University of the Western Cape

Efficacy of alcohol containing and alcohol-free chlorhexidine mouth rinse in reducing periodontal disease during prophylactic treatment.

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Thesis submitted in partial fulfilment of the requirements for the degree of Magister Chirurgiae Dentium in the discipline of Oral Medicine and Periodontics MChD (OMP)

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Key words

Antimicrobial

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Conventional management of periodontal diseases (i.e. scaling, root plan and polishing)

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Efficacy

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Oral rinse

Periodontal clinical parameters

ABSTRACT

Chlorhexidine has been established as the gold standard against which new chemical plaque control agents are tested (Jones, 1997). The addition of alcohol in a chlorhexidine mouthwash had been widely used, however the comparative efficacy of alcohol free chlorhexidine mouthwash had not fully been explored in this study, two chlorhexidine mouthwash preparations were tested to evaluate their comparative efficacy in the treatment of periodontal disease. **Aims:** To assess the efficacy of alcohol-free chlorhexidine mouth wash in comparison to alcohol containing chlorhexidine mouth wash.

Objectives: To determine pre- and post- operative clinical parameters and microbial load in the management of patients with chronic periodontitis.

Methodology: A double blinded randomised control trial was conducted. Patients diagnosed with active chronic periodontitis were included in the study and randomised to either a test (chlorhexidine without alcohol) or control group (chlorhexidine with alcohol). A total of 50 patients were selected for the study.

Results: The Wilcoxon Signed Rank test was used to test the difference between the pre-post pair per clinical indicator and Bana-Zyme. The differences between before and after treatment per indicator were significant at $P<0.001$ for respectively Paroex and Peridex. These values demonstrated the difference between the clinical parameters taken before the treatment and six weeks post treatment.

Conclusion: Both mouth wash solutions with and without alcohol had proven to reduce the microbial load as shown by the BANA-Zyme test, with the alcohol containing solution having been more effective.

DECLARATION

I hereby declare that *"Efficacy of alcohol containing and alcohol-free chlorhexidine mouth rinse in reducing periodontal disease during prophylactic treatment"* is my own work, that it has not been submitted before for any degree or examination in any university, and that all the sources I have used or quoted have been indicated and acknowledged by complete references.

I declare that I have no competitive interest in any of the companies that manufacture any of the tests used in this study. No shares and or stocks are held nor will I have any financial gain or loss with the publication of any manuscript and articles pertaining to this research.

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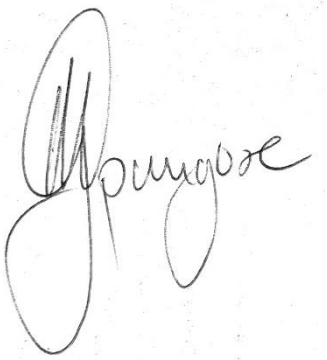
2018

Signed: 

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DEDICATIONS

Dedicated to my children (Asande and Lwazi)

LIST OF TABLES

Table #	Content	Page number
Table 1	Based on Paroex "Wilcoxon Signed Rank Test"	**32**
Table 2	Based on Peridex "Wilcoxon Signed Rank Test"	**33**
Table 3	Distribution of groups treated in the study	**35**
Table 4	Demonstrates the frequency score for the plaque index 6 weeks after treatment	**35**
Table 5	Demonstrates the frequency score for the gingival index 6 weeks after treatment	**36**
Table 6	Demonstrates the frequency score for the pocket depth 6 weeks after treatment	**36**
Table 7	Demonstrates the frequency score for the gingival recession 6 weeks after treatment	**37**
Table 8	Demonstrates the frequency score for tooth mobility 6 weeks after treatment	**37**
Table 9	Demonstrates the frequency score for Bana-Zyme test 6 weeks after treatment	**37**

LIST OF FIGURES

Figure #	Content	Page Number
	CHAPTER 2 (Literature review)	
Figure 01	Chlorhexidine molecule: two symmetrical 4-chlorophenyl groups and two bisguanides groups linked by a hexamethylene bridge between.	**05**
Figure 02	molecular structure of Peridex oral rinse	**10**
Figure 03	molecular structure of Paroex oral rinse	**10**
Figure 04	Periodontal risk – A patient centered paradigm	**16**
Figure 05	Proposal for the pathogenesis circa year 1997	**18**
	CHAPTER 4 (Research and Methodology)	
Figure 06	Brown unmarked bottles randomly assigned to the study group	**24**
Figure 07	Paroex mouthwash	**25**
Figure 08	Peridex mouthwash	**26**
Figure 09	Bana-zyme test machine and BANA-strips	**27**
Figure 10	Toothbrush and toothpaste	**27**
	CHAPTER 5 (Results)	
Figure 11	Two dimensional variation in the plaque and gingival indices in relation to the other parameters	**38**
Figure 12	Demonstrates a slightly higher value after treatment for gingival index as an isolated parameter	**39**

List of graphs

Graphs	Content	Page number
Graph 1	Represents the Plaque index before treatment and 6 weeks follow-up	**34**
Graph 2	Represents the gingival score before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)	**35**
Graph 3	Represents pocket depths before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)	**36**
Graph 4	Represents clinical attachment loss before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)	**36**
Graph 5	Represents teeth mobility & gingival recession before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue: Recession before treatment, Red: Recession after treatment, Grey: Mobility before treatment, Yellow: After treatment)	**37**
Graph 6	Represents average Bana-zyme hits on negative and positive values before and after treatment of periodontitis.	**37**
Graph 7	Representation of the plaque index between the two groups. Paroex shows greater reduction in the plaque	**41**

	index when compared to Peridex. (Blue is Paroex and Brown is Peridex)	
Graph 8	Representation of the gingival score between the two groups. Paroex shows greater reduction of this parameter when compared to Peridex. (Blue is Paroex and Brown is Peridex)	**41**
Graph 9	Represents the clinical attachment 6 weeks after treatment. Paroex show greater reduction in the clinical attachment loss when compared to Peridex. (Blue is Paroex and Brown is Peridex)	**42**
Graph 10	Represents the values of pocket depth 6 weeks after treatment. There is greater reduction in periodontal pockets in the group that received Paroex when compared with Peridex. (Blue is Paroex and Brown is Peridex)	**42**
Graph 11	Represents the average values of teeth mobility between the two test groups. Paroex group shows more reduction in the mobility of teeth following treatment when compared to Peridex. (Blue is Paroex and Brown is Peridex)	**43**
Graph 12	Represents recession in two test groups. No notable improvement of gingival recession was recorded in both test groups. . (Blue is Paroex and Brown is Peridex)	**44**
Graph 13	Graph 13: Represents the values of the BANA-Zyme test. The microbial load was reduced in both test groups equally. (Blue is Paroex and Brown is Peridex)	**45**

CONTENT PAGE

Title page……i

Key words……....ii

Abstract……....iv

Dedications……..vi

Acknowledgement……vii

List of tables……viii

List of figures……ix

List of graphs……x

Content list……xii

Chapter 1 (Introduction)……2

Chapter 2 (Literature review)……4

2.1 Chlorhexidine……...4

2.1.1 Chemical Structure……..5

2.1.2 Use in dentistry……....6

2.1.3 Mechanism of action……7

2.1.4 Toxicity and side effects……..8

2.2 Alcohol……..8

2.3 Peridex and Paroex……...10

2.3.1 Chemical structure……..10

2.3.2 Clinical pharmacology……11

2.3.3 Indications and usage……..12

2.3.4 Precautions and adverse reactions……...12

2.3.5 Dosage and administration…….13

2.4 Bana-Zyme……..……...…….14

2.5 Periodontitis…….....15

2.5.1 Clinical findings of periodontitis……18

2.5.2 Prevention and treatment (general overview)…….20

Chapter 3…….21

3.1 Aims

3.2 Objectives

Chapter 4 (Research and Methodology)……..23

4.1 Study design…….23

4.2 Study sample……24

4.2.1 Inclusion criteria……24

4.2.2 Exclusion criteria……25

4.3 Material……26

4.3.1 Paroex……26

4.3.2 Peridex……26

4.3.3 Bana-Zyme Kit……...26

4.3.4 Toothpaste and Toothbrush……27

4.4 Clinical Parameters……..27

4.5 Ethical consideration……...29

4.6 Conflict of interest……...30

4.7 Data collection and analysis……..…30

Chapter 5 (Results)…….….30

5.1 Values comparing before and after treatment……34

5.2 Values comparing the two test groups……...41

Chapter 6 (Discussion)…….45

6.1 Gingival index, bleeding on probing and plaque index……46

6.2 Gingival recession……..47

6.3 Pocket depth, Clinical Attachment Loss and Tooth Mobility……49

6.4 Bana-Zyme test……...51

6.5 Potential Limitations……...51

Chapter 7 (Conclusion)…….55

Chapter 8 (References)……..54

CHAPTER 1

INTRODUCTION

For the past few decades, mechanical and chemical measures have been adopted as the main option for the control of periodontal disease and caries. Such measures were mainly used as prophylactic options with minimal intervention from any professional dental care (Arweiler N.B., 2001).

Chlorhexidine had been the gold standard for evaluating new chemical plaque control agents (Jones, 1997). In this study, two chlorhexidine mouth wash preparations were tested to evaluate comparative efficacy in the treatment of periodontal disease. This was a comparative clinical study evaluating the efficacy of an alcohol-free chlorhexidine mouth wash with the widely used alcohol containing preparation.

Formulary preparation of chlorhexidine mouthwashes differ for all available solutions prepared for commercial use. These solutions contain varying concentrations of alcohol ranging between 5 to 15%. The addition of which is controversial because of its carcinogenic potential and tissue irritating properties (Herrera *et al*., 2003). However, there are non-alcohol containing solutions available for use. Previous studies examining different chlorhexidine preparations have shown a lack of consensus regarding the effect of additives on the antimicrobial efficacies of the different preparations (Arweiler *et al*., 2006 & Herrera *et al*., 2003).

This made it imperative to test any chlorhexidine formulation that contained additives against the well-studied and documented chlorhexidine formula.

Mechanical interventions (including tooth brushing and flossing) are known to cover a limited amount of surface area when it comes to prophylactic care. For

this simple reason, there has been greater interest in the use of chemical types of prophylactic interventions. (Kathrine R. 1998)

Chemical intervention is adjunctive to the mechanical part of prophylactic treatment. Traditionally, the available mouth washes in the market were known to be alcohol containing chlorhexidine (CHX) rinses and have been known to have a few adverse effects which may have contributed to premature cessation of usage by patients.

Use of mouth washes containing alcohol is usually contraindicated in patients with painful inflammation of the mucosa, those with ulcerated tissues including hypersensitivity to alcohol and patients with resins as restorative material. (Kathrine R. 2008)

The purpose of this study was to determine if alcohol free chlorhexidine mouthwash could be offered to patients as an alternative for prophylactic treatment.

CHAPTER 2

LITERATURE REVIEW

2.1 Chlorhexidine vs. the plaque biofilm

The plaque biofilm had been well established as the initiating aetiological factor for the development of periodontal diseases. The components of the biofilm are documented as a variety of microbial species especially bacteria, that are well protected by an extracellular polysaccharide matrix to ensure adherence to surfaces of teeth, oral soft tissues and other intraoral appliances.

Gram positive aerobic bacteria are collectively known as the primary colonizers and can be cultured in early plaque formation. As the biofilm matured, more Gram positive facultative anaerobes and spirochetes accumulated and at that stage these bacteria are known as secondary colonizers. (Moran J *et al*., 1995)

Further maturation ensured the development of a complex ecosystem and a protective biological advantage to all involved microorganisms, which included the facilitation of nutrient metabolism as well as waste product removal. Furthermore, the biofilm structure imparted a resistance to the diffusion of antimicrobial agents ensuring its survival. (Moran J *et al*., 1995).

The first scientifically proven study that demonstrated the anti-plaque effect of chlorhexidine was performed in 1970 by Loë and Schiott. They demonstrated that 10ml of 0.2% chlorhexidine as a mouth rinse twice daily for a minute was sufficient to prevent the build-up of dental plaque in the absence of mechanical plaque control over a period of 10 days. The result had thus prevented the development of plaque induced gingivitis within that period of time. (Loë and Schiott, 1970).

Chlorhexidine is known as a wide spectrum bactericidal and bacteriostatic agent within the dental setting. Though at differing minimum inhibitory concentrations (MIC), its effectivity against both Gram positive and Gram negative organisms had been established.

Gram negative cocci such as Veillonella are grouped under the most resistant strains of MIC while there low MIC species that included *Staphylococci*, *S. mutans, S. salivarius and E. coli* (Emilson, 1977).

2.1.1 Chemical structure of Chlorhexidine

In the middle of the 20th century, chemists were able to synthesize a compound known collectively as polybiguanides. It captured interest when it was observed to demonstrate a broad spectrum antimicrobial activity (Lindhe et al, 2008). Polybiguanides led to the synthesis of bisguanides which presented an even wider antimicrobial spectrum.

More studies were done on the newly found bisguanides, which led to the development of another compound which presented a much wider bacteriostatic and bactericidal effect. That new compound became (1, 6, bis-4, chloro, phenyldiguanidohexane), and was then termed chlorhexidine, a very strong cationic compound (Lindhe et al, 2008).

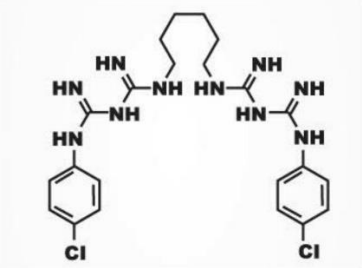


Figure 01: Chlorhexidine molecule, two symmetrical 4-chlorophenyl groups and two bisguanides groups linked between by a hexamethylene bridge. (Davies *et al*, 1954)

2.1.2 Use in Dentistry

Chlorhexidine and fluoride had valuable role in the prevention of dental caries by acting together and, thus, combining chlorhexidine and fluoride was an interest for research. (Gupta *et al*, 2012) It was extensively used during most dental surgical intervention including prophylactic type of treatments.

Chlorhexidine is used as an active ingredient in mouthwashes which were specifically created to control and minimise the amount of oral microflora and reduce plaque in the mouth. Its choice was because it lasted longer in the mouth compared to other mouthwashes and this was the main reason it was preferred over other treatments for gingivitis. (Gupta *et al*, 2012)

Chemical control of plaque was always used in combination with mechanical techniques as prophylactic treatment. Chlorhexidine was used to reinforce such mechanical means of plaque control by minimising accumulation of dental plaque on oral tissue surfaces. This was achieved by preventing adhesion of plaque on the surface.

Dental caries development was minimized using chlorhexidine on highly susceptible S. Mutans commonly implicated in the initiation and progression of caries. (Gupta *et al*, 2012)

Installation of dental prosthesis and orthodontic appliances increase surface area for microbes to adhere, thus, increasing microbial load in the oral environment. It was important to use chlorhexidine to keep microbes at minimum by immersing appliances in solutions of 0.2% chlorhexidine mouth wash when appliances were not used. (Gupta *et al*, 2012)

In cases of oral pathologic conditions, use of chlorhexidine was needed to heal and help regenerate oral tissues and maintain a stable oral hygiene. (Gupta *et al*, 2012) Such conditions includes gingivitis, periodontitis, general fungal infections and after extractions of teeth.

2.1.3 Mechanism of Action

Phosphate-containing proteins in the cell walls of bacteria presents a surface for adsorption of chlorhexidine molecules. Upon attachment to these phosphate-containing proteins, the chlorhexidine molecules penetrates and disrupts the cytoplasmic membrane of the bacteria resulting in leaking of its contents. This was achieved at bacteriostatic concentrations. (Lim K.S. 2008)

In turn, at a slightly higher concentration it provided a much potent effect on the microorganisms. Its bactericidal effects acted by forming an irreversible precipitate with intracellular adenosine triphosphate together with nucleic acids after damaging the cell membrane and entering the cytoplasm. (Lim K.S. 2008)

Chlorhexidine in its nature is known to be both bactericidal and bacteriostatic, fungicidal and fungistatic and some degree of destruction to viruses. (J.L. Leyes, 2002) The time and concentration of chlorhexidine action on a surface determined its potency. The longer the time on the surface the more destruction achieved by increased amount of perforation on the cell wall. Minimum inhibitory concentrations are lower for Gram-positive bacteria than for Gram-negative bacteria because chlorhexidine had an increased affinity for the cell wall of Gram-positive organisms. (Lorenz K. 2006) Prolonged exposure increased the bactericidal effect for most bacteria. (Christopher G. Jones. 1997)

2.1.4 Toxicity and side effects

Evidence of absorption of chlorhexidine by the gastrointestinal tract mucosa had not been found and it was believed it is non-existent. The hydrophilic nature of the cationic chlorhexidine molecule was the reason for failure of absorption by the mucosa. (Gupta et al, 2012)

As a result, all side effects of chlorhexidine were local reactions. These included the staining of teeth and the tongue observed after relatively short use (10-15 days) and taste alterations, particularly salty taste (Lindhe *et al*, 2008).

Desquamation of the epithelium could occur in some patients and soft tissue lacerations had been reported after prolonged exposure. Parotid salivary gland swelling had only occasionally been reported. Lastly, chlorhexidine may have had enhanced formation of supra-gingival calculus. Precipitation of salivary proteins was thought to attribute to formation and accelerating pellicle formation. However, when a chlorhexidine mouth wash was used appropriately it was generally considered to be safe (Gupta *et al*, 2012).

2.2 Alcohol

A large number of commercially available mouthwashes had a significant amount of alcohol as part of the ingredients. Addition of alcohol in the mouthwash presented with a few disadvantages.

Firstly, ingestion of such mouthwashes (especially by children) posed a risk for alcohol toxicity to some degree. (Herrera D. 2003)

Secondly, it can be argued that it may have contributed to carcinogenic effects since there is well documented link between tobacco smoking and alcohol consumption. It is suggested that the frequency of alcohol-containing mouthwash use might have increased the incidence of oral and pharyngeal cancer. (B.M. Eley. 1999)(Eleni G. 1995)

In alcoholics, the use of alcohol containing mouthwashes increased the risk of developing oropharyngeal cancer. (Winn et al, 1991) It was noted that, however, there was very weak evidence linking use of alcohol containing mouthwashes to carcinogenesis.

Thirdly, alcohol concentration of exhaled breath was also a part of concern to most patients as it was noted to change the reading on the breathalyser. This was, however found to be very insignificant in changing the readings. (Lorenz K, 2006)

Fourthly, the tissue irritating properties, which precluded its use in radiation or chemotherapy damaged epithelial surfaces (Ennibi *et al*, 2013). In patients under chemotherapy or radiation therapy, symptoms of mucositis -if present- were aggravated

Finally, Mouthwashes with alcohol had in some studies showed some degree of decreasing the tensile strength of the resin materials used for restoring teeth in dentistry. (Lorenz K, 2006) In various studies it was noted that composite resins gained weight by soaking in alcohol containing mouthwashes as compared to non-alcohol containing mouthwashes. This had been attributed to the absorption of alcohol by these composites, rendering it weak in structure. (E.L. Eley, 1999) In addition, the colour stability of composite resins was affected by prolonged use of mouthwashes that have alcohol as an ingredient.

2.3 Peridex and Paroex

2.3.1 Chemical Structure

Peridex™ (Chlorhexidine Gluconate 0.12%) Oral Rinse is constituted of 0.12% chlorhexidine gluconate (1, 1-hexamethylene bis [5-(p-chlorophenyl) biguanide] di-D-gluconate) in a base containing water, 11.6% alcohol, glycerin, PEG-40 sorbitan di-isostearate, flavor, sodium saccharin, and FD&C Blue No. 1.

Peridex is a near-neutral solution (pH range 5-7). Chlorhexidine gluconate is a salt of chlorhexidine and gluconic acid. (www.drugs.com)

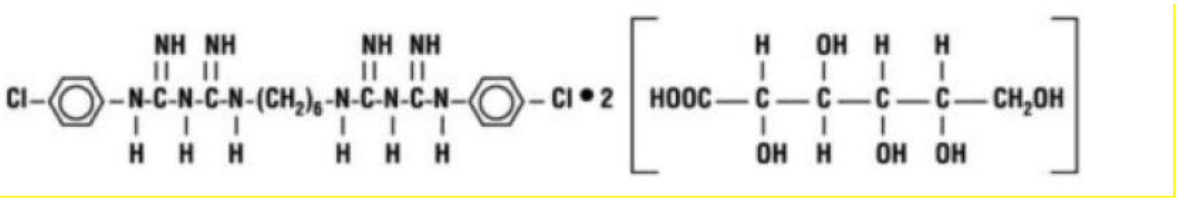


Figure 2: Shows the chemical structure of Peridex oral rinse (www.drugs.com)

Paroex™ is constituted of 0.12% chlorhexidine gluconate (1,1'-hexamethylene bis [5-(pchlorophenyl)biguanide] di-D-gluconate) in a base containing deionized water, propylene glycol, glycerin, polyoxyl 40 hydrogenated castor oil, mint flavor, potassium acesulfame, FD&C Red #40 and D&C Red #33. Paroex is a near-neutral solution (pH range 5-7). Chlorhexidine gluconate is a salt of chlorhexidine and gluconic acid. (www.drugs.com)

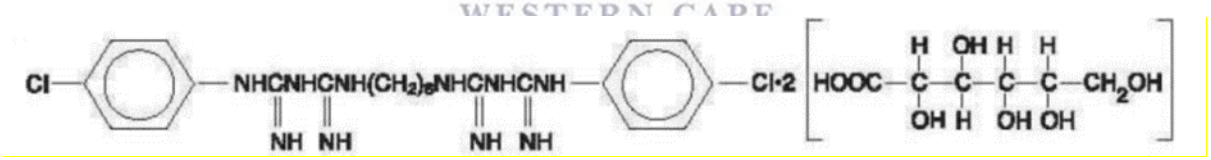


Figure 03: shows a molecular structure of Paroax (www.drugs.com)

2.3.2 Clinical Pharmacology

Peridex and Paroex provided an antimicrobial activity during oral rinsing. There was a lot of overlapping similarities between the two products given that they both had the same active ingredient, chlorhexidine gluconate, presented in an equal concentration.

The clinical significance of both solution's antimicrobial activities was not clear. General reduction of counts of bacteria during microbiological sampling of plaque had been observed in both aerobic and anaerobic microorganisms, ranging from 54 to 97 %. Six months clinical studies on both Peridex and Paroex did not yield any significant change in bacteria resistance, flourishing of opportunistic microorganisms or other adverse changes in the oral microbial ecosystem. The number of bacterial colonies had increased three months after the use of both solutions had been discontinued accompanied by return to baseline levels and resistance to plaque bacteria to chlorhexidine was levelled as to that at baseline.

Approximately 30% of the active ingredient, which was chlorhexidine gluconate, was retained by the soft tissues after rinsing. This was an advantage in reducing number of bacteria on the tissues as the drug was released slowly into the oral fluids. (www.drugs.com)

Chlorhexidine gluconate was poorly absorbed by the gastrointestinal tract. (Herrera *et al.*, 2003)

According to the product information, plasma levels of chlorhexidine gluconate reached a peak of 0.206mcg/g in humans 30 minutes after they ingested a 300mg dose of Peridex and Paroex. There was no detectable levels of chlorhexidine gluconate in the plasma of the subjects in the study 12 hours after administration of the compound. (www.drugs.com)

2.3.3 Indications and usage for Peridex and Paroax

Both Peridex and Paroex are indicated for use between dental visits in conjunction with professional program for the treatment and management of periodontal disease as classified by the WHO. However both the products had not been tested for use among patients presenting with necrotizing ulcerative

gingivitis (NUG) and necrotizing ulcerative periodontitis (NUP). (www.drugs.com)

2.3.4 Precautions and Adverse Reactions

General

Both solutions were found to cause staining of oral surfaces, such as tooth surfaces, restorations, and the dorsum of the tongue. However, not all patients experience a visually significant increase in tooth staining.

In clinical testing, approximately 56% of users exhibited a measurable increase in facial anterior stain, compared to 35% of control users after six months; 15% of users developed what was judged to be heavy stain, compared to 1% of control users after six months.

Unremoved plaque before the use of both solutions had a pronounced accumulation of staining. Stain can be removed from most tooth surfaces by conventional professional prophylactic techniques. (www.drugs.com)

Discretion had to be considered when prescribing to patients with anterior restorations with rough surfaces or margins. Some patients may experience an alteration in taste perception while undergoing treatment. (Arweiler NB., 2001)

Adverse Reactions

The most common side effects associated with chlorhexidine gluconate oral rinses were:

- increase in staining of teeth and other oral surfaces;
- increase in calculus formation; and
- alteration in taste perception,

- Oral irritation and local allergy-type symptoms have been reported.

The following oral mucosal side effects were reported during placebo-controlled adult clinical trials: aphthous ulcer, grossly obvious gingivitis, trauma, ulceration, erythema, desquamation, coated tongue, keratinization, geographic tongue.

The most frequently reported oral mucosal symptoms associated with Peridex and Paroex are stomatitis, gingivitis, glossitis, ulcer, dry mouth, hypesthesia, glossal edema, and paresthesia. Minor irritation and superficial desquamation of the oral mucosa were noted in patients who used Peridex. There have been cases of parotid gland swelling and inflammation of the salivary glands (sialadenitis) reported in patients who used Peridex. (Eleni G. *et al* 1995)

2.3.5 Dosage and Administration

The standardised recommended use was determined to be twice daily mouth rinsing for thirty seconds, in the morning and the evening after brushing of teeth. The usual dosage was 15ml (marked in cap) of undiluted solution. Patients were not to rinse with water or other mouthwashes, brush teeth or eat immediately after using either Peridex or Paroex. (www.drugs.com)

2.4 Bana-Zyme

The Bana-Zyme test has been used by many oral health clinicians in the quest for detection of enzymes in the tongue coatings, gingival crevicular fluids and plaque samples that hydrolyzes the synthetic peptide, Benzoyl-DL-arginine-B-napthylamide. It is a rapid test that can be done on the chair-side while the patient waits. . (N. Dhalla *et al*, 2015)

This enzyme is possessed by three anaerobic, periodontopathic species, i.e., Porphyromonas gingivalis, Treponema denticola and Tonnerella forsythia (formerly known as Bacteroides forsythus), that in vitro produce copious amounts of malodorous compounds. Plaque samples that were BANA-zyme positive invariably had one or more of these species present. (N. Dhalla *et al*, 2015)

It was widely accepted that periodontitis cases were largely made up of Gram negative anaerobes in the oral cavity. These microorganisms are responsible for producing numerous inflammatory biomarkers affecting the host directly. Plaque contribute to periodontal tissue breakdown and stimulation of mediator responses by the host, thus, contributing to the direct injury of the tissues. (N. Dhalla *et al*, 2015)

A strong relationship between a BANA positive reaction and elevated levels of plaque spirochetes had been demonstrated in clinical studies done by Loesche WJ in 1990. However, possibilities of other plaque species presence and host enzymes may have contributed to a positive result by the BANA-Zyme test kit.

BANA hydrolysis by plaque samples had the ability to be the marker of periodontal morbidity as assessed by probing depth measurements and by plaque proportions of microorganisms. (Loesche WJ, 1990)

When collecting a sample in the oral cavity, it was always a possibility that some organic material such as blood, saliva and ground clutter filter could contaminate the BANA strip but it had been demonstrated that neither of these products can hydrolyse BANA-zyme.

BANA positive test in sub-gingival plaque only occurs when there is elevated levels presence of spirochetes. This is because, a positive BANA test was designed to indicate more microbial load in a given localized area. (Loesche WJ, 1990)

BANA test did, however, come with a set of limitations. The major noted limitation was the fact that the kit cannot distinguish which of the three BANA positive species is detected in the plaque. It could only confirm to the clinician, presence of elevated anaerobic species.

Secondly, the BANA strips had a short shelf life and very technique sensitive. Thirdly, the unavailability of this kit in South Africa.

2.5 Chronic Periodontitis

Studies in periodontitis as a disease have come a long way in trying to understand how the condition develops and how it was sustained in the different oral environments amongst affected individuals. The disease could no longer be considered just as a simple bacterial infection leading to the destruction of the periodontium. Current look at periodontitis is seen as an entity represented by a collection of complex diseases involved purely as an interaction between the immune system and host inflammatory response, modifying environmental factors accompanied by the subgingival microbial colonization. (Bartold P et al, 2013)

It was important to note that colonization of microorganisms within the subgingival environment is generally observed as a commensal relationship between the host and the colony. This relationship was, however, in a state of homeostasis in normal healthy individuals.

A shift in this homeostasis, commensal relationship between the host and the microbiota towards a “pathologic state” could occur due to a variety of reasons to be discussed later. This shift was attributed to an overgrowth of microorganisms in subgingival environment leading to dysbiosis. (Darveau *et al*, 2012)

As illustrated in Figure 4, the microbial shift favouring pathogenic species required an environment that was conducive for microbial growth accompanied by a susceptible host for the disease to clinically manifest. Another way to look at periodontitis as a disease was considering that some patients may be rendered susceptible due to their genetic profiling and environmental factors as well. Such factors to consider can include examples like diabetes, chronic hyper-inflammatory conditions, polymorphisms in the gene for IL-1 and cigarette smoking. (Socransky *et al*, 1992)

Based on these considerations, it had become evident that the inflammatory nature of the disease gives rise to new opportunities for management of the disease and in this study, it helped in structuring the exclusion criteria.

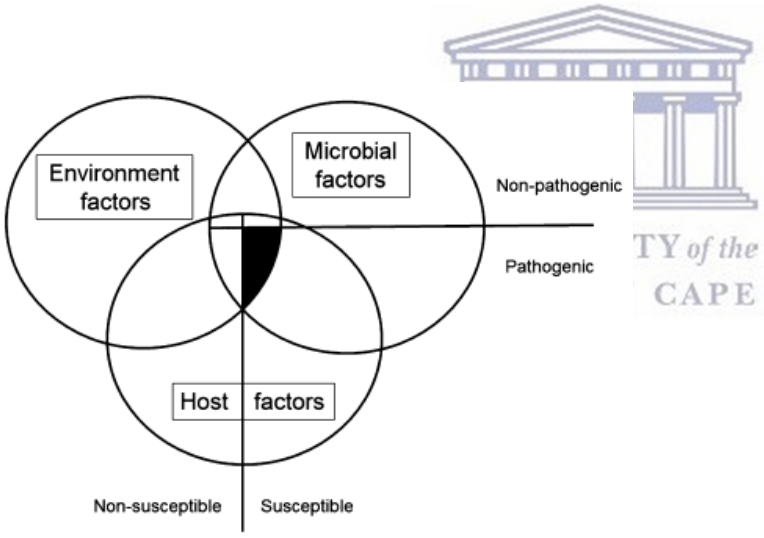


Figure 04: Periodontal risk – A patient centered paradigm. (Bartold P *et al*, 2013)

From the beginning of studies trying to understand the disease, researchers had come up with different hypotheses in defining the link between subgingival microbial colonization and pathogenesis initiation and progression. These hypothesis started from the "specific plaque hypothesis" demonstrated in the 1970s whereby the author described specific pathogenic microbes as being responsible for initiation of the disease. (Loë H *et al*, 1965)

Around the 1980s, researchers re-emerged the “Non-specific Plaque Hypothesis” that demonstrated that microbial mass in the subgingival microenvironment was responsible for initiating the disease. Following these hypotheses came the “Ecological Plaque Hypothesis” in which pathogenic periodontal microbes appear as a result of periodontal disease rather than being the cause of the disease. (Marsh PD, 2003)

In the more recent years the focus has shifted back to the bacteria as the central role to the disease development where it is proposed that specific microorganisms modulate the response by the host to impair the immune system and tip the homeostasis equilibrium of bacteria leading to microbial dysbiosis. This was then labelled as a “Keystone Pathogen Hypothesis”. (Darveau *et al*, 2012)

It was widely accepted that the shift in microbial load leading to dysbiosis of the subgingival microenvironment would lead to a response by the host and therefore, the disease was centred on degrees of inflammation on local tissues and may be sustained by secondary or primary chronic inflammatory conditions.

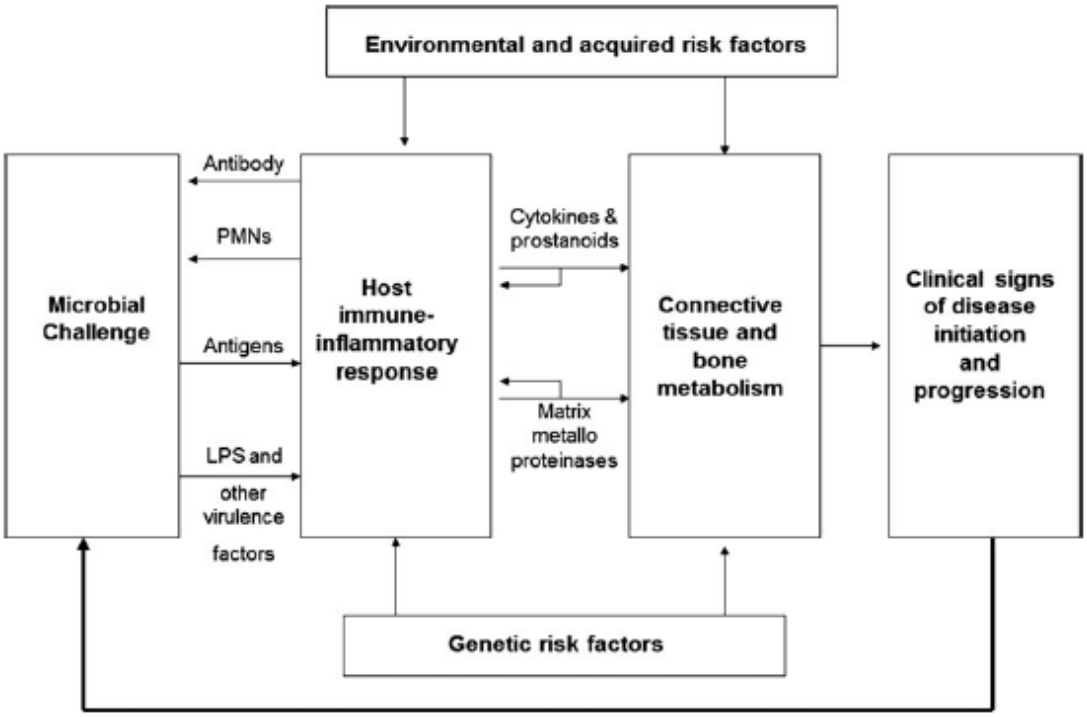


Figure 05: Proposal for the pathogenesis of periodontitis circa year 1997 (Bartold P *et al*, 2013)

2.5.1 Clinical findings of chronic periodontitis

In most cases, chronic periodontitis was treated at its advanced stages due to the fact that it took a long time (most times it took years) to develop and actually cause enough discomfort for a patient to look for professional care. The disease was asymptomatic at its early stages but as it progresses one could notice clinical changes that involve drifting of teeth, mobility of some teeth and eventually teeth could exfoliate spontaneously. Some other common clinical presentations included gross calculus accumulation, halitosis and even formation of abscesses that drains directly into the oral cavity through the periodontal pockets. (Lindhe, 2008)

Visual assessment, comprehensive radiographic examination and measurements of pocket depths were the main components that guided the clinician into getting to a clinical diagnosis. During a comprehensive clinical examination, pocket depths were measured at six sites around every tooth and the amount of

supragingival plaque or calculus, bleeding on probing, and exudate are recorded. All these records are used to come to a diagnosis and deciding the best suitable treatment while monitoring the improvement of the condition if there was any. (BL Pihlstrom *et al*, 2005)

As illustrated by Lindhe 2008, chronic periodontitis had a common presentation observed in almost all patients involved, including:

- the disease was generally seen in older individuals even though it may be present in younger patients
- destruction of the tissues was almost always directly proportional to factors including; oral hygiene, local predisposing factors and relevant risk factors for periodontal disease
- Although chronic periodontitis was initiated and sustained by microbial plaque, host factors determine the pathogenesis and the rate of progression of the disease.

Risk factors for periodontitis include oral microorganisms, genetic predisposition, smoking, nutrition, stress, diabetes and impaired host response. Systemic conditions including diabetes, osteoporosis and HIV/AIDS have been well identified to contribute in the rapid rate of progression of the disease even though not directly involved in the initiation of the disease. (Lindhe, 2008)

2.5.2 Prevention and treatment of chronic periodontitis

Emphasis on the prevention of chronic periodontitis was based on the management of risk factors linked to the cause of the disease and its pathogenesis. The widely accepted risk factor was the periodontal biofilm that forms on the teeth in the absence of effective oral hygiene.

However, various factors such as smoking, diabetes, ethnic origin, specific types of gram negative anaerobic bacteria in the periodontal biofilm, poor oral health education, irregular dental visits, genetic predisposition, increased age, male sex, stress, and depression had also been shown to be associated with loss of periodontal support, and are important considerations in the prevention and treatment of periodontitis. (BL Pihlstrom *et al,* 2005)

The biofilm was known to start forming after stopping all the oral hygiene procedures (including brushing and use of mouth washes) and this was all within 24 hours. Thorough tooth oral hygiene returns the gingiva to a healthy condition in about 7 days. (BL Pihlstrom, 2005; Lindhe *et al,* 2011)

Control of the periodontal biofilm with professionally administered oral hygiene can slow or stop periodontitis and tooth loss for many years. Tooth brushing, the use of dental floss and mouth washes to remove bacterial plaque from the teeth are the most common ways of disrupting the periodontal biofilm from teeth. Mouthwashes and dentifrices containing antibacterial drugs were used as adjuncts for controlling the biofilm. These combinations contain various biocides, surfactants, polymers, or other components that could reduce the biofilm and were generally not associated with the emergence of a resistant microbiota. (Lindhe *et al*, 2011)

Therefore, the treatment for periodontitis was aimed at establishing general periodontal health, arrest the progression of disease, prevent recurrence, and preserve the dentition in a state of health, comfort, function and pleasing aesthetics. This goal could be accomplished by various non-surgical and surgical therapies, depending on the specific treatment objective upon reaching a confirmed diagnosis. (Lindhe *et al,* 2011)

CHAPTER 3

AIMS AND OBJECTIVES

3.1 Aims

To assess the efficacy of alcohol-free chlorhexidine mouth wash in comparison to alcohol containing chlorhexidine mouth wash.

3.2 Objectives

1. Determine the pre-operative clinical parameters in patients with chronic periodontitis.
2. Determine the pre-operative microbial load before treatment of chronic periodontitis.
3. Determine the post-operative clinical parameters in patients with chronic periodontitis.
4. Determine the post-operative microbial load after treatment of chronic periodontitis.

   Compare the efficacy of alcohol containing chlorhexidine and alcohol free chlorhexidine on the clinical parameters and microbial load in patients with chronic periodontitis.

CHAPTER 4

METHODOLOGY

4.1 Study design

A double blinded randomised control trial was the study design employed. All subjects were, at the beginning of the study, diagnosed with active chronic periodontitis. (G. Armitage, 2004) The patients were randomly assigned, using Microsoft Excel Randomize tool, to either the test group (chlorhexidine without alcohol) and or control group (chlorhexidine with alcohol).

Unmarked 100ml brown containers (Figure 04) were used to dispense the two mouthwashes with 25 bottles filled with Paroex® and the remaining 25 filled with Peridex®. This process was done by an assistant who was not to be involved in the actual screening, treating and assigning mouthwash to the patient and, therefore, she was blind from the actual study and the clinician was also blinded since there was no knowledge of the container content. The containers were assigned only a number and on record the number had the information of what mouthwash is being used. This record was kept safe by the assistant and the clinician had no access to the information until completion of the study before data was analysed. At the clinic the clinician only received mouthwash containers with numbers and had to log the bottle number given to the patient selected.

At the completion of data collection the patients had the number similar to the container previously given and the names were then assigned to the mouthwash given, revealing which patients were in the control group and those who were in the test group.

A BANA test was conducted before periodontal treatment. Periodontal treatment will include a full mouth mechanical debridement and adjunct use of a mouthwash. They were shown a standardised (Modified Bass Method of tooth brushing) and given mouthwash to use twice daily for two weeks. All patients were then recalled in 6 weeks for re-evaluation.

BANA testing was also done first at the 6 week recall visit before all the parameters were measured.

Figure 06: Brown 100ml unmarked containers used to dispense the mouthwash

4.2 Study sample

Using the CheckMarker® sample size calculator it was determined that in order to come up with result confidence of around 95% and a marginal error less than 2%, a sample size of more than 49 participants was needed. Therefore, fifty three patient fitting the inclusion and exclusion criteria were invited to participate in the study

The study was done at the University of the Western Cape on patients presenting at the Oral Medicine and Periodontology department. It involved 50 patients over the age of 35 years and having an active chronic periodontitis.

4.2.1 Inclusion criteria

The study involved participants presenting with chronic periodontitis diagnosed at the department of Oral Medicine and Periodontology at the University of the Western Cape. Patients had to be dentate or partially edentulous and over the age of 35 years.

4.2.2 Exclusion criteria

- Patients on chemotherapy and/or radiation therapy
- Pregnant or lactating individuals
- Patients on antibiotic treatment 3 months prior to the study
- Cigarette smokers that use more than 15 cigarettes a day
- Patients that have underwent periodontal treatment in the past nine months prior to the study.
- Persons with active carious lesions
- Patients presenting with uncontrolled diabetes

4.3 Materials

Two commercially available mouthwashes were selected and BANA-zyme test kit was used.

4.3.1 Paroex

Figure 07: Paroex mouthwash. (0% Alcohol)

Paroex is an oral rinse containing 0.12% chlorhexidine gluconate in a base containing deionized water, propylene glycol, glycerin, polyoxyl 40 hydrogenated castor oil, mint flavour, potassium acesulfame, FD&C Red #40 and D&C Red #33. Paroex is a near-neutral solution (pH range 5-7).

4.3.2 Peridex

Peridex™ (Chlorhexidine Gluconate 0.12%) Oral Rinse is an oral rinse containing 0.12% chlorhexidine gluconate in a base containing water, 11.6% alcohol, glycerin, PEG-40 sorbitan diisostearate, flavour, sodium saccharin, and FD&C Blue No. 1. Peridex is a near-neutral solution (pH range 5-7).

Figure 08: Peridex mouthwash (11.6% alcohol)

4.3.3 BANA-zyme Kit

The BANA-Zyme Test is a rapid, 5-minute, Chair-side Test for the detection of an enzyme(s) in tongue coatings and plaque samples that hydrolyzes the synthetic peptide, Benzoyl-DL-arginine-B-napthylamide (BANA). This enzyme is possessed by three anaerobic, periodontopathic species, i.e., Porphyromonas gingivalis, Treponema denticola and Tonnerella forsythia (formerly known as Bacteroides forsythus), that in vitro produce copious amount sofmalodorous compounds. Plaque samples that are BANA-zyme positive invariably have one or more of these species present. Tongue -samples may have these and other malodorous bacterial species present.

Figure 09: A represents the BANA-zyme test machine and B shows a Bana-strip

2.3.4 Toothpaste and Toothbrush

Figure 10: Toothpaste and toothbrushes

4.4 Clinical Parameters

Probing pocket depth (PPD):

Probing pocket depths were measured using a graduated periodontal probe to the nearest millimetre. Measurements were taken from the base of the pocket to the gingival margin. (Lindhe *et al*, 2008)

Recession (RC):

Miller's classification for recession in 1985 is still a standard in recording levels of RC. A periodontal probe was used to measure RC from the cemento-enamel junction to the margin of the gingiva. (Lindhe *et al*, 2008)

Bleeding on probing (BOP):

A percentage bleeding score was recorded from the sum total of the teeth that were probed and bled, and then compared to the total number of teeth present to obtain a bleeding percentage score.

Clinical attachment level (CAL):

Attachment loss was measured with the use of a periodontal probe. The distance from the CEJ or the restoration boarder to the pocket base was recorded. (Lindhe *et al*, 2008)

Plaque index – (PI):

Silness & Löe in 1964 devised a scoring system by which the level of plaque can be categorised.

Index values – 0: No plaque visibility, 1: Plaque only visible on probing, 2: Plaque easily seen with naked eye, 3: gross plaque accumulation. (Lindhe *et al*, 2008)

Tooth mobility:

Mobility was measured according on the criteria stipulated by Miller in 1950. (Lindhe *et al*, 2008)

Degree 0: movement confined to physiological limitations. Between 0.1 to 0.3mm horizontally

Degree 1: tooth mobility of not more than 1mm in a horizontally.

Degree 2: Greater mobility of the tooth of more than 1mm in a horizontal movements.

Degree 3: Gross movement of the tooth in a horizontal and vertical motions (Linde *et al*, 2008).

These clinical parameters were recorded at the initial visit and repeated at the follow up visit after 6 weeks.

Oral health instructions were given to all participants. This included brushing all teeth in a circular motion with the use of soft bristle brushes for two minutes using provided standard toothpaste. (Figure 09) This was to be repeated twice daily (morning and night. Flossing was to be performed once daily.

Participants were to use only the prescribed mouthwash in order to maintain a constant clinical outcome. Verbal instructions were given in conjunction with written instruction. Written instructions were presented in two different languages commonly used by people in the area.

4.5 Ethical Considerations

Ethical approval was obtained from the University of the Western Cape, Dental Faculty. Patients consented to participation after reading an information sheet

and then signed for consent. The voluntary nature of the participation in this study was clearly explained to the participants, along with any potential advantage, disadvantage, complaints that might result due to taking part in this study. The researcher's contact details were available to all participants for further information about the study or its outcome. Participants could withdraw from their voluntary participation, with no prejudice, at any time during the study.

4.6 Conflict of interest

There was no conflict of interest in the proposed study. The study was self-funded and the researcher received no remuneration from any of the companies who sell the mouthwashes.

4.7 Data collection and analysis

Data was captured on a Microsoft Excel spreadsheet. Sample size, mean value and standard deviation was assessed by a statistician and the clinical data recorded was compared before and after the treatment.

The number allocated to each mouthwash container was merged with the patient information to reveal the two groups that were treated after the recall data was collected. The Wilcoxon Signed Rank test was used to test the difference between the pre-post pair per clinical indicator and Bana-Zyme tests. The interrelationships between the clinical indicators (post measurements) were explored with a categorical Principal Component Analysis (CATPCA).

CHAPTER 5

RESULTS

A total of 53 patients (23 females and 30 males) were treated in the study and re-evaluated 6 weeks post treatment. Data was collected on both visits for analysis. There were 25 patients who received Paroex and 28 received Peridex.

The Wilcoxon Signed Rank test was used to test the difference between the pre-post pair per clinical indicator and Bana-Zyme tests. The differences between before and after treatment per indictor are significant at $P<0.001$ for respectively Paroex (Table 2) and Peridex (Table 3). These values demonstrate the difference between the clinical parameters taken before the treatment and six weeks post treatment. The higher the sum of ranks value means the difference between initial parameter score and scores taken on re-evaluation of patients after treatment. A sum of ranks score of zero would have been an indicator of no difference between the initial parameter and post-treatment scores.

Therefore table 2 and 3 demonstrate that both solutions used in this study do have an impact on improving the oral health of the patients and this was achieved in both groups.

High value of the sum of ranks in plaque index, gingival index, pocket depth, clinical attachment and the Bana-zyme test indicated good improvement of periodontal status of the patients using both alcohol containing mouth wash and non-alcohol containing mouth wash. However, even though there was improvement in all seven tested parameters, tooth mobility and gingival recession showed mild improvement or remained the same.

Treatment					
		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Paroex	25	47.2	47.2	47.2
	Peridex	28	52.8	52.8	100.0
	Total	53	100.0	100.0	

Table 1: Distribution of patients treated in the study

Ranks				
		N	Mean Rank	Sum of Ranks
PI_A - PI	Negative Ranks	25[a]	13.00	325.00
	Positive Ranks	0[b]	.00	.00
	Ties	0[c]		
	Total	25		
GI_A - GI	Negative Ranks	24[d]	12.50	300.00
	Positive Ranks	0[e]	.00	.00
	Ties	1[f]		
	Total	25		
PD_A - PD	Negative Ranks	24[g]	12.50	300.00
	Positive Ranks	0[h]	.00	.00
	Ties	1[i]		
	Total	25		
Recess_A - Recess	Negative Ranks	10[j]	5.50	55.00
	Positive Ranks	0[k]	.00	.00
	Ties	15[l]		
	Total	25		
Clin_attac_A - Clin_attach	Negative Ranks	24[m]	12.50	300.00
	Positive Ranks	0[n]	.00	.00
	Ties	1[o]		
	Total	25		
Mob_A - Mob	Negative Ranks	15[p]	8.00	120.00
	Positive Ranks	0[q]	.00	.00
	Ties	10[r]		
	Total	25		
Bana-Zyne	Negative Ranks	22[s]	12.00	264.00
	Positive Ranks	1[t]	12.00	12.00
	Ties	2[u]		
	Total	25		

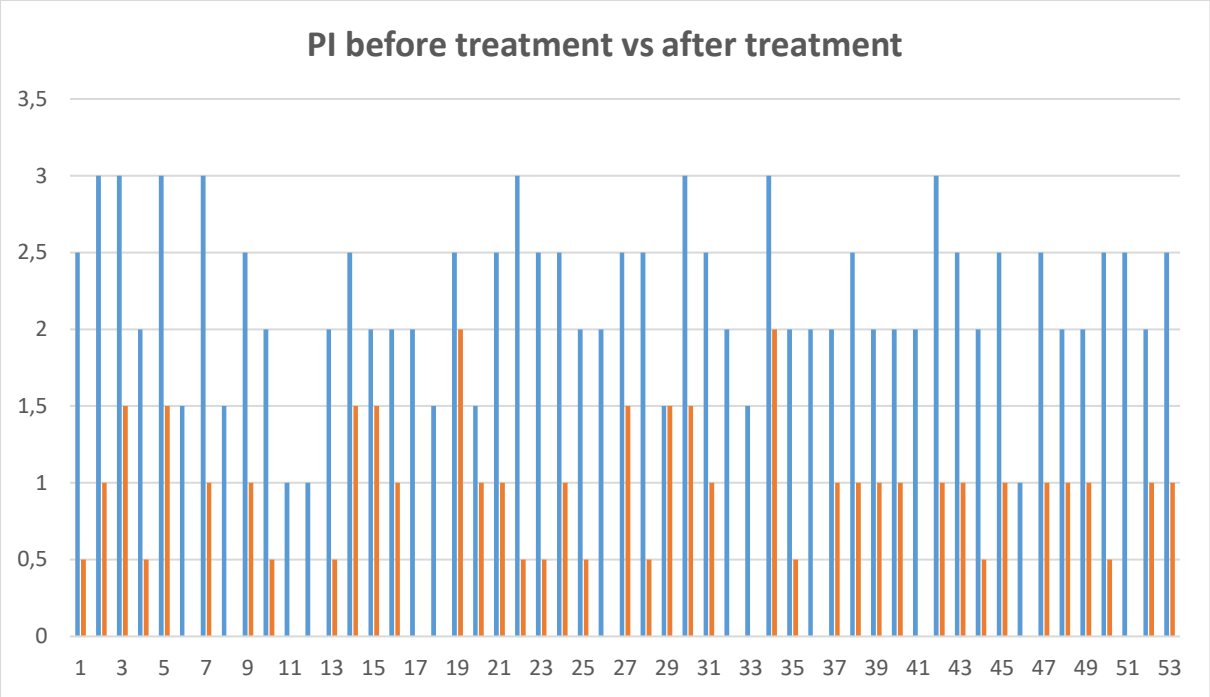
Table 2: Distribution of results based on Paroex.

Ranks				
		N	Mean Rank	Sum of Ranks
PI_A - PI	Negative Ranks	27[a]	14.00	378.00
	Positive Ranks	0[b]	.00	.00
	Ties	1[c]		
	Total	28		
GI_A - GI	Negative Ranks	20[d]	10.50	210.00
	Positive Ranks	0[e]	.00	.00
	Ties	8[f]		
	Total	28		
PD_A - PD	Negative Ranks	27[g]	14.00	378.00
	Positive Ranks	0[h]	.00	.00
	Ties	1[i]		
	Total	28		
Recess_A - Recess	Negative Ranks	12[j]	7.92	95.00
	Positive Ranks	2[k]	5.00	10.00
	Ties	14[l]		
	Total	28		
Clin_attac_A - Clin_attach	Negative Ranks	26[m]	13.50	351.00
	Positive Ranks	0[n]	.00	.00
	Ties	2[o]		
	Total	28		
Mob_A - Mob	Negative Ranks	7[p]	4.00	28.00
	Positive Ranks	0[q]	.00	.00
	Ties	21[r]		
	Total	28		
BZyme_A - BZyme	Negative Ranks	25[s]	13.00	325.00
	Positive Ranks	0[t]	.00	.00
	Ties	3[u]		
	Total	28		

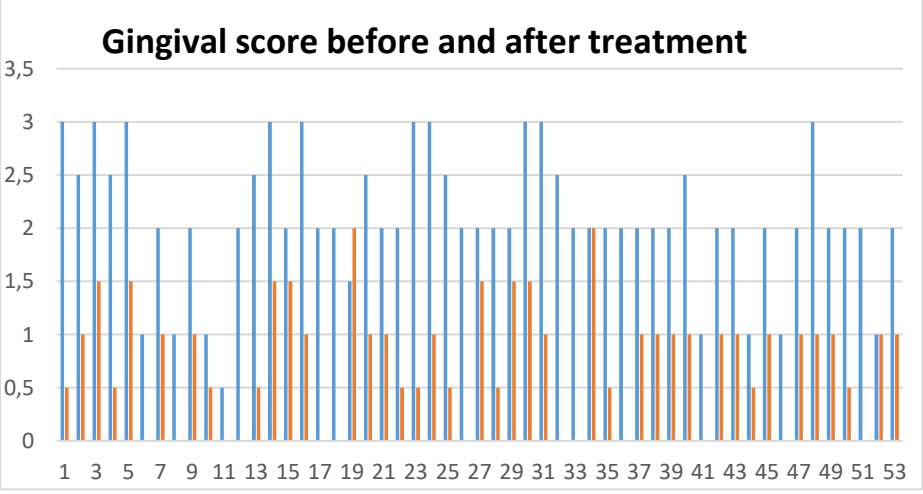
Table 3: Distribution of results based on Peridex

Next tables 3 to 9 present the frequencies of the scores of the indicators per Mouth Wash based on the post measurement values in the clinical parameters.

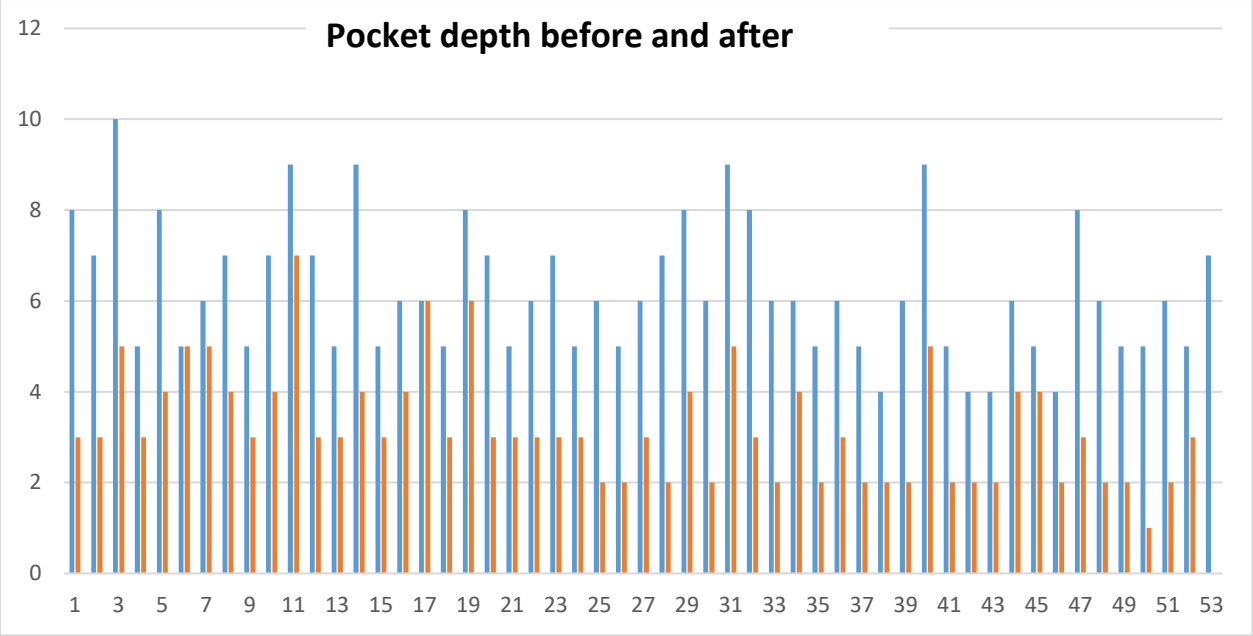
5.1 Values comparing outcomes before and after treatment.



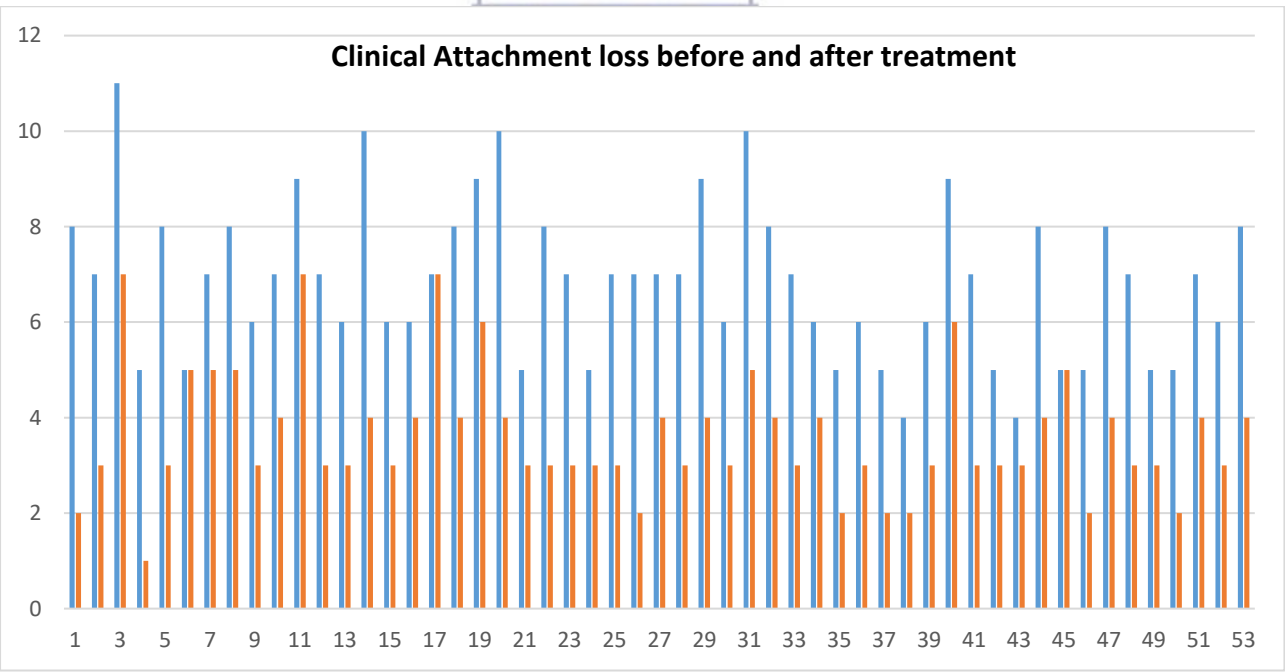
Graph 1: Represents the Plaque index before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)



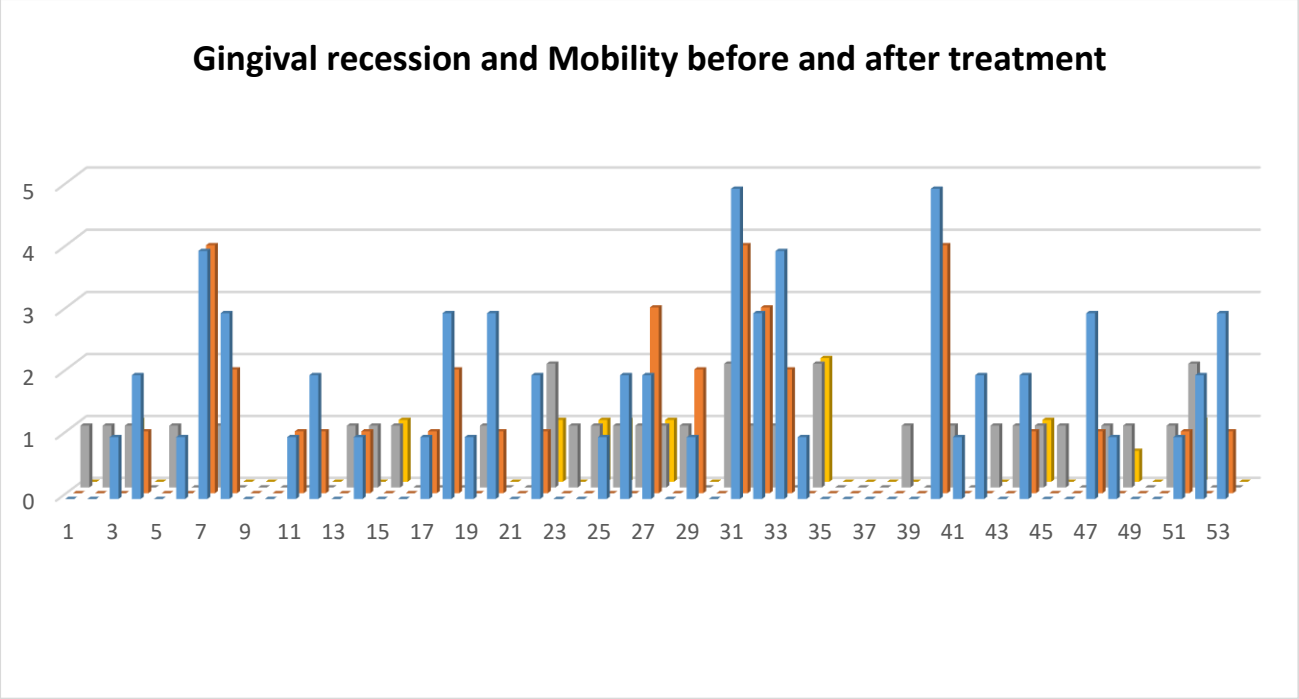
Graph 2: Represents the gingival score before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)



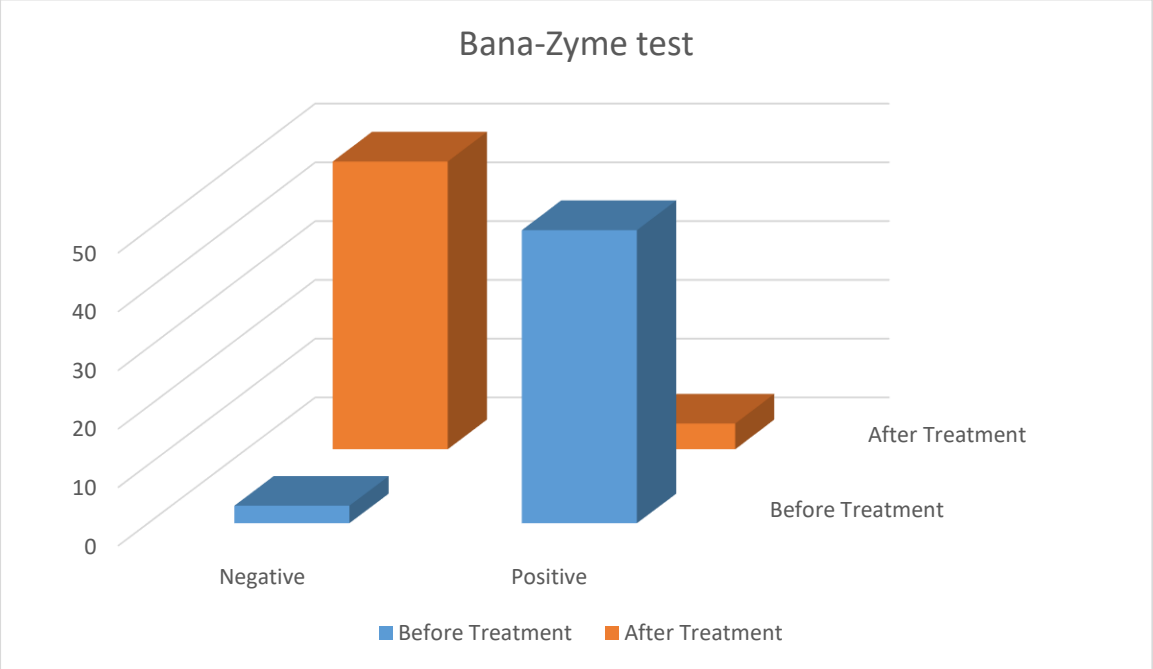
Graph 3: Represents pocket depths before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)



Graph 4: Represents clinical attachment loss before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue is representative of values before treatment)



Graph 5: Represents teeth mobility & gingival recession before treatment and 6 weeks follow-up after mechanical debridement and adjunct use of mouthwash (Blue: Recession before treatment, Red: Recession after treatment, Grey: Mobility before treatment, Yellow: After treatment)



Graph 6: Represents average Bana-zyme hits on negative and positive values before and after treatment of periodontitis.

PI_A				
PI value	Frequency	Percent	Valid Percent	Cumulative Percent
.00	13	24.5	24.5	24.5
.50	11	20.8	20.8	45.3
1.00	20	37.7	37.7	83.0
1.50	7	13.2	13.2	96.2
2.00	2	3.8	3.8	100.0
Total	53	100.0	100.0	

Table 4: The frequency score for the plaque index 6 weeks after treatment

GI_A				
	Frequency	Percent	Valid Percent	Cumulative Percent
.00	12	22.6	22.6	22.6
.50	11	20.8	20.8	43.4
1.00	17	32.1	32.1	75.5
1.50	6	11.3	11.3	86.8
2.00	7	13.2	13.2	100.0
Total	53	100.0	100.0	

Table 5: The frequency score for gingival index 6 weeks after treatment

PD_A				
	Frequency	Percent	Valid Percent	Cumulative Percent
1	1	1.9	1.9	1.9
2	16	30.2	30.2	32.1
3	18	34.0	34.0	66.0
4	10	18.9	18.9	84.9
5	5	9.4	9.4	94.3
6	2	3.8	3.8	98.1
7	1	1.9	1.9	100.0
Total	53	100.0	100.0	

Table 6: The frequency score for pocket depth 6 weeks after treatment

Recession _A				
	Frequency	Percent	Valid Percent	Cumulative Percent
0	33	62.3	62.3	62.3
1	11	20.8	20.8	83.0
2	4	7.5	7.5	90.6
3	2	3.8	3.8	94.3
4	3	5.7	5.7	100.0
Total	53	100.0	100.0	

Table 7: The frequency score for gingival recession 6 weeks after treatment

Mobility _A				
	Frequency	Percent	Valid Percent	Cumulative Percent
.00	41	77.4	77.4	77.4
.50	1	1.9	1.9	79.2
1.00	10	18.9	18.9	98.1
2.00	1	1.9	1.9	100.0
Total	53	100.0	100.0	

Table 8: The frequency score for tooth mobility 6 weeks after treatment

Bana Zyme _A					
		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	negative	49	92.5	92.5	92.5
	positive	4	7.5	7.5	100.0
	Total	53	100.0	100.0	

Table 9: Demonstrate the frequency score for Bana-Zyme test 6 weeks after treatment

The interrelationships between the clinical indicators (post measurements) are explored with a categorical Principal Component Analysis (CATPCA).

Tooth mobility appears a dimension on its own but the amount of variance it explains is less than its own variance. A two dimensional solution with the

remaining 5 clinical indicators explained 89% of the total variance with Eigenvalues D1= 2.53; D2=1.90 and revealed two clusters. Plaque Index and GI appeared to measure aspects that differ from aspect addressed by recession, pocket depth and clinical attachment. A cluster suggested a communality from elements that had, despite their association, unique contributions.

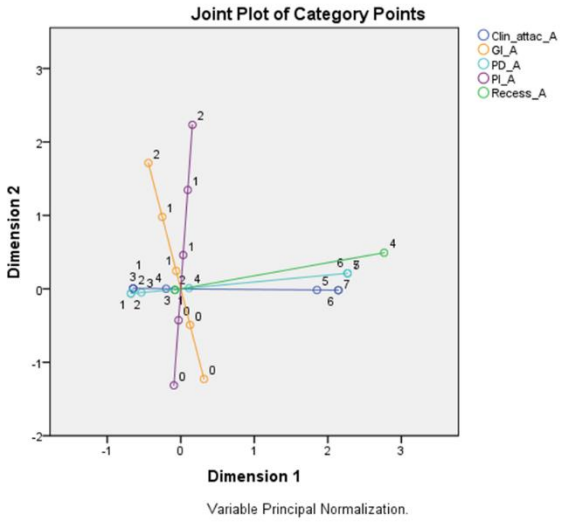


Figure 11: Illustrates the two dimensional variation in the plaque and gingival indices in relation to the other used parameters.

One approach would be to represent the underlying communality as a single scale variable: A one dimensional solution with Pi and GI explains 83%.

Adding the treatment variable, revealed some ordaining suggesting that Peridex is associated with slightly higher values than Paroex. The projection in one dimension, however, explains 62% and thus was unable to allocate 38% of variance.

Exploring Pocket Depth and Clinical attachment in a one dimensional analysis with the treatment variable explained 64% of the total variance. It also showed

that Pocket and Clinical attachment were unable to discriminate between both mouthwashes.

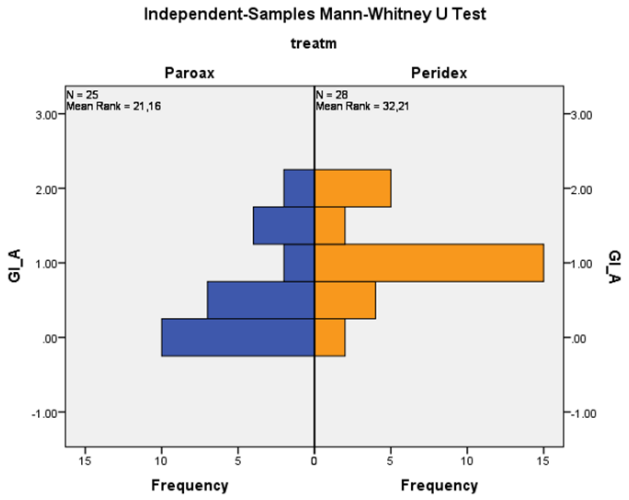
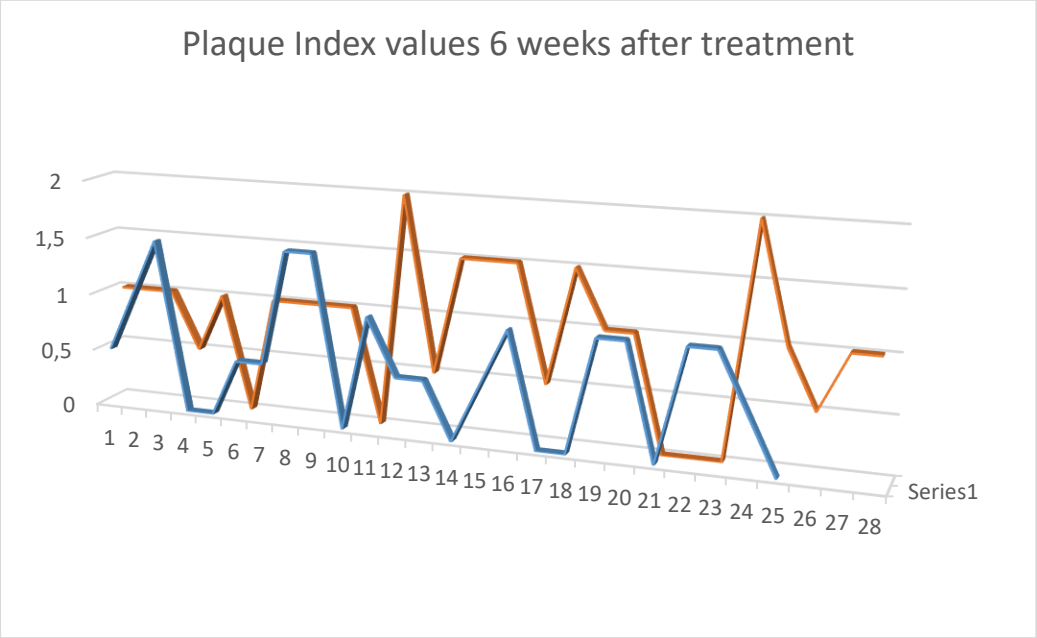


Figure 12: Demonstrates a slightly higher value after treatment for gingival index as an isolated parameter.

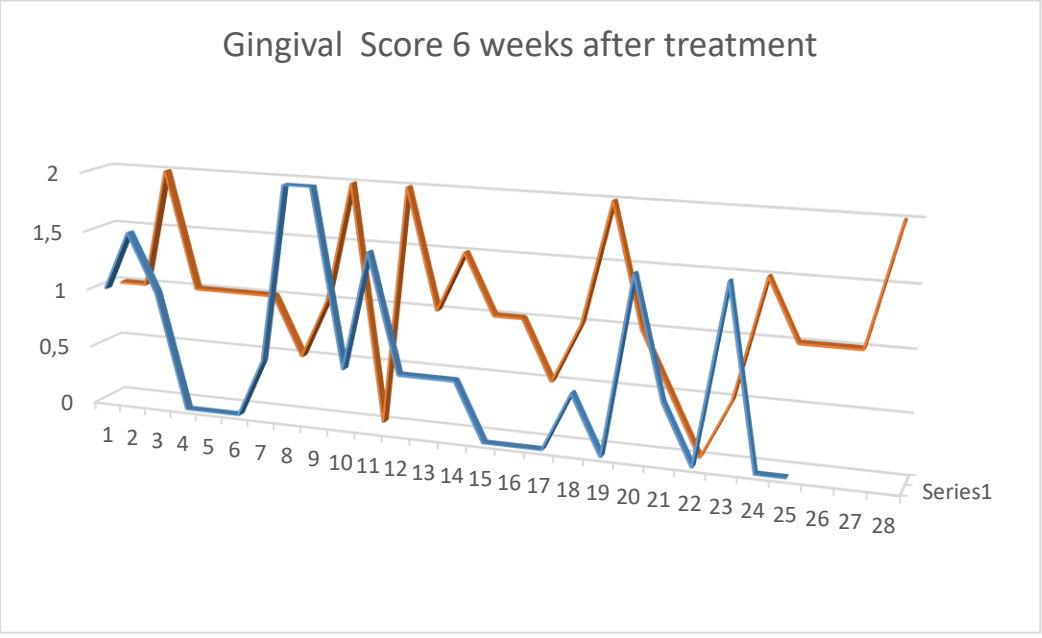
Although analysis did suggest that combined clinical indicators have common underlying factors except for PI and GI, the remaining indicators appear not affected by the treatment.

Singled out, only the GI indicator appeared to discriminate between the treatment: Paroex and Peridex significantly: Mann-Whitney U = 496, n1 = 25 n=28, P < 0.01 two-tailed. (The power of this results is small: 1-β=0.40).

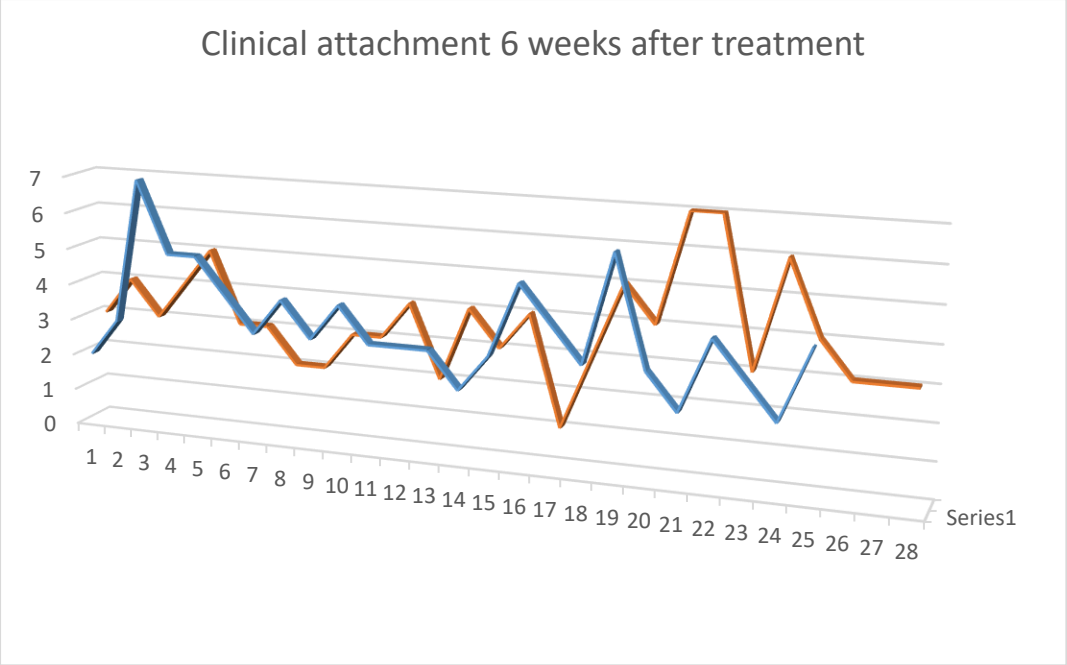
5.2 Values comparing the outcomes of the two test groups



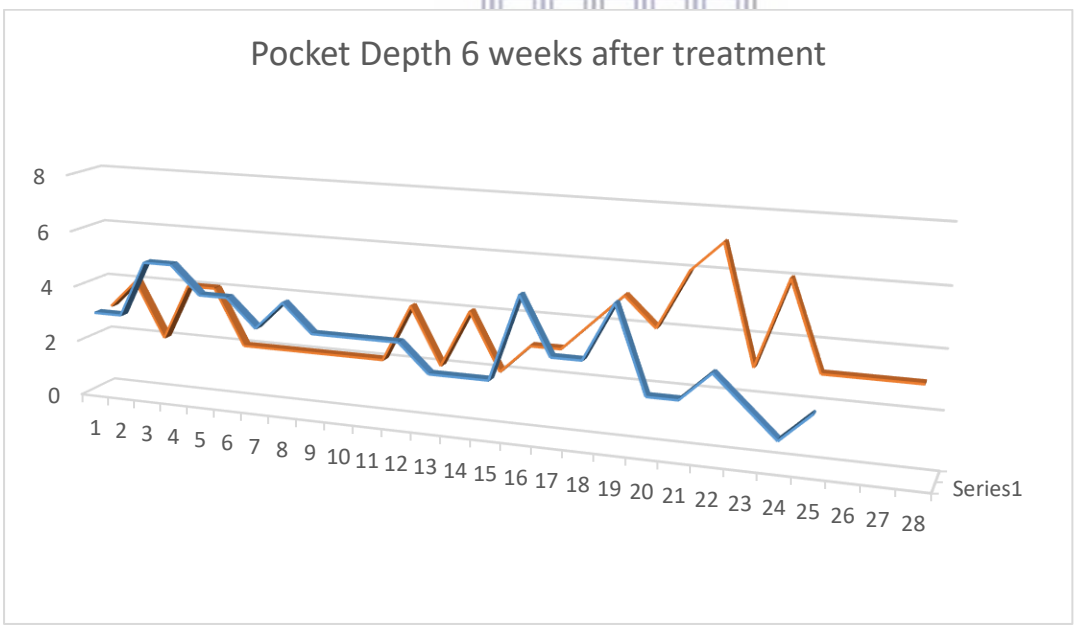
Graph 7: Representation of the plaque index between the two groups. Paroex shows greater reduction in the plaque index when compared to Peridex. (Blue is Paroex and Brown is Peridex)



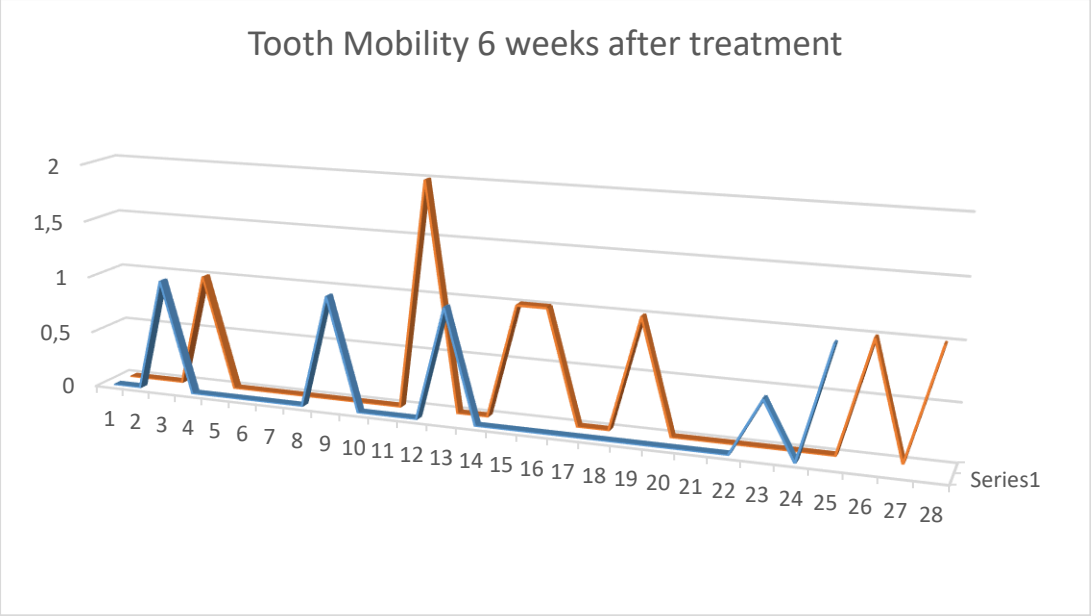
Graph 8: Representation of the gingival score between the two groups. Paroex shows greater reduction of this parameter when compared to Peridex. (Blue is Paroex and Brown is Peridex)



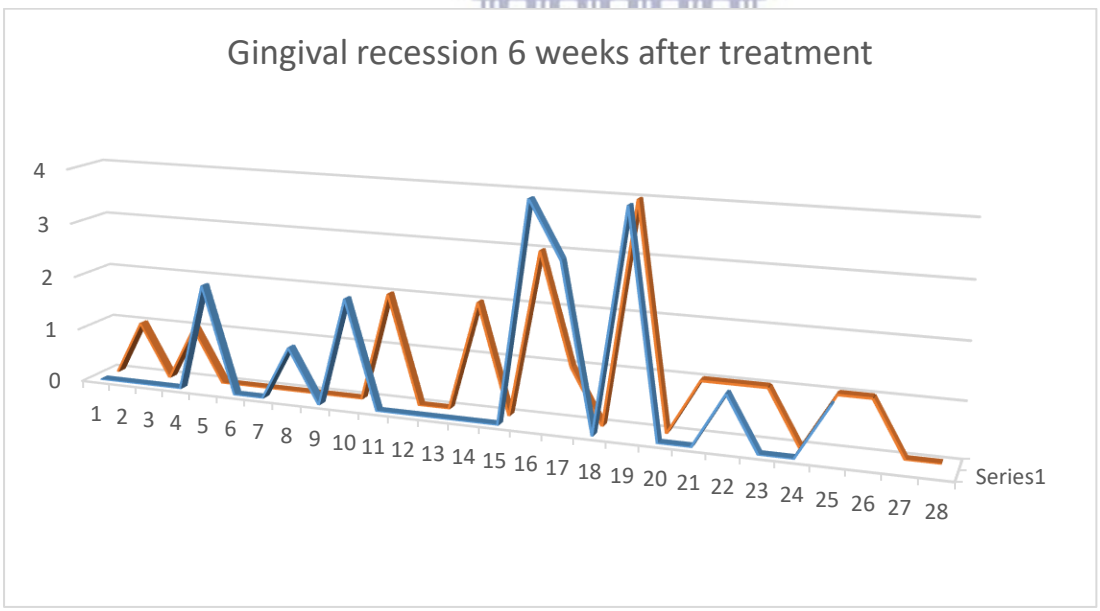
Graph 9: Represents the clinical attachment 6 weeks after treatment. Paroex show greater reduction in the clinical attachment loss when compared to Peridex. (Blue is Paroex and Brown is Peridex)



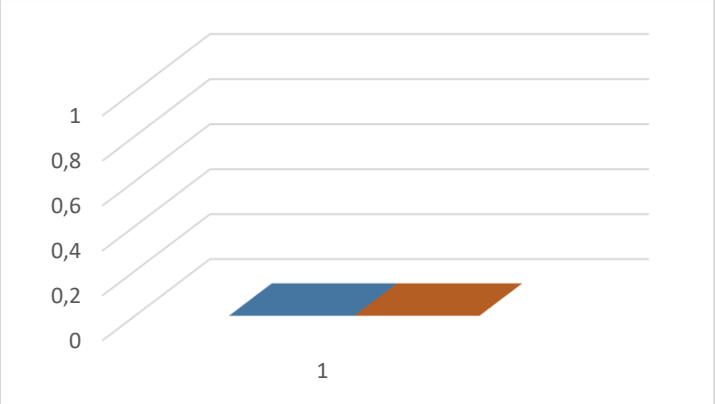
Graph 10: Represents the values of pocket depth 6 weeks after treatment. There is greater reduction in periodontal pockets in the group that received Paroex when compared with Peridex. (Blue is Paroex and Brown is Peridex)



Graph 11: Represents the average values of teeth mobility between the two test groups. Paroex group shows more reduction in the mobility of teeth following treatment when compared to Peridex. (Blue is Paroex and Brown is Peridex)



Graph 12: Represents recession in two test groups. No notable improvement of gingival recession was recorded in both test groups. . (Blue is Paroex and Brown is Peridex)



Graph 13: Represents the values of the BANA-Zyme test. The microbial load was reduced in both test groups equally. (Blue is Paroex and Brown is Peridex)

CHAPTER 6

DISCUSSION

The effect of both mouthwash preparations was investigated in a pre-post design. Subsequent analysis was conducted to test the difference between both mouthwashes with the post-data (after treatment).

Seven variables were used to measure the effects and differences between the liquids: Six clinical indicators and one variable testing the presence of enzymes, produced by microbes, with the Bana-Zyme test.

The analysis of a treatment with multi dependent variables requires insight in the interdependency of these variables. It is evident that in a clinical approach a total impression is based on the presence of the indicators (variables). In essence even though there were seven variables tested, a clinician would not need to confirm all seven to come to a diagnosis of chronic periodontitis.

However, there are parameters that are a constant in the presence of active chronic periodontitis including gingival index, plaque index, pocket depth and clinical attachment loss. For the purpose of the study all seven parameters were tested and analysed and this may, to a certain extent, give results that were difficult to interpret. The question would then be, should one look at all parameters and look at chronic periodontitis based on all or specify which parameters are constant and focus purely on those.

6.1 Gingival Index, Bleeding on Probing and Plaque index

The outcomes of the collected data demonstrated that both chlorhexidine with and without alcohol mouth-rinse groups to had exponential reduction in gingival index and plaque index after a full-mouth-disinfection and following up on recall consultations. The result are confirming the findings of a clinical systemic evaluation where two mouthwashes where compared in a clinical setup performed in a 25 year period. In this study it was found that chlorhexidine as a major ingredient in mouthwash solutions was effective in controlling plaque and periodontal disease following mechanical oral hygiene procedures. (Van strydonck *et al*. 2012)

Jones C G in 1997 mentioned chlorhexidine as the most investigated chemical agent in control of periodontal disease and that it still remained a gold standard antimicrobial agent. The well-known mode of action involving the rapid attraction chlorhexidine molecules that are positively charged to the negatively charged microbial cells membrane resulting in rupture and leakage of intracellular content and reduction of overall microbial load in the oral cavity. In addition, chlorhexidine binds to surfaces within the oral cavity including teeth, oral mucosa and tongue and then slowly released to continue its action even long after rinsing. This maintain an oral environment where the antimicrobial activity last for up to several hours depending on factors such as rinsing time, dosage and pH levels.

After 6 weeks of repeated rinsing, as per the prescribed oral hygiene instructions, salivary chlorhexidine levels were higher in both groups.

Chlorhexidine as a major ingredient in mouthwash formulation with and without alcohol has long been known to exert therapeutic effects against plaque and gingivitis. (Leyes Borrajo *et al*, 2002) In a randomised, double blind study,

done by Jenkins S et al. 1993, a 0.12% chlorhexidine rinse was used after mechanical debridement, resulted in significant improvements of periodontal disease. The results from this study has shown great improvement of the gingival and plaque indices. (Table 4 and 5)

Chaves ES *et al* in 1993 explored the relationship between bleeding on probing and gingival index bleeding as clinical parameters of gingival inflammation. Bleeding of the gingiva provides a very important clinical sign of inflammation that has been used as a key parameter in evaluation of gingival and periodontal status. Visual signs of inflammation in the gingival tissue accompanied by bleeding upon gingival stimulation are considered to be sensitive indicators of early gingivitis, thus gingival indices based on bleeding have been emphasized. Their paper concluded that GI bleeding and BOP are not interchangeable measures of bleeding tendency. However, the two parameters appear to evaluate distinct inflammatory conditions of the periodontium. Reduction in these parameters, therefore, indicates improvement of the periodontal status of patients as found to be the case in both mouthwash solutions with and without alcohol.

6.2 Gingival recession

Thrombelli L, 1998 describe gingival recession as the displacement of the gingival margins apical to the CEJ. Histologically, the destruction of the gingiva due to various factors is associated with the loss of teeth supporting periodontal structures including the periodontal fibers and the alveolar bone.

A review article by Tugnait A *et al* in 2001 mentioned that the mechanism by which gingival recession occur is still unclear even though the etiological factors have been identified. The paper further classify the etiological factors

into two groups: recession associated with non-pathologic alveolar bone loss and that associated with pathologic bone loss.

Recession associated with non-pathologic alveolar bone loss include: anatomy, tooth position, orthodontic tooth movement, mechanical trauma and local plaque retention factors such as poor restoration designs.

Recession associated with pathologic alveolar bone loss include: periodontal disease and smoking.

For the purpose of testing the two mouthwash solution, the inclusion criteria involved only the patients whereby the gingival recession was primarily thought to have been caused by the progression of the chronic periodontitis status of individuals in the study

Tugnait A *et al*, 2001 further postulate that the loss of alveolar bone is mostly attributed to the presence of ant form of periodontal disease. Therefore, loss of bone occur along with the loss of underlying connective tissue attachment followed by apical migration of the junctional epithelium. The recession associated with periodontal disease is may be observed affecting all the surfaces of the teeth together with loss of interdental papilla. As the condition progresses the patient will start seeking professional help with the main complaint being pain, poor aesthetics and the beginning of the mobility in the involved teeth. (Tugnait A. *et al*, 2001)

Measures to correct gingival recession defects will include correction and improvement of the oral hygiene, treatment of periodontal disease cessation of smoking and traumatic habits. (Thrombelli L, 1998) The use of both chlorhexidine solutions with and without alcohol did not show much improvement in the presence of recession because in most cases the condition will need surgical intervention to cover the defect. However, surgery is

determined or decided upon once the periodontal disease has been observed to be inactive. (Tugnait A. *et al*, 2001)

6.3 Pocket Depth, Clinical Attachment Loss and Teeth Mobility

Mdala *at al*, in 2014 tried comparing clinical attachment level and pocket depth for predicting periodontal disease progression in healthy sites of patients with chronic periodontitis with the aim to understand how degeneration of healthy periodontal sites progression in patients with chronic periodontitis. They concluded that the transition probability for periodontal disease were higher with clinical attachment loss plus bleeding on probing than it is for pocket depth plus bleeding on probing. The reduction of both the clinical attachment loss and the probing pocket depth seen in the results in the study can be attributed to the mechanical debridement and with adjunct use of mouth washes. Both the non-alcohol containing solution and the alcohol-containing solution does show reduction in these parameters with no significant difference between the two.

It has been illustrated earlier that formation of deep pockets and loss of clinical attachment is pathognomonic for periodontal disease. For this reason, the reduction of deep periodontal pockets and gain of clinical attachment are obvious clinical goals of successful periodontal therapy and traditionally pocket probing is the evident method for diagnosis and evaluation of therapy. However, measuring gingival recession, pocket depth and clinical attachment level simultaneously is redundant, since with any two of these parameters the third is also established. (Mombelli, 2005)

The information regarding the dynamic phenomena may be obtained by combining acquired data from repeated assessment. The ability to record clinical loss or gain of attachment depends on the reproducibility of a single

measurement. Use of manual probe (like the one used for this study) has a resolution of 1 mm. (Mombelli, 2005)

On the other hand there is increased tooth mobility which is a common symptom also of advanced forms of periodontal disease (Lindhe & Nyman, 1984). They conducted studies on animal models where periodontal disease was induced and observed that loss of connective tissue attachment together with bone were symptoms that occurred with increased tooth mobility. The authors, then concluded that the increase in mobility was mainly attributed to the apical displacement of supporting alveolar bone. The importance of the amount of alveolar bone volume for teeth stability was also emphasised by Lindhe & Nyman (1989).

Schulte et al. (1992) looked into the relationship between tooth mobility and some indices of periodontal disease. These included the radiographic bone level, pocket depth, recession and bleeding on probing index. He was able to show that the amount of bone loss was the parameter which was most highly correlated to the plaque volume score, followed by the periodontal probing pocket depth.

Clinical outcomes evaluating tooth mobility following periodontal therapy has been studies in several investigations. Treatment procedures restricted only to supragingival debridement fail to reduce events of tooth mobility in patients presenting with periodontitis. This finding is in total agreement with the observation that gingivitis alone cannot cause increased mobility of teeth. (Giargia M *et al*, 1997)

Selection of a treatment procedure which effectively controls events of inflammation in sub-marginal periodontal tissues often results in a reduction of increased mobility of teeth. (Lindhe & Nyman, 1975). In this study the selected treatment included sub-gingival debridement and as such improvement of

mobility in teeth was improved in both the alcohol-containing and the non-alcohol-containing mouthwashes as adjunct treatments.

6.4 Bana-Zyme

Nipun D et al, 2015 studied the use of Bana-zyme strips on periodontitis patients with the aim to detect the presence of micro-organisms before and after full mouth debridement in adults with chronic periodontitis. In their conclusion they had made a few suggestions for the use of Bana-Zyme test as a potential diagnostic tool which could be employed

From the observation of the study, the following conclusion was drawn suggesting that BANA-Enzymatic test™ may be a potential diagnostic tool, which could be employed:

- "As a reliable indicator of BANA positive species in dental plaque
- As a simple, chair side test to detect a BANA hydrolyses from P. gingivalis, *T. denticola* and *T. forsythia*, anaerobic bacteria associated with adult periodontal disease
- As an objective means of determining diseased sites, requiring some form of periodontal treatment"

In this study, however, the Bana-Zyme test has shown reduction in the microbial load following mechanical debridement and use of adjunct mouth washes of both solutions, with and without alcohol. (Shown in Graph 6) The tests have not been able to distinctly show superiority of one solution when the two were compared.

6.5 Potential Limitations

Treatment for periodontal disease was carried out in a span of six weeks including follow up assessments. Oral hygiene instruction needed to be adhered to as means of home care treatment and maintenance. Patients may have not strictly followed the instructions as recommended at the first visit.

On that note, constant communication was done either telephonically or by means of reminder short message service as means of participant motivation. Only two groups were tested in accordance with the study design. It would have served the study better to have had a third control group receiving only mechanical debridement without adjunct use of mouth wash.

Moreover, having a lot of variables to test in one study made it difficult to analyse the data. It was therefore difficult to produce a united outcome leading to statistician producing individual results on each of the seven variables that were tested.

CHAPTER 7

CONCLUSION

With the new and revised approach to treating and managing patients presenting with periodontitis, mechanical debridement with the aim of removing plaque and calculus (disturbance of the biofilm), the use of adjunct mouth washes with chlorhexidine as the main ingredient, remains the most effective method. (Arweiler N.B., 2001) This include both professional interventions by the clinicians and home-care by the patients.

Both mouth wash solution with and without alcohol have proven to reduce the microbial load as shown by the BANA-Zyme test, with the alcohol containing solution being more effective. The improvement in the clinical parameter of measuring periodontitis may be attributed to traditional mechanical debridement with adjunct mouth washes, irrespective of whether the mouth wash solution has or has no alcohol.

The two test group showed a significant reduction in the gingival index score. The test group that used Paroex showed an even greater results in the reduction of bleeding on probing. Generalised reduction of the inflammation due to mechanical debridement with the adjunct use of mouth wash has showed improvement in the status of periodontitis.

The prognosis of gingival recession after full mouth debridement and use of mouth washes alone was not as positive, thus, requiring surgical management of defects. Gingival recession is thought to be directly linked to the thickness of the gingiva accompanied by other important factors including the presence of enough keratinized mucosa around teeth and local frenum pull. (Hsun-Liang Chan *et al*, 2016) These factors together with other gingival recession predisposing factors impact on the outcome of defects following non-surgical periodontal therapy. Even though in this study no measurements on the amount

of keratinized mucosa and biotype, Hsun-Liang Chan noted that it may be very likely that patients presenting with thinner biotype and inadequate keratinization of the gingiva may be the reason for poor outcomes following non-surgical periodontal therapy. (Hsun-Liang Chan *et al*, 2016)

Full mouth mechanical debridement in the treatment of periodontitis with adjunct use of mouthwash either with or without alcohol will yield results of improved periodontal status. For reasons that may prompt the clinician to not use solutions with alcohol, the end result will produce reduction in the periodontal clinical parameters and therefore, improve the status of chronic periodontitis.

CHAPTER 8

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