EFFICACY, SAFETY AND POST-TREATMENT COMFORT OF CYANOACRYLATE AS AN ADJUNCT TO NON-SURGICAL PERIODONTAL THERAPY:

A PILOT RANDOMIZED CLINICAL TRIAL

by

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Abstract

Objectives: Non-surgical periodontal therapy (NSPT) can be associated with post-treatment complications of gingival tissue tenderness and teeth sensitivity that can negatively impact oral self-care thereby significantly affecting periodontal wound healing in its critical phase. The ineffectiveness of biofilm disruption can also further increase the sensitivity of gingival tissues and teeth resulting in patient’s reluctance in continuing regular oral self-care and professional therapy. This pilot split-mouth blinded randomized clinical trial assessed the efficacy of supra-gingival application of cyanoacrylate adhesive (PeriAcryl®90HV) in promoting periodontal wound healing and preventing increased sensitivity of teeth and gingival tissues following NSPT.

Methods: 24 participants with active periodontal disease were recruited. Two selected quadrants with symmetrical disease were randomly divided into experimental and control arms and received NSPT. The experimental sites additionally received an application of cyanoacrylate adhesive (PeriAcryl®90HV) extending 1-2mm along gingival margins. Primary and secondary endpoints included clinical attachment level (CAL), bleeding on probing (BOP), probing depth (PD) and plaque score (PI), from baseline to the final visit at 6-8 weeks following NSPT. Furthermore, teeth sensitivity and pain and gingival tissue discomfort at baseline, 1-week and the final visit at 6-8 weeks following NSPT were ascertained using the visual analogue scale (VAS) and daily journaling. Receptiveness to future application of the cyanoacrylate adhesive was also evaluated using VAS.

Results: The application of cyanoacrylate after NSPT resulted in no statistical changes between arms for CAL, PD, PI and sensitivity from baseline to the final visit at 6-8 weeks. BOP reduction...
was statistically significant (p=0.003*) in the experimental arm. The application of cyanoacrylate had no adverse events and was found highly acceptable (69.6%; n=16/23) by the subjects.

**Conclusions:** The reduction of BOP in the study indicates, that the application of cyanoacrylate with its antimicrobial traits and treatment acceptance, may serve as a useful adjunct to NSPT in reducing gingival inflammation.
Lay Summary

The accumulation of microorganisms causes periodontal disease in the form of dental plaque and calculus deposits, and the presence of risk factors in a susceptible individual lead to the breakdown of tooth-supporting structures. Professional cleanings of these deposits at regular intervals is the mainstay for management of periodontal diseases. This procedure can accompany with variable degrees of pain, discomfort and heightened sensitivity in the teeth and gums making it difficult to maintain optimum oral self-care. The use of cyanoacrylate adhesives applied to gum margins following professional cleanings was envisioned to provide an isolated environment for healing and serve as a barrier to external influences of trauma or infections. The results of the present human clinical trial showed that added application of this adhesive with its antimicrobial traits and subject acceptance was successful in significantly reducing inflammation as evident from significantly reduced bleeding from gums when evaluated with a dental probe.
Preface

This dissertation is an original intellectual product of the author, Dr. Rashi Chaturvedi. The Clinical Research Ethics Board (CREB) at The University of British Columbia approved the clinical trial involving human subjects; certificate number H15-03277. Rashi Chaturvedi was involved in protocol writing and obtaining ethics approval for conducting the experiment under the supervision of Clinical Associate Professor, Ms. Penny Hatzimanolakis. Dr. Chaturvedi was involved in the screening, assessing eligibility, obtaining informed consent and recruitment of the study participants. Dr. Chaturvedi coordinated the scheduling of participant visits, was responsible for completion of the patient reported assessment forms, taking intra-oral photographs and following up with the participants after treatment. Ms. Rebecca Kan was the examining clinician, and Ms. Mary-Ann McKinnon was the treating clinician in this trial. The primary author was also involved with ensuring that proper record keeping and reporting requirements are met with, the organization of data and its statistical evaluation. The statistical tests were performed under the guidance of Dr. Hsing Chi von Bergmann.
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List of Abbreviations

B: Mid- Buccal
BOP: Bleeding on Probing
CAL: Clinical Attachment Level
CI: Confidence Interval
cm: Centimetre
DB: Distobuccal
DH: Dentine Hypersensitivity
DL: Distolinguinal
g: grams
L: Mid- lingual
MB: Mesiobuccal
ML: Mesiolinguinal
mm: Millimetre
N: Newton
NS: Non- Significant
NSPT: Non-Surgical Periodontal Therapy
PD: Probing Depth
PI: Plaque Index
RCT: Randomized Clinical Trial
R: Recession
SD: Standard Deviation
SRP: Scaling Root Planing
VAS: Visual Analogue Scale
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Dedication

To my dear son Atharv

The last few years of our lives have been chaotic and extremely stressful, but despite all odds you have blossomed into an amazing kid. I am so proud of you sweetheart for being so supportive, loving and understanding in my most difficult times. I hope our struggles instill in you a sense of discipline, a hope to strive hard to attain your goals, your dreams. I will cherish all these memories and will make up for all lost time. May god shower his choicest blessings on you and keep you happy and smiling always. I love you so much my sunshine!
Chapter 1: Introduction

The oral cavity is a protective niche to a wide variety of microorganisms. This closed microbiome environment can result in periodontal disease. The American Academy of Periodontology in 2008 transitioned the concept of periodontitis as an “infectious disease” to inflammatory disease\(^1\). The release of inflammatory mediators from the diseased gingival tissues can induce widespread systemic effects on various organ systems. The diagnosis of periodontitis heightens the risk of acquiring or worsening other chronic systemic conditions by influencing the natural course, duration and outcome\(^2,3\). Severe periodontitis is the 6\(^{th}\) worldwide prevalent oral disease with an overall prevalence of 11.2%. The global burden of disease studies has identified an increase of 57.3% from 1990-2010\(^4\).

The primary objective of managing periodontal diseases is reducing or arresting the progression of inflammation, i.e. active disease. Periodontal pathogens employ the inflammatory process to enrich their environment by creating the etiologic biofilm\(^5\).

Mechanical disruption of the biofilm is the standard of care procedure in managing periodontal diseases. It is accomplished via periodontal debridement, which is a holistic approach to arresting disease progression and re-establishing overall oral health. Scaling and root planing (SRP) are fundamental professional instrumentation procedures utilized during periodontal debridement, which removes hard and soft deposits above and below the gingival margin. \(^6\). In this study, SRP and debridement are interchangeable. All the terms, SRP, debridement and mechanical disruption are various non-surgical periodontal treatment (NSPT) methodologies. Debridement is performed with either hand, powered driven devices (sonic or ultrasonic) or in combination to restore dental
tissues to a compatible biological state, i.e. non-inflammatory\textsuperscript{7}. However, post-treatment complications of SRP include gingival recession, soft tissue tenderness, teeth and root sensitivity and pain\textsuperscript{8,9}.

Researchers have assessed the discomfort and pain that patients perceived following various periodontal therapeutic procedures and have demonstrated that patients experienced significant duration and magnitude of pain following SRP\textsuperscript{10–12}. The heightened sensitivity leads to reduced compliance in maintaining oral self-care, thereby creating a vicious cycle of increased plaque deposits, the creation of an acidic milieu and increasing dentinal hypersensitivity\textsuperscript{13,14}. This further enhances the anxiety and apprehension and at times denial for subsequent hygiene maintenance visits, which can reduce the prognostic outcome and significantly influence oral and systemic health\textsuperscript{15}.

Conventionally, options such as administration of over the counter analgesics, topical anesthetic gels and desensitizing toothpastes have been utilized to minimize post-treatment discomfort following SRP. These treatment options have their own set of limitations such as lack of predictability, prolonged duration to obtain an adequate response, compromised quality of life due to side effects such as gastric irritation, numbness, altered taste sensations, and need for repeated use.

A demand, therefore, continues to exist for adjuvant therapy that may provide a more predictable protective barrier during the immediate and critical healing period following SRP. This may
assist patients with daily oral self-care procedures and for some individuals, provide the ability to complete their recommended periodontal treatment.

It is hypothesized that a novel adjunctive cyanoacrylate adhesive placed after SRP may assist with post-treatment complications.

Cyanoacrylates developed in the 1950's, and are FDA approved as antimicrobial tissue adhesives with decades of successful applications in both medicine and dentistry. Medical soft tissue applications of cyanoacrylates adhesives are standard and include repairs of traumatic lacerations, myocardial tears, inguinal hernia, and sealing of cerebrospinal fluid leaks. Intra-orally they are used as alternatives to sutures, for stabilizing free gingival grafts, cleft repairs, sinus membrane perforations repair, as hemostatic agents especially in patients on anticoagulant therapy and healing of extraction sockets $^{16-20}$.

The present study was designed to assess the application of cyanoacrylate adhesive after SRP procedure. This adjunctive placement may assist with post-treatment tissue healing during the immediate and critical period following SRP, the ability to complete comprehensive periodontal treatment and most importantly, patient's compliance with daily oral self-care procedures to obtain oral and overall systemic health.

Currently, no studies have assessed the efficacy of cyanoacrylate adhesives as an adjunct to SRP. This pilot split-mouth, parallel, randomized controlled clinical trial aims to explore the efficacy of cyanoacrylate adhesive as an adjunct to SRP in minimizing post-treatment discomfort of the
dental soft and hard tissues and encourage wound healing as assessed by the periodontal clinical parameters (clinical attachment, probing depths, bleeding on probing).
Chapter 2: Review of Literature

2.1 Periodontium in health and disease

2.1.1 Introduction

The periodontium is a complex orchestra of soft and hard dental tissues functioning to support the tooth. These harmonious structures comprise the periodontal ligament, alveolar bone, cementum and the gingiva.

The cementum forms the outer structure of the roots into which the periodontal ligament fibres insert providing anchorage of the tooth to the surrounding alveolar bone. The alveolar bone is further subdivided into the alveolar bone proper that forms the tooth socket and the supporting alveolar bone comprising of the inner and outer cortical plates as well as the cancellous trabecular bone between them. The periodontal ligament predominantly made up of collagen fibres in variable distribution patterns helps provide support to the teeth and bears the brunt of occlusal stresses during function and additionally provide formative, nutritive, sensory and proprioceptive functions. The gingival tissues, more specifically the oral or outer epithelium and its underlying connective tissues serve an essential function of protecting the underlying structures by bracing the masticatory forces. The gingival tissues attach to the tooth forming a seal and providing a barrier to the onslaught of infection. It adapts to form a ‘V’ shaped crevice surrounding the tooth and the gingival sulcus whose constituents provide a localized mechanism of immune response. A sulcular epithelium lines the lateral wall of the gingival sulcus in its coronal part, and junctional epithelium forms its apical extent that is first affected in response to the virulent mechanisms of the periodontopathogens. The attachment of the junctional epithelium to the tooth occurs at the dento-gingival junction via an epithelial attachment comprised of basal
lamina and hemidesmosomes. The junctional epithelium comprises an external basal lamina where it attaches to the underlying connective tissue and an internal basal lamina towards the tooth surface$^{7,21}$.

2.1.2 The oral microbiome

The oral microbiome comprises of a plethora of microorganisms that are present primarily as a host-associated biofilm in synergistic relations of varying capacities. Dental biofilms form via an ordered sequence of events, resulting in a structurally and functionally organized, species-rich microbial aggregation. The bacteria are retained in an intercellular matrix that is derived from saliva, gingival crevicular fluid (GCF) and bacterial products. These primarily comprise of organic constituents such as polysaccharides, proteins, glycoproteins, lipids and DNA and inorganic constituents such as calcium, phosphorus, sodium, potassium and fluoride. These microorganisms have been identified and clustered into various complexes based on their isolation from specific stages of the disease process.

A large number of plaque samples were analyzed through DNA-DNA hybridization technique the presence of specific microorganisms playing a role in periodontal infections$^{22}$. One of the key findings of this study was the definition of bacterial complexes, as opposed to individual bacterial species, that were associated with either periodontal health or disease. The early colonizers from the yellow, blue and purple complexes comprise primarily of gram-positive rods and cocci that were predominantly seen in periodontal health and included bacteria from the streptococcus, velionella and actinomyces species. The green complex included Capnocytophaga and Aggregatibacter Actinomycetamcomitans (serotype A) species. The orange complex
contained those organisms generally considered to colonize dental plaque later, i.e. Fusobacteria, Prevotella, and Campylobacter species. The presence of these organisms is now thought to facilitate colonization of mature dental biofilms (microbiome) by the red complex organisms either through the presentation of appropriate binding sites or by the creation of a suitable environment for the growth of these more particular species. The “red complex” is composed of three bacterial species; *Porphyromonas gingivalis*, *Treponema denticola*, and *Tannerella forsythia* and were found to be most strongly associated with severe periodontal disease.

This finding led to the concept that there exists a co-dependency or synergy between different bacterial species acting in concert as a specific complex. A pyramid was constructed to designate the various complexes and their association with plaque development.

Increased accumulation of these periodontopathogens overwhelms the host defences, leading to an increased accumulation of inflammatory cells and a proliferative response of the junctional epithelial cells. Elaboration of destructive enzymes and inflammatory mediators destructs the underlying connective tissue leading to an apical migration of the junctional epithelium and pathological deepening of the gingival sulcus forming a periodontal pocket.

### 2.1.3 Periodontal disease pathogenesis

Inflammatory and immune reactions to microbial biofilms are the predominant features of gingivitis and periodontitis. The interaction of the microorganisms with the host determines the course and extent of the resulting disease.
Classical studies by Loe et al.\textsuperscript{26} established the critical role of dental plaque in initiating gingival disease and few studies demonstrated that the response to biofilms was variable amongst individuals indicating the possible existence of an inherent host response\textsuperscript{27,28}.

Ultimately, criteria were devised to enable the identification of putative pathogens that cause periodontal disease. Pathogenic potential of bacteria within the biofilms varies from individual to individual and from site to site. Microorganisms may exert their pathogenic effects either directly by causing tissue destruction itself or indirectly by stimulating and modulating the host response\textsuperscript{29,30}.

The pathogenicity of microorganisms is related not only to the host's innate or immune-modulatory capability but also to the virulence of the bacteria themselves. The host response functions in a protective capacity, preventing the local infection from progressing to a life-threatening systemic infection. However, local alteration and destruction of host tissues, which is evident as periodontal disease, may result.

Haffajee et al.\textsuperscript{31} explained that in addition to understanding etiology and pathogenesis of periodontal disease, it is essential to recognize the site-specific nature of the disease. They explained the attachment level changes that occurred over the period of destructive periodontal disease progression and cited the models of periodontal disease to include the continuous, random burst or asynchronous multiple burst activity.
Inflammatory and immune processes in periodontal tissues are a response to a large number of microbes that reside outside the soft tissue and their products acting over an extended period. The composition of bacterial plaque associated with gingival health differs from that of plaque associated with different periodontal diseases. When specific bacteria within the plaque increase to significant numbers and produce virulence factors beyond the individual patient’s control threshold, the balance shifts from health to disease. The shift occurs in combination with the simultaneous reduction in the host defensive capacity.

Microbiological factors significantly influence the development of gingivitis and its progression to periodontitis. Classic theories for the role of dental plaque as an etiological agent in periodontal disease included the non-specific (quantitative plaque differences) and specific-plaque hypotheses (qualitative plaque differences). A relatively newer concept is the ecological plaque hypothesis that is based on the nonspecific plaque accumulation creating an environment change for the microbial shift of specific periodontal pathogens leading to disease progression.

On reviewing periodontal disease pathogenesis, it was argued that rather than the disease being caused by specific bacteria, the disease selects the bacteria adding more strength to the ecological plaque hypothesis.

More recently, the ‘keystone pathogen concept’ was introduced by Hajishengallis et al. according to which specific microorganisms were identified as being keystone pathogens such as P.
*gingivalis*, i.e. they played a critical role in periodontal disease pathogenesis and progression despite being present in relatively low abundance.

The stable synergistic interrelationship of the microorganisms in a dental biofilm is usually disturbed following invasion by a keystone pathogen or changes in the environmental milieu leading to dysbiosis, a predominance of opportunistic pathogens and onset of disease. The concept of ‘Polymicrobial synergy and dysbiosis’ was introduced and according to this model, a synergistically surviving heterogeneous population causes the periodontal disease and was in a state of homeostasis resisting the host immune response as a combined activity of the microbiome community. Disease becomes active and progressive with the activation of the keystone pathogens that interact with the accessory pathogens in the community and enable the breakdown of the host immune response and enhance the pathogenicity of the host-associated (biofilm) microbiome.

### 2.1.4 Stages of periodontal disease progression

Biofilms induced disease may be classified into three main categories of health, gingivitis and periodontitis. Page and Schroeder explained the development of gingivitis lesion based on histopathological and ultra-structural analysis and categorized the stages in the pathogenesis:

- **Initial Lesion**: Occurred within 2-4 days following the accumulation of dental plaque and clinically characterized by an enhanced GCF flow with an increased presence of serum proteins. Histologically, engorged blood vessels were seen with increased accumulation of PMN’s spell out and 5-10% loss of perivascular collagen.
• **Early lesion** developed within 4-7 days of plaque accumulation and T cells were the predominant infiltrating cells with evidence of cytopathic alterations of the fibroblasts and 60-70% of collagen loss. Vascular changes were extremely pronounced and clinically evident by the presence of increased bleeding on probing.

• **Established lesion** usually occurred within 14-21 days of plaque accumulation and was typically characterized by an abundance of B-lymphocytes and plasma cells without evidence of bone loss. Connective tissue loss was progressive with simultaneous proliferation, apical and lateral migration of the junctional epithelium.

• **Advanced lesion** included periodontal pocket formation and loss of supporting alveolar bone and marked the transition between gingivitis and periodontitis.

These stages till date explain the mechanism of pathogenesis of a periodontal lesion from its inception as gingivitis and progression to develop into a severe form of periodontitis lesion. Periodontal destruction as mentioned earlier usually occurs in an episodic fashion with periods of exacerbation followed by periods of quiescence or inactivity and are characterized by a conversion of a T-lymphocytic to a predominantly B-lymphocytic lesion\(^3^8\).

Immunohistochemical assessment demonstrated that periodontitis lesions had significantly higher numbers of macrophages, plasma cells and auto-reactive B1a cells as compared to long-standing established gingivitis cases. There was no significant difference found in the proportions of T and B-lymphocytes between the two lesions\(^3^9\).

Microbiologically this change is hallmarked by an increase in the number of unattached, motile, gram-negative anaerobic rods and cocci from the less pathogenic dense non-motile gram-positive
periodontopathogens. This transition is thought to be triggered by an invasion of the bacteria into the underlying connective tissues leading to the mounting of a host immune response\textsuperscript{40}.

**Summary:**

Microbial organisms, having co-evolved with the innate defence systems of their respective hosts, have developed strategies not only to overcome protective host barriers but also to manipulate these systems to their advantage. The current method of managing periodontal disease remains the mechanical disruption of non-specific dental plaque (biofilms). Most investigators have reported the presence of gingival inflammation due to the presence of this complex biofilm with variability noted in the composition at both individual and site-specific level. The relationship between the oral microbiota and the host is dynamic, and several intrinsic and extrinsic factors can perturb this exquisite balance. Therefore, an understanding of this relationship is critical for the effective clinical management of dental patients both during health and when managing disease.

### 2.2 Assessment of Periodontal disease

#### 2.2.1 Introduction

Diagnosing and regular monitoring of periodontal diseases necessitates the use of clinical periodontal parameters. These parameters provide baseline assessments and diagnostic statements to which future comparatives are drawn over the course of periodontal management. Traditional clinical parameters are still considered the gold standard to diagnose and classify periodontal diseases which include subjective assessment of soft tissues; changes in gingival color, contour, consistency and surface texture, and followed by a more objective assessment of clinical inflammation, i.e. bleeding on probing (BOP). The progression of infection is best
evaluated using a periodontal probe to determine the probing depth (PD), which is the distance between the gingival margin and the base of attachment (sulcus/pocket). PD serves as a surrogate endpoint to measure the disease process as well as response to periodontal therapy\textsuperscript{41}.

Periodontal disease is characterized by a pathological deepening of the gingival sulcus due to the host's microbiome inducing the breakdown of the underlying periodontal ligament fibers and connective tissue. The loss of periodontal tissue support is measured by the determination of clinical attachment level (CAL) which is the distance between the cementoenamel junction and the bottom of the sulcus/pocket. CAL measurements provide a more reliable estimate of the amount of destruction and a therapeutic endpoint. Additionally, various periodontal parameters such as the presence of gingival recession, plaque, calculus, suppuration, tooth mobility and furcations are comprehensively assessed to determine the periodontal status\textsuperscript{42}.

2.2.2 Periodontal probes: Generations\textsuperscript{43}

The word ‘probe’ is derived from the Latin word *Probo* that means ‘to test’. Periodontal probing to determine pocket depth and clinical attachment levels have been considered the single most effective method since times immemorial for diagnosing periodontal disease as well as for assessing changes following therapy. It helps to identify the location of the apical extent of a pathologically deepened gingival sulcus and helps quantify the loss of attachment of the periodontally affected teeth.
**First generation:** These include the conventional or manual probes that are neither pressure controlled nor allow for automatic data collection. These devices are readily available, inexpensive and color-coded for easier visual identification. The tactile sensitivity is preserved with these probes thereby enabling maneuverability around sub-gingival deposits/margins of restorations to provide accurate readings. However, these probes are not force controlled and may be subject to errors in visualization and transfer of readings. Some examples include Williams probe, CPITN, UNC-15, University of Michigan O probes.

**Second generation:** These are pressure sensitive probes designed with standardization of forces up to 20 g. However, no computer storage of data and the readings have to be entered manually e.g.: True pressure sensitive probe, Yaple probe.

**Third Generation:** These probes are pressure controlled as well as automated probes with computer-assisted direct data capture with digital readouts thereby eliminating errors in data transfer. They also enable estimation of the CEJ and have a higher resolution of 0.2 mm as compared to 1 mm in a manual probe. These are however lacking in tactile sensitivity that may lead to an underestimation of PD and CAL, especially in untreated cases. These also have a learning curve and require training and calibration before attaining proficiency in their use. Additionally, they may cause increased patient discomfort due to a standardized application of force and lack of patient-dentist feedback.

**Fourth Generation:** These are 3D probes that are being developed to image the pocket serially and sequentially. These are however invasive in their design.
**Fifth generation:** These probes use ultrasound imaging based on mechanical vibrations at frequencies above the limit of human audibility. This system has been studied to accurately determine the distance between the gingival margin and alveolar crest and tooth supporting structures. These are not only 3D but also non-invasive with ultrasonic waves detecting the apical extent of the periodontal tissues and are attached to computer readout, and automated data capture is attained. These are however expensive, need professional training and are still being investigated as useful adjuncts to conventional techniques to provide a more accurate diagnostic assessment of periodontitis\textsuperscript{44}.

**Automated pressure-controlled probes:**

Gibbs et al.\textsuperscript{45} worked in developing a novel periodontal disease recording instrument that was named the Florida Probe®️, since it was developed following research at the University of Florida. It was made to meet the requisite guidelines laid down by the NIDR (National Institute of Dental Research) protocol that included the following:

1. A precision of ± 0.1 mm
2. A range of 10 mm
3. Constant probing force
4. Non-invasive, lightweight and easy to use and learn
5. Should be able to access any site/location around all teeth
6. A guidance system to ensure reproducibility of reference for measurements
7. Possible complete sterilization of all its parts in contact with the oral cavity
8. No risk of bio-hazard or electric shock
9. Digital output directly to a computer-based interface
The introduction of computer-aided periodontal probing in dental offices reduces the time and stress associated with the full mouth traditional manual periodontal probing. It provides a constant point of reference for probe tip insertion that is usually the gingival margin or surface of a stent both being easily identifiable. This probing system combined the advantages of an automated and pressure controlled probing system with an additional ability of storage of data on the computer. The probe tip reciprocates back and forth through a sleeve, the edge of which rests at the gingival margin to obtain an accurate reading. To further enhance the reproducibility of the reading and also to establish a fixed reference point, use of an occlusal stent was suggested and the probe tip was suitably modified for this purpose. The probe applies a constant force of 15 g with a resolution of 0.2 mm. The connected probe automatically records into a digital workflow and colour-coded elements that are easy to understand and comprehend by the patient. The patient involvement is enhanced as they listen to voice call out and verbal warnings of their deep pockets making them more involved, aware and interested in following up with their treatment and improved compliance to oral care.

Several studies were conducted to compare its clinical usefulness to manual probes. Clark et al.\textsuperscript{46,47} conducted experiments using the pressure controlled automated probes to determine the range of measurement errors when compared to manual probes alone or used with auxiliary devices such as splints. The results showed a substantial reduction in the probing error ranging from 0.4-0.7 mm to 0.2-0.3 mm on using electronic probes\textsuperscript{46,47}. Hence, this provided a considerable gain in sensitivity of detecting changes. Outlier measurements were obtained even with the use of electronic probes, and hence in such cases, a mean value of two of the closely
matched readings was considered, and this option provided the most efficient in minimizing measurement errors.

Results from another in vitro experimental study comparing the effectiveness of manual vs. electronic probes indicated that automated probes depicted superior accuracy over conventional probes and that they matched the manual probes for their degree of reproducibility\textsuperscript{48}.

A systematic review to assess the reproducibility of electronic probes when compared to manual probes in measuring CAL in patients with untreated chronic periodontal disease included clinical trials with comparative assessments between the two generations of probing systems and also in which examiners were calibrated. Given only 2 studies were found suitable, no meta-analysis could be performed. Results revealed that reproducibility of manual probes was comparable to those of the electronic probes although the evidence was weak due to small sample size. However, the review emphasized the need for calibrating examiners in a clinical trial as well as blinding them to minimize bias and improve the strength of evidence\textsuperscript{49}.

\textbf{2.2.3 Bleeding on probing (BOP).}

The risk predictor BOP is an essential parameter for periodontal disease. The benefits of maintenance care at regular intervals in patients treated successfully for periodontal disease cannot be stressed enough. Lang et al.\textsuperscript{50} conducted a retrospective study to evaluate the prognostic value of bleeding on probing in identifying sites with periodontal disease activity in recall patients. The study revealed that when the mean BOP scores exceeded 16\%, there were significantly more residual pockets $\geq 5$ mm and significantly more sites losing $\geq 2$ mm of attachment. The key finding was the fact that the predictability values for losing probing
attachment increased exponentially from 1.5% in the absence of BOP to 30% with a BOP incidence of 100%. On the other hand, the absence of BOP showed an almost 100% predictability for health in the present study.

The eliciting of BOP during periodontal probing is also strongly governed by force applied. Another study by Lang et al.\textsuperscript{51} correlated the effects of increasing probing forces on the BOP scores and he found that mean BOP% increased from 7.1% to 41.5% on increasing the probing forces from 0.25N to 1N. The results of this study also demonstrated that the force applied to a probe with a standardized point diameter of 0.4 mm must not exceed 0.25 N if the traumatization of the healthy tissues has to be avoided.

A longitudinal assessment of absence of BOP as an indicator of periodontal stability showed an increased prevalence of BOP in sites with increasing pocket depths and greater attachment loss during the maintenance period. 86% of the sites with shallow pockets (1-3 mm) rarely exhibited BOP. Selecting a standard of $\geq 2$ mm for loss of attachment, the positive predictive value of BOP was only 3.8-5.8% whereas the negative predictable value for disease progression of BOP was almost 98.1%. Therefore, the absence of BOP has been identified as a good indicator for maintenance of periodontal stability\textsuperscript{52}.

2.2.4 Challenges in periodontal disease assessment

Some of the inherent challenges with the measurements of clinical parameters include the subjectivity in the assessment of categorical variables as well as the inherent variability both in
the intra and inter-examiner assessment. Moreover, periodontal disease is heterogeneous and shows an irregular pattern of presentation\textsuperscript{53}

Periodontal probing depths can be quite variable even at the same site due to many factors. These include operant variations, instrument manufacturing errors, the health of the periodontal tissue and measurement errors.

Operator related variability could include angulation of the probe, pressure applied, changes in working position and/or site of measurements; line angle vs. interproximal or differences in the anatomical contours of the tooth crown.

Probing forces as previously identified with BOP, can impact the results. Studies have shown a positive, linear relationship between probing depth and probing force\textsuperscript{54,55}. A gentle force of 0.25N or 25 g is the gold standard for probing depth assessment. The pressure exerted by the probe is directly proportional to the force on the probe and inversely proportional to the area at the probe tip. Doubling the force will increase the pressure by a factor of 2, and if the diameter is doubled, the pressure is reduced by a factor of 4\textsuperscript{56}. Hence, standardization of probing forces is essential for minimizing operator bias and improving reproducibility of measurements. A pressure controlled probing system enables application of a steady force and thereby provides higher accuracy.
The manufacturing errors include variations in the tip diameter, standardization of tine characteristics, parallel-sided vs. tapered probe and variances in the graduation scales in different batches produced leading to lack of instrument design standardization.

A dog model study showed that application of a 20 g force on the probe handle with a diameter of 0.6 mm would exert forces of 40-50 Ncm² and best measure sulcus depth by being placed at the coronal end of the junctional epithelium. The characteristics such as a parallel probe tine vs. tapered as well as the presence of a ball tip resulted in deeper probing depth measurements.

An opposing pressure from the underlying connective tissue attachment limits the advancement of the periodontal probe in the gingival sulcus. The presence of inflammation reduces this tissue tone considerably and results in a deeper penetration of the probe beyond the junctional epithelium and into the underlying connective tissues.

A histological study demonstrated a linear relationship between the inflammatory status of the gingival tissues and their resistance to probe penetration. A constant probing force of 30 g resulted in a mean penetration of 0.30 mm coronal to the connective tissue attachment when the gingival tissues were healthy with an index of 0 as compared to being 1.25 mm apical to the connective tissue attachment when the gingival index was 3 characterized by edema, color change and spontaneous bleeding from the gingival tissues due to presence of severe inflammation. The location of the probe tip in the gingival sulcus, therefore, depends on the pressure applied, the diameter of the probe tip as well as the health of the underlying tissues.
One of the primary reasons that lead to unpredictability and variability of the parameters assessed is the measurement error. Systematic errors refer to the differences in measurements by the same examiner across different time points. There is a definite under or over-estimation while recording probing depth and the presence or absence of this measurement bias is referred to as ‘accuracy’. The second measurement error is a random error that is not predictable and refers to the ‘precision’ of measurements. It is estimated from data obtained from duplicate measurement readings. The third is the presence of outlier readings that should be identifiable and excluded to improve data quality prior to analysis. Additionally ‘consistency of readings’ both intra and inter-examiner is also essential. It is therefore prudent to provide proper training both didactic and clinical to the examining team before beginning the study followed by their application to a subject population with similar inclusion criteria as the study. These could range from 3-5 subjects in periodontal clinical trials.

Measurements of the testing parameters are made on a single quadrant of each subject and then repeat measurements performed not earlier than 15 min from the completion of the first examination. This helps in minimizing the memory of the previously recorded readings. The reproducibility of the readings is then analyzed using percentage agreements within ± 1 mm for continuous variables such as probing depth and clinical attachment levels. These values should usually lie within this range >90% of the time^60.

These reproducibility readings help determine the variability of the examiners or within the same examiner that should ideally be minimized. Hence, calibration in clinical trials involves two critical steps; examiner alignment where the appropriate training in the index system or use of
specific equipment is provided, followed by examiner assessment under which the reproducibility study is performed.

It is always recommended to establish a baseline threshold for the change in parameters to classify as disease progression. This ensures that a cut-off value that will represent a clinically significant change has been determined before the beginning of the study. The standard deviation associated with measurement errors while recording clinical parameters are found to be in the range of 0.8-1 mm and hence to be able to identify a definitive change in PD or CAL, the value should exceed 2 mm\textsuperscript{24}.

Use of controlled force probes and taking a repeated set of readings are considered some ways to minimize these errors; however, these are not exempt from their own set of limitations. For example, the Florida Probe\textregistered automated pressured probe has a decreased tactile sensitivity and with the controlled force probe, the patient-dentist feedback is also not possible due to insertion of the probe in one motion and with a predetermined force.

**Summary:**

Periodontal disease diagnosis, prognosis and treatment planning strongly depend on the assessment of the disease using traditionally employed methods of measurement. Periodontal probing to ascertain pocket depth and attachment level continues to be the mainstay for diagnosing periodontal health. The long-term monitoring of periodontal health status to assess progression or stability of disease as well as response to treatment is also achieved with this tried
and tested methodology. Some of their limitations were thought to be resolved with the innovation of pressure-controlled probes that provided some degree of enhanced accuracy, resolution and reproducibility but were not wholly free of measurement errors. Despite these facts, the periodontal probes remain the best diagnostic tools to assess periodontal tissue health. Hence, appropriate training and examiner alignment are highly recommended to minimize bias and potential errors in measurements.

2.3 Non-surgical periodontal therapy (NSPT)

2.3.1 Introduction

Periodontal disease is a chronic infectious disease modified by a host associated oral microbial biofilm. Within the oral microbiome the removal of soft and hard deposits, as well as disruption of the symbiotic sub-gingival microbial relationships, has been found to be an essential modality for its treatment\(^61\). NSPT continues to remain the gold standard for phase I therapy as part of non-surgical and before surgical periodontal procedures\(^62\). According to Waerhaug\(^63\) the rough surface of calculus does not in itself induce inflammation but that its deleterious effects relate to its ability to provide an ideal surface for microbial colonization and hence the rationale behind its removal rests on removal of these surface irregularities that serve as a niche for plaque accumulation. Real time observation of the sub-gingival sites with the help of a dental endoscope/perioscopy system has enabled detection of residual calculus and presence of inflammation of the pocket wall that is clinically evident as BOP\(^64,65\). These calculus deposits were covered with a bacterial biofilm 100\% of the time causing a sustained inflammation in the adjoining tissues.
The calculus adheres to the underlying tooth structure through various mechanisms; via the dental pellicle, mechanical interlocking into resorption areas, grooves and tooth and root surface irregularities, direct attachment or the penetration of the bacteria into the underlying root structure\textsuperscript{66}.

These bacteria and their endotoxins invaded deep into the porous calculus extending to the underlying tooth structure and resulted in necrotic changes in the cementum. The removal of this necrotic cementum was believed to be essential to ensure complete elimination of microorganisms. Currently, this concept is refuted and the objective is to disrupt and remove the microorganisms rather than aggressively removing unnecessary cementum\textsuperscript{67}. Histological studies demonstrated not only healing of periodontal tissues around the surface of decontaminated calculus but also attachment of the junctional epithelium\textsuperscript{68}. Studies also confirmed that the lipopolysaccharides and endotoxins of the colonizing bacteria were not penetrating into the subsurface layer of cementum\textsuperscript{69}. The current consensus of a therapeutic endpoint of periodontal procedures is to achieve a biologically compatible surface to enable a healthy periodontal attachment, i.e. arresting inflammation\textsuperscript{70}. To eliminate the niche that favours bacterial accumulation and recolonization the removal of calculus that is embedded into the root surface cementum is required.

Supra-gingival plaque biofilm serves as a constant reservoir of microorganisms that migrate apically and repopulate the sub-gingival microenvironment. Therefore, both supra and sub-gingival debridement on a consistent basis are crucial for the long-term maintenance of periodontal health\textsuperscript{71,72}. 

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Patients usually remove the supra-gingival plaque accumulated on the teeth surfaces with daily oral self-care procedures such as brushing, flossing and use of other adjunctive oral hygiene devices. Brushing with the use of modified Bass technique has shown that the toothbrush bristles penetrate up to 1 mm into the gingival sulcus and disrupt sub-gingival plaque. Brushing however was not very useful in minimizing interproximal plaque removal and bleeding and this effect of 35% was found increased to 67% with the adjunctive use of dental floss. Dental floss has been found effective in disrupting sub-gingival plaque to a depth of 2-3.5 mm and was able to remove up to 80% of interdental plaque on diligent use. In patients with periodontal disease with presence of wider gingival embrasures, interproximal brushes are a more effective aid than flossing in plaque removal. Additionally, mouth-rinsing with antiplaque agents was only found to penetrate the sulcus depth to about 0.2 mm. The healthy or inflamed gingival tissues with sulcus depth up to 3 mm maybe maintainable to a great extent with oral self-care measures alone.

A systematic review showed moderate strength of evidence for repeated and effective oral self-care in patients with gingivitis to be as beneficial as repeated professional plaque removal. However, in patients with periodontal disease and pockets deeper than 3 mm, a consistent in-office professional periodontal debridement (instrumentation) is pivotal to bridge these deficiencies, as oral self-care is inadequate to sustain the long-term periodontal health without comprehensive professional intervention.

Clinical trials have shown that self-care programs alone are not effective in reversing gingival inflammation without the intervention of professional debridement. Cercek et al. assessed the
usefulness of plaque control strategies using oral physiotherapy as compared to professional instrumentation in its effectiveness after two years in treating patients with chronic periodontal disease. The results of the study showed that oral self-care methods although effective in minimizing inflammation were inadequate in improving periodontal health unless combined with professional debridement\textsuperscript{78}.

NSPT (debridement) aims to eliminate both living bacteria in the microbial biofilm and calcified biofilm microorganisms (calculus) from the tooth surface and adjacent soft tissues. The therapy leads to a decrease in the bacterial loads and hence lesser tissue inflammation and beneficial clinical changes. Also, it helps create an environment in which the host can more effectively prevent pathogenic microbial recolonization using daily oral self-care methods.

\subsection*{2.3.2 Limitations of NSPT}
A critical probing depth of 2.9 mm is considered a necessary pre-requisite for performing periodontal instrumentation (scaling/root-planing) procedures to attain gain in clinical attachment\textsuperscript{79}. Substantial probing depth reductions and clinical attachment gain are anticipated in deeper pockets ranging from 4-6 mm or \textgreater 7 mm vs. shallower sulcus of \textless 3 mm depth that would lose attachment following a NSPT procedure. A mean PD reduction of 1.29 mm and 2.16 mm and a mean gain of CAL of 0.55 mm and 1.25 mm has been noted in pockets ranging in depth from 4-6 mm and \textgreater 7 mm respectively\textsuperscript{61,80}. Initial shallow sulcus depths (1-3 mm) could reach a loss of 0.42 mm attachment from SRP.

Some of the drawbacks identified with non-surgical periodontal instrumentation include pain and discomfort in the gingival tissues; post scaling tissue trauma, swelling and bruising, gingival
recession with open interdental spaces and increased hypersensitivity[^81]. The effectiveness of NSPT also decreases with increasing pocket depths. Calculus free surfaces were found to be 87% in up to 3 mm sulcus depth and diminished to 43% and 32% with increasing depths of 4-6 mm and >7 mm respectively[^82]. The presence of furcation defects, root proximities, root grooves, mal-alignment of teeth further adds to complexities in attaining effective plaque and calculus removal with NSPT alone. The microorganisms in these sites also can invade into the underlying connective tissues that provide a niche for recolonization.

### 2.3.3 Pain and discomfort following NSPT

The International Association for the study of Pain definition for pain is “an unpleasant sensory and emotional experience associated with actual or potential tissue damage or described by the patient concerning such damage”. It not only represents the intensity of nociceptive stimulation but also includes the effects of fear and anxiety on pain reactivity[^83]. Painful stimulation not only activates the receptors and sensory pathways leading via thalamus to the cortical areas but also activates areas in the brain that are involved in emotional reactions. Pain is a subjective experience, and various ways of quantifying it include the visual analogue scale (VAS), numeric rating scale, verbal response scale, behavioral pain scale to name a few. Although significant bias exists in its measurements of self-reported responses, the VAS has been a time-tested and widely used tool for quantifying pain.

VAS is a uni-dimensional single item scale that is a straight 10cm line with verbal descriptors at either end defining extreme limits of the sensation to be measured as 0 being ‘no pain’ to 10 being ‘maximum pain’. The participant is asked to mark a vertical line at a point that they feel
represents their current perception of pain response. The VAS score is determined by measuring in millimetres from the left-hand end of the line to the point that the patient marks. There exist infinite points between these two ends for the patient to mark. This scale is reliable, valid and more straightforward to administer, and has been effectively used to quantify dental pain\textsuperscript{84,85}.

There are however some limitations with its use such as the difficulty for patients to accurately rate as there is no guidance available, which may result in greater variability. The addition of vertical lines and or lines with extra descriptors to the scale have offered more sensitivity for assessment to address this. A systematic review showed the numeric rating scale had better compliance, responsiveness, ease of use and acceptability compared to VAS\textsuperscript{86}.

Dental treatment has always been found associated with varying degrees of pain, and the perception of this pain may be significantly influenced by factors such as the level of anxiety and fear, previous dental experiences, socio-demographic variables and level of dental awareness. Studies have been conducted to assess the perception of pain and discomfort during periodontal probing, scaling, root surface debridement, periodontal surgery and maintenance treatment.

The NSPT procedure usually results in some amount of inadvertent tissue trauma to the gingival tissues. A study by Claffey et al.\textsuperscript{87} showed that 22\% of the sites lost >1 mm attachment following SRP due to injudicious instrumentation use. This trauma possibly triggers the activation of the local mechanoreceptors and polymodal nociceptors that eventually leads to release of prostaglandins; bradykinin and histamine that stimulate pain pathways to the central nervous system. Pihlstrom et al.\textsuperscript{11} reported pain and discomfort of various degrees in 90\% of the patients
following NSPT. 16% of the patients reported the pain to be moderate to strong and 23% patients self-medicated with analgesics to relieve post-procedural pain. The peak pain intensity was found to occur 2-8 hours following NSPT procedure\textsuperscript{11}. Several other studies have also shown the perception of varying degrees of pain amongst patients undergoing NSPT\textsuperscript{88-90}.

A study to examine the degree of pain experienced by patients during the NSPT procedure and the influence of dental anxiety on the patient’s perceived response to pain was conducted\textsuperscript{91}. The results of the study showed limited experience of pain during scaling and root planing procedure with a mean VAS score of 19.91±17.76 indicating a high degree of subject variability in the response. Women, non-smokers and patients in the age group of 30-40yrs were found to be more anxious during the dental procedures and this reflected in their higher VAS scores too; however, the correlation of anxiety to higher pain experience was statistically significant only in non-smokers\textsuperscript{91}.

A systematic review reported that dental anxiety between surgical and non-surgical periodontal treatment groups showed no significant difference. Dental anxiety however had a significant effect on expected pain before, during and after dental treatment\textsuperscript{92}. Van Wijk et al.\textsuperscript{93} showed in a study that patients who had a higher level of anxiety to pain response were at a significantly higher risk of ending up in a vicious cycle of anxiety, fear of pain and avoidance of dental treatment.

Measurement of pain is inherently difficult because of its dual aspects of physical and emotional components. One is entirely dependent on the patient’s response based on their perception and interpretation of pain. Other covariates such as age, gender, type of therapy and level of anxiety
all have modifying influences on the patient’s response to pain. Therefore, all these parameters need to be assessed to have a better estimation of pain.

2.3.4 Dentin Hypersensitivity

According to the Canadian advisory board in 2003 on dentine hypersensitivity, dentin hypersensitivity (DH) is defined as pain derived from exposed dentin in response to chemical, thermal, osmotic or tactile stimuli that cannot be explained as arising from any other defect or dental disease\textsuperscript{94}. To accurately diagnose DH, increased sensitivity due to caries, cracked tooth, micro-leakage or fractured restoration should be ruled out\textsuperscript{95}.

The various theories laid down for its pathogenesis include the neural theory, odontoblast transduction theory and the hydrodynamic theory. Amongst these the most accepted is the hydrodynamic theory that was laid down by Brannstorm 1964. According to this theory, when the uncovered dentinal surface is exposed to thermal, chemical, tactile or evaporative stimuli, the fluid flow within the dentinal tubules is expected to increase. This fluid movement causes an alteration in pressure and excites pressure-sensitive nerve receptors in the innervated pulp tissue, which infers, that the intensity of pain is directly correlated with the increase in number and diameter of the dentinal tubules\textsuperscript{96}.

The various methods of assessment of dentine hypersensitivity include the stimulus-based assessment or a subjective patient response-based assessment of the application of distinct stimuli. These stimuli-based assessments rely on the individual’s pain threshold to varying intensities of stimuli. One of these techniques involves the application of a calibrated probe with
increasing tactile pressure at the explorer tip in 10g increments until the patient experiences discomfort. Alternatively, a thermal or electric device may be used by an application of graded stimuli.

The response-based assessments measure pain severity following application of a standardized, constant and reproducible stimulus. Air-blasting the isolated buccal surface has been consistently used and measuring the response on a pain assessment scale where the type of scales could vary from verbal response to visual analogue or numeric rating scales each with its advantages and limitations.

Patients who have received NSPT frequently report an increased sensitivity of the teeth to hot, cold, evaporative and osmotic stimuli. This heightened response usually peaks during the first-week post therapy and then gradually subsides or disappears in the subsequent weeks.

A systematic review assessed the incidence of dentin hypersensitivity of 62.5-90% at day 1 following NSPT that decreased to approximately 52.6% at the end of week 1. A clinical trial was conducted to assess the incidence and severity of dentine hypersensitivity in 35 patients following NSPT for moderate to severe periodontal disease assessed over a 4-week period. The results showed that scaling and root planing resulted in transiently increased root dentine sensitivity in the immediate follow-up period after NSPT. In this trial, about 31% of patients reported an increase of VAS score >4cm for 1-2 teeth after SRP while only 6% reported this change for 5 teeth or more. When the threshold for VAS assessment was reduced to >2cm, 34% subjects reported sensitivity in 1-2 teeth while 26% of the subjects reported 3 or more teeth
affected. The sensitivity was found to reduce in intensity over the 4-week assessment period, but the incidence of affected teeth remained unchanged.

The inadvertent removal of cementum during SRP procedures results in the exposure of dentinal tubules that are subjected to hydrodynamic forces resulting in movement of the dentinal fluid and causing an increased response to stimuli that is eventually perceived by the patient as pain.

An acidic environment due to the continued presence of plaque deposits on the root surface has been shown to promote patency of dentinal tubules, favouring the dissolution or prevention of any peripheral deposition of minerals in the dentinal tubules\textsuperscript{98}. Microscopic studies have confirmed when these teeth and root surfaces are kept free from plaque during the maintenance period; they become highly mineralized and display mineral depositions at the peripheral ends of the dentinal tubules\textsuperscript{99,100}.

In an in situ model using dentine slabs placed in the oral cavity and later assessed through SEM evaluation for the effect of plaque control on the patency and occlusion of dentinal tubules. In this study, the absence of plaque control, the diameter of dentinal tubule orifices was found to increase by 390\% of baseline values within 3 weeks. In contrast, the tubule diameters significantly reduced to <20\% with the reinstitution of plaque control strategies\textsuperscript{101}.

Suge et al.\textsuperscript{14} demonstrated in a dog model that absence of plaque control resulted in increased incidence of open dentinal tubules by almost 3 times (from 3.51- 10.62\%) after 3 weeks compared to the initial value. The diameter of the dentinal tubules was significantly increased from baseline to within a 7-day period in the non-plaque control group and was also significantly higher compared to the plaque control group. Some of the open dentinal tubules in the plaque
control group also showed occlusion of the tubules with precipitated mineral deposits that were not seen in the absence of plaque control.

A human clinical trial conducted by Tammarro et al\textsuperscript{9} also confirmed that meticulous plaque control significantly reduced the mean VAS scoring over time. The natural recovery from DH is the deposition of salivary minerals calcium and phosphate, which is significantly reduced with residual mature biofilm on the dentine surfaces.

The importance of daily biofilm disruption is pivotal in maintaining gingival health and preventing this vicious cycle of increased DH due to the acidic milieu created following biofilm accumulation that serves as a deterrent to oral self-performed regimes.

2.4 Periodontal Dressings

2.4.1 Introduction

Periodontal wound healing is a sophisticated and coordinated series of events that restore the typical structure and function of the injured tissues. To enable undisturbed healing to occur the environment needs to be protected and kept free from extraneous factors. Periodontal dressing materials were developed with this aim of protecting the surgical sites from mechanical damage or infection and enabling a favourable milieu for unperturbed healing\textsuperscript{102}.

These dressing materials are usually applied at the surgical sites engaging the necks of the adjacent teeth to gain retention with the objectives of holding the periodontal flap in the desired position, protect the underlying healing tissues from trauma or infection, provide some degree of
hemostasis and wound clot stability and above all provide enhanced patient comfort. They have significantly evolved over the last few years and are classified into:

- **Eugenol containing dressing materials**: Ward’s WONDR Pak that comprised of zinc oxide and eugenol mixed and applied. The eugenol was found extremely useful in obtunding pain as well as reduce bacterial growth due to its antiseptic properties. Some of the drawbacks associated with this material included potential allergic reaction, served as an irritant to the oral mucosal tissues and also possible necrosis due to its cytotoxic effects.

- **Non- Eugenol dressing materials**: These are currently the most commonly used following periodontal surgical procedures. They include brands such as Coe-Pak, Peripac, Periocare, and Perio putty. Their constituents include a zinc oxide base with other non-eugenol containing rosins, gums and oils to provide a putty-like consistency. These have the advantage of being non-irritant to the tissues, are less allergenic, have a neutral taste and odour and possess better pliability and adhesive properties serving as a closely adapted barrier to saliva and oral bacteria.

- **Dressing containing neither eugenol nor zinc oxide**: This third group of dressing materials includes cyanoacrylates, light-cured dressings and collagen-based oral adhesives.

Additionally, the available dressing materials have been modified by the addition of antimicrobial agents such as Chlorhexidine, and antibiotics like Bacitracin and Tetracycline.

The applicability of periodontal dressing materials as a routine standard of care is controversial. Studies have reported both an improvement in periodontal health as well as no significant change
following their use post-surgically. Some of the drawbacks cited include plaque accumulation, adsorption of bacterial products, halitosis, and mechanical irritation to the surrounding tissues. Despite these, however, patient comfort is enhanced, postoperative sensitivity is reduced, and an increased psychological feeling of well-being is achieved.

2.4.2 Periodontal dressings following NSPT

NSPT procedures lead to varying levels of inadvertent tissue damage that creates small wound sites that eventually heal by formation of a long junctional epithelium. However, in this critical period of wound healing, it is essential for the clot to stabilize and the wound site to be protected from any mechanical, chemical or bacterial onslaught. The dressing materials not only stabilize and adapt the tissues to the underlying tooth structure but also prevents the colonization of the oral microorganisms. They serve as a barrier and help in creating an isolated microenvironment that favours enhanced wound healing as well as minimizes the hypersensitivity response of the teeth and gingival tissues.

This adjunctive application to NSPT has been attempted in a few clinical trials with positive results. Sigusch et al.\(^{103}\) introduced the concept of applying a periodontal dressing material following NSPT procedure in patients with aggressive periodontitis. Application of the dressing for 7-8 days resulted in significant improvement in the clinical periodontal parameters assessed when compared to the control sites. They also showed that these results remained stable over a 24-month period. Another split-mouth clinical trial showed that a 7-day application of a non-eugenol dressing (Coe-Pak) following SRP procedure and curettage within 24 hour period resulted in a significant reduction in PD, CAL and BOP at the 2-month re-evaluation visit in patients with moderate to severe chronic periodontitis. The dressing was removed at 7 days
following completion of the debridement procedure\textsuperscript{104}. Keestra et al.\textsuperscript{105} conducted a split-mouth RCT where periodontal dressing was placed at test sites following a one-stage full-mouth disinfection procedure. The results of the study showed a significant improvement in PD and CAL in moderately deep pockets in patients with chronic periodontitis over a period of 3 months. Additionally, the patients reported significantly lower intensity of pain perception at sites where the application of the periodontal dressing was performed compared to the control sites\textsuperscript{105}.

A recent systematic review concluded that additional placement of periodontal dressing materials following NSPT could be beneficial in improving clinical outcomes over a short period. However, more controlled and long-term clinical trials are essential to validate their use as a standard of care. All of the included studies in this review used zinc oxide containing eugenol-free dressing materials\textsuperscript{106}.

### 2.4.3 Cyanoacrylates Adhesives

Cyanoacrylates are tissue adhesives that were developed for their medical applicability in wound closure in the 1950s during World War II and have been successfully used as tissue adhesive dressings for several decades in both medicine and dentistry.

### 2.4.4 Structure and Chemistry

Cyanoacrylates are acrylic resins that are synthesized by condensation of a cyanoacetate with formaldehyde in the presence of a catalyst. (\textit{Figure 1}) These rapidly polymerize in the presence of water (specifically hydroxide ions) to form long strong chains. They have an alkyl side chain whose numbers can be increased from methyl (one) to ethyl (two) or the more recent n-butyl
(four), iso-butyl, iso-amyl (five) and 2 octyl (eight) structures. They undergo polymerization through an exothermic reaction that leads to the release of heat as well as thermal decomposition products that may include formaldehyde, hydrogen cyanide and oxides of carbon and nitrogen. Increasing the alkyl chain length significantly reduces toxicity by slowing the degradation rate of the molecule and reducing the metabolites released. Hence, the butyl and octyl cyanoacrylates are indicated for in vivo use\textsuperscript{107}.

These are maintained in their liquid state by addition of an acidic stabilizer that prevents the molecules from cross-linking. Their application to the tissue surface eliminates the inhibitor and exposure to the anionic molecules results in the polymerization\textsuperscript{108}.

Due to the proteinaceous nature of the tissues that contain many base residues, the potential for good wetting of proteins provides the adhesive qualities to the cyanoacrylates. Additionally, it has been shown to exhibit mechanical interlocking due to penetration of the adhesive into tissue surface irregularities. Polymerization of the material occurs within 10-15 seconds. Cyanoacrylates are biodegradable and undergo polymer breakdown by a hydrolytic attack on the C-C bond. These materials are not absorbable and are also sloughed from the surface of the skin and mucosa 7-10 days following surface application\textsuperscript{108,109}.
2.4.5 Therapeutic indications, dentistry and beyond

The use of cyanoacrylates of the methyl and ethyl groups was initially seen in technician laboratories for denture repairs. These formulations released higher levels of formaldehyde on their polymerization and were found to be histotoxic. These were later replaced with the longer alkyl chain containing cyanoacrylates such as the butyl and octyl versions and that found great usefulness in medicine and dentistry⁠¹⁶⁻⁰²⁰.

These adhesives are being extensively used in the field of medicine for soft tissue repairs such as traumatic lacerations, myocardial tears, inguinal hernia repairs, sealing of cerebrospinal fluid leaks, cosmetic rhinoplasty and several more.

Intra-orally, these adhesives have been used in several settings, such as, but not limited to, alternatives to sutures, for stabilizing free gingival and sub-epithelial connective tissue grafts,
cleft repairs, repairs of sinus membrane perforations, hemostatic agents especially in patients on anticoagulant therapy and healing of extraction sockets.

A study assessed the effectiveness of cyanoacrylate in stabilizing free gingival grafts in comparison to sutures of varying thicknesses. The cyanoacrylate adhesive resulted in a successful healing of the graft, significantly less graft shrinkage, less reported postoperative pain at the recipient site and significantly decreased surgical procedure time\textsuperscript{16}.

Apart from varied applications in the oral surgical field, cyanoacrylate adhesives have also found potential with endodontically treated teeth by sealing dentine; orthodontically by bonding brackets, and splinting for stabilizing traumatized teeth, prosthodontics by controlling micro-leakage at the tooth restoration interface, desensitizing teeth, pit and fissure sealants as well as to prevent infiltration of porosities in early carious tissues\textsuperscript{107}.

2.4.6 Applications and Limitations

Cyanoacrylates have found partiality amongst clinicians due to the simplicity and painless application, short operative time, quick adhesion to the hard and soft tissues, the ability to form an effective protective barrier, and the hemostatic and antibacterial properties. These materials are also self-shedding and biocompatible with minimal soft tissue reaction. These are more cost-effective due to the availability of multiuse vials and also reduce the risk of accidental injury from the use of suture needles.
Some of the limitations include the lack of tensile strength and hence a higher risk for wound dehiscence in high-tension areas; sites with heavy moisture and friction due to risk of premature detachment of the material; infected wounds; immune-compromised patients since they are at increased risk for poor and delayed healing as well as patients with known allergy to cyanoacrylates or formaldehyde. The setting through an exothermic polymerization reaction may lead to heat release with safety concerns about the degradation products\textsuperscript{111,112}.

### 2.4.7 Hemostatic Effects

Cyanoacrylates have been found to serve as excellent materials for attaining hemostasis in a surgical setting and have therefore been used for attaining wound closure. The tissue adhering properties help attain an approximation of the wound edges and providing bleeding control. They form a macro-film on the tissue surface causing mechanical obstruction to blood flow and serve as a surface agent that results in the activation of the clotting cascade. As an alternative use to sutures, the application has had varying degrees of success and eliminated the additional suture removal appointments.

To assess the hemostatic abilities of cyanoacrylate glue in surgical extraction wounds in patients on anticoagulant therapy, a clinical trial found the application of topical adhesive highly effective in obtaining hemostasis with no incidence of an adverse event\textsuperscript{20}. Several animal and human studies have found the use of tissue adhesives highly comparable to the use of sutures for attaining successful wound closure\textsuperscript{113–115}. 
2.4.8 Antimicrobial Effects

Cyanoacrylates have been found to possess some degree of antibacterial effects. Quinn et al.\textsuperscript{116} conducted a study to determine the antibacterial effects of cyanoacrylate (n-2- butyl cyanoacrylate) and the risk of contamination when reusing the same vial in multiple patients. The technique of application involves dropping the adhesive from a short distance to approximate the edges of the wound and thus avoiding contamination. Results showed the creation of two distinct zones surrounding the applied tissue adhesive. Zone 1 was a 2-3 mm circumferential area immediately adjacent to the cyanoacrylate. This zone was dehydrated due to the action of the released formaldehyde and cyanide in addition to the heat during the process of polymerization and hence did not support any microbial growth. The surrounding zone 2 depicted lack of growth of gram-positive bacteria possibly due to the action of an active double bond of cyanoacrylate that combines with the free amino or hydroxyl groups present in their cell wall. None of the vials showed any bacterial contamination in this study\textsuperscript{116}.

The antibacterial action of octyl-cyanoacrylate and was found to be effective against Methicillin sensitive Staphylococcus aureus (MSSA) and to a limited extent on Methicillin resistant Staph Aureus (MRSA) isolated from patients with chronic suppurative otitis media. The tissue adhesive is known to have an electronegative charge following polymerization and hence found effective against the positively charged cell wall of gram-positive bacteria\textsuperscript{117}. Ethyl cyanoacrylate as well as n-butyl cyanoacrylate were found effective in inhibiting Bacillus subtilis even when present in high concentrations\textsuperscript{118}. 2 octyl cyanoacrylate had antibacterial effects against gram-positive bacteria and the inhibition was found to continue for up to 10 days following the culturing\textsuperscript{119}. Samples taken from the inhibition rings did not produce bacterial
culture indicating a possible bactericidal action. The study, however, did not have a control group for comparison of the zones of inhibition\textsuperscript{119}. Additionally, another study showed that although 2 octyl-cyanoacrylate was antibacterial to only gram-positive organisms, 2-ethyl cyanoacrylate was found bactericidal to gram-negative bacteria such as E.coli\textsuperscript{120}. (Figure 2)

Apart from the antimicrobial properties of cyanoacrylate that helps decrease the bacterial count at the wound site, it also acts as an impervious microbial barrier that prevents the ingress of surrounding microorganisms. An in vitro study demonstrated the barrier action of 2-octyl cyanoacrylate, which was effective in warding off both gram-positive and gram-negative motile and non-motile bacteria\textsuperscript{121}. Another similar study has also demonstrated a similarly effective barrier function of this tissue adhesive except against for \textit{Pseudomonas aeruginosa}\textsuperscript{122}.

These studies reinforce the benefits of using cyanoacrylates in preventing postoperative surgical site infections; however better controlled randomized clinical trials are necessary to provide the strength of evidence.
2.4.9 Desensitizing Effects

Several techniques both over the counter and in-office have been utilized to manage dentinal hypersensitivity in patients. These include at home application of desensitizing kind of toothpastes containing salts of fluoride, strontium, potassium nitrate, potassium oxalate, arginine or in-office procedures such as the use of lasers to precipitate and coagulate proteins,
iontophoresis to actively introduce these salts to form bonds with the hydroxyapatite and seal the exposed dentinal tubules or even application of dental restorative materials. Cyanoacrylates can minimize dentinal hypersensitivity by forming a protective barrier over the exposed dentinal surface and minimizing the fluid displacement within the dentinal tubules in response to stimuli. Perez et al.\textsuperscript{123} showed that the application of cyanoacrylates was successful in 96.7\% of the enrolled subjects with no incidence of adverse events. However, this study lacked a control group. An older study by compared the use of cyanoacrylates to sodium fluoride (NaF) paste in the treatment of dentinal hypersensitivity\textsuperscript{124}. Cyanoacrylate resulted in an immediate desensitizing effect following a single application and was statistically more effective than NaF, which required repeated applications to be effective\textsuperscript{124}. More recently conducted RCT have shown cyanoacrylates to be equally effective as lasers for the treatment of dentinal hypersensitivity over short and long periods\textsuperscript{125,126}. It was also noted in a randomized controlled clinical trial that, cyanoacrylates and Galium-Aluminium –Arsenide (diode) laser were equally useful techniques to minimize dentinal hypersensitivity and improve the quality of life of the subjects including parameters such as physical pain, psychological discomfort and social disability. Hence, cyanoacrylates offer a highly effective, more accessible and lower cost procedure that may be provided to patients to safely manage DH.

2.4.10 Clinical and Histological Healing

Cyanoacrylates have demonstrated an ability to support ideal healing in tissue wounds. It is hypothesized that the adhesive forms a barrier on the wound surface preventing any seepage of fluids, thereby providing an isolated environment for undisturbed healing and also prevents
secondary infection. The superior surface of the adhesive serves as a scab, providing a moist surface for faster epithelial migration and wound closure\textsuperscript{127}.

One of the most widely used applications of cyanoacrylates in dentistry has been the alternative to suturing of intraoral and other maxillofacial wounds. Several studies have compared the healing variability between the two techniques and have found superior if not comparable results of cyanoacrylates to the use of sutures\textsuperscript{113–115}.

A study showed significantly lesser pain in the first 3 days post-operatively and significantly lesser bleeding following surgery with the use of cyanoacrylate as compared to sutures in the closure of minor intraoral surgical wounds such as surgical extractions of third molars\textsuperscript{114}. Another split-mouth study on alveoloplasty procedures reported a significantly greater number of subjects complaining of tenderness and demonstrating erythema up to 14 days following the surgical procedures\textsuperscript{128}.

Significant reduction in pain and edema following suturing may have been attributed to tissue trauma from insertion of suture needles that created multiple microsurgical wounds creating additional sites of inflammation and healing\textsuperscript{129}. This aligns with Kulkarni et al.\textsuperscript{130} who observed cyanoacrylate adhesives as an alternative to sutures in closure of periodontal flaps to have significantly lowered inflammation in the early healing period. 73.3\% of sutured sites had moderate and 13.3\% severe inflammation on the 7\textsuperscript{th} postoperative day vs. 13.3\% and 6.7\% with the use of cyanoacrylate adhesive respectively.
The medical cosmetic cyanoacrylate glue application in a randomized controlled clinical trial involving the closure of facial skin incisions, identified significantly reduced total surgical time; mean 69.50 ± 33.393 vs. 379.00 ±75.390 seconds with sutures and also superior ratings for cosmetic outcomes and patient satisfaction. There was no significant difference noted in the wound complications when compared to suturing\textsuperscript{115}.

At histological levels, primary closure with sutures found significantly greater inflammation with a denser distribution of neutrophils, lymphocytes, histiocytes and eosinophils as compared to wounds closed with cyanoacrylates only which had a significantly lower incidence of inflammation, reduced vascularity and better epithelialization of the wound at the 1-week post-operative time point\textsuperscript{128,129,131}. These differences became non-significant at the 2 or 3 weeks postsurgical time point.

One of the shortcomings of most of these comparative studies has been the use of silk sutures for wound closure. Silk sutures provide tensile strength and ideal knot tying characteristics; they are however, not ideal for healing. They are multi-filament demonstrating a wicking effect that adsorbs large quantities of fluids and plaque and creating an environment that impedes the rate of healing, which can exhibit a higher inflammatory response during healing as compared to some of the newer monofilament suture materials.

Providing a tension free approximation of wound margins as well as an isolated and protected environment considerably enhances wound healing following surgical procedures.
Cyanoacrylates are consistently justifying their clinical application by fulfilling ideal closure criteria and successfully evolving as tissue adhesives with effective wound repair characteristics.

2.4.11 Biocompatibility and safety

The adverse reactions or events attributed to cyanoacrylates are limited and are linked to a release of substances (i.e. trace amounts of formaldehyde and cyanoacetate) produced during an exothermic reaction following adhesive placement. To minimize this risk, the increasing length of the alkyl chain molecules has significantly reduced the degradation rate of cyanoacrylate; hence, isobutyl or octyl cyanoacrylates are found to be least histotoxic\textsuperscript{132–134}.

Some in vitro studies have assessed the cytotoxicity of methyl and ethyl cyanoacrylates in direct and indirect contacts with fibroblasts and osteoblasts cell cultures and found methyl derivatives to be more cell-damaging due to increased release of heat and toxic by-products\textsuperscript{133,135,136}. It was proposed that these adhesives might generate lipid hydroperoxides that activate the cyclooxygenase pathway and also oxidize and lyse cell membranes. The release of cytotoxic substances has been noted for almost a 2-week period following use of commercially available cyanoacrylates\textsuperscript{133}.

Toxicity in occupational exposure has been observed with the methyl and ethyl cyanoacrylates due to their volatility and chemical reactivity that creates a potentially hazardous environment for workers. These may be present as monomers in acrylic resins and have been found to be associated in rare instances with dermatological reactions such as contact dermatitis and airway hyper-responsiveness (i.e. asthma) \textsuperscript{107}. No true allergic type 1 hypersensitivity reactions have
been reported that necessitated hospitalization or emergency intervention. The type of hypersensitivity that has been associated is type IV\textsuperscript{137}. However, a study investigating the use of various acrylates (including ethyl cyanoacrylate; n= 86/275) via allergic-patch testing, reported no reactions whatsoever\textsuperscript{138}.

Adverse reactions associated with isobutyl and octyl cyanoacrylates are not widely reported and these have been safely used in both in-vitro and in-vivo environments. The US FDA and Health Canada have approved it for use in medicine and dentistry. In Canada, Periacyl (90/10 blend of N-butyl and 2-octyl cyanoacrylate) is considered a Class I device per the Medical Device Regulations. (Appendix I)

**Summary:**

Cyanoacrylates have evolved over the years as materials of choice for various dental and medical applications, the primary ones being to attain hemostasis, wound healing, primary closure and supported secondary closure, stabilization, barrier to enable adequate healing and for minimizing dentine hypersensitivity. Their ease of use and good tissue response has favourably led to their widened scope of use. Various off-label uses of this adhesive have been successfully seen such as to retain locally delivered antibiotics in periodontal pockets following their sub-gingival placement or to control hemostasis as well as at sites with inadvertent tissue damage following NSPT procedures. Due to its high degree of clinical effectiveness and lack of safety concerns there is a need to support various clinical applications and hence more controlled studies are necessary to establish its use as a standard of care in routine dental practice.
Chapter 3: Rationale, Hypothesis and Objectives

3.1 Rationale

The oral health status impacts the overall systemic health. Dental caries and periodontal diseases are two most prevalent oral infectious diseases that have a significant impact on individual’s health and quality of life. The worldwide prevalence of periodontitis in its severe form has been reported to be 11%. According to the latest National Health and Nutrition Examination Survey (NHANES 2009-2012) the prevalence of periodontitis in the United States has been found to be 46% in adults > 30 years old. An older survey by Health Canada 2007, reported that 21% of the adult population with teeth had moderate to severe forms of periodontal disease.

The standard of care for managing oral diseases is primarily to mechanically disrupt the oral supra and sub-gingival microbiome both professionally via NSPT and daily patient oral-self-care regimes. The NSPT focuses on disruption and thoroughly removing any irritants (biofilm, calculus, and necrotic tissues) on hard and soft dental tissues both above and below the gingival margin to return the periodontium to biological health i.e. no inflammation.

NSPT can be accompanied with some post-treatment complications, which include gingival recession, soft tissue tenderness, dentin sensitivity and pain, which can make oral self-care regimes like tooth brushing and interdental care challenging.

Researchers have assessed patient’s discomfort and pain following various non-surgical therapeutic procedures and have shown that tooth and root hypersensitivity following SRP
usually peaked around 3 hours with a duration of 1-3 weeks following NSPT treatment\textsuperscript{9,97}. Pihlstrom et al.\textsuperscript{11} also concluded that patients experienced pain of significant duration and magnitude following NSPT. The heightened sensitivity leads to patient’s ineffective biofilm disruption by either brushing or any self-care regime aversion. Recolonization of the biofilm, especially aciduric bacteria increase hypersensitivity, which leads to a vicious cycle of persistent and worsening symptoms\textsuperscript{13, 101}. Conversely, open dentinal tubules due to freshly planed root surfaces favourably mineralize in the absence of plaque\textsuperscript{100}.

A need was identified for novel therapeutic approaches, such as the adjunctive use of cyanoacrylate adhesives to NSPT to reduce immediate post instrumentation sensitivity and improve the effectiveness of oral self-care regimes. Daily biofilm disruption is paramount in achieving and maintaining long-term periodontal and overall health. With enhanced post-operative comfort, the patient’s ability to disrupt biofilm and maintain optimal daily oral self-care regimes could be achieved.

Reducing inflammation and promoting wound healing may also be enhanced with the adjunctive placement of cyanoacrylate dressing by creating a barrier to the pathogenic biofilm accumulation. This novel application was anticipated to obtain ideal periodontal therapeutic endpoints and positively impact overall health.
3.2 Hypothesis

The supra-gingival surface application of cyanoacrylate adhesive extending 2 mm apical and coronal to the gingival margins adjunctively to NSPT will promote periodontal wound healing and reduce the sensitivity of hard (teeth) and soft (gingiva) tissues.

3.3 Objectives

The purpose of this pilot study is:

1. To determine the efficacy of cyanoacrylate adhesive in preventing the increase of sensitivity of hard (tooth dentin) and soft (gingiva) tissues after NSPT, enabling better immediate oral self-care;

2. To determine the efficacy of cyanoacrylate in its ability to isolate the sub-gingival microenvironment & promoting periodontal wound healing;

3. To determine the safety of cyanoacrylate as a periodontal adhesive dressing after NSPT.

If the results are statistically significant, it may support future randomized clinical trials in determining protocols for clinical relevance.
Chapter 4: Material and Methods

4.1 Materials

1. Cyanoacrylate adhesive: high viscosity blend of 90/10 N-butyl and 2-octyl cyanoacrylate (PeriAcryl®90 HV, Blacklock Medical Products and GluStitch, Inc.) (Figure 5)
2. Automated pressure controlled probe: Florida Probe ® FP 32 version 5 (Figure 3,4)
4. Nabers periodontal probe, Hu-Friedy® to assess furcations
5. Hand and Power Instrumentation for Phase 1 NSPT.
6. Local anesthetic as required (lidocaine 2% with 1:100000 epinephrine, unless contraindicated to epinephrine)
7. Plaque Disclosing solution: Gum® Red Cote disclosing liquid
8. Camera for intra-oral photos

Figure 3 Florida Probe® System© (© 2018 Florida Probe Corporation, by permission)
Figure 4 Florida Probe® in use for measuring PD\textsuperscript{143} (© 2018 Florida Probe Corporation, by permission)

Figure 5 Cyanoacrylate adhesive: (PeriAcryl®90 HV)\textsuperscript{144} (© 2018 Glu Stitch Inc., by permission)
4.2 Research Methods

4.2.1 Study Design

This pilot study was a single-blinded split-mouth randomized controlled clinical trial to assess the efficacy of cyanoacrylate tissue adhesive as an adjunct to NSPT in adults with chronic periodontitis.

4.2.1.1 Study Enrolment

The protocol for this Clinical Trial was reviewed and approved by the University of British Columbia Clinical Research Ethics Board in Vancouver, Canada. The potential subjects fulfilling the study’s criteria and willingness to participate were assessed at the UBC Dental Clinic screening sessions. Inclusion was not limited by gender or race but subjects must be capable of giving informed consent and be 19 years of age or older. They were required to have a minimum of 18 teeth and at least 4 measurements sites with pocket depth of at least 5.0 mm with bleeding on probing in at least 2 different quadrants; be able to understand and communicate in English; and be willing and able to return for treatment and evaluation throughout the course of this study.

Exclusions to enrollment were any antibiotics or prescribed anti-inflammatory drugs in the last month; pregnant, nursing or planned to become pregnant over the course of the trial; active smoking history (tobacco or otherwise); sites with overt abscess, active caries or crown/root fractures; known adverse reactions or allergies to cyanoacrylates or formaldehyde; and any significant disease or medication(s) that, in the opinion of the investigator, interfered with the evaluation of safety or efficacy of cyanoacrylate and overall compliance.
All eligible subjects were given the informed consent (*Appendix 2*) form to review before accepting participation. Subjects were explained that they could stop their participation at any time without any consequences to their care.

**4.2.1.2 Calibration and training**

The clinician performing all the periodontal clinical assessments (examiner clinician) was trained and calibrated in the use of the automated Florida Probe® system before the RCT to minimize intra-examiner variability. The examining clinician was a registered dental hygienist with 30 years of clinical experience.

**4.2.1.3 Blinding**

The present study was a single examiner-blinded trial. The examining clinician recorded the clinical periodontal measurements at baseline and 6-8 weeks and was blinded to the treated and applied sites. Also, the subjects were directed not to disclose any information about the sites receiving the adjunctive cyanoacrylate application to the blinded examining clinician during the 6-8 weeks final assessment visit.

The treating clinician, a registered dental hygienist with over 30 years experience practicing in a periodontal clinic, performed all the NSPT. The assessment data obtained by the examining clinician did not disclose the experimental sites to the treating clinician. The experimental sites (application of the cyanoacrylate adhesive following completion of instrumentation) were only revealed to the treating clinician after completing instrumentation and opening a coded-sealed
envelope. In addition, the treating clinician performed VAS testing for teeth sensitivity (air blast test) and measured plaque scores at the 1-week visit for both experimental and control sites.

4.2.1.4 Randomization and Randomization codes

Each enrolled subject was assigned a unique identification number. This number is a chart number that is assigned to each individual when registering for treatment at UBC dental clinics. This number enables access to confidential patient information on UBC digital patient database only through secure login by authorized personnel.

Following the completed comprehensive periodontal assessment with the Florida Probe®, the printed odontogram identified two equally periodontally affected quadrants for each subject. The quadrants were selected based on the presence of sites with the greatest pathology that were defined based on the criteria of highest probing depths, the presence of bleeding on probing and or suppuration. A computer-generated pseudorandom algorithm was used to determine the experimental sites (application of the cyanoacrylate adhesive following completion of instrumentation) for each subject. A secured, sealed envelope with the subject’s identification, which only the principal investigator was aware of, was revealed at the periodontal therapy session to the treating clinician. The examining clinician was unaware of the randomization sequence or the experimental sites for any subject.

4.2.1.5 Study visits schedule

The study comprised of five visits (Appendix 3). The initial visit (Visit 1) included screening for potential applicants who were approached during UBC dental clinics. They were given an
informed consent form to review and consider their participation in the study. Upon providing written informed consent, the subjects were assessed for eligibility through a review of their medical/dental history and comprehensive oral examination. If eligibility was confirmed, they were enrolled and scheduled for the next visit (Visit 2).

Visit 2 comprised of a detailed baseline examination that included reviewing medical/dental history, intraoral photographs and comprehensive oral and periodontal examination by the examining clinician. Using the Florida Probe® the following periodontal clinical parameters were assessed; Probing Depth (PD), Recession (R), Clinical Attachment Level (CAL) and Bleeding on Probing (BOP). These parameters were measured at six sites per tooth; mesiobuccal (MB), mid-buccal (B), distobuccal (DB), mesiolingual (ML), mid-lingual (L) and distolinguval (DL). The PD, CAL and R readings were assessed in millimetre (mm). BOP score was calculated as a percentage using the Ainamo and Bay index\textsuperscript{145}. The Plaque Index (PI) scores were assessed following application of disclosing agent as being present or absent on four sites; MB, B, DB, and L was also calculated as a cumulative percentage separately for experimental and control sites (Plaque Control Record)\textsuperscript{146}. (Figure 6)

Dental hypersensitivity (DH) for baseline and post-treatment week-1 and weeks 6-8 was assessed using a response to an air blast stimulus and visual analogue scoring (VAS). An air-blast from an air-water syringe was directed to the exposed root surface for 1 s. The syringe was held perpendicularly 2–3 mm from the root surface. During testing the treating examiner’s gloved fingers shielded the adjacent teeth. After this stimulation, the subject was asked to score the discomfort\textsuperscript{147}. The perceived discomfort for each tooth was graded for each stimuli by using a 10
centimetre (cm) VAS, labeled at the two extremes with “no discomfort” at the zero extreme and with “maximal discomfort” at the 100 mm (10cm) extreme. Data from the VAS were recorded by measuring in cm the distance between the zero point and the sign marked by the subject on the 10-cm line\(^4\). \textit{(Appendix 4)}

This assessment was done on a separate sheet for each tooth to avoid bias from previous assessments. Several readings were randomly repeated for reproducibility assessments.

Any discomfort of the soft tissues (gingiva) or hard tissues (teeth) perceived by the subject at baseline or following therapy was also assessed using VAS scoring by the participant. \textit{(Appendix 5)}

The VAS scoring was further categorized into three categories of <1, 1-4, >4 to denote no perceived pain, mild pain and moderate to severe pain respectively for interpretation of the readings.

Additionally, the mobility of the teeth using the end of handles of 2 instruments, furcation involvements using Nabers periodontal probe and presence/absence of suppuration with digital application of pressure were also assessed. At the end of visit 2, eligible quadrants were identified and were randomly allocated to experimental or control arms, which were coded with the subject’s identification number and sealed in envelopes until visit 3.

Visit 3 involved treatments of both experimental and control sites with NSPT. The same treating clinician completed all debridement sessions with no time restriction. Following completion of
NSPT, the adjunctive application of cyanoacrylate adhesive at the gingival margins extending 1-2 mm above and below on both labial and lingual/palatal aspects were applied to the randomly selected experimental sites. The cyanoacrylate adhesive is dispensed in plastic containers in a monomeric liquid form, and upon contact with hydroxyl ions present in water, it rapidly polymerizes adhering to moist living tissue. The sites intended for application were isolated and dried following completion of NSPT. The adhesive was loaded in the pipettes and then carefully applied on the superficial gingival surface extending 1-2 mm coronal and apical to the gingival margin. Intraoral photographs were taken following completion of its application. *(Figure 8)*

Subjects were given post-operative instructions that included avoiding any unnecessary use of analgesics or antibiotics and making note if they had to, avoid the use of any over-the-counter mouth rinses and also to refrain from brushing and flossing of only the experimental sites for the 1-week post-operative period. They were guided to complete a daily journal for a week, which asked to reflect on their pain perception or discomfort in teeth or gingival tissues as well as their acceptability of having this material in their mouth using the VAS scoring criteria. *(Appendix 6)*

The subjects were followed-up at Day 2 or 3 following NSPT via phone or email to address any concerns or issues and to ensure compliance with completing the daily journaling.

Visit 4 was scheduled 1-week following the completion of NSPT where participants were assessed for any adverse events and assessed for any pain or discomfort in teeth or gingival tissues as well as their acceptability of having the material in their mouth. *(Appendix 7)*

PI and changes in teeth sensitivity in response to air blast stimulus using VAS scoring at both experimental and control sites was evaluated. Removal of any remnants of cyanoacrylate
adhesive was done and subjects were instructed to resume their oral self-care regimes. The treating clinician completed debridement of the remaining quadrants not included in the study.

Visit 5 was the final scheduled visit with the examining-clinician at 6-8 weeks following completion of NSPT. This visit included the same comprehensive periodontal clinical parameters evaluated at baseline as well as sensitivity assessment of each tooth using the response to the air blast stimulus using VAS scoring. The subject’s perception of discomfort in the soft tissues (gingiva), hard tissues (teeth), and their acceptability to the adjunctive adhesive dressing was determined using VAS scoring. Any adverse events were also reported during this period. *(Appendix 8)* Subjects maintained their oral self-care hygiene regimes during the study unless otherwise specified for the adjunctive adhesive sites.

**Figure 6 Intraoral photograph of application of plaque disclosing agent at baseline visit**
Figure 7 Intraoral Photographs of experimental sites at Baseline visit

Figure 8 Intraoral Photographs of experimental sites following application of adhesive after NSPT
Figure 9 Intraoral Photographs of experimental sites at 1-week follow-up after NSPT

Figure 10 Intraoral Photographs of experimental sites at final visit (6-8 weeks) follow-up after NSPT
4.2.2 Statistical methods

4.2.2.1 Study sampling
Since this was a pilot clinical trial, the sample size could not be determined using power based calculations. The sample size was decided to be a minimum of 20 subjects based on a study by Julius 2005\textsuperscript{148} who recommends a minimum of 12 subjects per treatment arm for pilot clinical trials.

4.2.2.2 Calibration study
A calibration study was performed on 7 participants with similar eligibility criteria for the disease. Intra-examiner error was determined using percentage agreement for measuring PD and gingival recession\textsuperscript{60}. In the present study, it was found to be 95.18 % within ±1 mm for PD and 98.8% within ±1 mm for R.

4.2.2.3 Endpoints of the study
In the present study, the unit of analysis was each site. The primary endpoints were

- Prevention of increased post-operative discomfort and hypersensitivity (hard and soft dental tissues) after NSPT with the placement of cyanoacrylate adhesive.
- Change in clinical attachment levels (CAL) after NSPT with the adjunctive placement of cyanoacrylate adhesive after 6-8 weeks.

The secondary endpoints were:

- Reduction in
  - Probing pocket depth (PD) after 6-8 weeks.
  - Bleeding on probing (BOP) after 6-8 weeks.
• Plaque scores (PI) after 6-8 weeks.

• Participants’ compliance and perception of having the cyanoacrylate adhesive post-operatively.

• Assess for any adverse reactions to the placement of cyanoacrylate adhesive at the supragingival margins.

4.2.2.4 Statistics applied

All statistical analysis was completed in consultation with a well-versed faculty trained in statistics from UBC Dentistry and using the IBM® SPSS Statistics program version 25. For all statistical analysis, a significance level of 5% (p<0.05) and 95% confidence interval were used.

The parameters of PD, R and CAL, VAS, PI and BOP were considered as continuous data as the Florida Probe® with a precision of 0.2 mm was used to assess them. The data were assessed for normality using the Kolmogorov–Smirnov and Shapiro Wilk tests. Histograms and Q-Q plots were also used to confirm normality of data.

As the data for PD, R and CAL, was not normally distributed in both control and experimental arms, non-parametric tests were applied. To compare these scores from baseline to final visit at 6-8 weeks, Wilcoxon Signed Rank Test was used for each set of data. To compare and determine differences between control and experimental arms (sites) for these parameters, Independent Samples Median Test was used. To control for potential outliers, Median Test was selected to compare the changes between the two data sets as it does not assume equal variance and is more robust to the presence of outliers. The data was graphically represented as box plots with an
interquartile range. Additionally, a data set containing sites with baseline PD ≥ 4mm were analyzed separately for PD, CAL and R.

The PI data was found normally distributed in both control and experimental arms, and parametric tests were applied. To compare these scores from baseline to week-1; week-1 to final visit at 6-8-weeks; and baseline to final visit at 6-8- weeks, a paired t-test was used for each data set. To compare the mean differences between control and experimental data sets, an independent t-test was used.

The BOP score and the VAS score data were not normally distributed in either the control or experimental arms; non-parametric tests were applied for both. To compare the BOP scores from baseline to final visit at 6-8 weeks and the VAS scores from baseline to week-1; week-1 to final visit at 6-8 weeks; and baseline to final visit at 6-8 weeks the Wilcoxon Signed Rank Test was used for each data set respectively. To compare and determine differences between control and experimental arms for BOP and VAS, Independent Samples Median Test was used.

The parameters of pain in the hard tissues (teeth) and soft tissues (gingiva) following NSPT procedures as well the acceptability of treatment provided were analyzed as descriptive data based on the participants’ perceptions scores on the VAS forms. In assessing the qualitative feedback obtained from the participants, content analysis was performed, and some common themes emerged that were presented graphically.
Chapter 5: Results

5.1 Subject Summary

This pilot study employed an in-vivo, examiner-blinded, split-mouth; parallel randomized controlled clinical trial design. Twenty-eight (28) subjects were screened and twenty-four (24) were found eligible and enrolled for participation. One subject was lost to follow-up due to medical concerns unrelated to the study. Twenty-three (23) subjects completed all 5 visits of the trial. For every 23 subjects, two equally periodontally involved quadrants, were randomly assigned to either an experimental or control sites. The experimental areas comprised of quadrants treated with NSPT followed by the application of cyanoacrylate adhesive to the gingival margins. The control sites were treated with NSPT alone. A site level analysis was performed. The CONSORT study flow diagram is detailed in Figure 11.
Figure 11 CONSORT flow diagram of the RCT
5.2 Demographics Summary

The study subjects had twelve (12) males and eleven (11) females with an age range of 38-96 years. The mean age was 61 years old. All enrolled subjects were medically fit to undergo the NSPT procedure. They were all non-smokers or former smokers with no active dental caries or overt abscesses. 12 out of the 23 subjects were on regular maintenance at UBC dental clinics but had not had an NSPT appointment in the last 6 months or longer and 11 subjects were new patients that had not had NSPT in the last 1 year or longer from the time of recruitment. All enrolled subjects had a minimum of 18 teeth with PD of at least 5 mm in 4 or more sites in at least 2 different quadrants.

5.2.1 Descriptive statistics of site distribution; PD, Recession, and CAL.

Six sites per tooth were measured to collect the periodontal clinical parameters (PD, CAL, R) with the Florida Probe®, which was set at a default of 0.2 mm precision recording and 15 g force. The 6 sites include the MB, B, DB, ML, L and DL. The number of sites (n) assessed was 918 and 882 for the experimental and control sites, respectively. The sites with baseline PD ≥ 4mm were excluded to obtain results in this category and the number of sites (n) assessed was 253 and 242 for the experimental and control arms, respectively.

5.2.2 Descriptive statistics of site distribution; BOP.

The BOP score was ascertained as a binary variable, i.e. present or absent following probing. The measurements were performed at 6 sites per tooth (MB, B, DB, ML, DL). A consolidated BOP score of positive percentage sites was calculated for the experimental and control arms.
individually and then statistical comparisons were drawn between different time points as well as between the two data sets.

5.2.3 Descriptive statistics of site distribution; PI

The plaque score was ascertained as a binary variable i.e. present or absent following application of a disclosing solution. Data were collected at 4 sites per tooth; MB, B, DB, and L. A consolidated plaque score of positive percentage sites was calculated for the experimental and control arms individually and then statistical comparisons were drawn between different time points as well as between the two data sets.

5.2.4 Descriptive statistics of site distribution: Teeth Sensitivity using VAS

The teeth sensitivity VAS scores were collected by using the air blast testing at 1 site per tooth; B surfaces. The total number of sites (n) assessed was 153 and 147 for the experimental and control arms, respectively.

5.3 Statistical Analysis of Qualifying Sites for all Endpoints.

5.3.1 Primary Endpoint Analysis for Qualifying Sites: CAL.

The mean and standard deviations of CAL for experimental and control data sets at baseline and 6-8 weeks is summarized in Table 1. The mean CAL at baseline for experimental and control arms (sites) was 3.68±1.67 mm and 3.59±1.69 mm respectively. The mean CAL at the final visit (6-8 weeks) was 3.67±1.60 mm and 3.61±1.65 mm respectively.
The mean differences in CAL for both data sets were statistically non-significant between the two-time points for both experimental (p=0.761) and control arms (p=0.893) and also between the experimental and control data sets (p= 0.878). (Table 2, Figure 12)

Table 1 Mean CAL scores (mm) ± SD at baseline and final visit (6-8 weeks)

<table>
<thead>
<tr>
<th>Arms (sites) (quadrants)</th>
<th>N (number of sites)</th>
<th>Baseline</th>
<th>Final (6-8weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
<td>Mean</td>
</tr>
<tr>
<td>Control</td>
<td>882</td>
<td>3.599</td>
<td>1.693</td>
</tr>
<tr>
<td>Experimental</td>
<td>918</td>
<td>3.679</td>
<td>1.668</td>
</tr>
</tbody>
</table>
Table 2 Descriptive of the number of sites exhibiting hard and soft tissue discomfort at various time points.

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final</th>
<th>‘p’ value</th>
<th>‘p’ value between data sets</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>-0.007±1.3448</td>
<td>0.893&lt;sup&gt;NS&lt;/sup&gt;</td>
<td>0.878&lt;sup&gt;NS&lt;/sup&gt;</td>
</tr>
<tr>
<td>Experimental</td>
<td>0.011±1.3276</td>
<td>0.761&lt;sup&gt;NS&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

NS= Non significant; p significant at <0.05

Figure 12 Box plot depicting the change in CAL (delta CAL) from baseline to final visit (6-8 weeks) in both control (0) and experimental data sets (1)
The mean and standard deviations of CAL for sites with baseline PD ≥ 4 mm at experimental and control data sets at baseline and 6-8 weeks is summarized in Table 3. The mean CAL at baseline for experimental and control sites was 5.40±1.299 mm and 5.43±1.319 mm respectively. The mean CAL at the final visit (6-8 weeks) was 4.64±1.604 mm and 4.76±1.67 mm respectively.

The mean differences in CAL for both data sets were statistically significant between the two-time points for both experimental (p=<0.000*) and control sites (p=<0.000*) but non significant between the experimental and control data sets (p = 0.356). (Table 4, Figure 13)

Table 3 Mean CAL scores (mm) ± SD at baseline and final visit (6-8 weeks) at sites with baseline PD≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (number of sites)</th>
<th>Baseline</th>
<th>Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Control</td>
<td>242</td>
<td>5.428</td>
<td>1.319</td>
</tr>
<tr>
<td>Experimental</td>
<td>253</td>
<td>5.400</td>
<td>1.299</td>
</tr>
</tbody>
</table>
Table 4 Mean change in CAL ± SD, P-values at final visit (6-8 weeks) and between data sets at sites with baseline PD≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final</th>
<th>‘p’ value</th>
<th>‘p’ value between groups</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
<td>‘p’ value</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>-0.66± 1.463</td>
<td>&lt;0.000*</td>
<td>0.356NS</td>
</tr>
<tr>
<td>Experimental</td>
<td>-0.76±1.445</td>
<td>&lt;0.000*</td>
<td></td>
</tr>
</tbody>
</table>

NS= Non significant; *= significant, p significant at <0.05
**Figure 13** Box plot depicting the change in CAL (delta CAL) from baseline to final visit (6-8 weeks) in both control (0) and experimental data sets (1) in sites with baseline PD ≥ 4 mm

5.3.2 Primary Endpoint Analysis for Qualifying Sites: Teeth Sensitivity VAS

The mean VAS scores for teeth sensitivity following air blast assessment at baseline, 1 week following NSPT and at the final visit (6-8 weeks) are summarized in *Table 5*. The mean VAS score for experimental and control sites at baseline was 0.40±1.14 cm and 0.60±1.44 cm respectively. The mean VAS score at the first follow-up visit (1 week) was 0.33±1.12cm and 0.28±0.95cm respectively. The mean VAS score at the final visit (6-8 weeks) was 0.48±1.24cm and 0.47±1.19cm respectively.
The mean VAS score was significantly reduced from baseline to 1-week post NSPT (Delta VAS 1) in the control sites (p=0.01) and non-significantly between baseline and final visit (Delta VAS, p=0.144), and between 1-week and final visit (6-8 weeks) (Delta VAS 2, p= 0.11). The VAS score was non-significant between the various time points in the experimental sites (Delta VAS 1, p= 0.287; Delta VAS 2, p=0.15 and Delta VAS, p=0.468) as well as between the experimental and control arms (sites) at all time points (Delta VAS 1, p= 0.368; Delta VAS 2, p=0.748 and Delta VAS, p=0.914). *(Table 6), Figure 14,15*

**Table 5 Mean VAS scores (cm) ±SD at baseline, 1 week and final visit (6-8 weeks)**

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>1-week</th>
<th>Final (6-8weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
<td>Mean</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Control</td>
<td>147</td>
<td>0.60</td>
<td>1.438</td>
<td>0.28</td>
</tr>
<tr>
<td>Experimental</td>
<td>153</td>
<td>0.40</td>
<td>1.140</td>
<td>0.33</td>
</tr>
</tbody>
</table>
Table 6 Mean VAS difference, P-values at 1 and 6-8 weeks and between arms.

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to 1-week (Delta VAS 1)</th>
<th>1-week to 6-8 weeks (Delta VAS 2)</th>
<th>Baseline to 6-8 weeks (Delta VAS)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
<td>Mean difference ± SD</td>
<td>Mean difference ± SD</td>
</tr>
<tr>
<td>Control</td>
<td>-0.32± 1.420</td>
<td>0.18± 1.332</td>
<td>-0.14± 1.421</td>
</tr>
<tr>
<td>'p' value</td>
<td>0.010*</td>
<td>0.110NS</td>
<td>0.144NS</td>
</tr>
<tr>
<td>Experimental</td>
<td>-0.08± 1.118</td>
<td>0.15±1.424</td>
<td>0.08± 1.334</td>
</tr>
<tr>
<td>'p' value</td>
<td>0.287NS</td>
<td>0.150NS</td>
<td>0.468NS</td>
</tr>
<tr>
<td>'p' value between arms</td>
<td>0.368NS</td>
<td>0.748NS</td>
<td>0.914NS</td>
</tr>
</tbody>
</table>

* = Significant, NS= Non significant; p significant at <0.05
Figure 14 Box plot depicting the change in VAS (delta VAS 1) from baseline to following NSPT in both control (0) and experimental (1) sites.
5.3.3 Secondary Endpoint Analysis for Qualifying Sites: BOP.

The mean and standard deviations of BOP for experimental and control arms at baseline and 6-8 weeks are summarized in Table 7. The mean BOP % at baseline for experimental and control sites was 25.59±16.76 and 19.91±16.71 respectively. The mean BOP% at the final visit (6-8 weeks) was 12.69±12.69 and 16.13±17.24 respectively.

The mean reduction in BOP was statistically significant between the two time points in the experimental (p=0.001) but non-significant in the control data sets (p=0.055). The mean difference between the arms was statistically significant with the experimental dataset depicting a more significant reduction (p=0.003) in BOP scores. (Table 8, Figure 16)
Table 7 Mean BOP % scores ± SD at baseline and final visit (6-8 weeks)

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
<td>Mean</td>
</tr>
<tr>
<td>Control</td>
<td>23</td>
<td>19.91</td>
<td>16.71</td>
</tr>
<tr>
<td>Experimental</td>
<td>23</td>
<td>25.59</td>
<td>16.76</td>
</tr>
</tbody>
</table>

Table 8 Mean reduction in BOP ± SD, CI and P-values from baseline to final visit (6 -8 weeks) and between the data sets.

<table>
<thead>
<tr>
<th>Arms</th>
<th>Baseline to Final</th>
<th>'p' value</th>
<th>'p' value between data sets</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>3.78± 8.942</td>
<td>0.055NS</td>
<td></td>
</tr>
<tr>
<td>Experimental</td>
<td>12.9±15.116</td>
<td>0.001*</td>
<td>0.003*</td>
</tr>
</tbody>
</table>

NS= non significant, *= significant; p significant at <0.05
5.3.4 Secondary Endpoint Analysis for Qualifying Sites: PI

The mean and standard deviations of PI for experimental and control data sets at baseline, 1-week and 6-8 weeks are summarized in Table 9. The mean PI (%) for experimental and control sites at baseline was 76.75±17.44 and 72.07±23.98 respectively. The mean PI at the first follow up visit (1-week) was 54.74 ± 26.47 and 45.17 ± 26.59 respectively. The mean PI at the final visit (6-8 weeks) was 69.73±18.86 and 62.47 ± 22.76 respectively.
The mean plaque score showed a statistically significant reduction between baseline and 1-week visit (Delta PI 1) in both experimental (p<0.000) and control arms (p<0.000). Following this a significant increase in PI was noted from 1-week to final visit at 6-8 weeks (Delta PI 2) in both groups, experimental (p= 0.018) and control (p=0.012). However, the reduction in PI between baseline and the final visit at 6-8 weeks (Delta PI) was statistically non-significant in both experimental (p= 0.074) and control groups (p=0.103). On comparison between the two data sets, the mean PI difference was statistically non-significant at all time points (Delta PI 1, p= 0.542; Delta PI 2, p=0.791 and Delta PI, p=0.705). *(Table 10, Figure 17)*
Table 9 Mean PI (%) scores ± SD at baseline, 1 week and final visit (6-8 weeks)

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to 1-week</th>
<th>1-week to 6-8 week</th>
<th>Baseline to 6-8 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
<td>SEM</td>
<td>CI (95%) (lower to upper)</td>
</tr>
<tr>
<td>Control</td>
<td>26.91±29.541 6.159</td>
<td>14.12-39.67 0.000*</td>
<td>-17.308±30.49 6.35 7</td>
</tr>
<tr>
<td>Experimental</td>
<td>22.01±24.173 5.041</td>
<td>11.55-32.46 0.000*</td>
<td>-14.991±28.21 5.88 2</td>
</tr>
<tr>
<td>‘p’ value b/n data sets</td>
<td>0.542NS</td>
<td>0.791NS</td>
<td>0.705NS</td>
</tr>
</tbody>
</table>
### Table 10 Mean PI reduction, CI and P-values at 1-week & 6-8 weeks and between data sets

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (number of subjects)</th>
<th>Baseline</th>
<th>1-week</th>
<th>Final (6-8weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
<td>Mean</td>
</tr>
<tr>
<td>Control</td>
<td>23</td>
<td>72.07</td>
<td>23.975</td>
<td>45.17</td>
</tr>
<tr>
<td>Experimental</td>
<td>23</td>
<td>76.75</td>
<td>17.436</td>
<td>54.74</td>
</tr>
</tbody>
</table>

NS= non significant, *= significant; p significant at <0.05
Figure 17 Clustered Bar graph depicting mean % reduction in PI (ΔPI1) from baseline to 1-week, (ΔPI2) from 1 week to final visit (6-8 weeks) and from baseline to final visit (ΔPI) (6-8 weeks) for control (0) and experimental (1) sites.

5.3.5 Secondary Endpoint Analysis for Qualifying Sites: PD.

The mean and standard deviations of all the PD for the experimental and control sites at baseline and 6-8 weeks are summarized in Table 11. The mean PD at baseline for experimental and control sites was 3.11±1.51 mm and 3.04±1.55 mm respectively. The mean PD at the final visit (6-8 weeks) was 3.11±1.39 mm and 3.06±1.45 mm respectively.

The mean differences were statistically non-significant between the two time points in each dataset (Experimental, p=0.45; Control, p=0.274) and between the two groups (p=0.767) for PD. (Table 12, Figure 18)
Table 11 Mean PD scores (mm) ± SD at baseline and final visit (6-8 weeks)

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>Final (6-8weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
<td>Mean</td>
</tr>
<tr>
<td>Control</td>
<td>882</td>
<td>3.037</td>
<td>1.552</td>
</tr>
<tr>
<td>Experimental</td>
<td>918</td>
<td>3.105</td>
<td>1.512</td>
</tr>
</tbody>
</table>

Table 12 Mean reduction in PD ± SD, CI & P-values at final visit (6-8 weeks) and between data sets.

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
</tr>
<tr>
<td>Control</td>
<td>-0.027±1.218</td>
</tr>
<tr>
<td>Experimental</td>
<td>-0.002±1.239</td>
</tr>
</tbody>
</table>

NS= non significant, *= significant; p significant at <0.05
Figure 18 Box plot depicting the change in PD (delta PD) from baseline to final visit (6-8 weeks) following treatment in both control (0) and test (1) sites.

The mean and standard deviations of only sites with baseline PD ≥ 4 mm at baseline for experimental and control arms at baseline and final visit at 6-8 weeks is summarized in Table 13. The mean PD at baseline for experimental and control sites was 5.06 ±1.03 mm and 5.07 ±1.08 mm respectively. The mean PD at the final visit (6-8 weeks) was 4.25 ±1.38 mm and 4.37 ±1.53 mm respectively.

The mean differences in PD for both data sets were statistically significant between the two-time points for both experimental (p=<0.000*) and control arms (p=<0.000*) but non-significant between the experimental and control data sets (p= 0.973). (Table 14, Figure 19)
Table 13 Mean PD scores (mm) ± SD at baseline and final visit (6-8 weeks) at sites with baseline PD≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>Final (6-8weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Control</td>
<td>242</td>
<td>5.070</td>
<td>1.076</td>
</tr>
<tr>
<td>Experimental</td>
<td>253</td>
<td>5.058</td>
<td>1.032</td>
</tr>
</tbody>
</table>

Table 14 Mean reduction in PD ± SD, CI & P-values at final visit (6-8 weeks) and between data sets at sites with baseline PD≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
</tr>
<tr>
<td>Control</td>
<td>-0.70± 1.402</td>
</tr>
<tr>
<td>Experimental</td>
<td>-0.813± 1.510</td>
</tr>
</tbody>
</table>

NS= non significant, * = significant; p significant at <0.05
Figure 19 Box plot depicting the change in PD (delta PD) from baseline to final visit (6-8 weeks) following treatment in both control (0) and test (1) sites with baseline PD ≥ 4mm

5.3.6 Secondary Endpoint Analysis for Qualifying Sites: R

The mean and standard deviations of gingival recession (R) for the experimental and control sites at baseline and 6-8 weeks are summarized in Table 15. The mean recession at baseline for experimental and control sites was 0.574±1.01 mm and 0.562±1.05 mm respectively. The mean R at the final visit (6-8 weeks) was 0.560±1.04 mm and 0.541±1.04 mm respectively.

The mean differences were statistically non-significant between the two-time points for each dataset (Experimental, p=0.215; Control, p=0.304) as well as between the two arms (p=0.540) for recession. (Table 16, Figure 20)
Table 15 Mean R (mm) ± SD at baseline and final visit (6-8 weeks)

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final (6-8 weeks)</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
<td>‘p’ value</td>
<td>‘p’ value between data sets</td>
</tr>
<tr>
<td>Control</td>
<td>0.021±0.735</td>
<td>0.304&lt;sup&gt;NS&lt;/sup&gt;</td>
<td>0.540&lt;sup&gt;NS&lt;/sup&gt;</td>
</tr>
<tr>
<td>Experimental</td>
<td>0.013±0.752</td>
<td>0.215&lt;sup&gt;NS&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

Table 16 Mean change in R (mm) ± SD, CI & P-values at final visit (6-8 weeks) and between data sets.

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Control</td>
<td>882</td>
<td>0.562</td>
<td>1.046</td>
</tr>
<tr>
<td>Experimental</td>
<td>918</td>
<td>0.574</td>
<td>1.007</td>
</tr>
</tbody>
</table>

NS= non significant, *= significant; p significant at <0.05
Figure 20 Box plot depicting the change in R (Delta recession) from baseline to final visit (6-8 weeks) following treatment in both control (0) and experimental (1) sites.

The mean and standard deviations of R for sites with baseline PD ≥ 4 mm at experimental and control data sets at baseline and 6-8 weeks is summarized in Table 17. The mean R at baseline for experimental and control sites was 0.342 ± 0.68 mm and 0.357 ± 0.812 mm respectively. The mean R at the final visit (6-8 weeks) was 0.391 ± 0.893 mm and 0.398 ± 0.897 mm respectively.

The mean differences in R for both data sets were non-significant between the two-time points for both experimental (p=0.464) and control arms (p=0.579) as well as between the experimental and control data sets (p=0.550). (Table 18, Figure 21)
Table 17 Mean R (mm) ± SD at baseline and final visit (6-8 weeks) at sites with baseline PD ≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>N (no. of sites)</th>
<th>Baseline</th>
<th>Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Mean</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Control</td>
<td>242</td>
<td>0.357</td>
<td>0.812</td>
</tr>
<tr>
<td>Experimental</td>
<td>253</td>
<td>0.342</td>
<td>0.684</td>
</tr>
</tbody>
</table>

Table 18 Mean change in R (mm) ± SD, CI & P-values at final visit (6-8 weeks) and between data sets at sites with baseline PD ≥ 4 mm

<table>
<thead>
<tr>
<th>Arms (sites)</th>
<th>Baseline to Final (6-8 weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean difference ± SD</td>
</tr>
<tr>
<td>Control</td>
<td>0.04± 0.658</td>
</tr>
<tr>
<td>Experimental</td>
<td>0.05 ± 0.666</td>
</tr>
</tbody>
</table>

NS= non significant, *= significant; p significant at <0.05
Figure 21 Box plot depicting the change in R (Delta recession) from baseline to final visit (6-8 weeks) following treatment in both control (0) and experimental (1) sites with baseline PD ≥ 4 mm.
5.4 Subject Reported Outcomes

5.4.1 Acceptability of treatment: Subject perception.

There were no clinically significant adverse side effects noted with the intraoral application of the cyanoacrylate adhesive. 48% of the subjects found it highly acceptable with no issues regarding its use. However, 52% of the subjects cited some concerns following the adhesive application along the gingival margins. These concerns have been graphically represented in Figure 22.

Figure 22 Subjects perception of acceptability and concerns to tissue adhesive (Periacryl® 90HV) following NSPT.

The acceptability of the adhesive application (Periacryl® 90HV) post-NSPT increased each day of the week. The percentage of subjects with a VAS score of ≤ 1 increased while the number of subjects scoring >4 on the VAS decreased. On day 1, following application of the adhesive, 30.4% subjects (n= 7/23) scored >4 and 43.5% (n=9/23) scored ≤ 1. However, at the first follow-
up visit at week-1, there were no subjects that scored > 4 on VAS and the number of subjects scoring ≤ 1 increased to 87% (n=20/23). (Figure 23)

At the second follow-up visit at weeks 6-8 (final visit) subjects were asked their future acceptability of a repeated adhesive post-NSPT. 69.6% (16/23) subjects reported a score of ≤ 1 and only 13% (n=3/23) marked a score of >4. (Figure 24)

**Figure 23 Daily record of subject perception of acceptability of tissue adhesive (Periacryl® 90HV) following NSPT for a 1-week period**
Figure 24 Patient perception of the acceptability of repeated application of tissue adhesive (Periacryl® 90HV) at the final visit (6-8 weeks).

5.4.2 Subject perception of hard tissue (teeth) and soft tissue (gingiva) discomfort.

Subjects were asked to report discomfort levels with hard tissues (teeth) and soft tissues (gums) using VAS forms. A reported higher number of subjects experienced tooth and gingival discomfort at the experimental sites as compared to the control sites (Table 19). 8/23 subjects reported discomfort following NSPT at the experimental sites, 6/23 at the control sites, 4/23 on both experimental and control sites and 7/23 complained of no teeth discomfort and 5/23 had no gingival tissue discomfort.
Table 19 Descriptives of the number of sites exhibiting hard and soft tissue discomfort at various time points.

<table>
<thead>
<tr>
<th>Site of discomfort</th>
<th>Hard tissue discomfort (teeth)</th>
<th>Soft tissue discomfort (gingiva)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Experimental</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>Control</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Both</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>Nil</td>
<td>7</td>
<td>5</td>
</tr>
</tbody>
</table>

It was found that a higher number of subjects experienced pain >4 (n= 6/23) on the VAS on Day 1 following NSPT with hard tissues (teeth). The number of subjects experiencing moderate to severe pain reduced over the week and on day 8 (1st follow up visit) assessment, no subjects were reporting any pain response of >4. (Figure 25)
Figure 25 Subject-reported discomfort in the hard tissue (teeth) at baseline, daily for a period of 1-week following NSPT and at final visit (6-8 weeks)

For the soft tissue discomfort (gums), a similar trend was observed, with an increased number of subjects reporting a score of >4 (n= 6/23) on the VAS on Day-1 following NSPT and reduced over the week with Day 8 having no subjects reporting any soft tissue discomfort of > 4 on the VAS. (Figure 26)
Figure 26 Subject reported discomfort in the soft tissue (gingiva) at baseline, daily for a period of 1 week following NSPT and at final visit (6-8 weeks).
**Chapter 6: Discussion**

Currently, best-practice modality in managing the primary etiology of periodontal diseases is thorough NSPT. It enables the supra, sub-gingival and within sulcus or pocket space, mechanical disruption of the oral microbiome (biofilm), the removal of its by-products and contributing or iatrogenic factors like retentive calculus deposits or ill-contoured restorations to achieve periodontal healing and repair, i.e. biologically compatible.

To accomplish thorough NSPT soft dental tissues, like inflamed gingiva, distended periodontal ligaments and hard dental tissues, like infected cementum, and opened dentinal tubules can lead to a period of post-therapy pain or discomfort. Daily oral self-care regimes are pivotal for successful therapy outcomes, which are profoundly diminished by the lack of daily mechanical disruption due to post-NSPT pain or discomfort.

Current studies have linked several systemic health issues to periodontal diseases, especially those of a chronic inflammatory condition. For several decades, bacteria were identified as the leading risk factor that linked the body’s diseases, but recent medical publications have strongly correlated with certain conditions having a causation effect, and inflammation as the primary link. In the oral cavity, inflamed periodontal tissues, especially noted with BOP, release inflammatory mediators into the bloodstream, which can progress to other organ-systems, and initiate a harmful cascade of events.

Although encouraging daily oral self-care prevention is beneficial, it is insufficient in preventing the continual periodontium breakdown. Thus, failure of consistent professional dental care, i.e.
thorough NSPT or completion of any phase I care and daily oral self-care prevention will initiate the body’s inflammatory process leading to an increase in periodontal and health risk factors. Previous dental pain experiences can lead to anxiety and fear of future visits, which can also diminish the effects of obtaining oral and overall health. Delaying dental treatments may lead to worsening of the diseases and thereby requiring potentially more invasive or painful treatment\textsuperscript{151,152}.

A survey-based analysis of a sample of 1036 subjects from a representative Australian population reported that dental fear was associated with avoidance of treatment visits, greater perceived treatment need and problem-oriented access to dental care. A vicious cycle pattern was established and it was found that 38.5\% of subjects with moderate to high levels of dental fear fit the hypothesized model vs. 0.9\% of subjects with no fear. 92\% of the subjects with moderate to high fear avoided the dentist as compared to only 50.5\% of non-fearful subjects. Additionally in subjects with moderate to high dental fear that avoided dental treatment, the prevalence of treatment need and problem-based visiting was 4.9 times higher (60.9\%) than those who did not avoid dental care (12.5\%)\textsuperscript{152}.

Chung et al.\textsuperscript{153} conducted a study on sample of 40 subjects that were split into 2 groups and assessed by 2 different hygienists for their pain perception following full-mouth periodontal probing and periodontal instrumentation procedures. It was observed that 20-33\% of patients had a significant pain experience and scored higher on their anxiety assessment prior to the NSPT procedure. A similar observation of a highly significant correlation between fear of getting “teeth cleaned” and pain experience on VAS scoring was also reported.
The present trial was designed to explore a possible mechanism of minimizing discomfort or pain in the perioperative and the immediate post-NSPT period, to encourage daily thorough oral self-care routines and maintain ongoing professional NSPT. During the NSPT, various forms of local anaesthetics and sedation techniques have been employed to minimize apprehension and discomfort. However, currently, there is no immediate placement of therapeutic modalities for postoperative pain management.

6.1 Periodontal wound healing; Impact on periodontal clinical parameters

Healing of periodontal tissues following non-surgical and surgical therapy is initiated with an inflammatory phase over a 24-48 hour period that involves the release of various biological mediators including histamine, leukotrienes, prostanoids and cytokines some of which are associated with causing a painful response\textsuperscript{154}.

Periodontal wound healing has been studied in great detail in both animal and human studies. The healing of the junctional epithelium and attachment to the tooth surface takes approximately 1 week to repair followed by continuing maturation of the granulation tissue and connective tissue fibres over the next 3-4 weeks. During this period, the fibrin clot should stabilize, to enable adequate and timely healing. Hence, the first week following NSPT is the most critical phase that requires adequate isolation from external trauma and seclusion from any source of plausible infection\textsuperscript{155}. The application of cyanoacrylate adhesive at the gingival margins was envisioned to create a biological barrier that would provide a secluded environment for healing of the epithelial attachment.
Cyanoacrylates are tissue adhesives that have been used since 1950’s as hemostatic dressings in varied applications in medicine and dentistry. The adhesive used in this study is a viscous blend of N-butyl and 2-octyl cyanoacrylate. It is dispensed in plastic containers as monomeric forms, and upon contact with hydroxyl ions present in water, it rapidly polymerizes adhering to moist living tissue. In Canada, PeriAcryl is a Class I device per the Medical Device Regulations and is indicated as a useful adjunct when performing procedures as gingivectomies, mucogingival surgeries, flap surgeries and biopsies. The higher viscosity material provides improved flow control allowing for precision application and less waste.

In the present study, the periodontal clinical parameters PD, CAL and R did not show a significant change from baseline to the final (6-8 weeks) visit. However, on excluding pockets ≥ 4 mm in depth a significant change was noted with regards to PD and CAL from baseline to final visit (6-8 weeks). Studies have demonstrated deeper pockets (> 4 mm) to have a greater change in PD and CAL as compared to shallower sulcus (1-3 mm)\(^6\). According to the 2015 taskforce update, the American Academy of Periodontology classified periodontitis with the presence of PD ≥ 4 mm with BOP\(^{156}\).

No significant difference in PD, CAL, and R was noted between the experimental and control sites. NSPT, being the treatment in favour of periodontitis, is also considered the reference standard and thus used as an active control for periodontal trials. There are few studies in which investigators compare NSPT with no treatment and, therefore, the strength of NSPT makes it challenging to demonstrate significant clinical benefits when compared to any adjunctive therapeutic modalities. Similarly, it was shown in various studies assessing the effectiveness of a
locally delivered antibiotic therapy that the primary improvements in clinical parameters are obtained through NSPT specifically SRP alone to the extent of 1.45 mm mean reduction in PD and adjunctive use of antibiotics only provided a modest benefit of 0.25-0.5 mm\textsuperscript{157,158}. Additionally, the small sample size may not have allowed for the statistical power to result in a mean difference between the experimental and control arms.

The clinical parameter BOP had a significant reduction in % BOP score in the experimental sites from baseline to the final 6-8 week visit. BOP reduction was non-significant in the control sites. The difference between the two arms was also statistically significant (p=003\textsuperscript{*}). BOP is not only an indicator of periodontal inflammation but also an important risk predictor of progression of periodontal disease. It is an objective clinical sign that is more sensitive and reliable than visual signs of inflammation\textsuperscript{159}. The diseased periodontal tissues have been shown histo-pathologically to have increased vasculitis, thinning and degeneration of the sulcular epithelium as well as perivascular collagen loss that predisposes the tissues to bleed on provocation when probed\textsuperscript{38}. Lang et al.\textsuperscript{50} have shown that in periodontal maintenance subjects assessed on 4 subsequent recalls that had sites with BOP on all recall visits vs. sites with no BOP, the incidence of CAL >2 mm was found to be 30\% vs. 1.5\% respectively. The subjects with mean BOP>16\% were found to have a significantly higher number of sites with residual PD>5 mm and CAL>2 mm. Lang et al.\textsuperscript{52} have also shown that the positive predictive value of BOP may be only approximately 6\%, but its negative predictive value is almost 98\% indicating that absence of BOP is a reliable indicator of the stability and health of the gingival tissues. BOP sites have shown on histopathological assessment an almost 3 fold increase in the number of inflammatory cells compared to sites with no BOP\textsuperscript{160}. Several studies have shown a significant and strong
correlation of BOP as compared to PD or CAL to C-reactive protein (CRP) levels in blood\textsuperscript{149, 150}. CRP is an essential inflammatory mediator that has been causally linked to various systemic diseases.

The results of the present study indicate that the adjunctive application of the cyanoacrylate adhesive did significantly reduce periodontal inflammation, which, can potentially translate to improvement in healing and minimizing the disease risk of the periodontium.

BOP scoring assessment may be affected by variables such as the width of the probe and the force of probe insertion. Lang et al.\textsuperscript{51} showed that increasing the probing forces from 0.25N to 1N increased the mean BOP\% from 7.1 to 41.5%.

The use of the automated pressure-controlled Florida Probe\textsuperscript{\textregistered} was intended to provide a standardized force (15 g) and precision of 0.2 mm during measurements, thereby minimizing the variability in clinical assessments. Each site was probed with a uniform and recommended force during the assessment of the clinical parameters including BOP at baseline and the final visit.

Both the arms in the present study individually demonstrated a significant reduction in plaque scores from baseline to 1-week (experimental p<0.000, control p<0.000) but with no significant difference between the sites (p=0.542). A significant increase in plaque accumulation was noted from 1-week to the final 6 to 8-week visit (experimental p<0.018, control p<0.012) but again this difference was non-significant between the two groups (p=0.791). This is understandable since NSPT results in significant removal of supra and sub-gingival plaque and biofilm deposits that eventually approach baseline values over a period of 2-3 months. Several studies have shown
that the sub-gingival bacterial plaque levels are reversed 9-11 weeks following their professional removal155,161. The reduction in plaque score at the experimental sites (adhesive application) was found comparable to the control sites despite instructions to avoid oral self-care (brushing and interdental cleaning). The experimental sites demonstrating a significant reduction in BOP and plaque score may be attributed to the antibacterial properties of the cyanoacrylate; heat and the trace amounts of toxic by-products are potentially bactericidal. This antibacterial response has been demonstrated for a period of up to 10 days following its application119. The cyanoacrylate application despite its plaque retentive texture was found capable of minimizing supra-gingival biofilm recolonization.

To improve healing in the immediate post-NSPT period, some previous studies have adjuncively used periodontal dressing materials following NSPT. Sigusch et al.103 conducted a longitudinal study on aggressive periodontitis patients where the initial supra-gingival scaling was completed and then the sub-gingival scaling and root planing and curettage session was followed up with application of a zinc oxide periodontal dressings at the test sites for a variable period of 3-4 and 7-8 days and compared to control sites with no dressing applied. All these subjects were administered systemic metronidazole at this visit. These patients were reassessed at 6 and 24 months. Results of the study showed a statistically significant improvement in all clinical periodontal parameters when compared to baseline but PD reduction and CAL gain at both time points in the test sites where the dressing was placed for 7-8 days was statistically significant as well the difference in reduction in BOP at 24 months when compared to the control group. Similar results of increased reduction in PD and gain in CAL were obtained in another
split-mouth study conducted by Genovesi et al.\textsuperscript{104} over a period of 2 months with the use of zinc oxide containing periodontal dressing following NSPT.

Keestra et al.\textsuperscript{105} conducted a split-mouth study on untreated patients with moderate to severe chronic periodontitis with one stage full mouth disinfection and additional application of periodontal dressing (Coe Pak) for a period of 1-week at the test sites. The results of the study showed that at three month re-assessment, a significant improvement in the clinical parameters of PD, CAL, PI and BOP and found them all to be significantly greater at the experimental vs. control sites. Additionally, at 1-week when the periodontal dressing was removed, the subjects were asked to grade their severity of pain in both test and control sites using a numeric rating scale. The pain experience was significantly lower at the experimental vs. control sites\textsuperscript{105}.

None of the above-conducted studies included subjects’ perception of acceptability of the dressing material placed. Only one study recorded the subjects’ pain experience, which was done at 1-week following NSPT. In the present study, an attempt was made to obtain daily subjects’ perception of pain and discomfort for a 1-week period following NSPT. The subjects’ perception of the intra-oral adhesive and the overall acceptance of the application were also assessed.

The concept behind the use of periodontal dressings was to protect the blood clot, stabilization of the periodontal tissues and provision of a barrier to external trauma or bacterial colonization. This may have led to the enhanced healing obtained in all these studies. On the contrary however, several clinical trials are not in favour of the use of periodontal dressings based on its limited effects on healing, increased propensity to accumulate dental plaque with subsequent
microbial invasion, greater pain experience, and difficulty in mastication and eating. Hence, due to lack of adequate evidence, clear conclusions regarding the benefit of the use of periodontal dressings are still lacking following both surgical and NSPT procedures. Future studies with larger sample size, longer follow up and better control is essential to validate their use as standard of care. Most periodontal dressing materials are bulky, can irritate the surrounding tissues, interfere with mastication and speech and can increase halitosis due to the adherence of bacteria. This might explain the non-compliance of clinicians and possibly a deterrent in use. In comparison, the cyanoacrylate adhesive used in this study is not bulky and easier to apply as a thin film with the necessary barrier function and additional haemostatic and antibacterial properties. Currently, there is no known published study clinically applying cyanoacrylate adhesive as an adjunct to NSPT.

6.2 Dentine hypersensitivity

Teeth sensitivity was also assessed in this study using VAS scoring. A decrease in sensitivity was noted in the experimental and control sites at 1-week following NSPT but this reduction was significant for the control sites (p= 0.01) and non-significant in the experimental sites (p=0.287). A subsequent change in VAS at the final (6-8 week) visit was non-significant for both experimental and control sites.

There are two plausible rationales for the decrease in sensitivity in the control sites at 1-week post NSPT; the control sites had a higher mean score of teeth hypersensitivity at baseline compared to the experimental sites, and daily mechanical disruption was more manageable at the control sites resulting in lower plaque accumulation than the experimental sites. Subjects were
advised to not mechanically disrupt the adhesive to ensure the barrier would remain intact. The greater accumulation of plaque at the experimental sites may have caused heightened sensitivity due to an acidic milieu. However, the mean difference between the sites was statistically non-significant at all time points. This is in concordance with a study by Tammaro et al.\textsuperscript{9} that showed an increase in the dentine sensitivity following scaling/root planing (NSPT) only in teeth that have high baseline sensitivity and have also shown a reduction in sensitivity within 2-3 weeks following meticulous plaque control.

\section*{6.3 Patient-reported outcomes}

Subjects reported outcomes have been studied in various parameters of health assessment and is emerging as an essential tool in analyzing the effectiveness and quality of clinical care provided. An opinion directly obtained from a patient is a successful method of assessing the patient’s experience as perceived by them. Subject reported outcome assessment techniques should be reliable, valid, sensitive enough to detect differences in treatment provided and be clinically interpretable\textsuperscript{162}. Given the value in incorporating these outcome measures\textsuperscript{163}, the present study attempted to obtain subjects’ perception of the pain experience following NSPT and the acceptability of the adjunctive treatment provided. The split-mouth study designs rather than a controlled placebo may have introduced a bias due to lack of subject blinding. To minimize this bias, the subjects were informed that the tissue adhesive applied may or may not have any active ingredient. The subjects were also strongly advised against discussing their experimental sites with the examining (blinded) clinician to maintain the sanctity of a blinded study. Moreover, the tissue adhesive applied is self-shedding and is gradually lost over a period of 5-10 days and this time factor may also be variable between subjects.
Other factors impacting the variability in the subjects’ perception results include: the reliability of subject’s scoring on the VAS scale, the level of awareness and understanding of the questionnaire, compliance to follow instructions, the reluctance to provide honest answers for the benefit of the study, and the genuine inability to differentiate between pain from hard tissues (teeth) or soft tissues (gingiva).

In the present study, efforts were made to have a simple questionnaire and a repeatable format. Diagrammatic representations were used to simplify the label sites of pain or discomfort. VAS scale was used for evaluating the subjects reported outcomes. Baseline assessments were performed under supervision in the dental clinic thereby familiarizing the subjects with the format and minimizing errors. To enhance subject compliance, follow-up communication during the week was performed to address any concerns and to support daily compliance with the questionnaires.

VAS has been successfully used in various health-based research studies to quantify pain perception. It requires little training and is very easily adaptable to subject use. It was found to be highly correlated to the 5-point verbal pain descriptor as well as the numeric rating scale. Its reliability has been considered to be more effective with literate and aware subjects. To further minimize the variability in marking in this study, in addition to the already existing verbal descriptors, vertical lines at 1 mm equal intervals were used without any verbal or numerical descriptors to provide spatial orientation on the 10cm line and without creating any bias.84 The VAS scoring in this study was further categorized to help interpret the results into scores ≤ 1; no pain perceived, >1-4; mild pain perceived and >4; moderate to severe pain perceived.
These categories were adopted based on various studies that have determined the cut-offs in studies using receiver operator characteristics (ROC) curves\textsuperscript{164,165}.

The resulting pain perception for both teeth and gingival tissues depicted a predictable increase in the number of subjects experiencing moderate to severe pain from baseline following NSPT and a reduction in their number at the 1-week follow-up visit.

Pain perception in the teeth was noted as being >4 in 26% (/23) subjects at day-1 following NSPT and at the 1-week visit follow-up, no subjects perceived any pain >4 on the VAS. Similarly, for pain perception in soft tissues, 26% (/23) subjects reported experiencing pain >4 on day-1 and this reduced to no subjects experiencing moderate to severe pain at the 1-week follow-up visit. These results should be interpreted with caution since it may be difficult for subjects to differentiate between pain in the teeth and gingival tissues. The trends, however, indicated a reduction of discomfort/pain for both teeth and adjacent soft tissues over the 1-week post-NSPT period. These results concur with a similar study conducted by Pihlstrom et al.\textsuperscript{11} to assess patient perception of pain following NSPT and they reported pain to be of maximum intensity in all subjects in the first 2-8 hours and found the pain returning to baseline levels by the next morning of the procedure.

The study was clinically relevant for the adhesive application protocol used. There were no reported adverse reactions, allergic reactions, medical emergencies or localized abscess formations following the application of the adjunctive adhesive. This material (Periacryl\textregistered\textsuperscript{90HV}) was found safe for the clinical application in the sample population studied.
However, some subjects (12/23) did complain of the adhesive feeling sticky, rough textured, causing altered taste perception and at times sticking to the adjacent tissues causing irritability. One subject was also unhappy about not being able to brush the site with the adhesive application and felt it be “unclean”. However, 11/23 subjects had no concerns with its application. Some of these comments could be attributed to the variability in the application of the adhesive, accessibility and individual patient acceptance and tolerance to having a new material intra-orally. The application of the adhesive does have a slight learning curve. The earlier applications by the trained treating clinician might have an impact on the subjects’ comments.

Incidentally, a greater number of subjects (8/23) complained of both hard and soft tissue discomfort at the test sites as compared to the control sites (6/23). However, this should again be interpreted with caution since the subjects may not have been able to differentiate pain from the discomfort of having the sticky material in their mouth. This is one disadvantage of not having a placebo on the control site with a similar texture that may have minimized the variability.

Overall the subjects were accepting of the adjunctive adhesive placement with 69.5% subjects (16/23) scoring <1 (acceptable) indicating a high acceptance and only 13% (3/23) subjects scoring >4 (not tolerable) indicating a reluctance in repeating this therapy. Further analysis on subjects acceptance of the adhesive application following their NSPT appointment, 30% (7/23 subjects) scored >4 on VAS scoring for their tolerability of the material, and this percentage consistently reduced over the following 1-week with no subject scoring >4 at the week-1 follow-
up visit. The material does self-shed within 5-10 days and provides within the first few days, the barrier function following NSPT that is critical for wound healing.
Chapter 7: Conclusions and Future directions

7.1 Conclusions

Periodontal disease is an inflammatory disease, and that burden not only causes localized effects but also leads to widespread systemic conditions. Inflammation rather than bacteria is the body’s link between oral and overall health status. BOP is an essential indicator of gingival inflammation and a reliable predictor of further periodontium destruction. The statistical and most importantly, the clinical significance of BOP reduction in the present study might suggest that application of cyanoacrylate adhesive may provide a secluded microenvironment to reduce inflammation.

In the present study when all sites were pooled there were non-significant changes noted with PD, CAL and R at both time points and between the two data sets. On excluding sites with baseline PD ≥ 4mm, statistical significance was noted with PD and CAL from baseline to final visit in both control and experimental arms. However, between the two treatment arms, no significant changes were noted.

No significant change in sensitivity of teeth was noted from baseline to 1 week and 6-8 week time points with adjunct application of cyanoacrylate adhesive and also on comparison to NSPT alone.

Pain and discomfort in both teeth and gingival tissues had the greatest incidence of being moderate to severe at Day 1 following NSPT and reduced to no subject reporting pain of that intensity at 1 week follow up.
No adverse events of clinical significance were noted following the application of cyanoacrylate adhesive during the entire course of the study.

In summary, oral health care providers are to impart comprehensive periodontal care that address the professional and daily mechanical disruption of the oral microbiome (biofilms) and its by-products, empathize with the dental fears and concerns, provide reasonable means for post-treatment comfortability and encourage consistent professional periodontal management.

Within the limitations of this present pilot study, it may be surmised that cyanoacrylate tissue adhesive with its antimicrobial traits and high acceptance rate may serve as a safe and beneficial adjunct to NSPT in reducing gingival inflammation as evident by reduction in BOP.

### 7.2 Future Directions

As with all study designs, the split-mouth has its advantages and disadvantages. Having an adhesive placebo with similar flow characteristics and texture but lacking the active ingredients to attain double blinding and minimize variability might be beneficial. Pre-testing of such a placebo material would be needed to ensure that it does not have any therapeutic effects by providing a barrier effect.

The sample size for human RCT can now be calculated using power analysis based on the results of this pilot study. It would be beneficial to have all subjects requiring phase I NSPT (initial therapy) or sub-categorize to determine the effect of the procedure on changes in all the periodontal clinical parameters.
The data for this study had clustering due to the multiple levels of assessments. It would be useful to conduct statistical tests using a linear mixed effect model for parameters of PD, CAL, R and VAS scores and logistic mixed effect models for data with binary variables such as PI and BOP. These can be done by assessing fixed effects such as the interaction of time and treatment and random effects such as assessing at the level of subject, quadrant, tooth and site.

The use of histopathological samples or levels of various inflammatory markers in the gingival crevicular fluid could be ascertained to evaluate the progress of healing following the use of cyanoacrylate adhesive. It would also be interesting to evaluate the bactericidal or bacteriostatic effects of cyanoacrylates against the common periodontal pathogens by pre and post treatment microbial sampling to understand its antimicrobial properties in the periodontal disease setting entirely.

The changes in pain and discomfort in the teeth and the gingival tissues need to be evaluated based on patient reported outcome (VAS scores) from baseline, daily journal for 7 days, and final visit (week 6-8) assessment.

The mean tooth and gingival tissue discomfort over time in experimental and control arms can be evaluated and plotted graphically (i.e. a plot for each treatment). A third plot may be considered for when patients reported discomfort in non-specific locations.

The patient reported outcomes are powerful tools to enable directed therapeutic intervention and improve quality of patient care and hence measures should be made from the results of the study to address the cited patient concerns and make suitable therapeutic alterations. It is essential to
continue to incorporate well-designed and reliable methods of documenting these responses in
the future studies as well.
Bibliography


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Appendices

Appendix 1: Health Canada approval of Periacryl® 90 HV

Manufacturer's Certificate to Cover Exports of Medical Devices

We, the undersigned, manufacturer of the following devices,

<table>
<thead>
<tr>
<th>MODEL</th>
<th>CATALOGUE</th>
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<tbody>
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<tr>
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<td>PerAcryl Unit of Use Clear High Visibility</td>
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</tbody>
</table>

We hereby certify that
(a) each device is manufactured, produced and sold in Canada in accordance with the requirements of Canada’s Food and Drugs Act and Regulations thereunder;
(b) tests have been conducted in respect of each device and that the tests indicate that the nature of the benefits claimed to be obtainable through the use of each device and the performance characteristics of each device are justified.

declaraons
(a) que chaque instrument est fabriqué, produit et vendu au Canada en conformité avec la Loi sur les aliments et drogues du Canada et du règlement qui en découle;
(b) que des essais ont été effectués pour chaque des instruments et que ces essais démontrent que les déclarations faites au sujet des avantages de l’utilisation des instruments en question et au sujet de leurs caractéristiques de performance sont justifiées.

Name of Manufacturer - Nom du fabricant:
Gilead Inc (Gilead Inc is covered under MDEL# 11-02 for Blacklock Medical Products Inc)

DEC 1 & 2014

John Eastwood
Notary Public
5058 - 47A Avenue
A Commissioner, Notary, BC V4K 1T3
604-916-8010

For office Use Only
Health Canada
Health Products and Food Branch

It is hereby certified that
(a) devices manufactured, produced and sold in the manner above described would not, by reason of the method of manufacture thereof, be in violation of the Food and Drugs Act of Canada and the Regulations thereunder;
(b) devices manufactured and sold in compliance with said Act and Regulations.

Il est attesté
(a) que les instruments fabriqués, produits et vendus selon les modalités susmentionnées ne contreviennent pas, du fait de leur méthode de fabrication, à la Loi sur les aliments et drogues du Canada et au règlement qui en découle;
(b) que les instruments fabriqués et vendus en conformité de la Loi et du dit règlement peuvent être exportés sans restriction.

2014
Canada

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Appendix 2: Participant Information and Consent Form

Study Title: The Efficacy of Cyanoacrylate (PeriAcryl®90 HV) in its Adhesive Post-Operative Properties on Periodontal Wound Healing and Patient Discomfort after Non-Surgical Periodontal Treatment: A Pilot Randomized Clinical Trial

Principal Investigator: Ms. Penny Hatzimanolakis, DipDH, BDSc, MSc, RDH
Department of Oral Biological & Medical Sciences
University of British Columbia
Telephone: 778-998-0924

Co-Investigators: Dr. Rashi Chaturvedi, BDS, MDS
Dr. Hannu Larjava, DDS, PhD, DipPerio
Dr. Edward E. Putnins, DMD, DipPerio, MRCD (C), MSc, PhD
Dr. Lari Hakkinen, DDS, PhD
Department of Oral Biological & Medical Sciences
University of British Columbia

1. Invitation
You are being invited to take part in this research study because you have been identified as having gum disease, and cleaning of your teeth has been recommended.
2. Your participation is voluntary

Your participation is voluntary. You have the right to refuse to participate in this study. If you decide to participate, you may still choose to withdraw from the study at any time without any negative consequences to the medical care, education or other services to which you are entitled or are presently receiving.

You should be aware that there is a difference for both you and your dental care professional between being a patient and being a research participant. As a patient, all dental procedures and treatments are carried out for your benefit only according to standard accepted practice. As a research participant, you and your dental care professional also must take into account the requirements for the research study. These may include procedures and treatments that are not part of standard practice or are not yet proven. This consent form describes the diagnostic and treatment procedures that are being carried out for research purposes. Please review the consent document carefully when deciding whether or not you wish to be part of the research and sign this consent only if you accept being a research participant.

If you wish to participate in this study, you will be asked to sign this form.

Please take time to read the following information carefully and to discuss it with your family, friends and doctor before you decide.
3. Who is conducting this study?

This study is being conducted by Ms. Penny Hatzimanolakis who is a Clinical Associate Professor with the UBC Department of Oral Biological & Medical Sciences. The study has been provided with cyanoacrylate adhesive (PeriAcryl®90 HV) as a gift in kind from Blacklock Medical Products and GluStitch, Inc.

4. Background

The first step in treating gum disease involves removal of the factors causing the disease; these include calculus deposits, plaque and the bacteria associated with it. Routine teeth cleaning removes these factors and smoothens root surfaces to minimize accumulations. However, this treatment is usually accompanied with some soft tissue tenderness and teeth sensitivity that may be uncomfortable making oral hygiene maintenance difficult for a few days. Depending on individual cases, additional therapy, such as with the use of cyanoacrylate adhesives (i.e. PeriAcryl®90 HV), may assist patients with daily oral self-care procedures enabling them to complete the recommended periodontal (areas around your teeth) treatment. Cyanoacrylates have been shown to have antimicrobial and enhanced wound-healing properties. This study is a pilot trial at UBC, and we intend to enroll 20 participants.

In Canada, the cyanoacrylate adhesive (PeriAcryl®90 HV) we will use in this study is a Class I device per the Medical Device Regulations, and Health Canada has approved its sale and use. PeriAcryl®90 HV is in wide use clinically, and this study will investigate this product prospectively within its current indication.
5. What is the purpose of the study?

This is a small pilot study to better understand the effectiveness of PeriAcryl®90 HV along with teeth and gum cleaning at sites of gum disease. This product is already widely used clinically for this purpose, but evidence for its use is lacking; this study will help contribute to its use through evidence-based oral healthcare.

A “pilot study” or “feasibility study” is being done to test the study plan and to find out whether enough participants will join a larger study and accept the study procedures. This type of study involves a small number of participants and so it is not expected to prove safety or effectiveness. The results may be used as a guide for larger studies, although there is no guarantee that they will be conducted. Participation in a pilot study does not mean that you will be eligible to participate in a future larger study. Knowledge gained from pilot or feasibility studies may be used to develop future studies that may benefit others.

6. Who can participate in this study?

You may be able to participate in this study if:

- You are willing to provide written informed consent and are 19 years of age or older;
- You have a minimum of 18 teeth and at least 4 measurement sites with teeth pocket depths at least 5 mm with bleeding on probing in at least 2 different parts (quadrants) of your mouth; and
- You must be willing and able to return for treatment and evaluation throughout the course of this study.
Participants must have a good understanding of English language, as the study is unable to fund a certified translator.

7. Who should not participate in this study?

You will not be eligible to participate in this study if:

- You have used any antibiotics or prescribed anti-inflammatory drugs in the last month;
- You have sites with overt abscess (such as ulcers, blisters, etc. in your mouth), active cavities or crown/root fractures;
- You are pregnant, nursing or otherwise plan to become pregnant over the course of the study;
- You are an active smoker;
- You have any known side-effects or allergies to cyanoacrylate or formaldehyde; and/or
- You have significant disease or take a medication that, in the opinion of the investigator, may interfere with your participation in this study.

8. What does the study involve?

Overall Design of the Study

If you choose to participate in this study, you will sign this form and have your pertinent medical/dental history reviewed. Your eligibility will be assessed in addition to your routine teeth cleaning evaluations. This is considered screening for the purposes of the study. You will have an assessment and examination with the researchers following this in 1 week. You will have photographs taken of your mouth as per routine periodontal care, and you will be randomized (like flipping a coin) to determine which part (quadrant) of your mouth will have
cyanoacrylate applied in another week. This is to get a clear baseline evaluation for the study. This study will involve a process called “blinding” where we will use an examiner in the study team who will not know which part (quadrant) of your mouth will have the cyanoacrylate. It is important that you do not tell the blinded examiner anything you may know or think you know about where the cyanoacrylate may be placed in your mouth. To further reduce bias in this study and although you will know which part (quadrant) of the mouth the adhesive (cyanoacrylate) is placed, the treating clinician will not inform you whether or not your adhesive contains the active cyanoacrylate. At the next visit, you will be assessed, and you will proceed with routine teeth cleaning (also referred to as scaling/root planing). You will have the cyanoacrylate placed along the gum lines at test sites of one of your quadrants of your mouth at this visit, and you will again have photographs taken over your mouth as per routine care. You will be instructed to not brush or floss the areas around where the cyanoacrylate was applied. You will be provided a journal to record your status on a daily basis for the first week once you have the cyanoacrylate placed along your gum line. You will also be requested to avoid any unnecessary use of over-the-counter pain medications (analgesics); , if you feel pain medication is absolutely necessary, then you will be asked to record the details of the medication use in your journal. You will be contacted after 48 hours following the procedure to check up on your general oral health status and remind you of the need to complete the journal on a daily basis.

In another week, you will be assessed again, and you will have photographs of your mouth taken as per routine care. The routine teeth cleaning will be completed at this time. The presence or absence of the cyanoacrylate adhesive will be assessed, and any remaining adhesive will be carefully removed paying particular attention to not agitate the gum tissues. At the end of this visit, you will be asked to resume routine oral self care at the test sites.
After approximately 6-8 weeks from starting the study, you will return for full assessment with the study team, and you will again have photographs of your mouth taken as per routine care. This will conclude the study. We expect participants to be in the study for approximately 2 months.

If you decide to join this study:
If you agree to take part in this study, the procedures and visits you can expect will include the following:

Screening Visit – Before you begin the study

(Visit 1: Week 0): 30 minutes
You will review this consent form and have your eligibility assessed.

With your consent, the study team will review information already gathered as part of your routine care. This will include the following:

- Review of your routine dental & medical history;
- Review of your routine comprehensive oral assessment, Periodontal Screening & Recording (PSR) (probing), generalized sensitivity identification; and
- Review of your routine photographs of your mouth.

The above procedures (other than your consent and eligibility) will be discussed by your dental professional team as part of your routine care.
Baseline & Randomization Visit
(Visit 2: Week 1): 45 minutes (30 minutes for routine care & 15 minutes for additional research procedures)

The study team will assess you in follow-up. You will have the following procedures:

- Review of your dental & medical history;
- New photographs of your mouth as per routine care;
- Randomization to identify which part (quadrant) of your mouth will have the cyanoacrylate adhesive placed and which part (quadrant) will be a matched control (i.e. to identify your “test” and “control” sites);
- Complete oral & periodontal examination with a blinded examiner with the study team; and
- Baseline assessment for overly sensitive teeth with the blinded examiner by applying a blast of air to the areas and also by using a visual scale (on paper) used by the study team.

Start of Cyanoacrylate Adhesive Placement
(Visit 3: Week 2): 60 minutes (50 minutes for routine care & 10 minutes for additional research procedures)

The study team will assess you in follow-up. The blinded examiner in the study team will not be present at this visit. You will have the following procedures:

- Review of your dental & medical history;
- Teeth cleaning (scaling/root planing with local anesthesia [to manage discomfort] as needed) on 2 opposite teeth sites by a treating dental care provider (who is not blinded);
• Placement of cyanoacrylate adhesive along the gum lines of selected “test” sites by the treating dental care provider (again who is not blinded);

• New photographs of your mouth as per routine care; and

• You will be given a journal to record in daily for the first week.

1st Follow-Up of Cyanoacrylate Adhesive Placement

(Visit 4: Week 3): 60 minutes (45 minutes for routine care & 15 minutes for additional research procedures)

The study team will assess you in follow-up. You will have the following procedures:

• Review of your dental & medical history;

• Complete oral examination;

• New photographs of your mouth as per routine care;

• Measuring sensitivity of “test” teeth using a blast of air to the appropriate areas;

• Measuring sensitivity of “test” teeth and soft tissues by using a visual scale (on paper) with the blinded examiner;

• Removal of any remaining cyanoacrylate adhesive;

• Assessment of plaque scores at test and control sites;

• Assessment of any side-effects or any other changes to your health; and

• Completion of teeth cleaning (scaling/root planing) by a treating dental care provider.
2nd Follow-Up of Cyanoacrylate Adhesive Placement & End of Study (EOS)

(Visit 5: Week 6-8): 45 minutes (30 minutes for routine care & 15 minutes for additional research procedures)

The study team will assess you in follow-up. You will have the following procedures:

- Review of your dental & medical history;
- Comprehensive oral examination;
- Complete periodontal examination;
- New photographs of your mouth as per routine care;
- Measuring sensitivity of “test” teeth using a blast of air to the appropriate areas;
- Measuring sensitivity of “test” teeth and soft tissues by using a visual scale (on paper) with the blinded examiner;
- Assessment of any side-effects or any other changes to your health.

9. What are my responsibilities?

The participants in this study are responsible for:

- Arriving for visits at the allotted time;
- Notifying the study team about a possible pregnancy;
- Reporting any changes in health, new symptoms, side-effects and medication changes (if any) over the course of the study;
- Notifying the study team about any antibiotics you may use or be prescribed; and
- Not disclosing the part of your mouth where cyanoacrylate adhesive has been applied.
Blinded evaluations will be done over the course of the study; therefore, it is important that you do not disclose the treatment assignment provided at randomization to the examiner.

10. What are the possible harms and discomforts?
The risks and discomforts of the standard teeth cleaning will be explained to you as part of your standard care. These include standard risks associated with tooth cleaning procedures such as:

- Redness of the gums;
- Sensitivity of the tooth and root surfaces to hot and cold stimulations;
- Swollen and tender gums;
- Slight pain; and/or
- Standard risks, such as numbing, as a result of a dental anesthetic (for pain management).

In rare instances, a delayed skin reaction (i.e. allergic-like reactions) and airway reactions (i.e. asthma) have been reported with the earlier-used cyanoacrylates for denture repair purposes. However, there have been no reported side-effects with the use of the cyanoacrylate products (PeriAcryl®90 HV) we will use in the study.

11. What are the potential benefits of participating?
You may benefit from additional oral healthcare (teeth cleaning) which would otherwise not be provided for at no charge. You may be responsible for covering the costs of oral healthcare (teeth cleaning) that is not part of this study. However, no one knows whether or not you will benefit from this study, specifically from the cyanoacrylate placement. There may or may not be direct benefits to you from taking part in this study.
We hope that the information learned from this pilot study can be used in the future to benefit other people with conditions.

12. What are the alternatives to the study treatment?
If you choose not to participate in this study or to withdraw at a later date, regular teeth cleaning may be available to you. You can discuss this with your dental care professional before deciding whether or not to participate in this research project.

13. What if new information becomes available that may affect my decision to participate?
If you choose to enter this study and at a later date a more effective treatment becomes available, it will be discussed with you. You will also be advised of any new information that becomes available that may affect your willingness to remain in this study.

14. What happens if I decide to withdraw my consent to participate?
You may withdraw from this study at any time without giving reasons. If you choose to enter the study and then decide to withdraw at a later time, you have the right to request the withdrawal of your information collected during the study. This request will be respected to the extent possible. Please note, however, that there may be exceptions where the data will not be able to be withdrawn; for example, where the data is no longer identifiable (meaning it cannot be linked in any way back to your identity) or where the data has been merged with other data. If you would like to request the withdrawal of your data, please let your study team know.
15. Can I be asked to leave the study?

If you are not able to follow the requirements of the study or for any other reason, the study team may withdraw you from the study and will arrange for your care to continue. On receiving new information about the treatment, your study team might consider it to be in your best interests to withdraw you from the study without your consent if they judge that it would be better for your health. If you are asked to leave the study, the reasons for this will be explained to you and you will have the opportunity to ask questions about this decision.

16. How will my taking part in this study be kept confidential?

Your confidentiality will be respected. However, research records and health or other source records identifying you may be inspected in the presence of the investigator or her designate by representatives of UBC Clinical Research Ethics Board for the purpose of monitoring the research. No information or records that disclose your identity will be published without your consent, nor will any information or records that disclose your identity be removed or released without your consent unless required by law.

You will be assigned a unique study number as a participant in this study. This number will not include any personal information that could identify you (e.g., it will not include your Personal Health Number, SIN, or your initials, etc.). Only this number will be used on any research-related information collected about you during the course of this study, so that your identity will be kept confidential. Information that contains your identity will remain only with the Principal Investigator and/or designate. The list that matches your name to the unique study number that is
used on your research-related information will not be removed or released without your consent unless required by law.

Your rights to privacy are legally protected by federal and provincial laws that require safeguards to ensure that your privacy is respected. Further details about these laws are available on request to your study team.

17. What happens if something goes wrong?
By signing this form, you do not give up any of your legal rights and you do not release the study team, participating institutions, or anyone else from their legal and professional duties. If you become ill or physically injured as a result of participation in this study, medical treatment will be provided at no additional cost to you. The costs of your medical treatment will be paid by your provincial medical plan.

In case of a serious medical event, please report to an emergency room and inform them that you are participating in a clinical study and that the following person can then be contacted for further information: Ms. Penny Hatzimanolakis at telephone number: 778-998-0924.

18. What will the study cost me?
All research-related dental care and adhesive application that you will receive during your participation in this study will be provided at no cost to you.
You will not be paid for taking part in this research study. However, you may be provided reasonable reimbursement for your extra costs, such as parking, up to $150 that you may need due to participation after having completed the trial at your last visit. Your study team will discuss how this will be done. Original receipts (except for meter parking) are required. Please give your receipts to a member of the study team.

19. Who do I contact if I have questions about the study during my participation?
If you have any questions or desire further information about this study before or during participation, or if you experience any adverse effects, you can contact Ms. Penny Hatzimanolakis at 778-998-0924.

20. Who do I contact if I have any questions or concerns about my rights as a participant?
If you have any concerns or complaints about your rights as a research participant and/or your experiences while participating in this study, contact the Research Participant Complaint Line in the University of British Columbia Office of Research Ethics by e-mail at RSIL@ors.ubc.ca or by phone at 604-822-8598 (Toll Free: 1-877-822-8598).

21. After the study is finished
You may not be able to receive the study treatment after your participation in the study is completed. There are several possible reasons for this, some of which are:

- The treatment may not turn out to be effective for you;
- Your caregivers may not feel it is the best option for you; and/or
- You may decide it is too expensive and insurance coverage may not be available
22. Signatures

**Study Title:** The Efficacy of Cyanoacrylate (PeriAcryl®90 HV) in its Adhesive Post-Operative Properties on Periodontal Wound Healing and Patient Discomfort after Non-Surgical Periodontal Treatment: A Pilot Randomized Clinical Trial

**Participant Consent**

My signature on this consent form means:

- I have read and understood the information in this consent form.
- I have had enough time to think about the information provided.
- I have been able to ask for advice if needed.
- I have been able to ask questions and have had satisfactory responses to my questions.
- I understand that all of the information collected will be kept confidential and that the results will only be used for scientific purposes.
- I understand that my participation in this study is voluntary.
- I understand that I am completely free at any time to refuse to participate or to withdraw from this study at any time, and that this will not change the quality of care that I receive.
- I authorize access to my health records as described in this consent form.
- I understand that I am not waiving any of my legal rights as a result of signing this consent form.
- I understand that there is no guarantee that this study will provide any benefits to me.
- I have read and understood all 9 pages of the consent form.
I will receive a signed copy of this consent form for my own records.

I consent to participate in this study.

<table>
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<tr>
<th>Participant’s Signature</th>
<th>Printed Name</th>
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<table>
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Obtaining Consent

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<th>Printed Name</th>
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My signature above signifies that the study has been reviewed with the study participant by me and/or by my delegated staff. My signature may have been added at a later date, as I may not have been present at the time the participant’s signature was obtained.
### Appendix 3: Study visits schedule

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<th>Visit 3</th>
<th>Visit 4</th>
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</table>
Appendix 4: Vas Assessment: Air Blast Testing

Tooth Number:

Kindly grade any discomfort with your teeth on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

Subject Code:
Date:

Patient Initials:
Appendix 5: Baseline Assessment

Subject Code: 
Date: 

1. Kindly grade any discomfort with your teeth on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

2. Kindly grade any discomfort with your gums on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’

Please indicate where you are feeling discomfort today.

3. Any additional comments or adverse events that you may want to share
Appendix 6: Daily Journal

Subject Code:
Date:

1. Kindly grade any discomfort with your teeth on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

No discomfort  |  Maximal discomfort

2. Kindly grade any discomfort with your gums on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’

No discomfort  |  Maximal discomfort

3. Kindly grade your acceptance with having this material present in your mouth, 0 being ‘acceptable’ and 10 being ‘not tolerable’.

Acceptable  |  Not Tolerable

Please indicate where you are feeling discomfort today.

Upper

Right

Left

Lower

4. Any additional comments or adverse events that you may want to share
Appendix 7: 1st Follow Up Visit

Subject Code:
Date:

1. Kindly grade any discomfort with your teeth on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

No discomfort | Maximal discomfort

2. Kindly grade any discomfort with your gums on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’

No discomfort | Maximal discomfort

3. Kindly grade your acceptance with having this material present in your mouth, 0 being ‘acceptable’ and 10 being ‘not tolerable’.

Acceptable | Not Tolerable

Please indicate where you are feeling discomfort today.

Upper

Right | Left

Lower

4. Any additional comments or adverse events that you may want to share

______________________________________________________________________________

______________________________________________________________________________

______________________________________________________________________________
Appendix 8: 2nd Follow Up Visit

Subject Code:
Date:

1. Kindly grade any discomfort with your teeth on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

2. Kindly grade any discomfort with your gums on a scale of 0 to 10, 0 being ‘no discomfort’ and 10 being ‘maximal discomfort’.

3. Kindly grade your receptiveness to the treatment provided, if it were to be repeated for you with ‘0’ being ‘accepting treatment’ and 10 being ‘not tolerating treatment’.

Please indicate where you are feeling discomfort today.

Upper

Right

Left

Lower

4. Any additional comments or adverse events that you may want to share