizing enamel lesions, but patients fail to follow dental professionals' advice. Compliance is of great concern because data demonstrate that more severe white spot formation occurs in fixed appliance therapy of over 24 months duration. In these situations, orthodontists are obligated on occasion to prematurely terminate therapy with noncompliant patients.

In a clinical investigation of 101 patients designed to study compliance with home fluoride therapy, Geiger *et al.* demonstrated a significant association between diminished decalcification with compliance. Patients with poor compliance developed white spot lesions in 64.7 percent of cases, while only 11.8 percent of those with excellent compliance developed decalcification. The same study reported greater than 50 percent of patients had poor compliance, while only 26.7 percent had excellent compliance.[47] A similar study exhibited a compliance rate of only 13 percent with patients asked to decrease their caries risk with daily fluoride mouth rinse. [41] Stratemann and Shannon also

had disappointing compliance rates in a study involving daily fluoride usage. Only 2 percent of compliant patients developed white spot lesions, compared to 66 percent of noncompliant patients. Unfortunately only 48 percent of patients were compliant. [17]

The slow progression of enamel caries offers the opportunity for dental professional to diagnose and manage caries before there is irreversible destruction of the tooth.[19] Therefore, decalcification can be reduced greatly if clinicians can establish motivational methods for instilling compliance. Unfortunately, patients with poor oral hygiene are the group least likely to comply with proper oral hygiene and fluoride regimens, despite motivational efforts. This is why advances in fluoride-releasing bonding materials that are not dependent upon patient compliance are essential to the orthodontic specialty.

Fluoride-containing bonding materials

Although exact quantities of fluoride released from bonding agents are not presently well defined, many bonding materials have been proven to reduce early caries formation. One of the first materials clinically established to reduce secondary caries formation was glass ionomer cement. Upon setting, glass ionomer cements exhibit an acid-base reaction, which releases a variety of ions, including fluoride. This fluoride is available to precipitate into tooth surfaces adjacent to orthodontic bands and brackets and offer protection from demineralization via stabilization of hydroxyapatite crystals. Glass ionomer adhesives have also been shown to take up fluoride from dentifrices and rinses, and then release this fluoride into solutions, including saliva and dental plaque. They have been proven clinically useful for cementation of orthodontic bands, while decreasing both enamel mineral loss and *Streptococcus mutans* levels.[48] However, when

used for bonding brackets to enamel, conventional glass ionomers exhibit low bond strengths and lengthy curing times when compared with composite resins. Therefore, hybrid (resin-modified) glass ionomers and fluoride-releasing composite materials have been developed in an attempt to duplicate the fluoride-releasing capabilities of conventional glass ionomers. These materials have higher bond strengths and more rapid curing times.[49]

Resin modified glass ionomers (RMGIs) demonstrate a sustained fluoride release, absorb exogenous fluoride from fluoridated dentifrices and rinses, and release fluoride over time. Fluoride release from RMGIs is somewhat less than that of conventional glass ionomers, but their bong strengths are significantly higher. Vorhies *et al.* showed teeth bonded *in vitro* with resin-modified glass ionomer cements demonstrated significantly more reductions in enamel lesion size adjacent to orthodontic brackets than teeth bonded with a non-fluoride-releasing composite resin. [49] Schmit *et al.* reported teeth bonded *in vitro* with RGMI cement showed a 50 percent reduction in lesion depth when compared to those bonded with composite resin. [43]

Recent manufacturing trends have incorporated fluoride-releasing capability into composite resin adhesives. Underwood *et al*. found a 93 percent reduction in lesion depth verses controls when using fluoride-exchanging adhesive *in vivo*. Fluoride-releasing resin composites demonstrate a rapid release of large quantities of fluoride followed by a slow-release of low levels of fluoride over time. Nevertheless, even small amounts of fluoride targeted directly at the site of orthodontic appliances show great potential.[50] Similarly, matrix-bound fluoride-releasing adhesives (MBF, Rely-a-bond, Reliance Orthodontic Products, Inc., Itasca, I11) provide sustained release of low levels of fluoride *in vitro*,

which could decrease demineralization in the immediate vicinity of orthodontic brackets. This occurs by forming a protective layer of calcium fluoride-like particles on the enamel surface. Recently, Cain *et al*. showed Light Bond (Reliance Orthodontic Products, Inc. Itasca, IL), a fluoride-releasing bonding resin, resulted in a 38 percent decrease in lesion depth *in vitro*. [51] Materials that demonstrate higher initial fluoride released tend to show significant inhibition of enamel demineralization.

Glass ionomers and some fluoride-containing adhesives have the unique ability to be recharged with fluoride ions when exposed to topical fluoride sources. Studies have found simply brushing with a sodium fluoride toothpaste significantly increases fluoride ion release *in vitro* from bracketed teeth bonded with resin-modified glass ionomer, fluoride releasing composite resin, and even light-cured composite resin (to a smaller extent) when compared to un-bonded controls. These findings suggest that bonding agents and bonded enamel take up and releases fluoride ions obtained from toothpaste and rinses. [51] Furthermore, fluoride-release from bonding adhesives is greater at a low pH environment rather than in a neutral pH environment. The rationale for it is that the low pH favors calcium fluoride formation due to more available calcium ions [5]

Fluoride-containing orthodontic materials

Not all fluoride products and treatment are equal. Different fluoride compounds, different vehicles and vastly different concentrations have been used with different frequencies and duration of application. These variables can influence the clinical outcome with respect to caries prevention and management. [52] Furthermore, fluoride-releasing sealants and ligature ties have been suggested as an alternative method to

protect the susceptible area beneath and adjacent to bonded attachments, independent of patient compliance. Resin-based dental sealants have proven to resist caries development on smooth enamel surfaces. Similar bonding materials designed for use on smooth surfaces have been tested in prior clinical trials. For example, Resilience M5 Protection Plus (Confidence Dental Products Co., Denver, Colorado 80010), a resin-based dental sealant, was applied adjacent to 112 previously bonded brackets *in vivo* with the goal of creating a fluoride-releasing caries-resistant barrier. [53] Recently, Cain *et al*. in an *in vitro* investigation found that Pro Seal (Reliance Orthodontic Products, Inc., Itasca, IL), a fluoride-releasing resin modified glass ionomer sealant, reduced mean lesion depth by 43 percent compared with untreated controls.[51]

In addition to sealants, fluoride-releasing elastomeric ligature ties have been developed that contain SnF2. These elastomeric ties provide a large burst of fluoride release for about one week, followed by diminishing release over time during *in vitro* studies. When changed at regular intervals, these ties could provide significant fluoride release and reduce *Streptococcus mutans* levels at bonding sites. However, in their present composition, these elastomers need to be replaced on a weekly basis to counteract the large quantity of bacteria adherent to the bracketed teeth in order to be effective.[54]

Amorphous Calcium phosphate

ACP technology was developed in 1991 by the American Dental Association's (ADA) Paffenbarger Research Center. ACP contains the same minerals found in hydroxyapatite and aims, in the presence of fluoride, to speed up remineralization. ACP technology is considered unstablized because a calcium salt and a phosphate salt are delivered separately (eg, through a dual-barrel syringe). This delivery system allows for

the precipitation of ACP at the tooth surface. Because it is not a premixed calcium phosphate compound, when ACP is introduced onto a tooth surface, a reservoir of calcium and phosphate ions forms. Rapid deposition of new mineral then may fill surface defects on the original tooth surface.[55]

ACP is available in a variety of products, including dentifrices, prophy pastes, fluoride varnish, fluoride gels, pit and fissure sealant materials, desensitizing agents, cements, and tooth whitening agents. In a dentifrice, ACP, with fluoride, enhances remineralization and forms a strong bond to the dentin, becoming an intrinsic part of the tooth.[56] Sealants containing ACP promote in situ remineralization of artificially induced carious lesions on smooth enamel surfaces, although not significantly more than sealants containing fluoride.[57] Prophylaxis paste with added calcium, phosphate, and fluoride has the potential to form ACP on the tooth surface.[55] ACP-containing orthodontic composite resins may reduce enamel decalcification in patients with poor oral hygiene without damaging the cement's shear bond strength.[58] The addition of ACP in carbamide peroxide whitening agents may reduce transient tooth sensitivity caused by the whitening process.[59]

Most studies in support of ACP are animal model, in vitro, or in situ caries model studies. Although the use of ACP to assist in the remineralization process shows promise, more clinical trial research is needed. One clinical trial demonstrated a significant decrease in root caries among 44 high-risk head and neck radiation patients with the use of a dual phase ACP dentifrice containing 1,100 ppm sodium fluoride in comparison to a toothpaste containing 1,100 ppm sodium fluoride only.[60]

Casein Phosphopeptide Amorphous Calcium Phosphate

Casein (milk protein) was first investigated as a way to reduce caries as early as 1946.[61] CPP-ACP is referred to as stabilized ACP and is a complex of casein phosphopeptides that stabilize an amorphous form of calcium phosphate to maintain the calcium and phosphate ions, ensuring their delivery into the tooth structure before they precipitate or crystallize. CPP-ACP readily binds to the surface of the tooth as well as to the bacterial plaque surrounding the tooth.[62, 63] The CPP-ACP complex also acts as a reservoir of bioavailable calcium and phosphate. Under acidic conditions, CPP-ACP releases calcium and phosphate to enhance remineralization.

The use of a CPPACP cream has demonstrated significant regression of white-spot lesions in post orthodontic populations.[64 65] A recent in situ study using CPP-ACP combined with 900 ppm fluoride found that the combination offered a higher remineralization potential than CPP-ACP alone.[66]

MI Paste Plus showed in recent reports to be useful in the reduction of white spots lesions.[12-14]. The active agent, casein phosphopeptide-amorphous calcium phosphate (CPP- ACP), is thought to stabilize and localize calcium, fluoride, and phosphate at the tooth surface in a slow-release amorphous form, thus enhancing deeper remineralization of WSLs.[67] A number of in-vitro and in-situ studies have demonstrated the remineralizing potential of casein phosphopeptide-amorphous calcium phosphate.[68-77]

Tri-Calcium Phosphate

TCP is a new hybrid material created with a milling technique that fuses beta tricalcium phosphate (ß-TCP) and sodium lauryl sulfate or fumaric acid. This blending results in a "functionalized" calcium and a "free" phosphate, designed to increase the

efficacy of fluoride remineralization.[14, 78] ß-TCP, which is commonly used in FDA-approved orthopedic applications to boost bone growth, is similar to apatite structure and possesses unique calcium environments capable of reacting with fluoride and enamel. While the phosphate floats free, these exposed calcium environments are protected, preventing the calcium from prematurely interacting with fluoride.[78] TCP provides catalytic amounts of calcium to boost fluoride efficacy and may be well designed to coexist with fluoride in a mouth rinse or dentifrice because it will not react before reaching the tooth surface.[79] When TCP finally comes into contact with the tooth surface and is moistened by saliva, the protective barrier breaks down, making the calcium, phosphate, and fluoride ions available to the teeth. The fluoride and calcium then react with weakened enamel to provide a seed for enhanced mineral growth relative to fluoride alone. Products available with TCP includes Clinpro™ 5000 toothpaste and Clinpro™ Tooth Crème.

Summary

Prevention of white spot lesion formation appears simple: educate patients towards improved oral hygiene, utilize fluoride mouth rinses and gels, and brush and floss following every meal and before bedtime. Despite the importance placed on performing adequate oral hygiene and preventative measures, relatively few patients place enough value on routine oral hygiene measures. Therefore, clinicians are faced with providing optimal orthodontic care while avoiding enamel decalcification that may progress into frank cavitation. Until all patients are compliant with oral hygiene measures or alternative

methods for plaque control are developed that are not dependent upon active patient cooperation, enamel decalcification remains a considerable problem for orthodontists.

There are no studies in the current literature in which these products are tested among each other during the orthodontics treatment.

CHAPTER 3

MATERIALS AND METHODS

Study Design:

Three prospective groups of 40 patients who are undertaking routine orthodontic treatment will be evaluated as part of this research protocol (total recruitment of 120 subjects):

- Group 1: Clinpro™ 5000
- Group 2: MI-Paste Plus
- Group 3: Clinpro™ Tooth Crème

The study called for three groups with random assignment to each treatment group. Each group was evaluated as a protocol for the reduction of white spot lesions at the start of the orthodontic treatment. Subjects were recruited through the Orthodontic Postgraduate Clinic at the University of Alabama at Birmingham School of Dentistry.

The Enamel Decalcification Index (EDI) (Appendix A) was used to determine the caries risk of all patients enrolled into the study. Patients were carefully selected for the study and included and excluded on the following criteria:

Inclusion criteria:

1. Permanent dentition,

2. Patients that in the opinion of the investigator will be compliant with the use of the paste,

3. Patients who have not used extensive fluoride regimes,

4. 12 years and older,

5. Subjects must use a non-fluoridated toothpaste (such as Tom's of Maine) for a one-week period prior to starting this trial.

Exclusion criteria:

1) Any medical or dental condition that in the opinion of the investigator could impact study results during the expected length of the study,

2) Patient is currently using any investigational drug,

3) Patient plans to relocate or move within six months of enrollment,

4) Patients who have or are currently undergoing fluoride treatment for white spot lesions,

5) Patients with IgE Casein Allergy or known allergies to fluoride or other components of the test materials,

6) Pregnant women.

Test Materials:

1. Clinpro ™5000 1.1% Sodium Fluoride Anti--Cavity Toothpaste with Tri--Calcium Phosphate (3M ESPE, Saint Paul, MN, USA)
2. MI Paste Plus(GC America, Alsip, Ill)
3. Clinpro™ Tooth Crème 0.21% Sodium Fluoride Anti--Cavity Paste with Tri--Calcium Phosphate (3M ESPE, Saint Paul, MN, USA)

For further details of the test materials please refer to Appendix F.

Instructions for Paste Delivery:

In order to fully evaluate each of the products, the base line was standardized to

all patients. Prior to start, patients were instructed to brush for one week with a non-fluoride toothpaste. Next, the selected product was brushed on for two minutes twice daily for 4 months. After brushing on the product, patients should not rinse their mouths with water. Rather, they should just expectorate (spit) so they don't clear out the actives from the product. Patient should also not eat or drink for 30 minutes following the treatment.

Subjects were reviewed every 4 weeks and EDI scores recorded.

Randomization Method And Assignment Of Subject Numbers:

Randomization for this study was performed using the freely available software package called "GraphPad Software, Quick calcs" (GraphPad Software Inc.)This software constitute of a random number generator, which is seeded, with the time of day so it works differently each time you use it. Each subject is first assigned to a group non-randomly. Then the assignment of each subject is swapped with the group assignment of a randomly chosen subject. This should suffice, but the entire process is repeated twice to make sure it is really random. This process was done to randomize our study products and distribution (i.e., Clinpro™5000, MI-Paste Plus, or Clinpro™ Tooth Crème).

The order of assignment of the different products was recorded (Appendix G). Then, upon their recruitment in the study, participants were assigned chronologically to each study group, according to this randomly established order (Group 1- Clinpro™5000, Group 2-MI--Paste Plus, Group 3- Clinpro™ Tooth Crème. Subject numbers were therefore assigned in chronological order to each subject upon enrollment into the study (starting with 1, 2, and so on).

Administration of the Study:

Re-call Visits:

Subjects were examined every 4 weeks when they come for their appointment. At the end of each appointment, the next appointment was scheduled. Reminders were sent to the participants to confirm their visits. At each visit, the indices were calculated and 3 intraoral photos were taken (frontal and buccal views). The brushing diary was also reviewed.

Records:

The following two forms of evaluations were carried out:

(a) Photographic records

(b) Clinical examination

Photographic records:

Figure Photographic records were used to determine the presence and resolution in the white spot lesions in each study group. A standard intra-oral photographic camera (EOS Rebel T3-18-55 Macro lens- 58mm- with Macro Ring Lite MR-14EX II) was utilized and the photographs were taken in a light controlled environment and photographs were captured in a pre-set photographic protocol. The Enamel Decalcification Index - EDI score (Banks and Richmond) was used to determine the number of white spot lesions present at each time frame. (2) (Appendix A)

(b) Clinical Examination

IRB Approval:

IRB approval was obtained prior to study initiation. (Appendix H)

Patient Compensation:

Patients were compensated for participating in the clinical trial by one-time issuance of a $25 Visa gift card.

Informed Consent and Health Insurance Portability and Accountability Act (HIPAA):

Prior to enrollment, the Investigator explained the nature and intention of the study, procedures, and the expected duration participation to each potential study patient. Individuals had the opportunity to read and consider the Informed Consent form (which includes HIPAA information) and had the opportunity to ask questions. After receiving satisfactory answers, interested individuals voluntarily signed and dated the consent form, thereby granting their permission to enter the study. The Investigator or impartial individual who witnessed the consent process signed and dated each patient's consent form. A copy of the signed and dated Informed Consent form was provided to each patient who enrolled into the study. All signed consent forms were maintained in the study file.

Case Report Forms (CRFs):

Case Report Forms (CRF) is designed to record data pertinent to each subject enrolled in the study (Appendix B). This form includes the patient's number, the visit number, patient's photographs, EDI, and the teeth evaluated.

In addition to this form, the patient had to fill a brushing diary (Appendix C) that

was attached to the CRF, visit by visit.

Monitoring:

This trial was monitored for compliance with this protocol and applicable regulations. All Informed Consent forms were reviewed for signatures and dates. Patient charts/records was reviewed to ensure that all enrolled subjects meet the study inclusion and exclusion criteria; charts were also monitored for any safety-related issues. All CRFs were reviewed for accuracy and completeness of data entry to ensure that the study protocol is being followed, and to perform source data verification against information contained in the patient charts.

Patients Lost to Follow-Up:

Patients lost to follow-up are those enrolled in the study who do not complete all of the follow-up assessments. The investigator made at least three attempts to contact the patient. Reason for loss of follow-up was documented in the patient's record.

Reasonable Risk Associated with Study Participation:

There is a potential for a study participant to experience digestive problems or an allergic reaction if that individual has sensitivities to any ingredient of the test products. Additionally, there is a possibility that subjects may experience minor gingival sensitivity or discomfort while brushing their teeth. This is primarily dependent on each subject's brushing technique and not expected to be related to the products under evaluation. These events are expected to be localized and transient in nature.

None of these events was experienced during the study.

Parameters Measured:

The following parameters were measured in this study:

1. Enamel Decalcification Index

The enamel decalcification Index, first reported by Banks and Richmond (1994) was employed as the measurement of decalcification. The facial surfaces of each tooth studied were divided into four areas – gingival, mesial, distal and occlusal. Each area was scored 0 (no decalcification), 1 (decalcification covering <50% of the area), or 2 (decalcification covering >50% of the area) at each time period. All 20 teeth for each subject were recorded for each time point. Two operators scored the photographs independently. The results of each product was analyzed individually and among themselves.

The sample is representative of the demographics commonly seeing in orthodontics practices in Alabama.

Statistical analysis:

Descriptive statistics were used to describe the changes of EDI for each dentifrice over time. A further analysis, the three-way analysis of variance (ANOVA) (StatView; SAS Institute, Cary, NC) was used to analyze the enamel decalcification index scores of the maxillary and mandibular right first premolar to the left first left premolar at the 0.05 level of significance. Fisher protected least significant difference intervals were used to compare mean enamel decalcification index scores.

Two operators scored the photographs independently. Their scores were compared by using the *t-test*; no statistically significant difference ($P \geq 0.05$) was found between the operators.

CHAPTER 4

RESULTS

Sample Size:

A total of 120 subjects were invited and consented to be part of the study. Of these numbers, 100 subjects were successfully recalled over the 4 study periods. A schematic representation of number of subjects distributed into the various treatment groups is shown on Table 2 (Appendix I).

A total of 1.885 teeth were scored and distributed as follows:

- Clinpro™ 5000 group - 634 teeth were evaluated over 35 patients
- Clinpro™ Tooth Crème group - 604 teeth were evaluated over 32 patients
- MI Paste group - 638 teeth were evaluated over 33 patients.

Enamel Decalcification Index (EDI) scores

The resulting dataset followed a systematic hierarchy: facial surface areas within a tooth, teeth within arches, arches within patients, across four time periods. Analysis was done at the tooth level, aggregating the enamel decalcification scores from all four areas, creating an enamel decalcification index (EDI) for each tooth ranging potentially from 0 to 8 (however our data has a maximum EDI of 6). Table 3

(Appendix J) displays distribution statistics for EDI scores of the three groups across the four time periods, and at study consent (T0).

The results showed that the majority of teeth maintained a "0" EDI score of 74% and above. EDI scores of "1" made up the next largest percentage and EDI scores of "6" made up the smallest percentage.

In the sample of 643 subjects using Clinpro™ 5000, 86.8% maintained an EDI score of 0 after 4 weeks. The highest percentage (7.2%) was found for EDI 1, decreasing to a low of 0.3% for EDI 6.

In the sample of 604 teeth evaluated with Clinpro™ Tooth Crème, 74.7% maintained EDI scores of 0. Excluding EDI 0, the majority was found on EDI 1 with 18.4% and lowest of 0.2% on EDI 6.

Finally, of the 638 using MI Paste, 79.6% maintained an EDI of 0 of the 4th week. Furthermore, 13.9% presented scoring EDI 1 and 0% had an EDI of 6.

The total analysis may be found in Appendix J

As a robustness check, EDI scores were compared between records complete over all time periods and the omitted records, and no statistically significant differences were found.

EDI Scores Compared to location and tooth

In order to determine if EDI scores could be affected by location in the oral cavity or tooth position, a 3 way ANOVA was performed. These independent variables were carefully coded for the purposes of our analysis. Maxillary teeth were dummy coded as

and 0 for mandible. Teeth were also coded categorically, with five categories, representing the incisors, canines, and bicuspids.

The was no evidence in our data that suggested that left side teeth differed from right side teeth so the data for the teeth were kept in five categories rather than ten – arch level differences were addressed using the aforementioned maxilla dummy variable. In our models, the central incisors were omitted and set as the reference group for the other teeth. Time was coded with values 1, 2, 3 and 4 to correspond with the time period of each data point. Our independent variable of interest, treatment, was categorically coded to represent the three toothpaste groups. Clinpro™ 5000 was designated as the reference group through omission in models: any Clinpro™ Crème and MI Paste results were in comparison to the Clinpro™ 5000 group.

Multi Regression Analysis (MRA)

Multi-level mixed effects Poisson regression was used to model the data. Figure 3 (Appendix K) shows the distribution of the EDI score data to have positive skewness, akin to that of a Poisson distribution with low lambda.

A visual examination of the data suggests that there are treatment differences over the course of the study. However, Appendix M and N respectively also suggest that EDI scores vary across other dimensions as well.

As a result, the study elected to use a multiple regression method to thoroughly answer the research question by controlling for other co-variants besides treatment alone. In order to do so, the study assigned random effects at the patient, arch and tooth levels to accommodate for the nested structure of the data.

Five models were created to develop the analysis. The first four models make use of all four 1-period lagged time spans (for example; time 0 to time 1, time 1 to time 2, time 2 to time 3, time 3 to time 4, and time 4 to time 5), providing an analysis set of 7540 total tooth observations.

The results are presented in Table 4 (Appendix O).

Tooth Type and Location in the Oral Cavity

In applying the MRA to determine tooth type and location, it was found that the EDI scores showed incremental change between periods. The upper arch exhibits higher enamel decalcification, as do the bicuspids when compared to the central incisor.

Time

As expected, higher EDI scores were noted in all groups as the study time progressed. (Model 2)

Dentifrice used

When the treatment variables, Clinpro™ Crème and MI Paste, in comparison to the omitted treatment group, Clinpro™ 5000 were placed into the regression analysis, the results provide support that treatment effects do exist, above and beyond the other variables controlled for.

Relative to Clinpro™ 5000, Clinpro™ Crème exhibits higher levels of enamel decalcification (significant at the 95% level) while MI Paste shows marginal significance (at the 90% level) in having higher EDIs than Clinpro™ 5000.

Time and Dentifrice

A significant finding was displayed for the base effects as well as the interaction terms: with combined results suggesting that Clinpro™ 5000 is associated with lower EDI relative to the other treatments, though the magnitude of the effect falls off with the progression of time as indicated by the negative interaction terms. This finding is somewhat in tandem with the Lowess curves illustrated in Figure 2, with Clinpro™ 5000 exhibiting a steep departure from the other two treatments earlier on.

CHAPTER 5

DISCUSSION

As the smile esthetics move up the priority list of a patient's anticipation of appliance removal it can only be threatened by unsightly damaged enamel at deboning. Poor hygiene may well destroy a great esthetic result otherwise by way of white spot lesions. White Spot Lesions are frequently encountered in orthodontic patients.[4-9] This damaged enamel shows up most frequently as a white, opaque area outlining the site of bracket bonding.[80] Considerable research has been devoted to fluoride delivery methods that reduce or minimize enamel demineralization and supporting mineralization is orthodontic patients. [4, 5, 17, 20, 47, 48] Despite an increased awareness by clinicians and their attempt to aid patients in the prevention of white spot lesions, enamel demineralization is still one of the major concerns for the orthodontic treatment. It has been reported to affect up to 97 percent of patient undergoing orthodontic treatment.[8, 81] Given that studies show that restoration of these lesions are very difficult, it is imperative to take all preventative measures necessary to prevent their formation. In this study, some alternative methods for prevention and reduction of white spot lesions were carried out.

The aim of this randomized double blinded clinical trial is to determine if Clinpro™ 5000, Clinpro™ Tooth Crème or MI-Paste has an effect on the formation and resolution of white spot lesions for patients undergoing orthodontic treatment. However, the MI Paste Plus showed in recent reports to be useful in the reduction of white spots lesions.[12-14]. The active agent, casein phosphopeptide-amorphous calcium phosphate (CPP- ACP), is thought to stabilize and localize calcium, fluoride, and phosphate at the

tooth surface in a slow-release amorphous form, thus enhancing deeper remineralization of WSLs.[67] A number of in-vitro and in-situ studies have demonstrated the remineralizing potential of casein phosphopeptide-amorphous calcium phosphate.[68-77] For this reason we decided to use this product as a control to this study, and consider it a randomized controlled clinical trial.

We used five models to develop our analysis. The first four models make use of all four 1-period lagged time spans (time 0 to time 1, time 1 to time 2, time 2 to time 3, time 3 to time 4, and time 4 to time 5), providing an analysis set of 7540 total tooth observations. (Appendix O).

Model 1 serves as a baseline model, controlling for 1-period lagged EDI, arch, and tooth type. Controlling for the 1-period lagged EDI models the incremental change between periods, and as expected the lagged EDI score is predictive of EDI in the following period. The upper arch exhibits higher enamel decalcification, as do the bicuspids when compared to the central incisor.

Model 2 introduces time, which is also positively associated with higher EDI.

Model 3 brings in the treatment variables, Clinpro™ Crème and MI Paste, in comparison to the omitted treatment group, Clinpro™ 5000. The results provide support that treatment effects do exist, above and beyond the other variables controlled for. Relative to Clinpro™ 5000, Clinpro™ Crème exhibits higher levels of enamel decalcification (significant at the 95% level) while MI Paste shows marginal significance (at the 90% level) in having higher EDI than Clinpro™ 5000.

Model 4 introduces interaction terms of treatment by time. Significance is displayed for the base effects as well as the interaction terms: with combined results

suggesting that Clinpro™ 5000 is associated with lower EDI relative to the other treatments, though the magnitude of the effect falls off with the progression of time as indicated by the negative interaction terms. This finding is somewhat in tandem with the Lowess curves illustrated in Figure 2, with Clinpro™ 5000 exhibiting a steep departure from the other two treatments earlier on. Our last models the change in EDI between the beginning of treatment (time 0) and the end of treatment (time 4). Thus, Model 5 uses a quarter of the observations as the first models: rather than aggregate 4 short time periods, Model 5 utilizes the data once across the span of the study, from start state to end state. The results are slightly different than in the previous models: arch differences are no longer significant, canine teeth also exhibit lower levels of EDI than the central incisor, and MI Paste is no longer even significant at the 90% level. Additional models were created changing the omitted treatment group in order to test for differences between MI Paste and Clinpro™ Crème: no significant differences were found.

In the present study, the sample is representative of the demographics commonly seeing in orthodontics practices in Alabama. When analyzing the results found in the models described, the comparison of the facial surface areas within a tooth shows that the final results after 4 weeks maintained the highest percentage with EDI 0 with Clinpro™ 5000 followed by the MI Paste Plus and Clinpro™ Tooth Crème. The results were further analyzed in different hierarchies across the four time points; comparing the teeth within the arches, it shows that the maxilla seemed to be more affected than the mandible. This result can be explained for the fact that the maxilla has more teeth surface than the mandible. The comparison of arches within patients, showed that the anterior teeth had higher EDI scores when compared to the posterior teeth. Anterior teeth usually have

bigger facial surface when compared with the premolars, and this fact could explain these findings. Altogether, the results lend strong support for Clinpro™ 5000 providing superior protection against enamel decalcification when compared to Clinpro™ Tooth Crème, and mixed support when compared to MI Paste.

Recent studies agree with the results of this study. They reported a beneficial effect from either supplemental fluoride or casein phosphopeptide-amorphous calcium phosphate. For example, a high-concentration fluoride varnish was shown to be effective in reversing WSLs at 3-month and 6-month follow-ups after debonding. In this study, varnish was applied every month during the first 6 months after debonding.[82] Another randomized clinical trial compared patients who used fluoridated chewing sticks impregnated with 0.5% sodium fluoride with non-fluoridated chewing sticks. The authors reported that fluoridated chewing sticks had a stronger remineralization effect on WSLs compared with the controls over a 6-week period. [83] In a randomized trial comparing casein phosphopeptide-amorphous calcium phosphate cream (Topacal) with a 0.05% sodium fluoride mouthwash and fluoridated dentifrice combination, the subjects in both groups demonstrated improvements. However, the casein phosphopeptide-amorphous calcium phosphate group was associated with a greater number of WSLs that totally disappeared after 12 months (63% of the sites, compared with 25% in the control group). [64] Another trial randomized 45 adolescents to 10% casein phosphopeptide-amorphous calcium phosphate paste (Tooth Mousse) or a control (placebo) cream. Over a 12-week period, the authors reported that 31% more WSLs had regressed with the remineralizing paste than with the placebo.[65] A final randomized trial assessed regression of WSLs during the initial period of orthodontic treatment. MI Paste was reported to significantly

reduce WSLs, compared with a placebo paste. [13] Prevention of white spot lesions is better than repairing lesions once they exist.

Moreover, a recent study using synchrotron radiation micro computed tomography(SR micro-CT) was used to access the densities of bovine enamel WSL. Their observations suggest Clinpro™ 5000, which contains 1.1% NaF, delivers remineralization benefits at the surface of the WSL better than the 0.21% Clinpro™ Tooth Crème.[84] Since penetration of fluoride into enamel is limited and decreases exponentially with enamel depth,[85] it might be possible that the functionalized TCP present in the Clinpro ™ dentifrices helps extend the depth of fluoride penetration, and therefore lesion remineralization [86] In a recent *in vitro* study, the effects of fluoridated, casein phosphopeptide amorphous calcium phosphate complex (CPP-ACP)-containing, and functionalized β-tricalcium phosphate (fTCP)-containing toothpastes on remineralization of white spot lesions (WSLs) was done by using Quantitative light-induced fluorescence (QLF-D) Biluminator™ 2. They found that fTCP- and CPP-ACP-containing toothpastes seem to be more effective in reducing WSLs than 1,000-ppm fluoride-containing toothpastes (conventional toothpaste).[87] In a recent *in vitro* study, the comparison of the same products used in our study (5000ppm sodium fluoride; GC MI paste plus and Clinpro™ tooth crème) were compared and evaluated under a scanning Electron Microscope for calculation of the percentage of occluded tubules. Similarly to our study, they concluded that sodium fluoride showed relatively greatest remineralizing and dentinal tubule occlusion property when compared with GC MI paste plus and Clinpro™ tooth crème.[88] Similar results were found in an *in vitro* study where casein phosphopeptide-amorphous calcium phosphate (CPP-ACP), casein phosphopheptide-

amorphous calcium phosphate fluoride (CPP-ACPF) and tricalcium phosphate fluoride (TCP-F) were compared. They found that remineralization efficacy was TCP-F > CPP-ACPF > CPP-ACP.[89] Also, results indicate that combining fluoride with tricalcium phosphate could provide more anti-caries benefits compared to using fluoride alone.[90]

In contrast to our results, one recent randomized clinical compared casein phosphopeptide-amorphous calcium phosphate with fluoride paste with a control paste, by using quantitative laser fluorescence images to measure WSL regression. The authors reported that, at 6 and 12 weeks, the size of the lesions did not change significantly over time or between the groups. They concluded that there was no clinical advantage for the use of the casein phosphopeptide-amorphous calcium phosphate with fluoride paste supplementary to normal oral hygiene over 12 weeks.[91] In a randomized trial conducted in Europe, 60 healthy adolescents with at least 1 WSL received either a daily application of casein phosphopeptide-amorphous calcium phosphate (Tooth Mousse) or standard fluoride toothpaste. The intervention period was 4 weeks, and the endpoints were quantitative laser fluorescence and visual scoring (Gorelick scale) from digital photographs. The mean areas of the lesions decreased by 26% to 58% in the study, but no significant differences were found between the casein phosphopeptide-amorphous calcium phosphate and the control groups.[92] Another randomized trial compared WSLs treated with a low-fluoride mouthrinse (50 ppm) to those treated with a nonfluoride mouthrinse. At 12 weeks, the lesions had decreased by 40% (SD, 14.5) in the treatment group and by 51.5% (SD, 12.3) in the control group, indicating no significant benefit from the low-level fluoride. [37] Moreover, a recent *in vitro* study compared the effectiveness of dentifrices containing tri-calcium phosphate or calcium phosphosilicate

in combination with fluoride to prevent the demineralization of overdenture abutments and root surfaces. They found that the addition of tricalcium phosphate or calcium phosphosilicate to fluoride-containing dentifrices (5000 ppm) does not significantly improve their ability to prevent demineralization of the cut dentin surface of overdenture abutments. [93]

Study limitations:

This clinical study had some limitations. One was that the patient's compliance could not be ascertained or ideally controlled. We attempt to overcome the compliance barrier giving the patients brushing technique instructions, standardizing the time and frequency to brush their teeth and using the brushing diary as a control of their actual compliance with the protocol. However it was observed plaque accumulation and gingival inflammation on a considerable amount of patients. Another limitation to our study design was a lack of blinding for patients and providers. However, we thought that blinding of the evaluators was the most important factor in preventing biased assessments. Also, the visual effect of decalcification was in some areas harder to be detected due the accumulation of plaque. However, white spot lesion development is usually difficult to predict early after orthodontic bonding. The best predictors for white spot lesions have been considered to be visible plaque and *mutans streptococci* (eg, the level of oral hygiene and thus the cariogenic challenge) around the fixed appliances.[94]

Overall the results suggest support that Clinpro™ 5000 provides marginally superior protection against enamel decalcification when compared to Clinpro™ Tooth Crème and mixed support when compared to MI Paste Plus. The results found for

Clinpro™ 5000 were marginally better than MI Paste Plus. However, in the context of the clinical application these were negligible.

CHAPTER 6

CONCLUSIONS

- EDI scores increased over the 4 time points in the study period.
- The use of Clinpro™ 5000, Clinpro™ Crème, and MI paste seem to lead to less formation of white spot lesions when compared to data in the literature
- Clinpro™ 5000 has a marginally better effect than the two other test pastes. However, this effect is clinically negligible.

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APPENDIX A

SAMPLE DATA SHEET (1)

EDI SCORING SHEET

Patient Number:_____

Visit number: _____

ENAMEL DECALCIFICATION INDEX

Upper Jaw

R L

	5	4	3	2	1	1	2	3	4	5
EDI Score										

Lower jaw

R L

	5	4	3	2	1	1	2	3	4	5
EDI Score										

Figure 1. Enamel Decalcification Index. Facial surface of each tooth was divided into four areas: g=gingival; m=mesial; d=distal; o=occlusal. A score was allocated for each area of each tooth: 0=No decalcification; 1=decalcification covering <50% of the area; 2=decalcification covering >50% of the area; 3=decalcification covering 100% of the area, or severe decalcification with cavitation. Total score per tooth calculated by summation of individual area scores for each tooth.
(Banks and Richmond, 1994)

APPENDIX B

SAMPLE DATA SHEET (1)

CASE REPORT FORM

Patient Number:_____

Visit number: _____

CASE REPORT FORM

Patient Photographs:

Right Buccal	Frontal	Left Buccal

EDI score: _____

ICDAS score: _____

Teeth studied:

Upper Jaw

R L

5	4	3	2	1	1	2	3	4	5

Lower jaw

R L

5	4	3	2	1	1	2	3	4	5

APPENDIX C

BRUSHING DIARY

Study Weeks ____________

Tooth Brushing Instructions:

- Wet the head of the toothbrush and place a 1-inch (2.5 cm) strip of toothpaste on the bristles.
- Brush twice daily for **2 minutes**, preferably in the morning and right before bed.
- After brushing, spit out the toothpaste, but **DO NOT RINSE**.
- Do not eat or drink for at least 30 minutes after brushing.
- Please enter the **Date** and **Time** (hour and minutes) each time that you brush your teeth.

  Return this diary and any remaining toothpaste at your next visit. The diary will be reviewed by the study team.

_______________ 2011

SUNDAY	MONDAY	TUESDAY	WEDNESDAY	THURSDAY	FRIDAY	SATURDAY
Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___
Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___
Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___
Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___	Date ______ A.M.___:___ P.M.___:___
Diary Reviewed by:						

Date of Next Appointment: ______________ Time: __________

Participant's signature: ___________________

APPENDIX D

SERIOUS ADVERSE EVENT / SERIOUS ADVERSE DRUG REACTION (SAE/SADR) FORM

TITLE OF CLINICAL STUDY: Comparison of Three Fluoride Dentifrice Products in the Prevention and Reduction of White Spot Lesions in Orthodontic Treatment
STUDY IDENTIFICATION NUMBER: 3M ESPE #CR-11-003
LOCATION AND PRINCIPAL INVESTIGATOR: University of Alabama – Birmingham / Dr. Chung How Kau

Subject's ID#:

DATE OF OCCURRENCE:		**TIME OF OCCURRENCE:**	
Intensity at onset:	☐ Mild Subject is aware of symptoms, but they are easily tolerated	☐ Moderate Symptoms restrict but do not prevent subject's daily activities	☐ Severe Subject is unable to perform usual daily activities
Related to Test Product?	☐ No	☐ Probably (Adverse Drug Reaction)	☐ Yes (Adverse Drug Reaction)
If related or probably related, was the ADR:	☐ Anticipated (refer to protocol)	☐ Unanticipated ☐ In nature ☐ In severity	
Is this a Serious Adverse Event (SAE)?	☐ No	☐ Yes* *Caused death, was life-threatening, required inpatient hospitalization, prolonged existing hospitalization, resulted in persistent or significant disability/incapacity, or resulted in congenital anomaly/birth defect	

** Notify the IRB(s) in accordance with their reporting requirements.*

Describe the AE/ADR in detail. Provide a chronology of events, actions taken, dates of hospitalization (if applicable), and outcome. Use additional paper, if necessary. Sign and date each attachment.
.....
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APPENDIX E

SERIOUS ADVERSE EVENT / SERIOUS ADVERSE DRUG REACTION (SAE/SADR) FORM, pg. 2

SUBJECT'S ID#:

DOES THE PATIENT HAVE A HISTORY OF THIS TYPE OF EVENT OR A SIMILAR EVENT?		
☐ NO	☐ YES (check appropriate box below)	
	☐ While on study	☐ While off study
	☐ While using another product (specify)	

MEDICAL TREATMENT OR OTHER CORRECTIVE ACTION FOR THE AE/ADR ?		
☐ NO	☐ YES (provide detail below)	
Start Date (dd/mmm/yy)	**End Date** (dd/mmm/yy)	Medication, treatment, and corrective action
		....
		....

WAS THE PATIENT WITHDRAWN FROM THE STUDY?		
☐ NO	☐ YES	Date of withdrawal: _____

COURSE AND OUTCOME OF THE AE/ADR		
☐ Subject Recovered **Date** (dd/mmm/yy) _____ **Time** (hh:mm) _____	☐ Adverse Event persists (Describe above)	☐ Fatal (Describe above)
Did the AE/ADE increase in severity?	☐ No	☐ Yes **Intensity** (compare with onset) ☐ Moderate ☐ Severe

PRINCIPAL INVESTIGATOR'S SIGNATURE:		
_____ Printed Name	_____ Signature	_____ Date
IRB SIGNATURE:		
_____ Printed Name	_____ Signature	_____ Date

FAX THIS FORM TO 3M AT **651-737-8114**, ATTN: **ROS RANDALL**
WITHIN 24 HOURS OF THE INVESTIGATORS' KNOWLEDGE OF THE EVENT

APPENDIX F

TEST MATERIALS:

1. Clinpro ™5000 1.1% Sodium Fluoride Anti-Cavity Toothpaste with Tri-Calcium Phosphate (3M ESPE, Saint Paul, MN, USA)

General Information:

Clinpro™5000 toothpaste is prescription-strength white toothpaste that contains 5000 ppm fluoride and an innovative tri-calcium phosphate ingredient with a vanilla mint flavor. The product is intended to be use once daily in place of conventional toothpaste, unless instructed otherwise by a physician or dentist.

Composition:

Clinpro™ 5000 toothpaste contains 1.1% sodium fluoride and an innovative tri-calcium phosphate ingredient which is sold exclusively through 3M ESPE. Each gram of Clinpro™ 5000 toothpaste
contains 5mg of fluoride ion in a neutral pH base consisting of water, sorbitol, hydrated silica, glycerin, polyethylene-polypropylene glycol, flavor, polyethylene glycol, sodium lauryl sulfate, titanium dioxide, carboxymethyl cellulose, sodium saccharin and tri-calcium phosphate.

Precautions:

DO NOT SWALLOW. Keep out of reach of children under 6 years of age.

Adverse Reactions:

Allergic reactions and other idiosyncrasies have been rarely reported.

Pre clinical testing:

A Diplomate of the American Board of Toxicology has assessed the safety of Clinpro™ 5000 as safe for its intended use.

2. MI Paste Plus (GC America, Alsip, Ill)

General Information:

MI Paste Plus is indicated for post-bleaching sensitivity, root planing and scaling, and during prophylaxis. It is also indicated as an alternative means of applying fluoride topically. The product may be applied to the teeth using a clean dry finger or a cotton tip. MI Paste Plus is indicated for once- or twice-daily use unless otherwise directed by a dental professional.

Composition:

This formulation contains 0.2% sodium fluoride, water, glycerol, amorphous calcium phosphate, D-glucitol, silicon dioxide, titanium dioxide, ethyl-4-hydroxybenzoate, butyl-parahydroxybenzoate, and propyl-4-hydroxybenzoate.

3. Clinpro™ Tooth Crème 0.21% Sodium Fluoride Anti-Cavity Paste with Tri-Calcium Phosphate(3M ESPE, Saint Paul, MN, USA)

General Information:

Clinpro™ Tooth Crème is a white crème that contains 950 ppm fluoride ion (like regular toothpaste) and a functionalized tri---calcium phosphate ingredient (fTCP) with a vanilla mint flavor. fTCP is a name given to the innovative, specially-prepared calcium--

based additive that co--exists with fluoride in an aqueous environment (i.e., in the toothpaste tube). In a special process, tri--calcium phosphate is "functionalized". Resulting in the formation of an organic calcium phosphate hybrid, in which calcium is protected from prematurely interacting with ionic fluoride while coexisting in the tube. As the toothpaste comes into contact with saliva during brushing, the protective barrier quickly breaks down and allows the calcium, phosphate, and fluoride to be readily available to the tooth. The tooth naturally absorbs these components, helping to prevent the initiation and further progression of demineralization and allowing remineralization to occur.

Clinpro™ Tooth Crème is a one--step product requiring patients to simply brush with it once or twice daily in place of their conventional toothpaste. Clinpro™ Tooth Crème was developed specifically for patients who need the benefits of a professionally--dispensed fluoride--containing preparation. It can be applied to enamel and exposed dentin as prescribed by a dental professional, dependent on each patient brushing to help remineralize demineralized enamel and to aid in the prevention of tooth decay.

Composition:

Clinpro™ Tooth Crème contains 950 ppm F^- (0.21% w/w) sodium fluoride in a neutral pH base and a mild abrasive to help remove dental plaque, debris, and stain. Each gram contains 0.95mg of fluoride ion. The other ingredients include water, sorbitol, hydrated silica, glycerin, polyethylene-polypropylene glycol, flavor, polyethylene glycol, sodium lauryl sulfate, titanium dioxide, carboxymethyl cellulose, sodium saccharin, and tri--

calcium phosphate.

Precautions:

DO NOT SWALLOW. Keep out of reach of children under 6 years of age.

Adverse Reactions:

Allergic reactions and other idiosyncrasies have been rarely reported.

Pre clinical testing:

A Diplomate of the American Board of Toxicology has assessed the safety of Clinpro™ Tooth Crème as safe for its intended use.

APPENDIX G

Assign subjects to groups:

A-Clinpro™ 5000

B-MI-Paste Plus

C-Clinpro™ Tooth Crème

Table 1

Subject #	Group Assigned
1	B
2	C
3	A
4	C
5	B
6	B
7	B
8	C
9	A
10	A
11	A
12	B
13	B
14	C
15	C
16	B
17	C
18	C
19	C
20	B
21	B
22	C
23	A
24	C
25	B
26	B
27	A
28	C
29	B
30	A
31	A

32	C
33	C
34	B
35	B
36	C
37	C
38	C
39	A
40	B
41	A
42	C
43	A
44	A
45	A
46	A
47	C
48	C
49	C
50	A
51	B
52	C
53	C
54	A
55	C
56	B
57	B
58	B
59	C
60	A
61	C
62	C
63	B
64	B
65	B
66	B
67	B
68	B
69	A
70	A
71	A
72	A
73	A
74	B
75	B
76	A

77	A
78	A
79	B
80	B
81	A
82	A
83	B
84	B
85	C
86	B
87	B
88	A
89	C
90	B
91	C
92	C
93	B
94	A
95	A
96	A
97	C
98	A
99	A
100	C
101	C
102	B
103	B
104	C
105	B
106	B
107	C
108	C
109	A
110	A
111	C
112	A
113	C
114	C
115	B
116	A
117	A
118	A
119	A
120	C

APPENDIX H

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THE FOLLOWING WERE APPROVED

INVESTIGATOR: Chung H. Kau Ph.D.
1530 3rd Avenue South
SDB 305
Birmingham, Alabama 35294

BOARD ACTION DATE: 12/30/2011
PANEL: 6
STUDY APPROVAL EXPIRES: 01/14/2013
STUDY NUM: 1122614
WIRB PRO NUM: 20102132
INVEST NUM: 162722
WO NUM: 1-700763-1
CONTINUING REVIEW: Annually
SITE STATUS REPORTING: Annually
INST. NUM: W101201002

SPONSOR: 3M ESPE Dental Products
PROTOCOL NUM: None
AMD. PRO. NUM:
TITLE:
Comparison of Three Fluoride Dentifrice Products in the Prevention and Reduction of White Spot Lesions in Orthodontic Treatment

APPROVAL INCLUDES:
Study and Investigator for an additional continuing review period. This approval expires on the date noted above.

WIRB APPROVAL IS GRANTED SUBJECT TO:

WIRB HAS APPROVED THE FOLLOWING LOCATIONS TO BE USED IN THE RESEARCH:
UAB Orthodontics Clinic, School of Dentistry Building third floor, SDB 305, 1919 7th Avenue South, Birmingham, Alabama 35294

If the PI has an obligation to use another IRB for any site listed above and has not submitted a written statement from the other IRB acknowledging WIRB's review of this research, please contact WIRB's Client Services department.

IF YOU HAVE ANY QUESTIONS, CONTACT WIRB AT 1-800-562-4789
This is to certify that the information contained herein is true and correct as reflected in the records of the Western Institutional Review Board (WIRB), OHRP/FDA parent organization number IORG 0000432, IRB registration number IRB00000533. WE CERTIFY THAT WIRB IS IN FULL COMPLIANCE WITH GOOD CLINICAL PRACTICES AS DEFINED UNDER THE U.S. FOOD AND DRUG ADMINISTRATION (FDA) REGULATIONS, U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES (HHS) REGULATIONS, AND THE INTERNATIONAL CONFERENCE ON HARMONISATION (ICH) GUIDELINES.

Full AAHRPP Accreditation

Theodore D. Schultz, J.D., Chairman

12/30/2011
(Date)

This document electronically reviewed and approved by Luk, Hsiang-Ning on 12/30/2011 1:18:45 PM PST. For more information call Client Services at 1-360-252-2500.

APPENDIX I

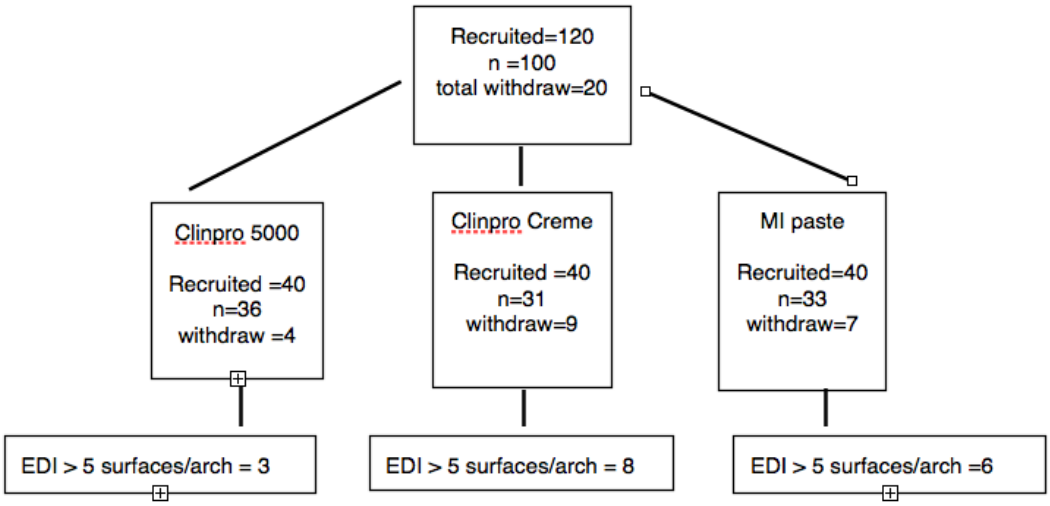

Table 2: Schematic representation of number of subjects broken into the various treatment groups

APPENDIX J

Clinpro 5000		EDI 0	EDI 1	EDI 2	EDI 3	EDI 4	EDI 5	EDI 6
	T0	100.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
N: 643 Teeth	T1	99.8%	0.2%	0.0%	0.0%	0.0%	0.0%	0.0%
over 35 Patients	T2	95.2%	3.0%	1.1%	0.8%	0.0%	0.0%	0.0%
	T3	87.7%	6.5%	3.4%	1.7%	0.3%	0.0%	0.3%
	T4	86.8%	7.2%	3.7%	1.7%	0.3%	0.0%	0.3%
Clinpro Crème		**EDI 0**	**EDI 1**	**EDI 2**	**EDI 3**	**EDI 4**	**EDI 5**	**EDI 6**
	T0	97.7%	2.0%	0.3%	0.0%	0.0%	0.0%	0.0%
N: 604 Teeth	T1	91.9%	6.0%	1.2%	0.2%	0.8%	0.0%	0.0%
over 32 Patients	T2	87.4%	9.1%	2.3%	0.3%	0.7%	0.0%	0.2%
	T3	79.1%	14.9%	3.8%	0.8%	1.2%	0.0%	0.2%
	T4	74.7%	18.4%	4.5%	1.2%	1.2%	0.0%	0.2%
MI Paste		**EDI 0**	**EDI 1**	**EDI 2**	**EDI 3**	**EDI 4**	**EDI 5**	**EDI 6**
	T0	99.8%	0.0%	0.2%	0.0%	0.0%	0.0%	0.0%
N: 638 Teeth	T1	94.5%	4.9%	0.6%	0.0%	0.0%	0.0%	0.0%
over 33 Patients	T2	88.7%	8.2%	1.9%	0.6%	0.6%	0.0%	0.0%
	T3	83.4%	11.0%	3.4%	1.6%	0.6%	0.0%	0.0%
	T4	79.6%	13.9%	3.4%	2.2%	0.6%	0.2%	0.0%

Table 3: Total number of teeth in each group and the EDI scores.

APPENDIX K

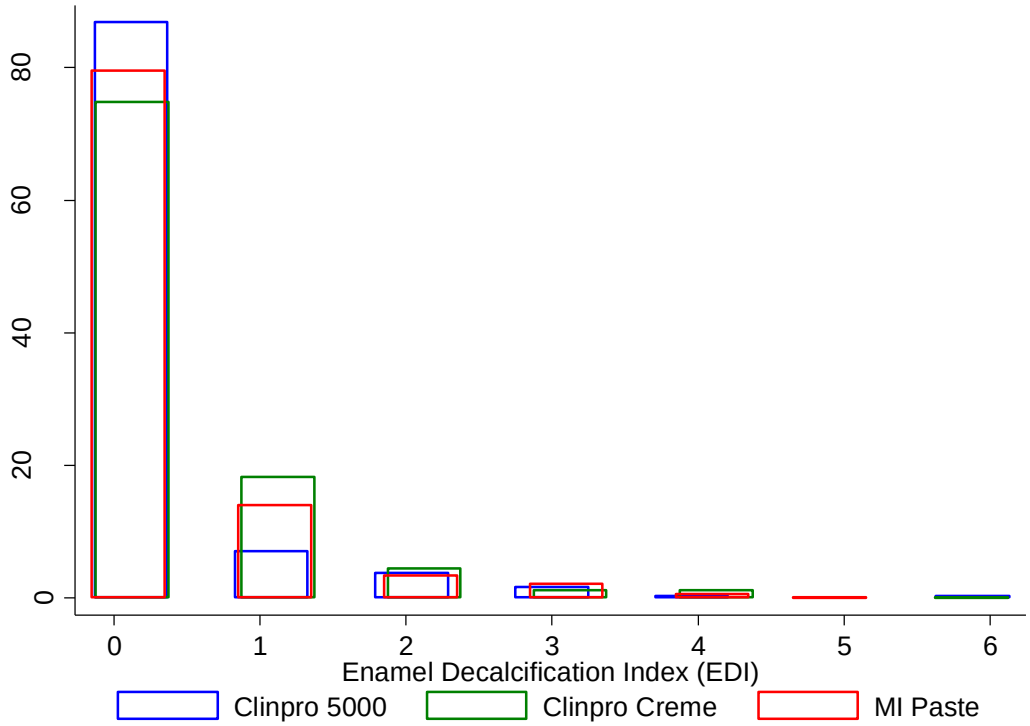

Figure 3: Percentage Distribution Histograms of EDI Scores at Time 4 by Treatment Group

APPENDIX L

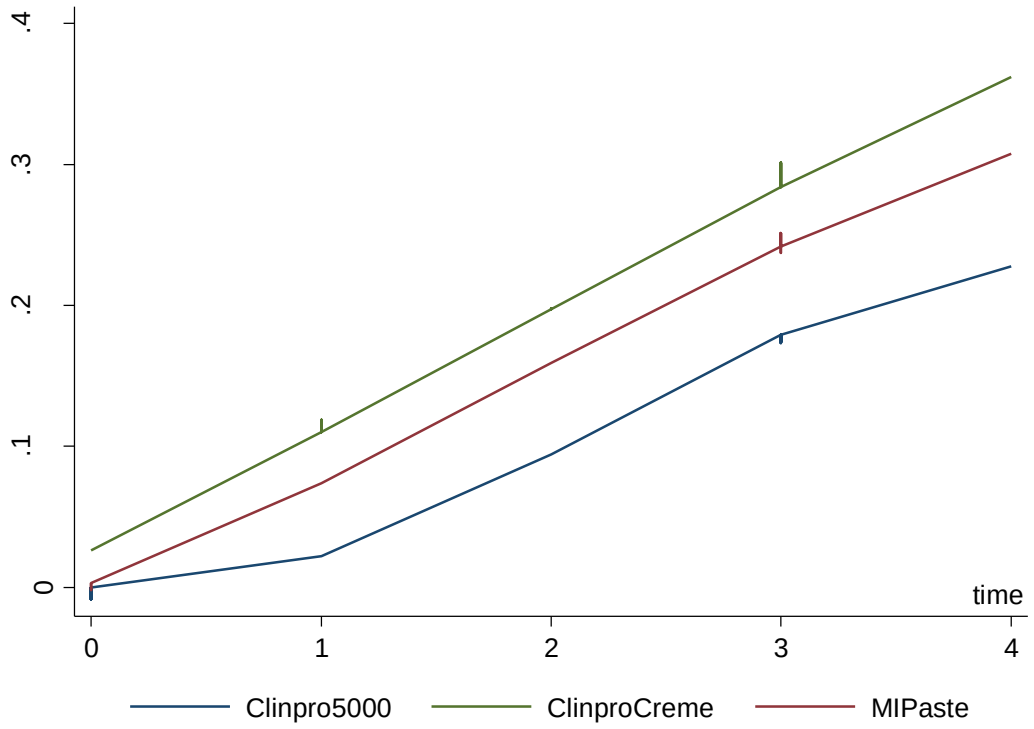

Figure 4. Lowess Curves of Mean EDI Scores across Time by Treatment Group

APPENDIX M

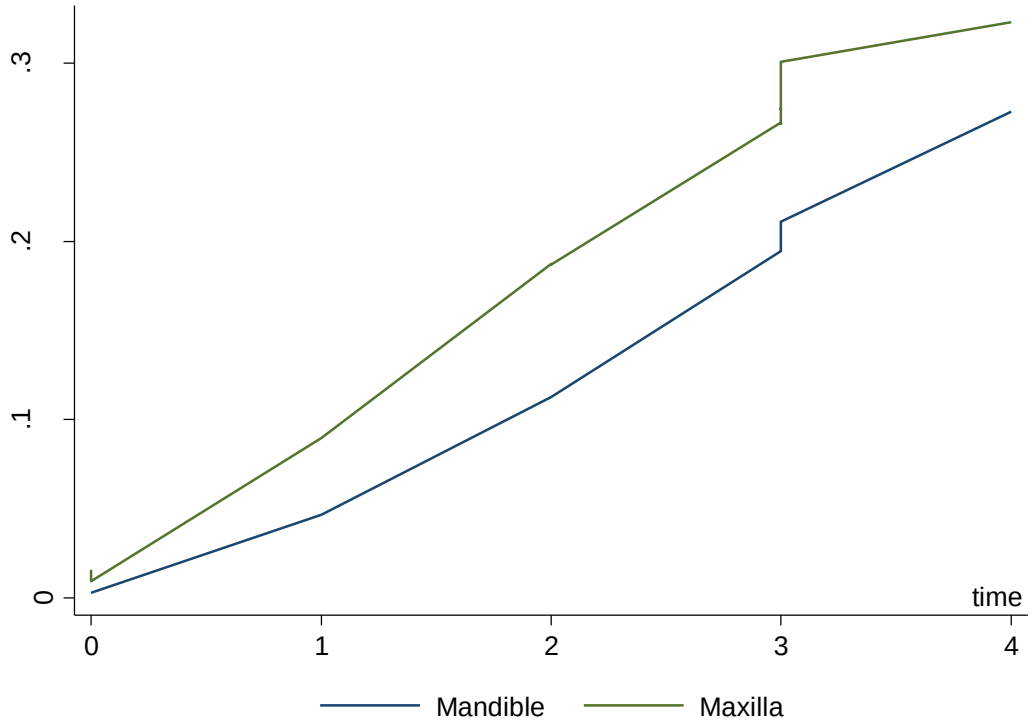

Figure 5: Lowess Curves of Mean EDI Scores across Time by Arch

APPENDIX N

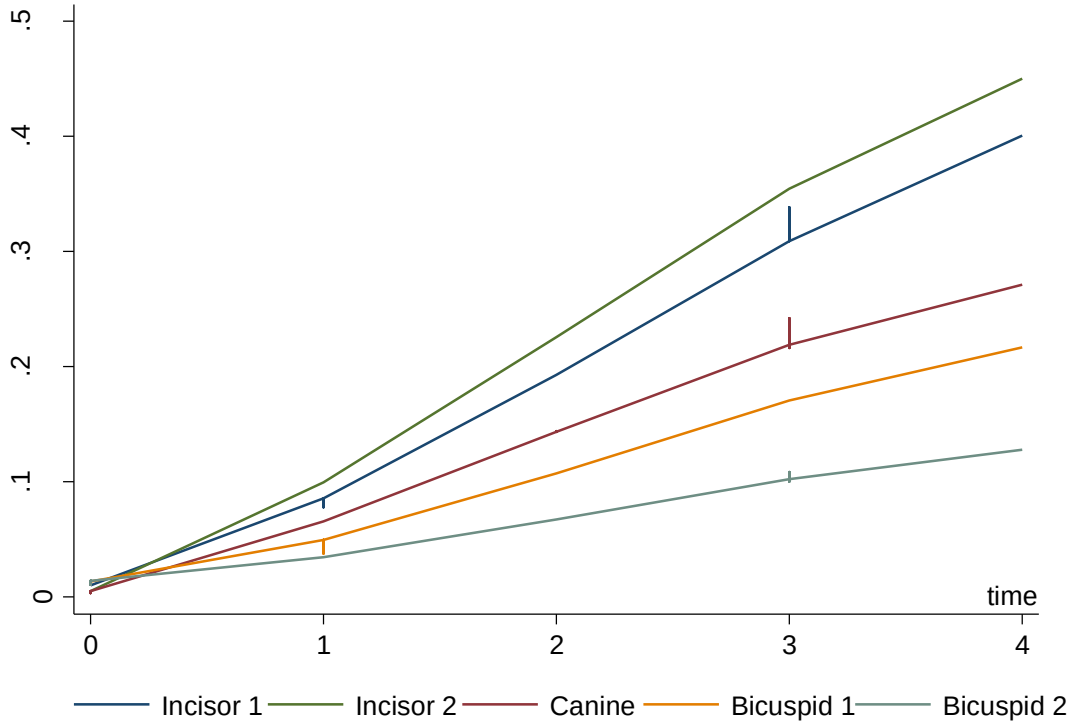

Figure 6: Lowess Curves of Mean EDI Scores across Time by Tooth Type

APPENDIX O

	Model 1	Model 2	Model 3	Model 4	Model 5
Lagged EDI (1 Time Period)	0.454***	0.234***	0.234***	0.209***	
	(0.029)	(0.034)	(0.034)	(0.034)	
Lagged EDI (4 Time Periods)					1.320***
					(0.367)
Maxilla	0.548**	0.591**	0.589**	0.595**	0.256
	(0.194)	(0.207)	(0.207)	(0.208)	(0.188)
Incisor 2	0.136	0.149	0.148	0.150	0.127
	(0.155)	(0.177)	(0.177)	(0.180)	(0.132)
Canine	-0.191	-0.217	-0.217	-0.219	-0.298*
	(0.164)	(0.186)	(0.186)	(0.189)	(0.149)
Bicuspid 1	-0.558**	-0.634**	-0.634**	-0.643**	-0.692***
	(0.174)	(0.197)	(0.197)	(0.200)	(0.160)
Bicuspid 2	-1.255***	-1.423***	-1.423***	-1.445***	-1.242***
	(0.203)	(0.227)	(0.227)	(0.230)	(0.188)
Time		0.344***	0.344***	0.544***	
		(0.031)	(0.031)	(0.065)	
Clinpro Crème			1.892*	2.730**	1.495*
			(0.841)	(0.875)	(0.697)
MI Paste			1.505+	2.113*	1.006
			(0.838)	(0.873)	(0.697)
Clinpro Crème X Time				-0.274***	
				(0.072)	
MI Paste X Time				-0.197**	
				(0.074)	
Constant	-4.718***	-5.782***	-6.892***	-7.527***	-3.961***
	(0.412)	(0.446)	(0.698)	(0.727)	(0.579)
Patient R.E. Constant	1.044***	1.089***	1.053***	1.059***	0.842***
	(0.126)	(0.126)	(0.126)	(0.126)	(0.131)
Maxilla R.E. Constant	-0.265	-0.221	-0.222	-0.217	-0.312+
	(0.168)	(0.168)	(0.168)	(0.168)	(0.190)
Tooth Number R.E. Constant	-0.238*	-0.050	-0.052	-0.028	-0.940***
	(0.119)	(0.101)	(0.101)	(0.099)	(0.238)
N	7540	7540	7540	7540	1885
Degrees of Freedom	6	7	9	11	8
Wald Chi-square	362.918	458.440	464.131	464.840	87.765
Prob > Chi-square	0.000	0.000	0.000	0.000	0.000

Table 4: Statistical Representation of Results