Efficacy of Mechanical Decontamination Strategies in the Treatment of Peri-implantitis

Doctoral thesis by

Sadia Khan

Department of Prosthetics and Oral Function
Institute of Clinical Dentistry
University of Oslo
Norway

2024

Series of dissertations submitted to the
Faculty of Dentistry, University of Oslo

ISBN 978-82-8327-090-7

Cover: UiO.
Print production: Graphic center, University of Oslo.

Truth is more valuable if it takes you a few years to find it.

Jules Renard

Contents

LIST OF FIGURES 6

ACKNOWLEDGEMENTS 7

ABBREVIATIONS 9

LIST OF PAPERS 10

BACKGROUND 11

OSSEOINTEGRATION 13

PERI-IMPLANT HISTOLOGY AND ANATOMY 13

THE DENTAL IMPLANT 14

BACTERIAL COLONIZATION OF THE IMPLANT SURFACE 15

PERI-IMPLANT HEALTH 15

PERI-IMPLANT DISEASES 15

ASSESSMENT OF PERI-IMPLANT TISSUES 17

AETIOLOGY OF PERI-IMPLANTITIS 18

PERI-IMPLANTITIS IN A HISTORICAL PERSPECTIVE 18

PREVALENCE OF PERI-IMPLANT DISEASES 19

PERI-IMPLANTITIS VERSUS PERIODONTITIS 20

GENERAL RISK FACTORS AND INDICATORS FOR PERI-IMPLANTITIS 20

LOCAL RISK INDICATORS AND FACTORS FOR PERI-IMPLANTITIS 22

PERI-IMPLANTITIS AND SYSTEMIC HEALTH 24

ANATOMY OF PERI-IMPLANT DEFECTS 24

THE GOALS OF PERI-IMPLANTITIS TREATMENT 26

TREATMENT OF PERI-IMPLANTITIS 27

PERI-IMPLANTITIS TREATMENT STRATEGY 27

DECONTAMINATION OF IMPLANT SURFACES 31

THESIS AIMS 34

MATERIALS AND METHODS 35

STUDY DESIGN 35

IN VITRO MODELS FOR INVESTIGATING DECONTAMINATION OF DENTAL IMPLANTS 35
CLINICAL STUDIES TO EVALUATE THE EFFICACY OF NON-SURGICAL TREATMENT OF PERI-IMPLANTITIS 39
DATA ANALYSIS 41

RESULTS **43**

DISCUSSION **47**

DISCUSSION OF THE MAIN FINDINGS 48
METHODOLOGICAL CONSIDERATIONS 56

ETHICAL CONSIDERATIONS **68**

CONCLUSIONS **69**

CLINICAL IMPLICATIONS **70**

FUTURE PERSPECTIVES **71**

REFERENCES **72**

APPENDICES **82**

List of Figures

Figure 1. Histological micrograph showing normal bone approximately 500 nm from the titanium surface. 13

Figure 2. Schematic illustration of anatomy in healthy tissues around a tooth versus a dental implant. 14

Figure 3. Modern dental implants in a historical perspective. 17

Figure 4. Peri-implant defect morphology as presented by Schwarz et al. 25

Figure 5. An illustration of peri-implantitis defect anatomy Monje et al. 25

Figure 6. A 3D resin model, representing anatomical features incorporating an advanced peri-implant defect. 35

Figure 7. Patients underwent an examination at baseline and at 1, 3, 6, 9, and 12 months. 39

Figure 8. Characteristics of occlusion spray on the buccal surface of implants. 49

Figure 9. Two different occlusion sprays were tested on SLA titanium discs. 58

Figure 10. The occlusion spray is composed of microscale particles embedded within a polymeric matrix, resembling the 3D structure of a biofilm. 58

Figure 11. Radiographic measurement of bone level. 67

Acknowledgements

The research presented in this thesis was performed at Institute of Clinical Dentistry, University of Oslo (UiO), and at Faculty of Odontology, Complutense University of Madrid. I express my sincere gratitude to both universities for their support and resources. Additionally, I want to thank the Norwegian Research Council (256756) for funding the clinical studies, and to Labrida AS for supplying chitosan brushes, and Straumann AG for providing dental implants.

I am profoundly thankful for the path of exploration and learning that has led to the completion of this doctoral thesis. I want to extend my heartfelt appreciation to my principal supervisor, Associate Professor Carl Hjortsjö, for your tremendous support and guidance throughout this entire period. Your generosity in sharing your knowledge and your caring attitude has been invaluable to me. To my co-supervisors, Associate Professors Odd Carsten Koldsland and Caspar Wohlfahrt, your dedication and enthusiasm for research has been incredibly motivating. I sincerely appreciate your encouragement and support. I also want to express my sincere gratitude to Professor Jan Eirik Ellingsen and Associate Professor Hans Jacob Rønold for presenting me with this intriguing opportunity. Additionally, I'm grateful to Associate Professor Erik Saxegaard for introducing me to academic writing. Furthermore, working alongside Sonni Mette Waaler, the first female Norwegian professor in prosthodontics, has been truly inspiring. Many thanks for leading the way.

A special acknowledgement goes to Professor Mariano Sanz for sharing your extensive wisdom gained through many years of academic accomplishments, providing invaluable guidance through the planning phase, overseeing the progress of the work, and facilitating the study in Madrid. I am also grateful that you introduced me to the outstanding researchers in the ETEP (Etiology and Therapy of Periodontal and Peri-Implant Diseases) group: Honorato Ribeiro-Vidal, Leire Virto, Enrique Bravo, and Paula Virginia Nuevo Gutiérrez.

My appreciation extends to Associate Professor Hanna Tiainen for all your unwavering support, invaluable assistance, and instruction in countless details, as well as for nurturing a passion for science and effective communication. Your availability whenever I needed guidance has meant a lot to me.

I extend a sincere thanks to my co-authors, Anders Verket, Ann-Marie Roos-Jansåker, Ibrahimu Mdala, Anna Magnusson, and Eirik Salvesen, for your significant contributions to the work in this thesis. Many thanks to Ibrahimu for infusing vitality into the challenging world of statistics.

I am sincerely grateful to Dental technician Mateusz for your invaluable contribution to the design and production of models utilized in this project. Your exceptional work capacity and dedication to enhance dental education are truly commendable.

I extend my heartfelt appreciation to Gerald Torgersen for calibration and blinding of radiographs, to Alejandro Barrantes for performing profilometry, to the National Center of Electron Microscopy at Complutense University Madrid for their assistance with microscopy, and to Anne Kristoffersen for facilitation of the clinical studies.

A warm acknowledgement goes to my dear friend and great prosthodontist, Sigurd. Thank you for paving the way for me and allowing me to work in an inspiring environment.

Special gratitude is extended to my esteemed colleagues, Lisa Printzell, Bjørn Einar Dahl, and Homan Zandi, who paved the way on the Ph.D. journey before me. Your support and encouragement have meant a lot. I admire not only your professional achievements but also the exceptional individuals you are. Thank you for being there for me every step of the way.

To my cherished friends outside the dental field, thank you for all your patience, love, and support. And to my piece of heart, Aniba (late), I did this for us. To my parents and brothers, you are my bedrock. Thank you for always letting me fly freely. To my dear mother-in-law, Greta, I admire your values, your positive attitude, and big heart. To my beloved Hans, thank you for enriching each day, making me remember that life is now, and never letting me walk alone (YNWA). Your presence means the world.

My MAC survived the sand!

With humility and gratitude

Sadia Khan

Oslo, June 2024

Abbreviations

BI	Bleeding Index
BoP	Bleeding on Probing
CFU	Colony-Forming Units
EDTA	Ethylenediamine Tetra Acetic Acid
ITT	Intention-to-Treat
mBI	Modified Bleeding Index
OCB	Oscillating Chitosan Brush
OR	Odd Ratio
PI	Plaque Index
Pi	Peri-Implantitis
PiM	Peri-Implant Mucositis
PP	per Protocol
PPD	Probing Pocket Depth
qPCR	Quantitative Polymerase Chain Reaction
RBL	Radiographic Bone Level
RCT	Randomized Controlled trial
SD	Standard Deviation
SEM	Scanning Electron Microscopy
SLA	Sandblasted, Large-grit, and Acid-etched
SoP	Suppuration
TC	Titanium Curette
US-PEEK	Ultrasonic Device with PEEK-tip
VAS	Visual Analogue Scale

List of Papers

This thesis is based on the following papers:

I. Anatomical 3D model with peri-implant defect for *in vitro* assessment of dental implant decontamination

Sadia Nazir Khan, Odd Carsten Koldsland, Hanna Tiainen, Carl Hjortsjö

Clin Exp Dent Res, 2024. DOI: 10.1002/cre2.821

II. The efficacy in decontaminating dental implants of an oscillating chitosan brush compared with an ultrasonic PEEK-tip. An *in vitro* study using a dynamic biofilm model

Sadia N. Khan, Honorato Ribeiro-Vidal, Leire Virto, Enrique Bravo, Paula Virginia Nuevo Gutiérrez, Odd Carsten Koldsland, Carl Hjortsjö, Mariano Sanz

Submitted for publication

III. Non-surgical treatment of mild to moderate peri-implantitis using an oscillating chitosan brush or titanium curette — a randomized multicentre controlled clinical trial

Sadia N. Khan, Odd Carsten Koldsland, Ann-Marie Roos-Jansåker, Johan Caspar Wohlfahrt, Anders Verket, Ibrahimu Mdala, Anna Magnusson, Eirik Salvesen, Carl Hjortsjö

Clin Oral Impl Res, 2022; 33:1254-1264. DOI: 10.1111/clr.14007

IV. Non-surgical treatment of mild to moderate peri-implantitis with an oscillating chitosan brush or a titanium curette — 12-month outcomes of a multicenter randomized controlled clinical trial

Sadia N. Khan, Odd Carsten Koldsland, Ann-Marie Roos-Jansåker, Johan Caspar Wohlfahrt, Anders Verket, Ibrahimu Mdala, Anna Magnusson, Eirik Salvesen, Carl Hjortsjö

Clin Oral Impl Res, 2023; 00:1-14. DOI: 10.1111/clr.1407

Background

The clinical problem

Oral health challenges impact a staggering 3.5 billion individuals globally, with approximately 267 million suffering from tooth loss.[1] Often attributed to dental trauma, periodontal disease, or dental caries, these conditions compromise the aesthetic and social aspects of the individuals' life and impede mastication and speech. A particularly concerning oral health affliction is the complete absence of teeth, commonly referred to as edentulism, which, despite being preventable, remains a prevalent issue globally.[2]

Dental implants have become the preferred method of tooth replacement, with 12 million implants being inserted globally each year.[3] Direct contact between bone and titanium was observed in animal studies and introduced in scientific reports as the concept of osseointegration by Prof. Brånemark and co-workers in 1969.[4] This seminal discovery became a clinical breakthrough and laid the foundation for rehabilitating partial and complete edentulism with implant-retained prosthetics. The specific number of implants inserted in Norway annually is currently unavailable, but data from the Norwegian Health Economics Administration suggests an annual increase in reimbursement for implant treatment.[5]

One notable advantage of replacements with dental implants compared to tooth-retained fixed prosthetics is that implant-retained replacements eliminate the need to remove sound tooth structure. Additionally, implant-retained replacements can be maintained as individual units allowing easier access for hygiene compared to multi-unit restorations.

The field of implant dentistry is constantly evolving, with a continuous introduction of new implant types and components to meet the needs of both patients and dentists. Advancements include the use of different implant materials, improved implant designs, computer-assisted treatment planning, and 3D printing. Furthermore, advances in implant surface properties and coatings have enhanced the implant integration with bone.[6]

The efficacy of implant treatment has been assessed through several long-term follow-up studies, with outcomes reported in terms of implant success, survival, complications, or failures.[7, 8, 9] According to the report from the International Congress of Oral Implantologists (ICOI) Pisa Consensus Conference,[10] an implant is considered to have survived if it remains in the mouth, exhibits no mobility or pain upon function, and has bone loss not exceeding 1/2 of the implant length. Conversely, an implant is classified as failed if it displays mobility, experiences pain upon

function, presents uncontrolled exudates or has significant bone loss.[10] As defined by Albrektsson et al.,[11] success criteria include no mobility, no peri-implant radiolucency, no pain, discomfort or infection, and bone loss less than 0.2 mm annually following the first year of loading. This definition is outdated, as it considers a bone loss of 2 mm after ten years as a "healthy condition". By a more current consensus, survival is characterized by the presence of the implant or reconstruction.[12] Conversely, success is defined by the presence of the implant or reconstruction, with the absence of any complications.

After ten years of function, long-term survival rates for implant treatment exceed 95%.[13] Survival rates for tooth-retained fixed dental prostheses and implant-retained crowns remain high over a period of five and ten years, showing comparable survival rates.[14] However, despite high survival rates, implant treatment is accompanied by biological, technical, and esthetical complications.[13, 15]

Implants, like teeth, rely on the health and stability of supporting soft- and hard tissues. Several medical and oral factors may impact the biological attachment between the implant and bone, potentially leading to the destruction of the supporting bone. The anatomical differences between the supporting tissues of teeth and implants contribute to a difference in the host response to trauma and disease.[16, 17]

The biological complications associated with dental implants can be classified into two types: those that disrupt the osseointegration process, leading to early loss of supporting tissues and eventually leading to early failure, and those that cause an inflammatory process affecting the soft- and hard tissues at a later stage when the implant is loaded with the prosthetic component.[18] It is well-established that bacterial biofilm contributes to peri-implant inflammation.[19, 20] A reduction in the bacterial load is a pivotal aspect in achieving peri-implant health.[21, 22] Effective removal of multispecies biofilm seems important to ensure lasting resolution of inflammation in the peri-implant tissues and to prevent further disease progression.[21] The ultimate objective is to diminish the bacterial burden to a level below the individual's immune system threshold, facilitating the resolution of the disease.[23]

Decontamination methods may be too gentle and thus not adequately address the removal of biofilm from the implant surface. Conversely, more intensive strategies, though proficient in mitigating biofilm presence, might unintentionally compromise the topographical integrity of the implant surface.[24] Additionally, mechanical approaches alone may be insufficient for effective decontamination.[21] Methods that balance the effective removal of biofilm with the preservation of implant surface integrity constitute a challenge in the maintenance and treatment of peri-implant diseases.

Osseointegration

In 1952 Dr. Brånemark discovered the direct interaction between titanium implants and vital bone, leading to clinical studies conducted in 1965.[25] He initially defined osseointegration as "a direct contact, on the light-microscope level, between living bone tissue and the implant" in 1984. This definition evolved to describe "a direct structural and functional connection between ordered living bone and the surface of a load-carrying implant".[26] In subsequent years, as more experience was gained with dental implants and osseointegration, several definitions of the term were launched.[11, 27]

Osseointegration refers to the "inseparable incorporation between vital bone and titanium"[28] and is primarily a histological definition. Specifically, the interface comprises newly formed cortical bone connecting with the titanium implant surface,[28] apart from the last layer approaching the titanium surface.[29] Histologically, ultrastructural analysis performed with a transmission electron microscope has shown a 100-500 nm zone of irregularly arranged collagen bundles and a 20-40 nm amorphous area containing proteoglycans and glycosaminoglycans at the titanium surface and bone interface (Figure 1).[29]

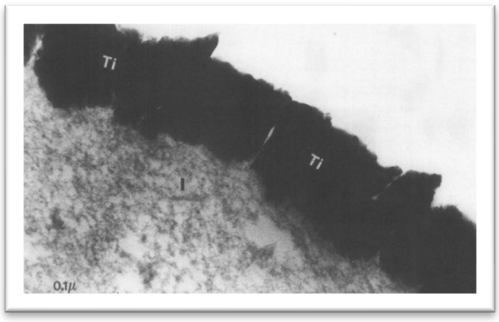


Figure 1. Histological micrograph showing normal bone approximately 500 nm from the titanium surface. The zone between titanium and bone is filled with an amorphous zone consisting of 20-40 nm proteoglycans and glycosaminoglycans (towards titanium) and a zone of randomly arranged collagen filaments. Derived from Albrektsson et al. 1994.[29] © 1994 JOHN WILEY AND SONS

Peri-implant histology and anatomy

In healthy conditions, the peri-implant mucosa and gingiva exhibit a comparable pink colour and firm consistency, albeit there are clinical and histological similarities as well as differences between the two (Figure 2). Soft tissue conditions around dental implants have been studied in animal studies.[30-32] Histologically, the peri-implant mucosa comprises connective tissue lined with oral epithelium, and a junctional epithelium facing the implant surface.[30, 31] On average, the peri-implant mucosa reaches a height of 3-4 mm about eight weeks after the implant placement.[33] Even in

healthy conditions, small clusters of inflammatory cells are present at the interface between the connective tissue and the epithelium.[34]

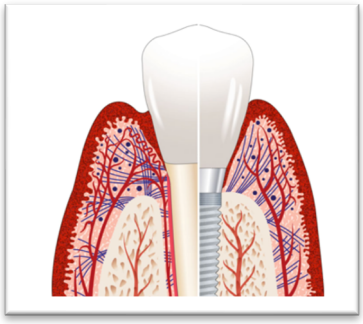


Figure 2. Schematic illustration of anatomy in healthy tissues around a tooth versus a dental implant.[35] © 2017 JOHN WILEY AND SONS

The dental implant

The dental implant complex comprises three integral components: the implant, the abutment, and the implant-retained supraconstructions. Titanium, zirconia, and titanium-zirconia alloy are primary materials for dental implants, with titanium being the most prevalent choice due to its biocompatibility.[6] At present, six distinct variations of titanium are utilized as implant biomaterials. Among them, four are classified as grades of commercially pure titanium (CPTi) (Grade I, Grade II, Grade III, and Grade IV), exhibiting purity levels ranging from 98-99.6%.[36] Additionally, titanium alloys, including variations of Ti-6Al-4V (6% aluminium and 4% vanadium), are also utilized.[36, 37] Each grade and alloy possess unique corrosion-resistance, strength, and ductility characteristics. Grade IV CPTi is the preferred alloy as the high oxygen concentration (0.4%) entails mechanical strength.[36, 37] Grade IV titanium has a low modulus of elasticity and is corrosion resistant.[37] In comparison, Ti-6Al-4V possess superior mechanical properties when compared to Grade IV titanium.[37, 38]

Zirconia was introduced as a metal-free option for patients with allergies, offering an aesthetically pleasing solution.[39] Subsequently, implants with a titanium-zirconia alloy were developed to improve mechanical strength and biocompatibility.[40]

Furthermore, the implant surfaces undergo nanostructure modifications to promote bone growth and facilitate osseointegration. Examples of such surface modifications include TiUnite, Osseospeed, and Sandblasted with Large grit and Acid-etched (SLA).[41, 42]

Bacterial colonization of the implant surface

Bacterial colonization and the formation of a complex biofilm can be observed within one hour following the placement of dental implants.[43, 44] The biofilm develops into a more intricate structure within two weeks.[43, 44] The adhesion of bacteria to non-shedding oral surfaces is a complex process that involves bacterial transport towards the implant surface, followed by the initial adhesion, lasting attachment, and subsequent colonization.[45] Surface roughness is a characteristic that influences the initial bacterial adherence.[45] The accumulation of bacterial plaque on dental implant surfaces may lead to an inflammatory process in the peri-implant tissues.[19, 22, 46, 47] Studies have identified periodontopathogen bacteria in clinically healthy and diseased peri-implant sites.[48]

The peri-implant environment provides an ecological niche for colonizing and proliferating anaerobic bacterial species.[49] A literature review reveals that peri-implant diseases manifest predominantly as mixed anaerobic infections.[50] The microbial composition closely resembles the subgingival flora observed in chronic periodontitis, with a predominance of Gram-negative bacteria.[50-52, 53] Furthermore, elevated levels peptostreptococci and staphylococci characterizes the peri-implant microbiota.[50] Compared to biofilms in periodontitis, those in the peri-implant region exhibit significantly less heterogeneity with a simpler structure.[53]

In peri-implantitis (Pi), the total bacterial load of seven species is approximately four times higher as compared to healthy implants.[51] The bacterial species identified in association with Pi are *Tannerella forsythia*, *Porphyromonas gingivalis*, *Treponema socranskii*, *Staphylococcus aureus*, *Staphylococcus anaerobius*, *Staphylococcus intermedius*, and *Streptococcus intermedius*.[51]

Peri-implant health

Peri-implant health is defined as the absence of all clinical signs of inflammation, as outlined in the consensus report from the World Workshop on the Classification of Periodontal and Peri-Implant Diseases and Conditions in 2017.[54] Specifically, for a peri-implant site to be considered healthy, it must exhibit the absence of erythema, bleeding on probing (BoP), swelling, or suppuration (SoP).[16] At health, the peri-implant tissues resemble the periodontal tissues.

Peri-implant diseases

Peri-implant diseases is a collective term for two biofilm-associated inflammatory conditions affecting the peri-implant supporting tissues: peri-implant mucositis (PiM) and Pi.[54]

Peri-implant mucositis

Histologically, PiM is characterized by an infiltrate with a flush of vascular structures.[31] Clinically, PiM is manifested by BoP, increased probing pocket depths (PPD), erythema, swelling, and SoP.[55] The presence or absence of the same clinical parameters is registered to evaluate treatment outcomes. PiM arises from the previously healthy peri-implant mucosa due to the accumulation of bacterial biofilm.[22, 56] A cause-and-effect relationship between the experimental biofilm build-up around titanium dental implants and a subsequent inflammatory response has been demonstrated.[22, 56, 57] Studies suggest that PiM is the precursor to Pi, and patients with untreated PiM are at a greater risk of developing Pi.[47] Therefore, it is crucial to provide adequate treatment for affected implants to prevent the onset of Pi.[58]

Salvi et al. compared the effects of experimental biofilm build-up for three weeks on peri-implant mucosa and gingiva in humans.[22] The results showed more bleeding sites in the peri-implant mucosa than in the gingiva. Interestingly, plaque incidence was higher at tooth sites than at implant sites after the same time with no oral hygiene. However, gingival inflammation at tooth sites increased less compared to mucosal inflammation at implant sites, suggesting that a similar bacterial load caused a severe inflammatory response at the implant sites. In experimental studies of PiM, achieving resolution of inflammation required biofilm control for more than three weeks for complete recovery at the clinical level.[55]

Peri-implantitis

Pi is an inflammatory process in soft- and hard tissues surrounding osseointegrated dental implants, leading to progressive destruction of the peri-implant bone.[59] In the case of Pi, radiographic bone loss is evident in addition to the mentioned parameters for PiM.[60]

The histopathological features associated with Pi encompass lesions beyond the junctional/pocket epithelium, housing large quantities of plasma cells, macrophages, and neutrophils.[17] When compared histologically to periodontitis lesions, the Pi sites exhibit larger inflammatory lesions.[48]

According to the consensus report of World Workshop on the Classification of Periodontal and Peri-implant Diseases, the diagnostic criteria for Pi are based on the availability of baseline data.[54] Changes in the radiographic bone level (RBL) combined with increased PPD compared to baseline data indicate a destructive process in the peri-implant tissues. In cases where previous examination data is accessible, the diagnosis necessitates the identification of bleeding and SoP upon probing, increased PPD compared to previous records, and bone loss changes surpassing the initial bone remodelling. Conversely, in cases where baseline data is absent, the diagnosis requires the detection

of bleeding and SoP on gentle probing, PPD ≥6 mm, combined with bone levels measuring ≥3 mm "apical of the most coronal part of the intraosseous portion of the implant".

Until the Pi diagnosis criteria were established in 2018 (Figure 3), inconsistencies were found in the definitions of the disease.[61] Furthermore, a standardized classification system categorizing the severity of the disease has not been established. While terms such as "moderate" and "severe" are referenced in the literature,[62] a consensus regarding their precise definitions is yet to be established.

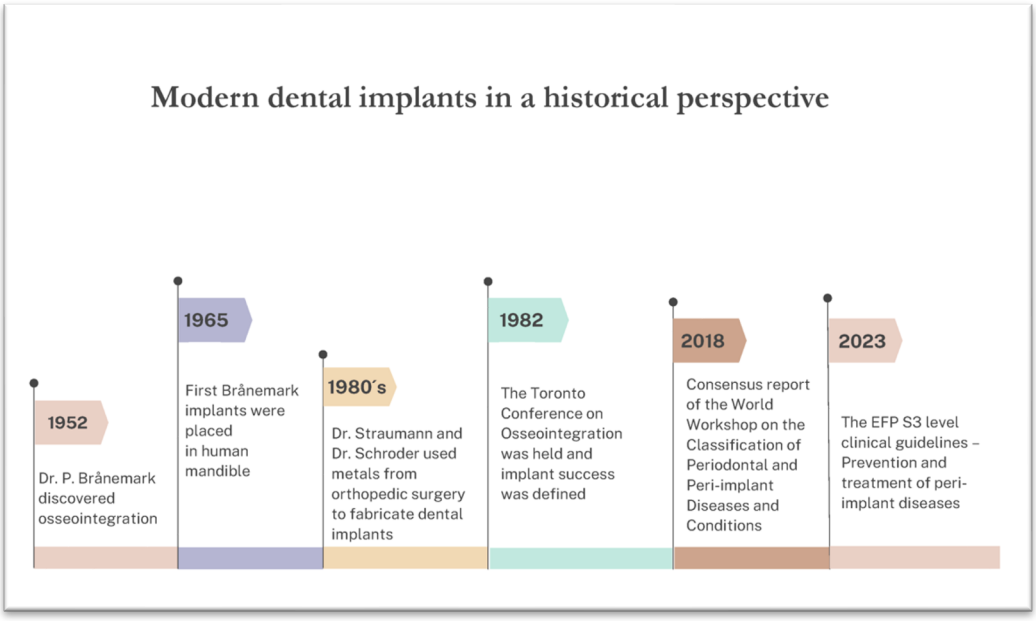


Figure 3. Modern dental implants in a historical perspective. Almost seventy years after the discovery of osseointegration, the first consensus reports were published, defining diagnosis criteria and clinical guidelines on treatment for peri-implant diseases.

Assessment of peri-implant tissues

A detailed assessment is recommended to evaluate peri-implant tissues to detect inflammation. This includes visual inspection, probing of the peri-implant tissues, and measuring peri-implant pocket depths.[63] The diagnosis of peri-implant conditions relies on a combined assessment of radiographic bone level changes and clinical parameters, emphasizing the importance of a comparative approach.[54] However, it is important to note that the diagnostic sensitivity of clinical parameters decreases without previous records, leading to an increased rate of false negative cases, particularly in the early detection of PiM and Pi.[64]

When evaluating peri-implant tissues, several parameters are vital. Plaque assessment and monitoring of oral hygiene habits are essential, as plaque accumulation can lead to peri-implant bone destruction.[65, 66] Regular clinical evaluations should include registration of oral hygiene, routine visual evaluation, and probing of dental implants, with particular attention to BoP.[67, 68] BoP, accompanied by increased PPD over time, indicates inflammation and is associated with a

reduction of peri-implant bone.[68, 69] PPD, BoP, and the presence of SoP are reliable parameters for diagnosing peri-implant health and disease.[70-72] Furthermore, radiographic evaluation, including baseline intra-oral radiographs and assessing changes in bone levels, is necessary to differentiate between health and disease states.[73, 74]

The assessment of peri-implant tissues encompasses three steps: Baseline probing within three months of prosthesis delivery, obtaining a baseline intra-oral radiograph after physiological remodelling, and subsequent radiographs in case of increased PPD and BoP to evaluate marginal bone levels.[75] These steps ensure accurate diagnosis and early detection of peri-implant diseases.

Aetiology of peri-implantitis

Extensive research, encompassing animal[19, 46, 76] and human studies,[22, 56] has provided insights into the aetiology of Pi, particularly concerning the reactions of peri-implant soft tissues to plaque formation. The studies have consistently demonstrated that plaque accumulation leads to inflammation in the peri-implant soft tissue.[48, 54] In a study involving individuals with intact dentition and undergoing controlled plaque accumulation for three weeks, it was observed that peri-implant soft tissues exhibited a more pronounced inflammatory response compared to gingival tissues in natural teeth.[22] The presence of plaque around dental implants, resulting from inadequate oral hygiene, has been linked to PiM.[77] It should be noted that PiM does not always progress to Pi.[54]

Peri-implantitis in a historical perspective

In a French publication in 1965, Levignac was the first to describe inflammation in peri-implant soft tissue with subsequent destruction of peri-implant bone.[78] Later, the infectious pathological condition was coined as "Peri-implantitis".[66] During the 1st European Workshop in Periodontology in 1993, it was agreed upon that the term should refer to destructive inflammatory processes around functional osseointegrated implants, exhibiting loss of peri-implant bone and formation of deeper peri-implant pockets.[59]

A study published in 2016 highlighted that the understanding and knowledge regarding PI during the 1980s were limited.[79] At that time, discussion primarily revolved around achieving successful osseointegration, with little attention given to the potential loss of implants due to PI. Implant failures were predominantly attributed to early loss related to poor surgical technique or occlusal overload.

Contrary to the widely accepted view, an alternative perspective challenges the notion that marginal bone loss around dental implants is due to infection, suggesting instead that it might be a foreign body response.[3] According to this viewpoint, marginal bone loss can be attributed to an exacerbated foreign body reaction disrupting the foreign body equilibrium. Various factors, including insufficient clinical handling, unfavourable patient factors, remnants of cement, or changes in loading conditions, may contribute to this disruption. While there is a suggestion that restoring the foreign body equilibrium is achievable, the absence of such balance may result in progressive bone loss, ultimately resulting in implant failure.

Prevalence of peri-implant diseases

Prior to 2018[54], the absence of consensus regarding diagnostic criteria for Pi resulted in varied definitions of the condition. Consequently, the inconsistency in case definitions has resulted in variations in the reported prevalence rates of Pi.[62, 80, 81] The prevalence of Pi also differs depending on whether it is assessed on implant level or patient level,[82] the severity of disease,[62] and across different patient cohorts.[69, 82-84]

Due to the heterogeneity in case definitions, the reported prevalence of Pi ranges widely, from as low as 1% to 47%.[85] A meta-analysis weighted a mean prevalence of Pi on the patient level to be 22%.[85] At the third EAO consensus conference in 2012, it was acknowledged that the prevalence of Pi within a 5–10-year timeframe following implant placement is approximately 10% at the implant level and 20% at the patient level.[84, 86] Furthermore, in a nine-year follow-up study involving a large patient cohort in Sweden, the findings revealed that 45% of the patients exhibited Pi, characterized by BoP/SoP and bone loss greater than 0.5 mm.[62] Among the patients, 14.5% had moderate or severe Pi, with BoP/SoP combined with bone loss exceeding 2 mm.

A high frequency of peri-implant diseases has been observed among individuals who smoke, with an estimated prevalence of 36.3%.[87] The number of implants affected by Pi has been reported to be greater in smokers in comparison to non-smokers.[88] Furthermore, patients with history of periodontitis exhibit a high prevalence of Pi.[89]

Risk indicators for Pi are generally not deemed an absolute contraindication for implant treatment, and a considerable number of patients with such indicators receive replacements for missing teeth with dental implants. Is it plausible to anticipate a potential escalation in the prevalence of Pi in the coming years as a result?[79]

Peri-implantitis versus periodontitis

Despite the resemblances in clinical characteristics, aetiology and progression, there are crucial histopathological differences between periodontitis and Pi.[17, 90] While periodontitis causes a general loss of supporting tissue around teeth, the destruction observed in Pi is more localized to specific implants.[20] Additionally, the destruction at peri-implant sites occurs more rapidly, described as a "non-linear and accelerating pattern".[54]

In human biopsies obtained from sites affected by Pi and periodontitis lesions, large inflammatory cell infiltrates (ICT) were observed laterally to the pocket epithelium.[17] The apical extension of the ICT was more pronounced in Pi biopsies compared to periodontitis.[60] Furthermore, the cellular composition of the lesions also differed. Plasma cells and lymphocytes were the predominant cell types in both lesions.[91] However, the proportions of neutrophil granulocytes and macrophages were higher in Pi than in periodontitis.[92] In addition, in Pi lesions, neutrophil granulocytes were observed in peri-vascular compartments away from the pocket area.

Moreover, evidence suggests that the composition of the peri-implant microenvironment, despite many similarities, may differ somewhat from the microbiome around teeth.[52] *In vitro* studies have demonstrated that *S.aureus* has a preference for titanium surfaces.[93] However, *S.aureus* is not strongly linked to chronic periodontitis. Furthermore, uncommon oral microorganisms such as *S.epidermis* are more prevalent in Pi lesions than in periodontitis lesions.[94] Most studies indicate that the subgingival microbial composition, in health and disease, is comparable between implants and natural teeth.[66, 94-97]

General risk factors and indicators for peri-implantitis

A risk factor is a well-established factor that directly contributes to the worsening of a disease or increases the chances of developing it. In contrast, risk indicators may also be potential risk factors, but the evidence supporting their association is less robust and often based on retrospective and cross-sectional studies.[74] Risk factors, on the other hand, are identified by prospective longitudinal studies.[68] Evaluating a patients' risk factors is crucial for planning preventive strategies, early disease detection, and providing a personalized treatment and maintenance program.

Factors such as history of periodontitis, inadequate plaque control and lack of regular maintenance follow-up visits are associated with risk of peri-implant diseases.[48] Furthermore, there is limited evidence supporting that tobacco smoking, diabetes, and submucosal cement may increase the risk of developing peri-implant diseases.[48, 54, 98]

- Periodontitis

  Patients diagnosed with periodontal disease have been found to exhibit a higher prevalence of Pi when compared to patients without periodontitis.[48, 62, 99] A retrospective study identified a history of periodontal disease as a risk for the development of Pi with an odd ratio (OR) of 3.63.[82] Furthermore, in a patient cohort with a longer follow-up time (9 yrs.), the OR of developing Pi was 1 for periodontally healthy patients and 4.1 for those with periodontitis.[62] Further, supporting these findings, the periodontally compromised group showed a higher prevalence of Pi in a study comparing periodontally compromised patients with healthy counterparts.[100] Specifically, implants in patients with residual periodontitis displayed deeper PPD and more bone loss compared to those without residual periodontitis.

  While the survival rates of implants in patients with a history of periodontitis are reported to be high in the first years after installation, a statistically significant decline is described after six to eight years of function.[99]

- Smoking

  Smokers exhibit significantly higher levels of nicotine in the gingival crevice fluid compared to levels detected in blood plasma, with a nearly 300-fold difference.[101] Numerous studies have identified the potential of nicotine to hinder the wound healing process.[102, 103] Additionally, it has been evidenced that smoking has a detrimental impact on the humoral immune defense,[104] and inhibits the proliferation and function of both B and T cells.[105]

  Based on the findings in the current literature, it is evident that dental implants inserted in smokers are associated with a considerably higher risk of failure when compared to non-smokers.[106, 107] Additionally, smokers display a higher mean marginal bone loss.[106]A significantly higher risk of Pi has been reported.[108, 109] However, the role of smoking as a potential risk remains inconclusive, as indicated in the consensus report on Pi for the World Workshop on the Classification of Periodontal and Peri-implant Diseases and Conditions and in a recently published systematic review and meta-analysis.[48, 110]

- Diabetes

  Diabetes is a chronic disease that occurs when the pancreas does not produce enough insulin or when the body cannot effectively use the insulin that it produces. According to the Public Health Report of 2017, an estimated 245 000 individuals in Norway had received a diabetes diagnosis.[111] Diabetes has long been recognized as a factor in dental implants due to its impact on wound healing[112] and the body's capacity to mount effective responses

against infections.[113] Maintaining proper control of blood sugar levels, known as glycaemic control, is important for individuals with diabetes. There is evidence suggesting a connection between glycaemic control and the occurrence of vascular complications.[114, 115]

Patients with poorly controlled diabetes are at a higher risk of experiencing Pi.[110, 116, 117] However, when diabetes is well-controlled, the success rates of dental implants are similar to those in non-diabetic patients.[75, 116] While some data suggest a correlation between diabetes and the risk of peri-implant disease, the available literature does not provide statistically significant differences in dental implant failure rates between diabetic and non-diabetic patients.[110, 118]

- Maintenance follow-up

  Several studies have highlighted the importance of peri-implant maintenance compliance in preventing Pi and sustaining the stability of peri-implant tissues.[47, 119, 120] Monje et al. specifically emphasized the need for a compliance rate of more than two yearly visits for healthy patients.[120] The study revealed a statistically significant association between compliance and peri-implant disease, with 86% fewer cases of Pi among patients in the compliance group ($p < 0.05$). In line with this, Gay et al. reported a 60% reduction in failure rates compared to patients with less than one visit per year.[121]

  In a 5-year follow-up study conducted by Costa et al., the incidence of Pi was examined in individuals with PiM who either received maintenance care or no maintenance therapy.[47] A higher incidence of Pi among patients with no maintenance follow-up was observed, with approximately 40% compared to 20% in the group receiving maintenance care.

Local risk indicators and factors for peri-implantitis

- Oral hygiene and dental plaque

  The impact of plaque control and oral hygiene in the development and progression of peri-implant diseases have been reported in the literature.[47, 83, 122, 123] Furthermore, a dose-dependent relationship between plaque scores and peri-implant disease has been suggested.[123] Specifically, individuals with higher plaque scores showed more severe peri-implant disease. In line with this, Vignoletta et al. assessed the association between plaque and peri-implant disease, showing an odds ratio of 1.8 for the presence of plaque, demonstrating an increased risk.[83] Collectively, these results demonstrate the importance of effective preventive strategies to maintain optimal oral health.[110]

- Prescence of keratinized mucosa

  Keratinized mucosa (KM) surrounding dental implants as a preventive measure against peri-implant diseases has been assessed in multiple studies.[64, 124-126] The evidence shows that implant sites with less than 2 mm of KM exhibit higher levels of brushing discomfort, increased plaque accumulation, and BoP.[124] Furthermore, less than 1 mm of KM at the implant level has been identified as a risk indicator for PiM.[83] Moreover, a minimum of 2 mm of KM appeared to be a protective factor against Pi.[127] The impact of KM dimensions seems related to plaque control rather than a direct correlation with soft tissue anatomy.[81] Furthermore, when pathological alterations are observed in the peri-implant mucosa, it is recommended that the KM dimensions are corrected surgically.[128]

- Occlusal overload

  The link between occlusal overload and the development of peri-implant bone loss has been investigated in several studies.[69, 129-131] While some authors suggest an association, the overall literature does not provide conclusive evidence supporting occlusal overload as a definitive risk factor for peri-implant diseases.[69] However, evidence indicates that occlusal overload may expedite the progression of peri-implant disease in the presence of inflammation.[130]

  To minimize occlusal stress and reduce extensive mechanical forces on implants, splinting is often recommended to minimize strain on both the implant and the peri-implant tissues. The concept behind splinting is to achieve a more balanced distribution of forces, aiming to alleviate the potential for overloading. However, the existing evidence regarding the effectiveness of splinting on stress and mechanical forces remains inconclusive.[129] Further research is warranted to establish more robust conclusions.

- Poor prosthetic fit and design

  Various prosthetic factors have been identified to influence the risk of developing Pi. Kordbacheh et al. reported an overrepresentation of cemented prostheses and poorly designed or ill-fitted restorations among affected implants.[82] Other studies also highlight the correlation between cement-retained prostheses and peri-implant disease.[82, 98, 122] However, an ill-designed or ill-fitting prosthesis emerged as the most influential risk factor, emphasizing the consequences of persistent plaque retention and poor access for plaque removal at the interface between the implant and the supraconstructions.[82, 132, 133]

In a literature review evaluating the prevalence of peri-implant disease in patients with screw-retained versus cemented supraconstructions, a higher prevalence of peri-implant diseases was observed when there was residual cement excess at the crown-abutment interface.[122] The type of cement used was found to play a role in the development of inflammation. Furthermore, screw-retained prosthetic constructions directly attached to the implants were found to carry a higher risk of developing peri-implant disease compared to supraconstructions designed for abutment level. Additionally, a more progressive Pi has been associated with fixed implant-supported compared to removable prostheses.[132]

Peri-implantitis and systemic health

As Pi shares similarities with periodontitis, Pi potentially correlates with initiating an immunological reaction and the development of systemic diseases. In a pilot study involving a limited study sample of otherwise healthy patients diagnosed with Pi, a low-grade inflammatory state, evidenced by elevated levels of white blood cells and interleukin-10 (IL-10), alongside an elevated presence of total cholesterol and low-density lipoprotein (LDL) cholesterol concentrations, was observed.[134] Additionally, results from an animal study conducted by Chaushu et el. support the impact of Pi on both inflammatory- and hepatic serum biochemical parameters.[135] Induced experimental Pi resulted in elevated levels of markers such as C-reactive protein (CRP), alanine aminotransferase (ALT), and aspartate aminotransferase (AST). These markers may indicate the systemic effects of peri-implant disease. Notably, following surgical debridement, a decrease in systemic inflammation, as demonstrated by the reduction of inflammatory and hepatic serum biochemical parameters to their baseline values. This suggests that effective treatment can restore systemic inflammation associated with Pi.

Anatomy of peri-implant defects

Pi leads to the destruction of the surrounding peri-implant bone structure, resulting in the formation of bone defects. The defects display variations in size and shape and may be classified as either Class 1, 2 or 3 according to the specific configuration.[132, 136] Class 1 defects exclusively manifest as intraosseous defects, whereas Class 2 defects involve horizontal bone loss (Figure 4).[136, 137] Class 3 defects exhibit combined characteristics from Classes 1 and 2.[132] Additionally, both classes are further divided into subclasses based on the anatomical characteristics of the respective bone defects.[136]

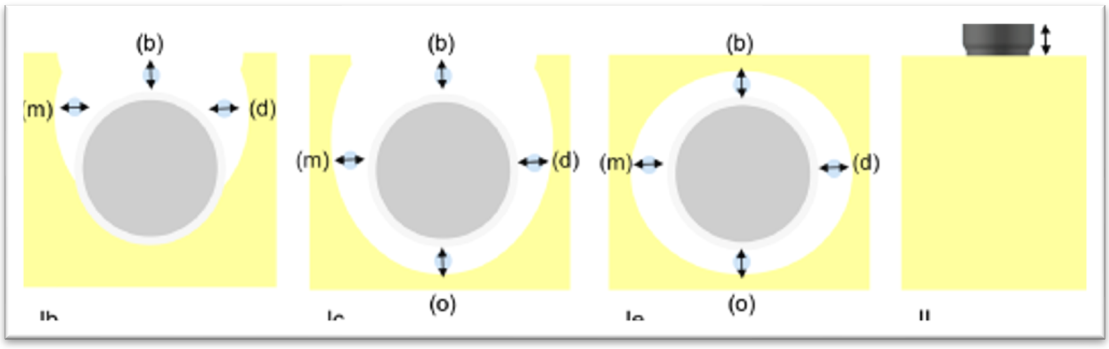


Figure 4. Peri-implant defect morphology as presented by Schwarz et al.[137] Class 1b refers to buccal dehiscence with semi-circular bone resorption up to the mid-section of the implant body. Class 1c presents a buccal dehiscence while the lingual wall is intact. Class 1e encompasses circular bone resorption but retains both buccal and oral bone. 1=intrabony component (marked as blue circles). Class 2 is identified as supra-alveolar component (highlighted as an arrow). For orientation, m: mesial; d: distal, b: buccal; and o: oral. © 2017 JOHN WILEY AND SONS.

Furthermore, the severity of bone loss has been suggested for categorization, employing the following grading system: Grade S (slight, <25% of the implant length), Grade M (moderate, ≥25%-50% of the implant length), and Grade A (advanced, >50% of the implant length) (Figure 5).[132]

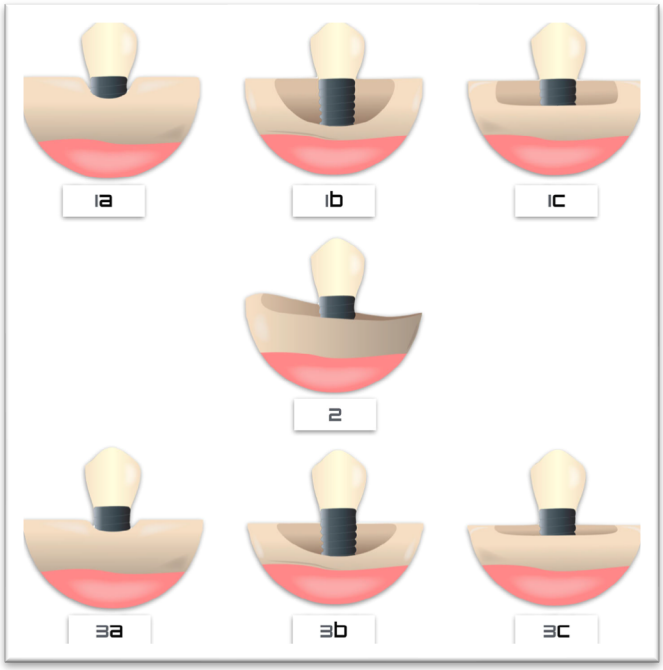


Figure 5. An illustration of peri-implantitis defect anatomy Monje et al.[132] The defects are based on their anatomical characteristics: ***Type 1***: Infra-osseus defect; *Type 1a*: dehiscence limited to the buccal bone; *Type 1b*: bone defect that maintains 2-3 intact walls; *Type 1c*: complete circular bone defect; ***Type 2*** defect category is characterized by horizontal bone loss. Defects in ***Type 3*** merges characteristics from Type 1 and 2. *Type 3a* is a defect with buccal dehiscence, while *Type 3b* is a bone defect with 2-3 intact walls, and *Type 3c* is a circular bone defect, all Type 3 defects have a horizontal bone loss component in addition. © 2019 JOHN WILEY AND SONS

Clinical and animal studies indicate a high prevalence of intraosseous circumferential defects.[48, 136] In human and dog studies, Class 1 defects were most commonly observed.[136] This observation was further corroborated by a cone-beam computed tomography (CBCT) study on Pi defects, which found that intraosseous defects, specifically with two to three walls, represented the most common defect configuration (55%).[132]

Monje et al. evaluated the influence of patient- and implant-related characteristics on peri-implant defect morphology.[132] The findings indicate that patient-, implant-, and site-specific factors were associated with distinct morphological features and the severity of the defects. Smokers exhibited more severe bone defects compared to non-smokers and former smokers. Moreover, Pi cases in implant-supported fixed prostheses displayed more advanced defects compared to patients with removable prostheses.

The bone defect morphology plays a crucial role in the decision-making process when considering treatment procedures.[138] The bone configuration impacted the mean PPD and clinical attachment level 6 and 12 months after surgical regenerative treatment performed with natural bone mineral combined with a collagen membrane.[137]

The goals of peri-implantitis treatment

Effective management of Pi requires strategic interventions to address the underlying causes and promote tissue healing. Mombelli et al. emphasizes the importance of mechanical and chemical treatments in disrupting the peri-implant biofilm and recognizing the role of microorganisms in the disease process.[50] A key aspect of managing Pi is the decontamination of the implant surface.[21] The interventions play a vital role in eradicating the source of infection and promoting the resolution of Pi lesions. Additionally, infection control is crucial to establish a healthy peri-implant environment.

Several consensus statements have articulated the end goals of Pi treatment.[23, 75, 139] To achieve successful Pi treatment, the ultimate goal, as stated by Heitz-Mayfield et al., is the resolution of the disease.[140] This entails the absence of pus or BoP and no further radiographic bone loss. However, if a complete resolution is not attainable, Heitz-Mayfield et al. suggested a reduction in clinical inflammation as an intermediary goal. This can be achieved by decreasing peri-implant PPD, minimizing BoP, and creating an unfavourable environment for biofilm growth. Furthermore, Heitz-Mayfield et al. suggested success criteria for Pi therapy as no peri-implant PPD ≥5 mm, no BoP upon light pressure, no SoP, and no additional bone loss.

According to the recently published clinical guidelines on prevention and treatment of peri-implant diseases, Herrera et al. emphasize the monitoring of residual inflammation and SoP alongside PPD.[75] The guidelines suggest using residual PPD of ≤5 mm with BoP registered as a spot bleeding from a maximum of one site and without SoP as endpoints of therapy. A re-evaluation is advised between 6 to 12 weeks after the non-surgical treatment.

It is essential to underscore that no specific PPD can be considered compatible with health.[54] As per the latest clinical guidelines on Pi treatment, clinical outcomes should be assessed by pre-determined success criteria to decide whether further treatment is warranted.[75]

Treatment of peri-implantitis

While there are distinctions in the care required for Pi and periodontitis, it is essential to recognize that the theoretical foundation of Pi treatment is built upon the efficacious approaches developed for treating periodontitis.[75] Therefore, a systematic, progressive approach similar to the one recommended for periodontitis treatment has been suggested to address Pi.[23, 141]

Professionally administered treatment may be non-surgical or surgical, performed alone or combined with adjunctive treatment. Both treatment strategies have shown improvements in inflammatory and microbial parameters.[141, 142] However, the ability to halt the progression of peri-implant bone loss over time remains unpredictable.[54, 143, 144] It is unclear whether the partial treatment effect is related to poor access with current devices or limited focus and information about the defect morphology in the non-surgical context.

Surgical treatment is considered superior to non-surgical treatment in cases with advanced peri-implant lesions displaying insufficient access.[86] However, in the presence of reduced marginal bone level and deep residual pockets, the risk of Pi recurrence remains even after surgical intervention.[144]

Peri-implantitis treatment strategy

Addressing Pi has proven challenging, primarily due to the previous lack of agreement on diagnostic criteria and the absence of definitive clinical treatment guidelines.[54, 75] However, primary prevention emerges as a key focus and has been emphasized in several consensus reports.[23, 58, 110] Patient-administrated mechanical plaque control, using manual or powered toothbrushes, has proven to be an effective preventive measure.[58] Additionally, professional intervention, comprising oral hygiene instructions and mechanical debridement, has shown a reduction in clinical signs of inflammation.[56]

A stepwise treatment approach based on the stage of implant therapy is suggested.[23, 75, 141] The recommendations, according to the recently published clinical guidelines, comprehensive treatment sequence for Pi is proposed.[75] This includes a pre-treatment phase involving thorough assessment and diagnosis, risk factor reduction, and addressing prosthetic issues limiting access to plaque control, if necessary.[23] Non-surgical debridement is advocated to maximize biofilm removal, supplemented with adjunctive chemicals if needed.[141] Surgical access is considered if non-surgical methods prove ineffective. Surgical treatment may include regenerative or resective approaches, with thorough implant surface decontamination considered vital.[23]

The clinical guidelines suggest a treatment management approach based on different stages of implant therapy.[75] Three clinical scenarios are outlined: patients awaiting implant rehabilitation, those undergoing it, and patients with existing dental implants. Preventive measures are divided into primordial, primary, and secondary prevention, emphasizing controlling risk factors, and maintaining peri-implant health. Furthermore, Heitz-Mayfield et al. and Herrera et al. highlight the importance of regular monitoring and supportive maintenance therapy to prevent disease progression and maintain peri-implant health.[23, 75]

- Non-surgical treatment of peri-implantitis

  Recent clinical guidelines on the prevention and treatment of Pi emphasize that upon Pi diagnosis, a decision must be made regarding the treatability of the affected implant.[75] If deemed treatable, the initial step shall involve non-surgical treatment, encompassing sub-marginal instrumentation.

  A 2008 literature review found that mechanical non-surgical therapy constitutes an effective approach to treat PiM lesions.[145] The efficacy was further enhanced when combined with antimicrobial mouth rinses as an adjunct. However, non-surgical mechanical treatment displayed limited effectiveness, when applied to Pi lesions. In such cases, adding chlorhexidine as adjunctive treatment yielded effects on clinical and microbiological parameters. In contrast, a more promising outcome was observed when local or systemic antibiotics were combined with mechanical non-surgical debridement, resulting in reduction of BoP and peri-implant PPD.[145]

  Conversely, a consensus report highlighted that non-surgical management of Pi can effectively reduce inflammation, improving clinical parameters.[61] These improvements were evident through reduced BoP and a decrease in peri-implant pocket depths.

Non-surgical therapy is advocated as the primary intervention as it allows the clinician to assess tissue healing and the patients' commitment to maintaining optimal oral hygiene.[75] In some instances, combining mechanical therapy with oral hygiene practices proved adequate in controlling the infection without necessitating surgical intervention.[141] Selected Randomized Controlled Trials (RCTs) combining non-surgical mechanical treatment with laser or chemical adjunctive are summarized in the Appendix.

A two-centre RCT comparing the efficacy of surgical and non-surgical debridement Pi treatment demonstrated that both approaches yielded comparable clinical outcomes.[146] Thus, for mild to moderate Pi cases, non-surgical treatment was suggested as the preferred initial treatment. Nevertheless, relying solely on non-surgical mechanical decontamination may yield unpredictable outcomes, especially in moderate to advanced Pi cases. Consequently, a surgical approach was suggested to enhance the chances of success in such cases.[147]

- Surgical treatment of peri-implantitis

Surgical intervention for Pi has shown long-term effectiveness, particularly for implants with non-modified surfaces.[148] Nonetheless, there remains a risk for disease recurrence, especially in cases where implants exhibit residual PPD of ≥6 mm 1 year post-surgery.[144] It is advisable to consider surgical treatment for Pi in patients with persistent signs of inflammation following non-surgical therapy, characterized by the presence of deep peri-implant pockets along with BoP or the presence of pus.[75]

Several surgical techniques have been documented in the literature. Among these, access flap surgery has shown promising results in reducing inflammation, BoP, SoP, and PPD.[141, 149] The approach involves raising a flap to provide adequate visibility and access for thorough decontamination of the implant surface.[75] Furthermore, resective surgery has been demonstrated effective in cases with uneven residual bone.[141] This technique removes diseased tissue and reshapes the bone to promote better healing and a healthier peri-implant environment. For specific defect morphologies, regenerative techniques have been recommended.[120] The approach aims to promote the regeneration of lost bone and soft tissue around the implant site.

Maintenance and post-surgical care are crucial for the long-term success of surgical treatment. Effective submucosal cleaning procedures have been identified as necessary during the maintenance phase after surgical treatment.[150] Regular maintenance therapy has

been shown to maintain stable peri-implant conditions, but residual pockets may indicate a higher risk of disease progression.[151]

- Adjunctive treatments in the treatment of peri-implantitis

  Additional measures to complement mechanical non-surgical, and surgical approaches has the aim to enhance treatment outcomes by reducing peri-implant inflammation. In a systematic review conducted by Linares et al., the potential benefits of incorporating local and systemic antimicrobials as adjunctive measures in non-surgical treatment were investigated.[152] The research revealed that systemic antimicrobials, especially in cases with initially deep sites, led to a reduction in peri-implant PPD and BoP. These findings underscore the potential impact of adjunctive measures on enhancing non-surgical interventions and align with findings in a systematic review conducted by De Waal et al. focusing on chemical surface decontamination.[153]

  Moreover, the role of systemic metronidazole was assessed in an RCT.[154] The study demonstrated that including systemic metronidazole as an adjunct to non-surgical treatment in Pi treatment led to additional improvements in clinical, radiographic, and microbiological parameters at the 12-month follow-up.

  Carcuac et al. evaluated the adjunctive effect of systemic antibiotics and local chlorhexidine combined with mechanical implant surface decontamination in surgical Pi treatment.[155] A positive influence of systemic antibiotics on treatment success, particularly in implants with a modified surface, was demonstrated. Furthermore, several adjunctive treatment methods combined with surgical intervention for Pi have been investigated clinically. Laser decontamination and implantoplasty have shown some potential to yield improved clinical outcomes when used alongside conventional treatment, albeit with weak supporting evidence. While these adjunctive treatments offer certain advantages, systemic antibiotics in combination with surgical therapy remain a subject of debate due to the limited evidence supporting the definitive benefits.[75, 149]

  Long-term follow-up studies are instrumental in understanding the sustainability of treatment outcomes. Carcuac et al. provided a 3-year follow-up on surgical treatment with various adjunctive measures.[156] Their findings indicated that the benefits of systemic antibiotics were limited and not sustained over the 3 years.

  Focusing on PiM and Pi treatments, Ramanauskaite et al. conducted a systematic review and meta-analysis.[157] The findings indicated that while alternative measures were superior

in reducing bleeding in non-surgical treatment of Pi, they provided no beneficial effect in resolving PiM. This emphasizes the importance of tailoring treatment strategies to the specific condition being addressed.

Furthermore, in the recently published clinical guidelines on Pi treatment, the need for further research to establish definitive evidence supporting the use of adjunctive measures such as photodynamic therapy, antiseptic desiccant solution, locally administered antimicrobials, and systemically administered antibiotics in non-surgical Pi therapy, is warranted.[75]

Decontamination of implant surfaces

Effective removal of microbial biofilm from implant surfaces poses a challenge in Pi treatment. A complex biofilm, encompassing various bacterial species and hard deposits, on contaminated implant surfaces necessitates successful removal to permanently resolve inflammation in the peri-implant tissues and to halt disease progression. The ideal goal is to suppress the bacterial load below the individual threshold level manageable by the host immune system, ultimately leading to disease resolution. Ensuring a delicate balance between decontamination effectiveness, accessibility to the contaminated surface, and the prevention of any remnants that could provoke an immune response is paramount during the decontamination process. Equally critical is the safeguarding against adverse events or any potential damage to the implant surface while performing decontamination procedures. This intricate balance is vital to achieving successful and safe management of Pi.

Various mechanical debridement methods have currently been employed to remove the biofilm on implant surfaces, such as plastic, carbon fibre, and metal curettes,[158-160] titanium brushes,[24, 161] ultrasonic instruments,[158, 159, 162] air-powder abrasive systems,[163] lasers,[159, 163] and the more recent oscillating chitosan brush (OCB).[164] The existing decontamination methods have undergone *in vitro* and clinical evaluation.

The *in vitro* effectiveness assessment has been performed on titanium objects, including discs, coins, sheets, and healing abutments, which are used to simulate dental implant surfaces.[165-168] Additionally, various studies have evaluated the decontamination effectiveness on dental implants alone or when inserted into 3D blocks with different bone defect configurations.[24, 169-171] In recent studies, an evolution has been witnessed wherein 3D blocks, incorporating anatomies of bone defects, teeth, and soft tissue, have been introduced.[172] However, it is crucial to acknowledge that real-life situations may be difficult to replicate *in vitro*. In clinical practice, challenges may arise due to differences in defect morphology at the site level and the presence of supraconstructions, making

access to specific implant surfaces more difficult than others. The use of less clinically realistic *in vitro* models can result in a disparity between findings from *in vitro* and *in vivo* studies.

The decontamination methods may prove too delicate, falling short in effectively removing biofilm and calculus from the implant surface. On the other hand, more aggressive approaches, while capable of tackling the biofilm, may inadvertently alter the integrity of the implant surface.[24] Achieving an optimal balance between adequate debridement and preserving implant integrity remains a significant challenge in implant decontamination.

Metal-free alternatives in Pi treatment have emerged in hand instruments, such as plastic and carbon curettes, and mechanical instruments like ultrasonic devices with polyether-ether-ketone tip (US-PEEK) instruments. Introducing these alternatives aims to provide gentler options that can better preserve the integrity of the implant surface. This is important, given the reported recurrence following both non-surgical and surgical treatments,[75, 144] where repeated interventions raise the risk of implant surface damage.

US-PEEK has been evaluated in several *in vitro* studies.[24, 161, 173] Cha et al. conducted an *in vitro* study to investigate the impact of five decontamination protocols, including using a US-PEEK, on the surface topography of dental implants.[24] The results showed that the use of US-PEEK left remnants of the instrument tip on the implant surface, particularly in the threaded area. Furthermore, the US-PEEK instrument induced microscopic alterations during decontamination.[174] Concerning the cleaning ability of the tip, complete removal of an implant surface covered with ink was not achieved.[161]

In a systematic review conducted by Louropoulou et al., the effectiveness of various mechanical instruments in cleaning contaminated implants was evaluated.[168] The review included studies that assessed machined and polished discs or implants and employed rotary and handpiece instruments for debridement. The authors highlighted that mechanical debridement alone cannot achieve complete biofilm removal. The constant need for effective implant surface decontamination without causing damage has driven the development of new equipment.

The OCB bristles are made of chitosan, a polysaccharide, holds widespread application and is derived from the shells of marine crustaceans such as lobsters, crabs, and shrimps. The process begins with demineralization, deproteinization, and discoloration of the shells, resulting in a substance known as chitin.[175] However, chitin is insoluble, necessitating further conversion by deacetylation to convert it into chitosan. Due to a broad spectrum of benefits, different forms of chitosan have long been used in cosmetics, food products, pharmaceuticals, and various other

industries.[176] Chitosan possesses favourable characteristics like biocompatibility and biodegradability and has, therefore, been of interest for biomedical applications.[175] In particular, controlled biodegradability has been used for controlled delivery of pharmaceuticals to avoid toxic reactions and to deliver low medication dosages over time.

In dentistry, chitosan's favourable characteristics have led to its incorporation in fabricating scaffolds used for bone, periodontal, and dentin-pulp regeneration.[176] This opens new possibilities for enhancing tissue regeneration and healing processes in dental treatments. Furthermore, chitosan improves the efficacy of various antimicrobial agents, making it a promising candidate for use in dental care products. Chitosan and its derivates show a wide range of antimicrobial activity against gram-negative and gram-positive bacteria.[177]

A clinical evaluation conducted by Costa et al. revealed that chitosan mouthwash effectively reduced streptococci and enterococci levels after 30 minutes of use.[178] The study aimed to verify the safety and validate the biological activity of the chitosan mouthwash *in vivo*. The findings indicated the safety of the chitosan mouthwash, demonstrating lower cytotoxicity compared to the control mouthwash. Moreover, it effectively reduced viable counts of bacteria significantly.

With an understanding of the many benefits of chitosan, an implant surface decontamination brush with bristles composed of chitosan was introduced in a recent innovation. The use of the OCB for the treatment of PiM and Pi has been evaluated *in vitro* and in clinical studies.[179, 150, 164, 180] While the efficacy of the brush has been assessed through an RCT in supportive Pi treatment following surgery, the efficacy for Pi treatment has not been validated through an RCT.[150]

Step-by-step efficacy testing stands as a cornerstone in medical and dental advancements. Commencing with realistic, translational *in vitro* studies, this approach ensures a foundation for further investigation. *In vitro* studies allow for controlled experimentation and provide insights into potential mechanisms and effects. Once initial evidence is established, the transition to *in vivo* models becomes crucial, bridging the gap between laboratory settings and real-world scenarios. Clinical studies further validate findings, providing insights into human responses and treatment outcomes. Moving forward, the strongest evidence lies in RCTs, where interventions are tested against placebos or standard treatments. The RCT design minimize biases, ensuring the highest level of evidence for efficacy. This systematic progression, from *in vitro* investigations to clinically relevant studies culminating in RCTs, establishes a framework for assessing and validating novel treatments, ultimately elevating medical and dental treatments to higher standards of care.

Thesis Aims

The primary overall objective of this thesis was to evaluate mechanical decontamination methods in the treatment of Pi.

The general hypothesis of this thesis was that mechanical decontamination employing an OCB would significantly show greater efficacy of dental implant decontamination compared to conventional methods, both *in vitro* and in clinical settings.

Overall research question:

Which mechanical decontamination protocol is the most effective for Pi management?

Aims of the studies:

(1) to develop and assess an *in vitro* 3D model with peri-implant defect to evaluate the efficacy and accessibility of devices for the decontamination of dental implants (***Study I***)

(2) to determine the *in vitro* efficacy of removal of multispecies dynamic biofilm on dental implants using an oscillating chitosan brush or polyether-ether-ketone tip for ultrasonic device (***Study II***)

(3) to assess the clinical and radiographic outcomes of non-surgical peri-implantitis treatment using an oscillating chitosan brush or titanium curettes at six months (***Study III***)

(4) to assess the clinical and radiographic outcomes and evaluate the migration between bleeding index (BI) stages of non-surgical peri-implantitis treatment using an oscillating chitosan brush or titanium curettes over a 12-month treatment period (***Study IV***)

Materials and Methods

Study design

This thesis consists of four studies: two *in vitro* experiments and two clinical studies. The first *in vitro* study evaluated the efficacy of an innovative 3D printed model featuring a simulated peri-implant defect (***Study I***). In the second *in vitro* study, a true, validated, dynamic multispecies biofilm on titanium dental implants installed in the 3D model was decontaminated using either an OCB or a US-PEEK (***Study II***). The clinical studies were designed to evaluate the efficacy of non-surgical treatment of Pi at three, six (***Study III***), and twelve months (***Study IV***).

In vitro models for investigating decontamination of dental implants

Anatomically realistic in vitro 3D model (Studies I & II)

In ***Study I***, an *in vitro* 3D printed model of teeth and an edentulous area with a dental implant with a peri-implant defect was constructed using computer-aided design software (Figure 6). A phantom model was scanned, a peri-implant defect was designed, and the model was separated vertically into two parts at the occlusal surface of the teeth and the edentulous area prior to 3D printing. Dental implants with SLA surface were installed, the printed parts were assembled, and the implants were sprayed with coloured occlusion spray to mimic biofilm.

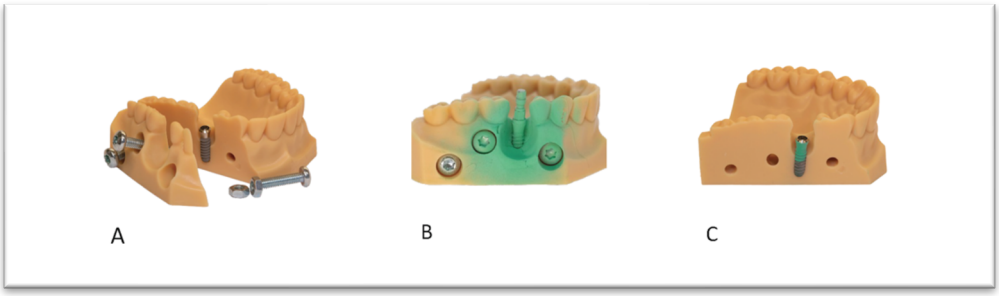


Figure 6. A 3D resin model, representing anatomical features incorporating an advanced peri-implant defect. The model was designed to facilitate convenient access for implant removal. *A:* The dental implant placed into the open model. The components were secured using screws. *B*: Following assembly, the visible implant was coated with coloured occlusion spray. *C*: The front section of the model was disassembled for extraction of the implant. The application of coloured spray was confined to the exposed implant area.

The function of the model was tested by evaluating the accuracy of the model, accessibility to the implant surfaces at site level, the decontamination efficacy, and alterations to the implant surface topography following decontamination with OCB, US-PEEK tip, OCB combined with water, blank gel, chlorhexidine gel, or OCB combined with chlorhexidine gel. Subsequently, image analysis and fluorescence spectroscopy were used to quantify remnant occlusion spray, while

scanning electron microscopy (SEM) and optical profilometry were employed to analyse implant surface alterations. In ***Study II***, implants seeded with biofilm were inserted into the validated 3D model before decontamination treatment.

Image analysis (Study 1)

Following decontamination, the implants were removed from the 3D model and transferred to a custom-made holder, employing a transfer piece to affix them, and subsequently subjected to photography. The photographic documentation encompassed the buccal, distal, palatal, and mesial implant surfaces, utilizing a single-reflex camera equipped with a Nikon 105 mm f/2.8D AF Micro Nikkor macro lens (Nikon D3200). To facilitate this process, the buccal side of the transfer piece was marked before the implants were removed from the model. Imaging occurred at a consistent angle, perpendicular to the implant's height axis, and adhered to fixed imaging parameters and lighting conditions. Subsequently, the images were calibrated for white balance and contrast, accomplished through Adobe Lightroom Classic. Furthermore, the images were binarized using ImageJ based on their colour histograms.

The image scale was calibrated using the implant diameter as a reference. Based on image examination, the extent of the area still covered by the green occlusion spray at the site level was determined by employing ImageJ®. In order to account for potential variations in implant orientation and imaging angles between experiments and test groups, all quantitative findings from the image analysis were presented as the ratio of the area covered with green occlusion spray to the total projected implant area for each implant, reported as a percentage. The image analysis process was repeated three times for each group.

Optical profilometry (Study I)

The implant surface topography of the decontaminated implants was evaluated using optical profilometry. Specifically, three distinct, non-overlapping areas on the buccal surface of the machined implant collar and the SLA surface were selected. Images were captured employing confocal mode with an EPI 20× objective, and the process was repeated three times (n=3). Subsequently, image processing was performed. Surface parameters were extracted and quantified using SensoMap Standard 7.4 provided by Sensofar.

Fluorescence spectroscopy (Study I)

Following decontamination, the total amount of occlusion spray remaining on the implant surface was assessed by placing the unmounted implants in 1.7 ml microcentrifuge tubes. The remaining occlusion spray film was removed by immersing the implants in 1 ml of isopropanol and followed

by a 20-minute sonication process at 60°C. The solvent, which contained the dispersed occlusion spray particles, was gathered, and used to estimate the remaining occlusion spray film quantity using a fluorometer (Qubit 4, Invitrogen) that measured fluorescence values through blue LED excitation at 470 nm and recorded emission at far-red wavelengths (665–720 nm). These values were subsequently transformed into weight/volume concentration using a standard curve generated from serial dilutions of known concentrations of fluorescent occlusion spray particles dispersed in the same solvent.

Scanning electron microscopy assessment of implant topography (Study I)

Imaging of clean implants was performed through a tabletop scanning electron microscope (SEM; TM-3030, Hitachi). Backscattered electrons at a 15 kV acceleration voltage were employed for imaging. A magnification of 2000x was applied.

Development of multispecies biofilm on implant surfaces (Study II)

Building on results from ***Study I***, in the second *in vitro* study (***Study II***), the mechanical implant surface decontamination efficacy of an OCB and a US-PEEK-tip was assessed by combining the validated 3D model and a validated dynamic multispecies biofilm model using six bacterial strains (*Streptococcus oralis, Veillonella parvula, Actinomyces naeslundii, Fusobacterium nucleatum, Porphyromonas gingivalis,* and *Aggregatibacter actinomycetemcomitans*). Dental implants were seeded with biofilm before decontamination treatment. Decontamination efficacy was evaluated using quantitative PCR (qPCR) and SEM.

Six different bacterial strains, namely *Streptococcus oralis* CECT 907T, *Actinomyces naeslundii* ATCC 19039, *Veillonella parvula* NCTC 11810, *Fusobacterium nucleatum* DMSZ 20482, *Porphyromonas gingivalis* ATCC 33277, and *Aggregatibacter actinomycetemcomitans* DSMZ 8324, were utilized for the study. These bacteria underwent cultivation on blood agar plates (Blood Agar Oxoid No 2; Oxoid) enriched with 5% (v/v) sterile horse blood (Oxoid), 5.0 mg/L hemin (Sigma, St. Louis, MO, USA), and 1.0 mg/L menadione (Merck) at a temperature of 37°C for 24-72 hours under anaerobic conditions (comprising 10% H_2, 10% CO_2, and a N_2 balance). Straumann® Tissue Level Standard titanium dental implants were employed in the study.

Each bacterial strain's pure cultures were nurtured under anaerobic conditions at 37°C for 24 hours in a modified brain-heart infusion (BHI) medium. Subsequently, the growth of bacteria in each pure culture was assessed using spectrophotometry. A mixed inoculum containing 10^6 colony-forming units (CFU)/mL for all six bacterial strains was created.

This mixed inoculum was introduced into the reaction chamber of the Lambda Minifor bioreactor (LAMBDA Laboratory Instruments). Planktonic growth inside the bioreactor was sustained for 12 hours and continued in a continuous culture mode with a flow rate of 30 mL/h. A peristaltic pump was employed to facilitate the flow of the bacterial culture through two customized Robbins devices containing the sterile implants. The implants within the Robbins devices were maintained at 37°C within anaerobic conditions, allowing bacterial growth and biofilm formation for 72 hours.

Quantitative polymerase chain reaction (qPCR) (Study II)

In ***Study II***, a quantitative evaluation of remaining bacteria post-decontamination utilizing qPCR was performed. Before DNA isolation, the implants underwent a sequential triple rinse in 2 mL of sterile PBS (with an immersion time of 10 seconds per rinse). The rinsing was performed to eliminate non-adherent bacteria from the implant surface. Subsequently, the disruption of biofilms was carried out via vortexing at room temperature for 2 minutes, employing 1 mL of sterile PBS. DNA extraction was accomplished employing a commercial kit (MolYsisComplete5; Molzym GmbH & CoKG) in strict adherence to the manufacturer's instructions, with the protocol for bacterial DNA extraction.

The detection and quantification of bacterial DNA were accomplished utilizing the hydrolysis probes 5´nuclease assay PCR technique. The primers and probes, provided by Life Technologies Invitrogen, Applied Biosystems, and Roche (Roche Diagnostic GmbH), were designed to target the 16S rRNA gene. A negative control of 2 µL sterile water [non-template control (NTC)] (Water PCR grade, Roche) was included. The samples were subjected to an initial amplification cycle involving 95°C for 10 minutes, followed by 40 cycles at 95°C for 15 seconds and 60°C for 1 minute.

Scanning electron microscopy assessment of biofilm (Study II)

SEM was used to assess the amount and structure of biofilm on the control implant and following decontamination treatment. Implants were extracted from 3D models and subjected to a gentle triple rinse in 2 mL PBS, each immersion lasting 10 seconds, to dislodge unattached cells. The implants were immersed in a fixative solution composed of 4% paraformaldehyde (Panreac Química) and 2.5% glutaraldehyde (Panreac Química) at 4 ºC for 4 hours. Subsequently, they underwent PBS and sterile distilled water rinse, each lasting 10 minutes, followed by dehydration through a graded ethanol series (30, 50, 70, 80, 90, and 100%), with each immersion lasting 10 minutes. The specimens were brought to the critical drying point and gold sputtered. The implants were analysed at the National Centre of Electron Microscopy (ICTS, University Complutense of

Madrid) through a JSM 6400 SEM equipped with a backscattered electron detector, operating at a 25 kV image resolution (JSM6400, JEOL).

Clinical studies to evaluate the efficacy of non-surgical treatment of peri-implantitis (*Studies III & IV*)

Randomized controlled clinical trial

Studies III and ***IV*** were designed as single-blinded, prospective, multicentre RCT with six- and twelve-month duration. The studies aimed to evaluate the clinical and radiographic outcome of the non-surgical treatment of Pi with a chitosan brush attached to an oscillating handpiece or a titanium curette (TC). Pi was defined as 2-4 mm radiographic bone loss, PPD ≥4 mm, and BI ≥2. During the six-month study (***Study III***), four screening sessions were scheduled during the six-month study: at baseline, one, three, and six months. Six clinical assessments were planned in the 12-month study (***Study IV***). Treatment was performed at baseline and repeated during the subsequent follow-up visits for cases displaying persistent inflammation (BI >1 and PPD ≥4 mm). ***Studies III*** and ***IV*** consisted of the same patient cohort. Algorithms employed in the studies are illustrated in Figure 7.

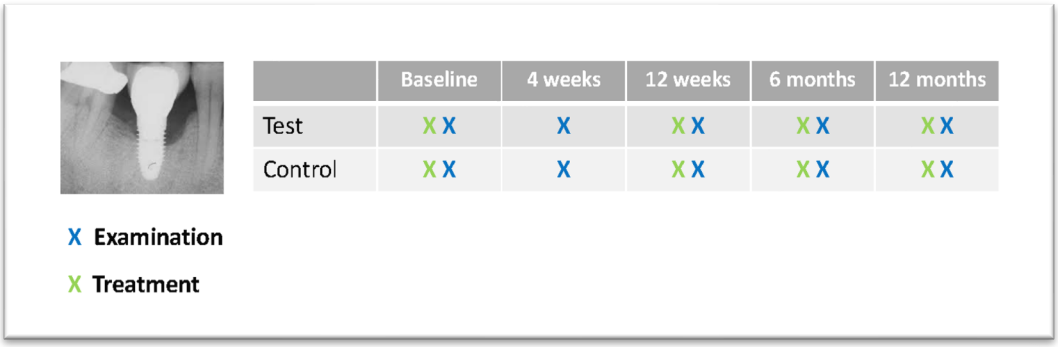


Figure 7. Patients underwent an examination at baseline and at 1, 3, 6, 9, and 12 months. Treatment was performed at baseline and repeated every 3 months in cases with persistent inflammation (defined as BI ≥1 and PPD ≥4 mm).

The primary outcome variable used in the study was changes in PPD. Secondary outcome variables included BI/pus, changes in RBL, and pain reported during treatment.

Patients with Pi, as defined, were included in ***Studies III*** and ***IV*** when:

1. implants had been functionally loaded for more than 12 months
2. age above 18 years
3. eligible for outpatient dental clinic treatment (ASA I and II)
4. full-mouth plaque scores ≤20% before inclusion
5. informed consent
6. commitment to all follow-up visits

Patients were excluded from the studies when:

1. radiographic peri-implant bone loss >4 mm
2. prosthetically impossible to access the implant for clinical measurements
3. technical complications that had contributed to Pi and were not possible to resolve
4. implant mobility
5. active periodontal disease
6. implants previously treated for Pi with grafting materials
7. medications with mucosal hyperplasia risk
8. systemic antibiotics treatment <three months prior to inclusion
9. medical conditions with an unwarranted risk and that would limit the patients' ability to participate in the study
10. unwillingness to undergo treatment
11. advanced, untreated, and/or uncontrolled Pi on neighbouring implants
12. patients with prosthetic constructions resulting in non-balanced traumatic occlusion
13. previous or ongoing radiotherapy in the head-neck region
14. chemotherapy
15. corticosteroid treatment

Detailed information about patients' general health, medications, and smoking habits was documented prior to the baseline examination. Pain experienced during intervention was reported using a visual analogue scale (VAS) at the three-month follow-up. Furthermore, full-mouth plaque score (%) was reported at baseline. In addition, the following clinical and radiographic parameters were measured at baseline and at 3, 6, and 12 months: plaque index (PI), BI, PPD, pus, and height of KM. RBL was assessed at baseline, 6, and 12 months (Table 1).

Table 1. Overview of self-reported, clinical, and radiographic parameters

Self-reported outcomes	**Description**
Background characteristics	Gender Age
General health	Diseases Medications
Smoking habits	Never Former Current
Pain experience during intervention	Visual analogue scale

Table 1 continues

Clinical and radiographic outcomes	
Bleeding index (BI)	Recorded 30 seconds following probing. Bleeding scores were classified into four categories: score 0 indicating no bleeding, score 1 denoting isolated bleeding spots, score 2 representing blood forming a red line, and score 3 indicating profuse bleeding.
Keratinized mucosa (KM)	KM height was measured at the midbuccal region using a periodontal probe.
Plaque index (PI)	Presence or absence of plaque was registered by running the probe along the marginal surface of the implant
Probing pocket depths (PPD)	Measured at six sites per implant
Pus or suppuration (Pus, SoP)	Presence or absence of pus
Radiographic bone level (RBL)	RBL was determined by measuring the distance from the implant neck to the point with bone to implant contact

Data analysis

Data analysis was performed using *SPSS* (IBM SPSS Statistics 25.0, IBM Corporation, Armonk, NY, USA), *Stata Statistical Software*, Version 16.1 (StataCorp.2001. Statistical Software: Release 7.0. College Station, TX: Stata Corporation), and *R* (R app 4.0.3 GUI Mac OS, R Foundation for Statistical Computing). Statistical significance was established at $p < 0.05$ for all four studies.

In ***Study I***, the proportion of the implant area covered with residual occlusion spray relative to the total implant surface was quantified as a percentage. All image measurements were repeated three times, and the averages and standard deviations (SD) were calculated. Similarly, the means and SD of fluorescence measurements were computed for each decontamination group and the control implants. As for the optical profilometry data, means and SDs were determined for the machined implant collar and the SLA surface. Variations between the decontamination groups and implant sites were assessed using a one-way ANOVA.

The primary variable in ***Study II*** was quantifying total bacterial counts of individual bacterial strains through qPCR analysis, quantified in CFU per millilitre (CFU/mL). The data was presented as

both means and SD. An analysis was undertaken at the experiment level for each study parameter (n=9).

A per-protocol (PP) analysis was conducted on patients who completed assessments at all designated time points in ***Study III***. Frequency and percentage statistics for categorical variables were employed and presented in means with SD for continuous data to describe patient and implant characteristics within the study groups. The patients were included across five different clinics, and to account for potential data interdependence among participants nested within these clinics, three-level linear and partial ordinal multilevel analyses were conducted. A multilevel partial ordinal logistic model was employed using gologit2 for the BI data. To assess the differences in PPD and BI between the groups at each study time point, a two-way interaction of time with the groups was extracted. Additionally, estimates of ICC to quantify the variability in PPD and BI attributed to differences between patients and clinicians were computed.

Study IV utilized the same primary- and secondary outcomes and statistical analysis as ***Study III***. In addition, Markov models were utilized to estimate the transitions between different states of BI.

Furthermore, the data in ***Study IV*** underwent analysis through two distinct approaches. Firstly, a PP analysis assessed complete patient cases across all time points. In addition, the intention-to-treat (ITT) principle was applied, encompassing all randomized patients in the study. For missing data, multiple imputations were generated using the R software (Version 4.0.3 GUI Mac OS, R Foundation for Statistical Computing).

Results

The efficacy of in vitro, anatomical, 3D models with peri-implant defect to evaluate devices for decontamination of dental implants (Study I)

The 3D models showed consistency with minimal variation across their components, facilitating precise implant insertion and ensuring uniform exposure of the implant threads. The occlusion spray, applied to simulate a biofilm, had a thickness of 9.6±1.4 μm and was consistent in its coverage. In composition, the occlusion spray resembled a biofilm grown on a titanium implant surface.

Implant surfaces in the control group showed intact occlusion spray upon removal. The occlusion spray was effectively removed from the machined implant collar for the OCB group, although it was inefficient on threaded areas. In the threaded region, a dark green shade of occlusion spray was observed on all implant sites following decontamination with OCB, indicating that the OCB efficiently reached all implant surfaces. The US-PEEK group partially removed the occlusion spray from both the implant collar and the threaded areas.

No method completely removed the occlusion spray. The OCB and US-PEEK groups showed statistically significant occlusion spray removal on buccal and mesial surfaces compared to the control group. The water group displayed minimal spray removal, almost resembling the control group. Chlorhexidine gel minimally impacted the occlusion spray. However, when combined with OCB, the gel significantly enhanced the decontamination outcomes. Fluorescence spectroscopy results further indicated that the OCB combined with isopropanol gel achieved promising outcomes.

Implant surface assessment showed that the US-PEEK group introduced alterations, such as scratches and disruptions. In contrast, the OCB- and water spray groups did not demonstrate any visible surface texture changes. No significant difference in surface roughness, skewness, kurtosis, or core fluid retention was observed across the machined or SLA surface groups.

In vitro efficacy of decontamination of multispecies dynamic biofilm on dental implants using an oscillating chitosan brush or polyether-ether-ketone tip ultrasonic device (Study II)

SEM imaging of biofilm structure and morphology showed that untreated implants displayed a robust and thick biofilm. The location and surface roughness played a role in biofilm growth, with rougher surfaces harbouring more bacteria. Specifically, the threaded surfaces were dominated by

spindle-shaped bacteria, whereas smooth surfaces mainly had scattered spherical and oval-shaped bacteria.

When treated with the US-PEEK, there biofilm, particularly in the fusiform bacteria, was markedly reduced across all implant areas. On the other hand, using the OCB demonstrated an effect on biofilm in machined areas. However, no decontamination effect on biofilm on rough surfaces was observed.

qPCR assessments post-decontamination revealed a significant reduction in bacterial colonies for the US-PEEK-treated group for all six bacterium types compared to the control group. Decontamination with the US-PEEK method achieved reductions ranging from around 83% to 93% for these bacteria. Compared to the untreated control group, the reductions were statistically significant for *V. Parvula*, *A. actinomycetemcomitans*, *P. gingivalis*, and *F. nucleatum*, ($p < 0.05$). However, the OCB method showed minor reductions in bacterial numbers, none of which were statistically significant.

Clinical and radiographic outcomes of non-surgical treatment of peri-implantitis using an oscillating chitosan brush or titanium curettes at 6 months (Study III)

A total of 45 patients were evaluated for study eligibility and randomly allocated to the test or control group. ***Studies III*** and ***IV*** flow charts were reported according to Consolidated Standards for Reporting Trials (CONSORT). After assessing the eligibility criteria, 39 patients were included in the study. Data was complete for 38 individuals, split into 21 for the test group and 17 for the control (***Study III***). Both groups displayed no statistically significant differences in baseline patient and implant characteristics.

Furthermore, no significant differences in PI severity between the groups was observed. The mean baseline KM was 2.8 mm and 2.5 mm for the test and control groups, respectively. At baseline, 36.8% of the test group implants and 54.5% of the control group implants had a KM of ≤2 mm.

While a reduction in PPD was noted for both groups, the between-group result was not statistically significant at any examined time point (baseline, 1, 3, or 6 months). Similarly, for BI, no statistically significant differences between groups were observed at any study time point. Although the likelihood of BI grade 3 diminished from the baseline to 6 months, the odds ratio of BI grade 0 rose during this timeframe. At the 6-month follow-up, pus was found in 33.3% of test group implants and 64.7% in the control, although the between-group difference was not statistically significant. Average RBL from baseline to 6 months remained stable: 2.5 mm for the test and 2.6 mm for the control group.

Two implants in the test group and one in the control group showed disease eradication at 6 months. Furthermore, one implant from the OCB group showed disease progression at 3 months and was consequently removed for surgical intervention.

Pain evaluation was reported for 22 patients, resulting in a 57.9% response rate. Seven participants underwent anaesthesia and, therefore, did not fill out the VAS form. Data was missing for nine due to oversight in form collection. Average VAS scores were 2.9 for the test group and 3.4 for the control, with no statistically significant difference.

Clinical and radiographic outcomes of non-surgical treatment of peri-implantitis using an oscillating chitosan brush or titanium curettes at 12 months (Study IV)

Of the initial cohort, 31 patients with Pi were examined over the 12 months. No adverse reactions related to the treatments were reported. Eight patients were excluded from the study by the clinicians. In the test group, one patient was excluded during the 3 to 6-month follow-up, and another three patients were excluded between the 6 to 12-month interval. For the control group, all exclusions were made between 6 to 12 months.

Employing PP analysis, a statistically significant reduction in PPD at 3, 6, and 12 months was observed in both groups compared to the baseline measurements. Furthermore, a statistically significant reduction in PPD between 6 and 12 months was observed in the test group. However, statistically significant differences between the groups were not identified at any examined interval.

For BI, a statistically significant reduction in BI 2 and BI 3 versus BI 0 and BI 1 at the implant level from the baseline to 12 months was observed in both study groups. However, no statistically significant differences between the groups were noted at any time.

Furthermore, a marked decrease in implants showing pus from baseline to 12 months was observed. No statistically significant differences between the two groups were identified.

An RBL of ≥3 mm was registered for a few patients in both groups at baseline. Radiographically, a stable bone level was observed throughout the study. No statistically significant changes in bone levels from the baseline to the 12-month follow-up were seen in either group.

At 12 months, disease eradication, defined as PPD <4 mm, BI 0, and no reduction in RBL compared to baseline, was shown for one implant in the test group. Treatment success, defined as ≤1 implant site with BI ≤1, absence of pus, PPD ≤5 mm, and no progressive bone loss, was seen in three implants in the test group and one implant in the control group, with no statistically significant differences identified between the two.

At baseline, KM ≥2 mm was observed for 70% of implants in the test group and 60% of in the control group. At the 12-month follow-up, both groups showed a decrease.

No significant differences in plaque scores were observed between the study intervals, and no association between BoP and plaque presence was found.

ITT analysis showed a stable RBL in both groups throughout the study. No statistically significant differences between the groups were observed in regarding PPD, BI, pus presence, or RBL.

Migration between bleeding index stages during a treatment period of 12 months with an oscillating chitosan brush or a titanium curettes (Study IV)

The multi-state Markov model described the progression and regression of peri-implant inflammation over 12 months (n=31). BI 0 was defined as a healthy state, BI 1 was an intermediary state between health and disease, and BI 2/3 represented disease.

For the OCB group, sites initially in the healthy BI 0 state, 60% managed to maintain the healthy status throughout the study period. However, 20% progressed to the intermediary BI 1 stage, and an equal percentage escalated to the disease states of BI 2/3. When originally in the BI 1 state, 50% reverted to a healthy BI 0 state, while 18.3% progressed to the BI 2/3 category. Meanwhile, those in the BI 2/3 state showed improvements, with 31.9% transitioning back to the healthy BI 0 state and 15% moving to the intermediary BI 1 state.

In the TC group, 66.7% remained in BI 0, 8% transitioned to BI 1, and 25.3% moved to the BI 2/3 states. In BI 1, 38.9% reverted to BI 0 state, and 24.1% progressed to BI 2/3. For those in the BI 2/3 state, 18.13% returned to BI 0, and 18.13% shifted to BI 1.

Discussion

In addressing Pi, the objective of mechanical decontamination is to reduce biofilm and thereby reduce inflammation.[140] However, the efficacy of existing treatment methods and protocols has been limited and resolution of inflammation has been infrequently achieved post-treatment.[35] Whereas earlier *in vitro* testing was performed on titanium objects such as coins and discs, contemporary research has shifted to employing anatomical models with titanium dental implants resembling the clinical conditions.[161, 172] From a clinical perspective, initiating treatment with non-surgical techniques is recommended as the primary phase, as it allows for control of inflammation before potentially advancing to more invasive surgical interventions.[75, 141] Furthermore, few clinical studies compare the treatment methods in studies with control groups.[181]

The overall research question, "Which mechanical decontamination protocol is the most effective for Pi management?" was addressed. The cumulative evidence presented in this thesis consistently corroborated the findings reported in the literature.[160, 182-188] When the outcomes of ***Study I*** were interpreted as measures of efficacy, comparing two mechanical devices in their capacity to remove occlusion spray—an analogue for biofilm—the parallels with the findings from ***Study II*** were evident. In the latter, the same devices were evaluated for their efficacy in removing a validated, dynamic biofilm from dental implant surfaces. Results in both studies agreed that achieving complete decontamination of the implant surfaces remains a challenge. Moreover, the studies collectively demonstrated efficient decontamination of smooth implant surface compared to the threaded implant surface when utilizing OCB. Conversely, using a US-PEEK tip emerged as a promising method in decontaminating threaded implant surfaces, although it altered the implant surface. With repeated mechanical debridement, damage prevention of the implant surface is an important key element, as alterations of the implant surface have been observed with prolonged debridement time.[189]

Non-surgical strategies for managing peri-implant diseases have proven effective at diminishing inflammation. However, achieving a complete resolution of the disease remains an uncertain outcome.[75, 190] A limited number of RCTs comparing exclusively on non-surgical, mechanical treatment of Pi have been conducted (Appendix). However, trials that combine mechanical treatment with adjunctive therapies, including laser or chemical antimicrobial interventions, seem more common.[181] Clinical investigations in ***Studies III*** and ***IV*** revealed a statistically significant decrease in PPD and BI ($p < 0.05$), alongside no further reduction of RBL in both groups, as observed at the six and twelve-month evaluations. Despite these advancements, inflammation remained as a prevalent observation at the end of the studies. It was also noted that there was no

statistically significant difference between the groups in reduction of PPD, BI, or the composite outcome. When applying the composite outcome criteria for determining disease resolution as advocated by Sanz and Chapple in 2012,[139] minimum number of implants met these criteria at the 6 and 12-month follow-up appointments.

Discussion of the main findings

Computer-aided design/computer-aided manufacturing (CAD/CAM)

In ***Study I***, a CAD/CAM fabricated 3D model was developed to assess the efficacy of mechanical or combined mechanical and chemical decontamination methods. The function of the model was tested by the removal of occlusion spray mimicking biofilm from titanium dental implants. Minimal variations were observed in the parts of the 3D printed models. A precise fit among the components was consistently demonstrated across all models. The insertion of implants was performed uniformly, revealing an identical number of threads on the respective implant sites (buccal, distal, palatal, and mesial). No occlusion spray on the non-exposed surfaces of the implants suggested a close seal between the model parts.

Additionally, the uniformity in the number of threads coated with spray across different sites on the models served as further evidence of the accuracy of the defects and the exactness of fit among the various models. The digital assessment of accuracy of the printed 3D models showed minimal variation between models (μm). These minor discrepancies could arise from the shrinkage during the models' final curing process.[191]

Manufacturing facilitated by computer technology can take the form of a subtractive method, as seen in milling processes, or employ an additive approach. Additive methods include techniques such as sintering, used with alloy materials, or 3D printing when working with resin-based materials. CAD/CAM milling and 3D printing techniques demonstrate comparable levels of precision.[192] In 3D printing, the level of accuracy is, amongst others, affected by the construction time. Furthermore, reducing the number of slices increases layer thickness and may result in a loss of accuracy.[191] *In vitro* models of Pi have been produced using CAD/CAM technology and 3D printing.[161, 172, 193] However, these models have been employed to test the efficacy of decontamination methods without prior validation of the models.[169, 171, 172, 194]

Validation of the anatomical 3D model

The 3D model was validated by assessing the accuracy of the 3D model, verifying occlusion spray as a biofilm analogue, and evaluating two machine-driven devices for mechanical decontamination.

The selection of the devices was based on their difference in mechanism of action. Moreover, a group in which implants were exposed to water, blank gel, and chlorhexidine gel (isopropanol-containing gel) were included to evaluate whether the occlusion spray was affected. In addition, the OCB was combined with water to evaluate whether or not the outcomes in the US-PEEK group could be attributed to the simultaneous use of water. The OCB was combined with a blank gel and an isopropanol-containing gel to evaluate whether the decontamination efficacy could be enhanced. The anatomical design allowed evaluation of accessibility and decontamination at the site level.

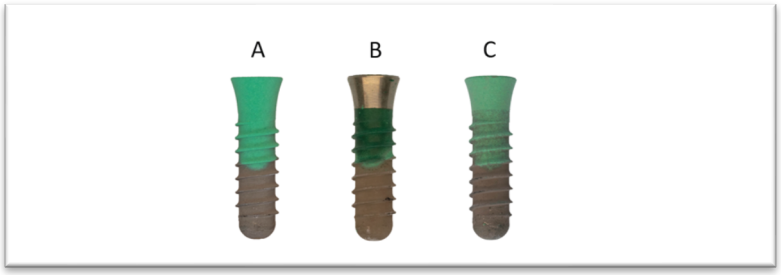


Figure 8. Characteristics of occlusion spray on the buccal surface of implants. *A* is the control implant, demonstrating the appearance of the intact spray. *Implant B*, subjected to decontamination with an oscillating chitosan brush, exhibits a clean machined area. On the threaded surface, the spray manifests as darker ground compared to the control implant, indicating that the spray particles are more tightly packed due to the decontamination process. *Implant C*, exposed to water spray, reveals a thinner spray pattern, allowing the implant surface to be visible through the spray.

The occlusion spray exhibited four distinct characteristics during application and removal (Figure 8): ***1)*** A pronounced green hue when densely accumulated by the instrument. ***2)*** The intensity diminished as the spray layer was reduced during decontamination, allowing the visibility of the implant surface through the spray layer. ***3)*** Partial spray removal maintained complete visibility of some implant areas, and ***4)*** Complete removal of the spray layer restored full visibility of the implant surface. However, when used on implant-retained supraconstructions, either the constructions were covered with the spray or partially removed,[193] as it appeared in the smooth implant surfaces in ***Study I***. This finding suggests that spray on rough surfaces displays more characteristics compared to smooth surfaces. Corroborating this, similar observations have been reported in studies employing ink as a substitute for biofilm, with either complete coverage by ink or partial exposure of the implant surface.[173, 194, 195] However, the distinctive visual inspection due to colour change, as seen with the occlusion spray, is not a feature reported in ink studies.[161, 173, 195] The colour transition indicates that while the instrument is in contact with the implant surface, it is ineffective in cleaning. These observations provide valuable perspectives when assessing the efficacy of mechanical decontamination combined with chemical agents designed to target biofilm and calculus. The assessment of accessibility, as determined by the amount of ink removed from

the implant surface by Sahrmann et al., may not only depend on the instruments' ability to reach the ink-stained area but also on its effectiveness in removing the ink.[173]

Additionally, the SEM images of the occlusion spray revealed solid inorganic particles embedded in a polymeric matrix. The composition morphologically resembled multispecies biofilm grown on titanium implant surface as demonstrated by Sanchez and co-workers.[166] The occlusion spray may serve as an effective *in vitro* simulation of true biofilm. It is important to note that while biofilm can be imitated with colour for study purposes, true biofilm is invisible to the operator.

The methods in ***Study I*** were employed to validate the anatomical 3D model. Analysis of residual occlusion spray on the implant surface indicated that the model was suitable for testing the accessibility and efficacy of decontamination protocols, whereas electron microscopy and optical profilometry could be used to reveal alterations in the implant surface topography following decontamination.

The possibility of quantifying residual occlusion spray at the site level was attributed to the anatomical design of the model and the marked surfaces on the implants. Further investigations of the implant surface confirmed that there was no damage to the portion of the implant embedded within the model, as the models' design allowed it to be opened prior to extraction and analysis. In contrast, with other models used for testing—whether anatomical or not—the implants were fixed in place and may sustain damage during removal.[161, 169, 173]

The removal and quantification of residual spray from the implant surface following decontamination was achieved. Analyses of implants treated with the OCB indicated that the fluorescence values were high for this group, corroborating the dark green hue observed during the visual examination post-decontamination. Fluorescence analysis was possible by dissolving and collecting the residual spray in an isopropanol solution. The reinforcement of findings by two separate analyses exemplifies the additional advantage of this methodology, a feature not documented in studies employing different biofilm simulation techniques to evaluate decontamination efficacy.[161, 195] Furthermore, fluorescence analysis could be used to train operators to apply a consistent amount of occlusion spray to implant surfaces. Achieving a uniform ink coating may be more feasible with dipping, provided the duration of immersion and the viscosity of the ink are maintained uniformly, as opposed to the potential irregularities when spraying the implant surface. However, a scientific comparison of the two methods has yet to be conducted.

A limitation of the model is that artificial mucosa was not included, which deviates from the non-surgical context. Additionally, because the model was not fixed within a phantom head, it does not

entirely replicate the conditions experienced in clinical practice. The outcomes of the anatomical model have not been compared with those from traditional methods using non-anatomic carriers for implants. However, a distinction is that the anatomical model allows for reporting results at the site level.

In vitro efficacy of decontamination methods on implants contaminated with multispecies biofilm

Mechanical decontamination of implants with a dynamic multispecies biofilm, as assessed in ***Study II***, showed a decrease in the bacterial presence. The qPCR results revealed that implants decontaminated with US-PEEK experienced a statistically significant reduction in four out of six bacteria: *S. oralis* (92.4%), *A. naeslundii* (85.8%), *V. parvula* (93.1%), *A. actinomycetemcomitans* (83.73%), *P. gingivalis* (84.32%), and *F. nucleatum* (83.64%) ($p < 0.05$). Similarly, Tong et al. conducted a comparative analysis of five treatment groups to evaluate the decontamination efficacy on implants explanted due to Pi.[97] The groups included saline irrigation, air-polishing with glycine powder, air-polishing with glycine powder combined with 17% EDTA, US-PEEK, and US-PEEK combined with 17% EDTA. Corroborating the outcomes in ***Study II***, Tong et al. demonstrated through post-decontamination SEM evaluation that US-PEEK effectively cleaned the implant surface. Furthermore, the decontamination efficacy of mechanical instruments applied on titanium discs seeded with a monospecies biofilm of *S. mutans* revealed superior decontamination for the US-PEEK group.[158] Additionally, electrochemical treatment of biofilm, aligning with the findings of ***Study II***, effectively reduced certain bacteria in a multispecies biofilm, though not uniformly across all species.[196]

Conversely, the OCB group displayed a marginal decrease in overall bacterial count compared to the control group, with the reductions lacking statistical significance for the bacteria studied. However, biofilm reduction was mainly noted on the machined collar for the OCB group, with minimal to no change in the threaded areas. This pattern of biofilm removal in the OCB group is consistent with findings in ***Study I***, where coloured occlusion spray removal was noted on the machined implant collar but not the threaded surfaces. This indicates that while the OCB reached all implant areas, its cleaning efficacy varied, being less efficient on the rougher, threaded surfaces. In contrast, an *in vitro* study assessed the decontamination of SLA implants contaminated with a static monospecies biofilm.[164] The study employed various methods, including OCB. The outcomes indicated a significant reduction in biofilm for all methods compared to the control. However, no significant difference in biofilm reduction was observed among the decontamination groups. While all instruments effectively accessed and cleaned the implant surfaces, the TC method induced alterations to the implant surface.

Surface alterations and material remnants are commonly observed following decontamination with instruments employed in treating peri-implant diseases.[24, 97, 162, 164, 174] In ***Study I***, SEM results showed surface alterations following decontamination with US-PEEK. In contrast, another *in vitro* study found that the implant surfaces remained intact after US-PEEK decontamination.[24] However, residues of PEEK were detected on the implants, a phenomenon consistent with findings from other *in vitro* studies.[162, 188]

In assessing implant decontamination efficacy, a distinction between results obtained for smooth and threaded implant surfaces is not always reported.[169, 173] Although, the impact of implant roughness and geometry on the decontamination process may provide important knowledge. Addressing this, a study conducted by Duarte et al. compared the bacterial adhesion of *S. sanguinis* to smooth and SLA surfaces after decontamination.[103] The investigation revealed alterations, particularly in smooth surfaces treated with metal curettes, resulting in surface changes and subsequently influencing biofilm adhesion. Despite the observed alterations, the biofilm adhesion on treated smooth surfaces remained markedly lower when compared to rough implant surfaces.

Moreover, the impact of combined mechanical and chemical decontamination on titanium discs contaminated with intraorally grown biofilm was assessed by Charalampakis et al., revealing that the remaining biofilm on the rough implant surface exhibited greater complexity and was characterized by a firmer attachment to the surface.[197] In contrast, the biofilm on the turned surface was described as more dispersed and less organized. These observations highlight distinctions in the biofilm characteristics that remain after decontamination on different implant surfaces and were also observed in ***Study II***, where treatment with US-PEEK led to a substantial reduction in biofilm quantity across all surface areas, encompassing threads and machined implant collar. In contrast, decontamination using OCB did not influence the structure and quantity of biofilm formed on rough surfaces. Conversely, OCB treatment resulted in a reduction of biofilm on the machined collar area, presenting scattered bacterial accumulations without a structured form as described by Charalampakis et al.[197] Moreover, in a study involving non-modified and modified titanium discs contaminated with *S. gordonii*, four different techniques were employed for instrumentation.[162] The non-modified titanium discs showed no residual bacteria across all treatment groups. However, significantly higher mean numbers of residual bacteria were observed on the modified discs.

The findings of ***Study II*** cannot be directly applied to clinical scenarios because of the *in vitro* nature of the study. Although an anatomical model was used, prosthetic supraconstructions and mucosa

were not added. Decontamination was performed on a bench, limiting the study's applicability to real-world clinical conditions.

Clinical outcomes of non-surgical peri-implantitis treatment

In ***Studies III*** and ***IV***, the OCB and TC groups exhibited PPD reductions at the six- and twelve-month follow-up. Similarly, as indicated by BI scores, BoP improved from baseline to six months and further to twelve months, with a decrease in sites with severe bleeding (BI 3) and an increase in no-bleeding sites (BI 0). At the six-month evaluation, pus was observed in 33.3% of the implants in the test group (OCB) and in 64.7% of the control group (TC). The presence of pus is indicative of ongoing bone destruction.[198] Moreover, the extent of pus has been linked to other markers of disease, including marginal bone loss and PPD.[199] In ***Studies III*** and ***IV***, pus was registered as present or absent without a grading scale based on severity. At the 12-month follow-up, both groups exhibited a statistically significant decrease in the presence of pus ($p < 0.05$). This observation suggests that while inflammation was frequently detected at six months, subsequent management of the inflamed sites mitigated inflammation over the 12-month period. However, the differences in PPD, BI, and pus improvements between the OCB and TC groups did not reach statistical significance, implying comparable efficacy of the treatments.

Few randomized clinical trials evaluate non-surgical mechanical treatment alone,[184, 186, 187] while laser or combined mechanical and chemical treatment is more frequently reported.[154, 182, 183, 185, 200-206] Assessing the efficacy of combined mechanical and chemical interventions does not allow for isolated evaluation of the impact of mechanical debridement.

A comparison of the efficacy of two mechanical methods in treating Pi was performed by Renvert et al.[184] Similar to ***Study III***, a manual approach (TC) was compared to a machine-driven instrument (ultrasonic Vector device) over a 6-month follow-up period. The study involved 17 patients in the TC group and 14 in the ultrasonic vector device group. While reductions in plaque and bleeding scores were observed compared to baseline, there was no change in PPD. The group differences were not statistically significant.

When reviewing RCTs focused on non-surgical treatment, with or without adjunctive therapies, a recurrent absence of statistical significance between the intervention and control groups is observed, as demonstrated in the Appendix. Additionally, maintaining a positive outcome one-year post-intervention appears to be challenging.[207]

A systematic review by Hashim et al. assessed the relationship between BoP and Pi.[208] Analysis of the included studies revealed a 24% probability of Pi in implants with bleeding. Bleeding was often

documented even when Pi was absent, suggesting factors such as mucosal trauma resulted from probing or PiM. Hence, using BoP as a diagnostic tool for Pi is associated with a considerable false-positive rate. Employing radiographic evaluation of bone alterations combined with clinical assessments to ensure accurate identification of peri-implant conditions and reporting a composite outcome is advised.[54, 139]

Sanz and Chapple advised reporting a composite outcome characterized by the absence of deep probing depths, no bleeding and no SoP.[139] However, despite the recommendations, the reporting of a composite outcome is often omitted in RCTs.[182, 183, 187, 201, 209] In ***Study III***, disease resolution was defined as the absence of peri-implant PPD ≥4 mm, no bleeding (BI 0) or SoP and no radiographic evidence of bone loss between baseline and six months. This criterion was reached for two patients in the test group and one in the control group. Additionally, in ***Study IV***, treatment success criteria were defined as ≤1 implant site with BI ≤1, absence of pus, PPD ≤5 mm, and the absence of progressive bone loss, and disease eradication was defined as PPD <4 mm, BI 0, and no increase in RBL compared to baseline. Treatment success criterium was met for three implants in the test group and one in the control group at 12 months. Although a reduction in inflammation, as seen through reduced PDs, BI, and pus, was commonly observed at 12 months, only one implant in the test group demonstrated complete disease eradication, with none in the control group.

In ***Study IV***, missing data was estimated through statistical imputation. Analyses of PPD and BI were conducted following the original study protocol using complete data sets and an ITT approach that included imputed data. Both analysis methods—the PP and ITT—yielded the same conclusion: There was no statistically significant difference between the treatment groups regarding PPD and BI reductions.

<u>Radiographic outcomes of non-surgical peri-implantitis treatment</u>

Radiographic assessments play a crucial role in assessing bone levels around dental implants. When presenting results from clinical studies evaluating the efficacy of non-surgical treatment methods, clinical findings are integrated with radiographic analysis.[182, 183, 186, 200, 210]

While incorporating radiological and clinical evaluations is advised for reporting in clinical research[139], a systematic review article assessing 159 studies demonstrated that 89% of the studies included data on PPD and 87% on BoP. However, radiological data was only reported in approximately half of the studies, at 49%.[211]

RBL assessments were conducted using peri-apical radiographs at baseline, and comparisons were made at the six- and twelve-month intervals. The criterion for study inclusion was a RBL ranging

from 2 to 4 mm. In ***Study III*** (n=38), the mean baseline RBL was recorded as 2.4 mm (±0.5) for the OCB group and 2.6 mm (±0.6) for the TC group. At the six-month follow-up, the mean RBL was observed to be 2.5 mm (±0.5) for the OCB group and 2.6 mm (±0.7) for the TC group. In ***Study IV*** (n=31), baseline RBL values were 2.4 mm (±0.7) for the OCB group and 2.9 mm (±0.5) for the TC group, with a change to 2.5 mm (±0.5) for OCB and 3.1 mm (±0.7) for TC at the 12-month evaluation. The periapical technique has been reported to result in an overestimation of RBL measurements by 0.3-0.4 mm.[212]

Study III showed a slight increase in RBL over six months. ***Study IV*** observed a similar baseline RBL for the OCB group but a higher initial RBL for the TC group at 2.9 mm. After 12 months, the OCB group increased by 0.1 mm, while the TC groups' RBL increased by 0.2 mm. Corroborating with results in ***Studies III*** and ***IV***, additional RCTs examining non-surgical treatment modalities demonstrate stable bone levels with minimal alterations in RBL at the six- and twelve-month follow-up.[182, 183, 185, 187]

Visual analogue scale

Pain during the first two days after Pi treatment, whether surgical or non-surgical, is a common response.[213] In ***Study III***, patients reported pain experience following the three months treatment using a VAS. On this scale, a score of 0 indicated no pain, while 10 represented the most severe pain imaginable. The form provided to participants included only a VAS for the patients to indicate their pain level. Despite research showing that shorter questionnaires typically yield higher response rates,[214] a low rate of responses were reached for VAS (22 out of 30 patients, or 57.9%). When analysing data with a low response rate, the results shall be interpreted cautiously due to potential weaknesses. A low response rate may reduce statistical power, increasing the risk of a Type II error.[215] Moreover, a low response rate could introduce *non-response bias*, suggesting that those who responded differed significantly from non-responders. Evaluating pain, it is a possibility that patients experiencing severe pain may be underrepresented in the response or the opposite. Although a statistical comparison of responders' and non-responders' characteristics is advised to assess differences among the groups,[216] was not performed in ***Study III***.

Among the non-respondents, the reason for non-response was ascertained for 7 out of 16 patients; it was noted that they were not inquired regarding VAS scores owing to anaesthesia administration pre-treatment. The underlying motivations for their preference for anaesthesia, whether attributable to prior dental experiences or pain during the intervention, were not reported. The factors contributing to the lack of response from the remaining nine individuals were not reported.

The VAS scores from two different studies suggest that the type of treatment for Pi may influence the level of pain experienced by patients.[186, 217] The mean VAS scores for the OCB and TC group in ***Study III*** were 2.9 (±1.93) and 3.4 (±2.09), respectively. While these differences were not statistically significant, a potential reason could be the low response rate affecting the results. Comparably, Hentenaar et al. reported that using erythritol powder was associated with less pain compared to piezoelectric ultrasonic scaling, although the difference was not statistically significant.[186] Meanwhile, Merli et al. showed that using glycine powder decontamination compared to mechanical debridement resulted in higher VAS scores for pain, which were noticeable during the procedure and a week later.[217]

Drop-out rate

A sample size calculation is fundamental to RCT design as it influences the reliability and ethical aspects of the study.[218] It ensures that the study reaches the power to detect a clinical effect when it exists. Without adequate power, a study may be unable to distinguish between the absence of an effect and the inability to detect the effect. From an ethical standpoint, it is important to enrol enough participants to have robust results but not to expose more participants than necessary to the risks of the trial.[219]

The RCTs in ***Studies III*** and ***IV*** were designed to ensure 80% power to detect a PPD change of 1 mm between the study groups. At six months, one patient was excluded from the test group for surgical intervention, while all patients in the control group completed the six-month follow-up. At the twelve-month follow-up, 18 patients were examined in the test group, while only 13 were examined in the control group. A comparative analysis of the baseline characteristics and clinical and radiographic data between patients who completed the study and those who withdrew was performed to address the low completion rate. The purpose of this comparison was to identify any consistent differences between the two groups. A comparison is reported in Tables 1 and 2 in ***Study IV***.

Additionally, an ITT analysis incorporating data imputation was carried out. The outcomes of this analysis were compared with those obtained from the complete case analysis (per protocol). It was observed that both the PP analysis and the ITT approach yielded similar results.

Methodological considerations

In ***Study I***, the primary objective was to evaluate a novel test method for decontamination efficacy. Despite bearing similarities to existing models, the proposed model distinguished through the split design, which facilitated easy removal of implants and subsequent reuse of the model. Moreover,

the model permitted an evaluation of decontamination, encompassing the extent of the implant surface covered by the spray and the analysis of residual spray by dissolving it from the implant surface. Established techniques such as image analysis, fluorescence analysis, SEM, and profilometry were employed to investigate the efficacy of the model. Similarly, the methodologies implemented in ***Studies II, III,*** and ***IV*** were validated and routinely applied in *in vitro* and clinical research.

The 3D model in ***Study I*** was created using the CADCAM technique and printed using resin material. ***Study II*** replicated the same model using autoclavable resin to demonstrate production variability. However, the autoclaved resin was not examined for bacterial contaminants. The CADCAM technique was selected because it allows easy customization and digital design modification. While plaster models have long been a standard in dentistry, 3D printing offers advantages in precision and the integration of a digital workflow.

The Pi defect was digitally designed following a recognized defect classification. [132] In future studies, an intraoral clinical scan of a Pi bone defect could provide a more realistic simulation. In the context of ***Studies I*** and ***II***, the resin models allowed for model reuse, unaffected by mechanical or chemical decontamination. The split design, easily assembled and opened, is another advantage enabled by resin, a feature not possible with the plaster models.

The use of an anatomical 3D printed resin model for *in vitro* decontamination assessment has been previously reported by Korello et al. However, a split design was not featured in their model.[172] In contrast to the model fabricated for this thesis, which excluded soft tissue simulation, Korello et al. included gingiva imitation, enabling the evaluation of non-surgical decontamination of the implant surfaces. Notably, the defect size chosen for the 3D model would typically be subject to surgical treatment in a clinical setting. This specific design was selected to demonstrate that asymmetrical defects can be created using the CAD technique. Additionally, the CAD approach facilitated precise implant placement.

The selection of occlusion spray as a biofilm analogue was based on its prior application in a decontamination study assessing prosthetic decontamination.[193] Two occlusion sprays, Okklufine® and Vita Cerec®, were applied to and tested on titanium discs. Okklufine® was selected over Vita Cerec®, as Vita Cerec was water-soluble and thus not suitable for testing decontamination methods involving irrigation (Figure 9). Additionally, an SEM analysis of a titanium disc coated with Okklufine was performed, revealing that the composition of the spray layer resembled biofilm (Figure 10).

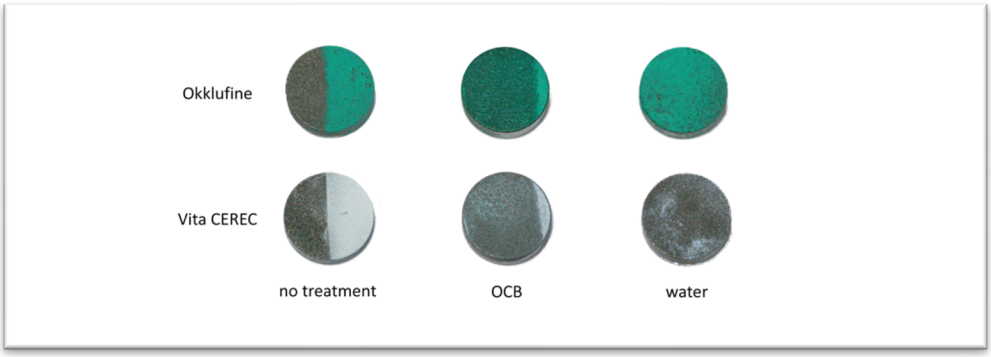


Figure 9. Two different occlusion sprays were tested on SLA titanium discs. Both sprays created a uniform spray layer and enabled assessment of decontamination with OCB. However, water irrigation removed the Vita CEREC layer almost completely and would therefore not allow assessing methods that incorporate water irrigation.

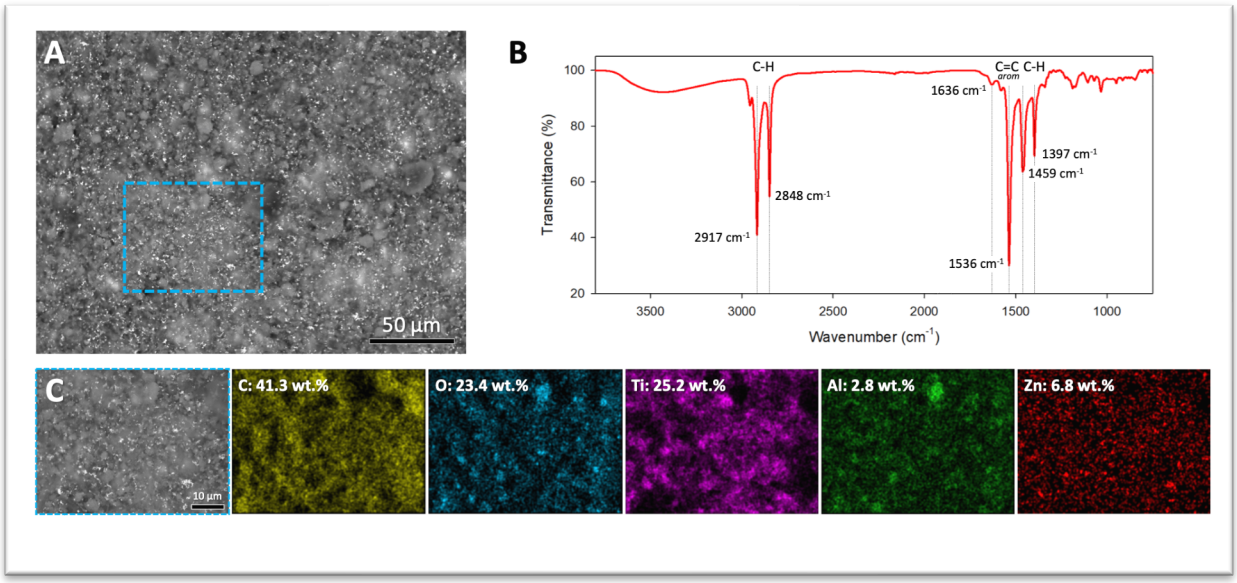


Figure 10. The occlusion spray is composed of microscale particles embedded within a polymeric matrix, resembling the 3D structure of a biofilm. *A*: Electron microscopy image of the occlusion spray on SLA titanium surface. *B*: Fourier-transform infrared (FTIR) spectrum of the organic matrix of occlusion spray, indicating that the polymeric matrix is composed of vulcanised styrene-butadiene rubber[220, 221] *C*: EDX mapping of the elemental composition of the occlusion spray within the area highlighted in *A*. In addition, small amounts of sulphur (0.28 wt.%) and chlorine (0.16 wt.%) were detected.

Decontamination of implant surfaces seeded with biofilm is a commonly applied method for *in vitro* efficacy evaluation.[158, 160, 222, 223] For this purpose, discs, coins, or implants have been cultivated with mono- or multispecies biofilm. While the early models consisted of static biofilms, the models have developed into multi-dimensional, dynamic biofilm models over time.[169, 224] The method employed in ***Study II*** has undergone a gradual refinement, evolving into a validated and widely adopted approach.[166, 169, 222, 225, 226] The aim of developing a dynamic, multispecies biofilm was to *in vitro* simulate conditions faced by oral bacteria, including factors such as temperature, saliva, pH, and other chemical conditions.[226, 227] Six bacterial species from the subgingival microbiota were included in the biofilm. These six bacteria consist of early, intermediate, and late colonizers.[227] The

multi-dynamic biofilm has been characterized and validated through SEM, confocal laser microscopy, bacteria culture, and qPCR.[166, 227, 228] In all experiments in ***Study II***, test implants were incorporated to assess the biofilm quality. Despite the proven efficacy of the dynamic multispecies method, the cultivation process remains sensitive, necessitating verification.

Four regions of interest were decided for SEM imaging of test and control implants: the machined implant collar, the interface area, the threads, and the valleys. Bermejo et al. further divided the implant threads into specific regions and demonstrated variations in microbial types depending on the location of the thread.[169] ***Study II*** did not conduct a specific analysis, where threads were divided into distinct surfaces.

The decision to utilize tissue-level implants was influenced by the compatibility of the bioreactor with narrow neck implants and the versatility of the implant, enabling the assessment of machined surface without threads and moderately rough threaded surface. Implants seeded with biofilm were mounted to the anatomical 3D models before decontamination treatment. The 3D models were printed in autoclavable resin and autoclaved before use, ensuring no bacterial contaminants on the models.

The objective of integrating two validated methods was to replicate the clinical scenario better. However, it is crucial to recognize that the clinical situation encompasses numerous components that cannot be fully simulated in *in vitro* testing.

In ***Studies III*** and ***IV***, the primary objective was to assess the efficacy of non-surgical treatment of Pi using a chitosan brush. The examination interval adhered to the consensus report on clinical research on Pi, aligning with the recommended short-term follow-up of 1-3 months post-treatment, a subsequent long-term follow-up of 6-12 months, and a recall interval of six months or shorter.[61, 139] Clinical evaluations at 3, 6, and 12 months are recommended to include PPD, BoP, and SoP assessment.[75, 139] The examination intervals in ***Studies III*** and ***IV*** align with other RCTs comparing the efficacy of non-surgical treatment methods for Pi.[154, 200, 229, 230] A three-month follow-up interval has been reported in RCTs assessing the efficacy of Pi treatment.[206, 231, 232]

The longitudinal study design may be affected by the Hawthorne effect, characterized by increased patient compliance, encompassing lifestyle modifications and enhanced oral hygiene throughout the study duration.[233] To mitigate this effect, patients may be blinded to treatment allocation. This blinding strategy may be implemented to control the potential influence of patient behaviour during the study.

A study with a 12 month follow-up may introduce the risk of publication bias if the study is fragmented to highlight positive short-term outcomes while postponing less favourable results at a subsequent stage.[234] The risk is particularly relevant when complete data is available, yet only short-term data is reported. Conversely, the advantages of short-term data are evident when reaching out to clinicians in instances where novel treatments exhibit adverse effects, and initial findings are important in discontinuing an intervention, such as in the case of pharmaceutical interventions. Nevertheless, the OCB had already been tested in a 6-month multicentre study, which included 63 patients diagnosed with initial Pi.[179] In ***Study III***, the data obtained at the 6-month follow-up was analysed while the study was still ongoing, reducing the likelihood of introducing bias.

Disease classification

Pi was defined as an osseointegrated implant with 2-4 mm RBL, BI ≥2, and PPD ≥4 mm. The study protocol was established before the publication of consensus on case definitions and diagnostic criteria for peri-implant diseases in 2018.[54] According to the guidelines, in cases where initial radiographs and probing depths are unavailable, the presence of RBL ≥3 mm and PD ≥6 mm, along with bleeding, indicates Pi.

As patients included in ***Studies III*** and ***IV*** were referred to specialist practices for investigation and treatment, in many instances, baseline data was missing. The disease definition for cases with missing initial data would have been applied if the study had been designed after the publication of the consensus report. This implies that the applied Pi definition may represent a less severe Pi in comparison to the current classification. Additionally, Pi was categorized as mild to moderate. However, no classification system currently divides the disease based on severity.

Comparing to existing literature, RBL change has been reported in terms of the count of implant threads experiencing bone loss rather than indicating the direct measurement of bone change.[235-237] Furthermore, Pi has been defined as RBL values ranging from as low as 0.5 mm,[62, 238] to 5 mm as reported by Zetterqvist et al.[239] The practice of delimiting RBL within specific upper and lower values appears to be infrequent.[62, 237, 240] However, delimiting hinders the impact of extreme values on mean calculations. A common strategy to address this involves establishing a specific threshold for RBL values and designating higher values as an exclusion criterion.

For PPD, including PPD ≥5 mm seems to be most frequently applied in Pi studies.[236, 237, 239, 241] Additionally, the criterion of PPD ≥4 mm seems commonly used.[80, 242, 243] A number of disease classifications do not incorporate PPD evaluation within the criteria.[81, 235, 244, 245]

In ***Studies III*** and ***IV***, the primary outcome was a change in PPD. The secondary outcome variables included changes in BI, pus, and RBL. Additionally, patient-reported pain during treatment was a secondary outcome in ***Study III***. A recent systematic review evaluating outcome measures in clinical research on peri-implant diseases reported that a primary outcome was not specified in more than half of the studies.[211] Among studies that did incorporate a primary outcome, the majority focused on one primary outcome. Consistent with ***Studies III*** and ***IV***, the primary outcome most commonly reported was PPD, followed by BoP and plaque. The studies were designed before the consensus reports published in 2018. In retrospect, it was realized that BoP would have been a better primary outcome as bleeding/no bleeding allows to evaluate inflammation, while changes in PPD indicate disease severity.

Sample size estimation

A sample size calculation was performed based on 80% power to detect a 1 mm PPD change between the test and control group before the study start. Including a 20% dropout rate, 17 patients were advised pr group. A limitation in ***Studies III*** and ***IV*** lies in the methodology used to calculate sample sizes, which primarily relied on group differences assessed through mean values and t-tests. However, a multilevel analysis was used to compare the groups. Calculating sample sizes for multilevel analyses, such as ANOVA, introduces a higher level of complexity due to the inclusion of additional factors in the computation.[219]

In the context of a two-sample t-test, the calculation incorporates effect size (1 mm difference in PPD), the significance level representing the likelihood of a Type I error (set at 0.05), the desired power indicating the probability of detecting an actual effect (80%), and the standard deviation for both groups. Conversely, multilevel analyses necessitate the inclusion of additional parameters, such as the intraclass correlation for repeated measures and the number of groups in the study.[246] The calculated sample size included insufficient number of patients to reach 80% power. Interpretations of the findings from underpowered studies should be approached with caution, given the elevated risk of not being able to detect an actual effect if present.[219]

The sample size calculation based on a 1 mm difference in PPD could be debated given that the benefits of non-surgical treatments tend to be modest and frequently fall below a 1 mm PPD reduction. Detecting smaller effect sizes and reaching statistical significance, a larger sample size is required. The selected effect size of 1 mm aligns with previous studies,[230, 247] while, Renvert et al. employed 0.6 mm difference in PPD for sample size calculation.[184]

Exclusion criteria

Conducting studies with broad inclusion criteria promotes the generalizability of results to diverse patient populations. The research question influences the selection of eligibility criteria in RCTs. However, excessive use of restrictive exclusion criteria and the exclusion of prevalent comorbidities may lead to selection bias and pose a risk to the external validity of the trial.[248] The CONSORT guidelines recommend to state eligibility criteria under the method section of the study.[249]

In ***Studies III*** and ***IV***, 13 exclusion criteria were defined. Initially, type 2 diabetes was proposed as an exclusion criterion. However, it was removed because type 2 diabetes does not serve as a contraindication for dental implant treatment. Excluding this patient group from the RCT would impact the study's generalizability.

Exclusion criteria 2, 3, and 12 were applied, targeting cases where clinical assessments were inaccessible due to prosthetic limitations, unresolved technical complications contributing to Pi, and prosthetic constructions leading to non-balanced traumatic occlusion. In managing Pi, a pre-treatment phase that reduces risk factors and improves plaque-accumulating prosthetic design is emphasized.[75] Resolution of technical contributing factors is essential before inclusion as the non-hygienic prosthetic form may potentially lead to attachment loss around the implant. Consequently, patients with poor prosthetic design and occlusal overload — considered local risk indicators impacting plaque control and exacerbating Pi — were excluded. Similarly, John et al. excluded patients with prosthetic risk indicators.[247] Furthermore, they implemented strict inclusion criteria, excluding patients with risks such as KM <2 mm and untreated periodontal disease. Similarly, patients with active periodontal disease were excluded from ***Studies III*** and ***IV***. Using general and local risk indicators and -factors to define exclusion criteria seems common in clinical studies.[154, 202, 206, 247]

Multicentre RCT

Studies III and ***IV*** were conducted in five dental specialist practices. Each clinic had a specialist in periodontology and a dental hygienist who participated as an examiner and an interventionist, respectively. The five clinics involved in the study were Public Dental Health Services in Kristianstad; Public Dental Health Services in Örebro; Hälsohögskolan, Jönkøping University; Spesdent; Bjerke Tannmedisin; and Oris Dental Madla.

Recruiting patients from a broader population is one of the main advantages of a multicentre study design.[250] Particularly for rare conditions, including adequate number of patients to achieve statistical power can be challenging.[139] In many cases, single-centre trials suffer from the recruitment of an insufficient number of participants, and thereby, the significant risk of failing to demonstrate treatment differences when they do exist, known as type II errors.[215] Additionally, the

participation of a broader range of dental staff across different centres, including multiple contributors in reviewing the study protocol, enhances the validity of the results and improves generalizability.[251,250]

However, the use of multicentre studies to evaluate the efficacy of invasive treatments or medical devices, the reliability has been questioned because of variations among clinicians.[251] Furthermore, conducting multicentre trials presents complexities in coordination, quality control, and data management compared to single-centre trials.[251] Adherence to the study protocol, established prior to the study's initiation, is crucial. However, achieving these objectives can be challenging due to the large number of clinicians involved and the geographical distances between centres. Meetings with the co-investigators during the data collection period were planned for ***Studies III*** and ***IV*** to ensure quality. These meetings were set as a platform for addressing any queries or challenges raised by participants, ranging from technical challenges with the online form to specific inquiries about patient inclusion, screening, and treatment. Additionally, the participants could contact the coordinators for assistance or clarification.

Consistent execution through practical calibration is vital in studies involving multiple practitioners, examiners, or repeated measures.[180, 252-254] Inter- and intra-examiner calibration is important in maintaining accuracy. Before the commencement of ***Studies III*** and ***IV***, a calibration meeting was conducted to review the study protocol. However, practical calibration training was not performed, as the examiners were experienced board-certified periodontologists and because of the geographical distance. Nevertheless, an assessment of intraclass correlation (ICC) was carried out for the obtained data. For BI, 10.3% of the variability was attributed to differences between clinics, while 22.2% of the variability was explained by patients nested within clinics. In the case of PPD, 7.7% of the variability was associated with differences between clinics, and a significant 51.2% of the variability was nested within patients.

Moreover, blinding of the interventionists was not possible in ***Studies III*** and ***IV***. This absence of blinding raises concern regarding potential treatment bias, given that the clinicians' personal preferences or dislikes may affect their performance, consequently impacting the study outcomes and validity of the findings. Treatment bias occurs when the knowledge of the treatment being administered influences the behaviour of those involved in the study.

Random assignment improves the internal validity of the study by ensuring the absence of systematic differences among participants in each group. Although a difference in baseline radiographic level was observed between the groups, 2.4 mm in the OCB group and 2.9 mm in the TC group, the CONSORT guidelines advise not to perform significance testing of baseline data.[249]

Moreover, the allocation led to an uneven composition of the groups, particularly evident at the 12-month follow-up, with 18 patients in the OCB group and 13 in the TC group. The multicentre trial design aspect was considered during the preparation of the randomization lists. However, the randomization process was not actively monitored by the administrator. In addition to the low power, the imbalance in group sizes introduces bias that complicates the interpretation of the results and is a limitation in ***Studies III*** and ***IV.***

Peri-implant assessment

At baseline and to assess the outcomes of non-surgical Pi treatment, residual inflammation was monitored at follow-up visits. Probing was performed to assess the presence of plaque, BoP and PPD. Lang et al. conducted an animal study assessing probe penetration in clinically healthy peri-implant tissues, PiM, and Pi.[70] The study suggested that probing is a valuable indicator for assessing the health or disease status of tissues around implants, supporting it as clinical diagnostic procedure for monitoring peri-implant tissues.

The implant-retained supraconstructions were not removed before clinical evaluation or treatment. Although baseline data and follow-up results were achieved the same way, not removing the supraconstructions presents a limitation for both assessment and treatment accessibility, potentially leading to inaccurate measurements.

Clinical assessment of peri-implant tissues was performed according to recognized methods. A probe with a 0.5 mm diameter tip and probing force of 0.2N is advised to assess peri-implant tissues.[75] ***Studies III*** and ***IV*** examined plaque accumulation, BI, and PPD at six distinct sites per implant, consistent with analogous studies evaluating peri-implant tissues.[182, 187, 254] However, measuring six surfaces per implant may pose challenges due to the anatomical differences between the supraconstructions and the circular implant. In contrast, ***Study I*** conducted assessments at four implant sites. Despite utilizing the same anatomical 3D model in ***Studies I*** and ***II***, a site-level analysis was not performed in ***Study II***. Nevertheless, a site-level analysis may be incorporated in future studies.

BoP was reported as BI, including four stages. In ***Studies III*** and ***IV***, Pi cases with BI ≥2 were included. As the BI measures the severity of BoP, it was decided to ensure the inclusion of cases where the inflammatory component was unequivocal, thereby avoiding the potential inclusion of cases with a single site with an isolated bleeding spot (BI 1), which may result from the probing itself. Spot bleeding is not considered a definitive sign of inflammation and might be a result of trauma to the peri-implant tissues. A pressure-sensitive probe was not employed, introducing a potential risk of trauma. Aligning with the consensus report of World Workshop on the

Classification of Periodontal and Peri-Implant Diseases and Conditions, caution is advised in interpretating bleeding spots.[67] Implants with BI 1 were retreated at three months in cases with PPD ≥4 mm was present. Active treatment combined with supportive treatment is reported in several other studies.[206, 255] According to the study protocol, the two modalities of active treatment have been merged with supportive treatment over the study periods.

In ***Studies III*** and ***IV***, bleeding was reported using a graded BI with scores between 0 and 3. A three-level logistic regression model was employed (level 1=time, level 2=patients, and level 3=clinics). Within- and between-group changes in BI were analysed using a two-way interaction of time with groups. Furthermore, ***Study IV*** utilized a three-state Markov model to follow the changes across different BI states. BI 0 represented health, BI 1 denoted an intermediate state between health and illness, and BI 2 and BI 3 were consolidated into a single state indicative of disease. Each of these states were treated as temporary. The Markov analysis was performed at the site level to follow changes in BI. Site-level analysis was chosen as BI data was recorded as an ordinal categorical variable and reported on the site level. In addition, transitions analysis on implant level was performed based on the highest BI value at baseline and 12 months. At the site level, a transition between all states was observed during the study period. Despite receiving treatment and follow-ups, some sites progressed from healthy to inflamed in both groups, as depicted in Figure 5 in ***Study IV***. Transitions in BI stages at the implant level from baseline to the 12-month interval, the implants were represented with the highest BI score. The results showed that a limited number of implants decreased in BI from stage 2/3 to BI 0, signifying an absence of bleeding. Conversely, the majority of implants registered a progression towards BI 1, denoting a shift to a less severe, transient inflammatory state. Furthermore, while the OCB and TC groups showed tendencies for progression and regression of peri-implant bleeding, distinct variations between the groups emerged. The TC group had a higher retention of the initial healthy state but also showed a substantial progression from BI0 directly to the disease states. The OCB group, in contrast, exhibited more of a gradient transition, with noticeable movement from BI 0 through BI 1 and onto BI 2/3.

Although logistic regression analysis indicated a decrease in the number of implants at the BI 3 and an improvement to BI 0, the Markov model revealed a more detailed pattern. It demonstrated that BI values fluctuated despite ongoing treatment and follow-up, moving not only towards healthier stages but also towards inflammation.

While Markov models has not been employed to analyse changes in clinical parameters in Pi treatments, it has been used to assess the economic evaluation of the treatments. The cost-

effectiveness of strategies for preventing and treating Pi by considering the financial implications and balancing the treatment costs against the likelihood of shifting between health states over time has been evaluated.[256] The idea is to integrate the economic aspects with clinical decision-making.

BoP was documented as an ordinal categorical variable featuring four distinct categories; this data was used for transition analysis, showing the transitions between the various stages (Markov models). Given that BI was recorded at the site level, the Markov analysis was conducted at the site level. Moreover, implant-level analysis was incorporated, driven by its clinical relevance. While Markov analysis in ***Study IV*** focuses on the immediate transition of health states post-treatment, it has previously been employed to evaluate the risk factors associated with periodontitis disease progression[257] and the cost-effectiveness of treatments in relation to their clinical effectiveness in Pi treatment.[256] Collectively, these studies demonstrate the versatility of Markov models in dental research, enabling the analysis of transitions between health states, risk factor identification, and economic evaluations of treatment strategies.

Radiographic assessment

Radiographic methods, including panoramic tomography and intraoral radiography are reliable for assessing marginal bone levels.[258] In ***Studies III*** and ***IV***, RBL was evaluated at three time points: baseline, six, and 12 months. The measurement of RBL was conducted on calibrated peri-apical radiographs. To ensure accuracy, digital radiographic measurements were performed using an ImageJ plugin developed for semiautomatic assessment for radiographic attachment level.[259]

To maintain objectivity, the investigators were blinded to the radiographs, and information regarding group affiliation or the timing of radiograph acquisition was eliminated. Thus, the investigators remained unaware whether the examined radiographs corresponded to the baseline, 6-month, or 12-month time point. To enhance reliability, three investigators independently evaluated all radiographs three times. There was a time interval of 14 days between the evaluations. The distance between the implant-abutment junction and the most coronal bone to implant contact was measured, as illustrated in Figure 11. However, panoramic tomography and intraoral radiographs obtained with long-cone parallel technique, do not allow the assessment of the buccal and palatal/lingual bone.

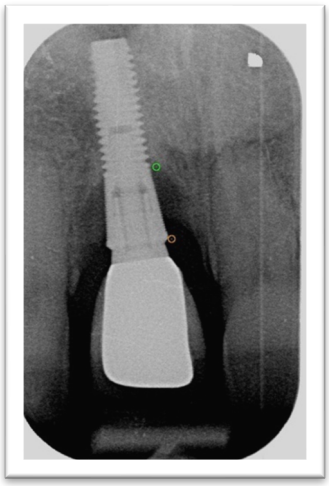


Figure 11. Radiographic measurement of bone level. The implant-abutment junction is denoted by the red circle, while the most coronal bone-to-implant contact is indicated by the green circle. Shared by patients' permission.

A limitation of the study pertains to the lack of standardization of radiographs taken at different study time points as reported in other RCTs.[187, 254]Although, the literature describes the use of individualized design for radiographic film holders, ensuring consistency across all recordings, it was not used in the present study. In addition, due to the multicentre approach, the radiographs were obtained by many operators and X-ray equipment.

Ethical Considerations

Studies I and ***II*** were preclinical, *in vitro* investigations. Implants in both studies were assigned numbers, ensuring investigators were unaware of the group assignments during analysis.

Studies III and ***IV*** complied with the principles of the Declaration of Helsinki and Good Clinical Practice (GCP) guidelines for medical devices. Ethical approvals were obtained from the Regional Committee for Medical and Health Research Ethics, South-East Norway (REK sør-øst 2017/710) and the Swedish Ethical Review Authority, Linköping, Sweden (EPN 2017/36-31).

The studies were registered on clinicaltrials.gov (12/08/2017, NCT03373448). All examination data, except radiographs and VAS forms, were digitally stored using standardized forms. Services from Tjenester for Sensitive Data (TSD, University of Oslo) were utilized to ensure confidentiality and secure handling of sensitive information. Encryption protocols were applied to align with personal data processing regulations.

Before the baseline examination, patients were informed about the study aim and objectives, and a written consent was signed. The possibility to withdraw from the study at any point was communicated. Examiners were instructed to complete a termination form whenever they excluded patients. This form required to specify the reason for exclusion and to note any adverse effects that may have occurred.

Decontamination with TC was chosen as the control treatment to maintain ethical standards, as administrating no active treatment to the control group would be unethical. The selection of TC as the control treatment was based on its widespread use by clinicians for non-surgical Pi treatment. It is important to emphasize that the test and the control treatment employ fundamentally different mechanisms of action, thereby potentially resulting in varying treatment outcomes. Specifically, the chitosan brush is attached to a machine-driven, oscillating handpiece, while the TC is a purely manual instrument devoid of any machine component.

Dr. Caspar Wohlfahrt holds a patent and has financial interests as a shareholder in the investigated medical device. This conflict of interest was reported in ***Studies III*** and ***IV***. Furthermore, the reporting adhered to Consolidation Standards of Reporting Trials (CONSORT) guidelines for RCTs.[249]

Conclusions

The general hypothesis of this thesis was partly rejected. While the OCB device demonstrated effective decontamination of the machined implant collar *in vitro*, it did not show superior efficacy in decontaminating the threaded implant area covered with multispecies biofilm compared to US-PEEK. However, clinical assessments at six and twelve months following non-surgical Pi treatment showed comparable efficacy for OCB and TC. Nonetheless, OCB did not exhibit statistically significant better outcomes compared to TC.

Specific conclusions:

- employing the 3D model enabled a realistic simulation of implant surface decontamination using a coloured occlusion spray as a biofilm mimic (***Study I***)
- the application of the 3D model allowed studying the accessibility and alterations of the implant surfaces following both mechanical and chemical decontamination (***Study I***)
- the 3D model permitted post-decontamination analysis of implants at site level (***Study I***)
- alterations in the occlusion spray provided a means to identify regions with incomplete decontamination or inaccessible surfaces (***Study I***)
- Mechanical decontamination of implant surfaces using US-PEEK demonstrated reduction of dynamic, multispecies biofilm in an *in vitro* 3D model. The OCB device exhibited biofilm reduction on machined implant surfaces. (***Study II***)
- At six months, non-surgical treatment of Pi with OCB or TC exhibited comparable efficacy, with no statistically significant distinction between the interventions. Achieving disease eradication remained uncertain for both groups. (***Study III***)
- At 12 months, non-surgical treatment of Pi using OCB or TC revealed no statistically significant differences between the groups. Both groups exhibited clinical improvements, including disease resolution in some cases. (***Study IV***)

Clinical Implications

In this thesis, a novel instrument was implemented and compared to commonly used clinical approaches in mechanical, non-surgical treatment of Pi. Despite multiple treatments, Pi resolved in a limited number of cases after 12 months. Stable radiographic bone levels suggested no disease progression, yet inflammation signalled the need for continuous evaluation and treatment.

The developed anatomical 3D model, facilitating site-level analysis of non-symmetrical defects, proved beneficial. Visual inspection and qualitative analysis describe access in instances where the instrument fell short of achieving complete decontamination.

It is crucial to have validated *in vitro* models that closely mimic the oral cavity. These models support clinically relevant environments and may aid development of medical devices and allowing for refinements before clinical application.

Future Perspectives

The management of Pi presents considerable challenges. Despite the efforts of diligent patients to maintain a plaque-free environment and the implementation of a planned treatment approach by skilled professionals, relapse remains a recurring issue. Given the high prevalence of peri-implant diseases and the high rates of both technical and biological complications related to implant treatment, it is strongly advised to preserve natural dentition for as long as possible. Therefore, it becomes paramount to prioritize patient education and guidance regarding preventive measures, minimally invasive prosthetic techniques, and biologically driven therapies to prolong the lifespan of natural teeth.

In the context of implant treatment, an approach to mitigate the risk of peri-implant diseases involves the identification and exclusion of patients who possess a predisposing risk profile. Additionally, precise, prosthetic-driven implant placement while adhering to hygienic design principles during the fabrication of implant-retained supraconstructions is important. These measures form a comprehensive strategy to reduce peri-implant disease prevalence. Furthermore, it is imperative to continue exploring novel methods and combinations that effectively decontaminate the implant surface and do not leave remnants while ensuring the integrity of the implant surface.

To enhance the clinical translation of the *in vitro* studies, 3D models with supraconstructions and insertion of the model into a phantom head should be considered prior to decontamination. Based on the findings from the present *in vitro* studies, RCTs that combine non-surgical mechanical treatment with adjunctive therapies should be designed. Future clinical studies should consider longer follow-up periods to evaluate the long-term efficacy. Additionally, studies should aim for larger sample sizes to enhance the statistical power.

References

1. Whiteford HA, Degenhardt L, Rehm J, et al. Global burden of disease attributable to mental and substance use disorders: findings from the Global Burden of Disease Study 2010. *Lancet* 2013;382:1575-1586.
2. Roberto LL, Crespo TS, Monteiro-Junior RS, et al. Sociodemographic determinants of edentulism in the elderly population: A systematic review and meta-analysis. *Gerodontology* 2019;36:325-337.
3. Albrektsson T, Dahlin C, Jemt T, Sennerby L, Turri A, Wennerberg A. Is marginal bone loss around oral implants the result of a provoked foreign body reaction? *Clin Implant Dent Relat Res* 2014;16:155-165.
4. Brånemark PI, Adell R, Breine U, Hansson BO, Lindström J, Ohlsson A. Intra-osseous anchorage of dental prostheses. I. Experimental studies. *Scand J Plast Reconstr Surg* 1969;3:81-100.
5. Stein AL, Lygre GB, Reichhelm I, Eggum E, Bull VH, Gjengedal H. Data fra Helfo og Norsk pasientskadeerstatning gir liten informasjon om kvalitet og omfang av behandling med tannimplantater i Norge. *Nor Tannlegeforen Tid* 2019;130:776-782.
6. Ellingsen JE, Thomsen P, Lyngstadaas SP. Advances in dental implant materials and tissue regeneration. *Periodontol 2000* 2006;41:136-156.
7. Albrektsson T, Donos N. Implant survival and complications. The Third EAO consensus conference 2012. *Clin Oral Implants Res* 2012;23 Suppl 6:63-65.
8. Heitz-Mayfield LJ, Huynh-Ba G. History of treated periodontitis and smoking as risks for implant therapy. *Int J Oral Maxillofac Implants* 2009;24 Suppl:39-68.
9. Salvi GE, Carollo-Bittel B, Lang NP. Effects of diabetes mellitus on periodontal and peri-implant conditions: update on associations and risks. *J Clin Periodontol* 2008;35:398-409.
10. Misch CE, Perel ML, Wang HL, et al. Implant success, survival, and failure: the International Congress of Oral Implantologists (ICOI) Pisa Consensus Conference. *Implant Dent* 2008;17:5-15.
11. Albrektsson T, Zarb G, Worthington P, Eriksson AR. The long-term efficacy of currently used dental implants: a review and proposed criteria of success. *Int J Oral Maxillofac Implants* 1986;1:11-25.
12. Lang NP, Berglundh T, Heitz-Mayfield LJ, Pjetursson BE, Salvi GE, Sanz M. Consensus statements and recommended clinical procedures regarding implant survival and complications. *Int J Oral Maxillofac Implants* 2004;19 Suppl:150-154.
13. Pjetursson BE, Heimisdottir K. Dental implants - are they better than natural teeth? *Eur J Oral Sci* 2018;126 Suppl 1:81-87.
14. Pjetursson BE, Brägger U, Lang NP, Zwahlen M. Comparison of survival and complication rates of tooth-supported fixed dental prostheses (FDPs) and implant-supported FDPs and single crowns (SCs). *Clin Oral Implants Res* 2007;18 Suppl 3:97-113.
15. Chackartchi T, Romanos GE, Sculean A. Soft tissue-related complications and management around dental implants. *Periodontol 2000* 2019;81:124-138.
16. Araujo MG, Lindhe J. Peri-implant health. *J Clin Periodontol* 2018;45 Suppl 20:S230-s236.
17. Berglundh T, Zitzmann NU, Donati M. Are peri-implantitis lesions different from periodontitis lesions? *J Clin Periodontol* 2011;38 Suppl 11:188-202.
18. Berglundh T, Persson L, Klinge B. A systematic review of the incidence of biological and technical complications in implant dentistry reported in prospective longitudinal studies of at least 5 years. *J Clin Periodontol* 2002;29 Suppl 3:197-212.
19. Berglundh T, Lindhe J, Marinello C, Ericsson I, Liljenberg B. Soft tissue reaction to de novo plaque formation on implants and teeth. An experimental study in the dog. *Clin Oral Implants Res* 1992;3:1-8.
20. Monje A, Insua A, Wang HL. Understanding Peri-Implantitis as a Plaque-Associated and Site-Specific Entity: On the Local Predisposing Factors. *J Clin Med* 2019;8.
21. Monje A, Amerio E, Cha JK, et al. Strategies for implant surface decontamination in peri-implantitis therapy. *Int J Oral Implantol (Berl)* 2022;15:213-248.
22. Salvi GE, Aglietta M, Eick S, Sculean A, Lang NP, Ramseier CA. Reversibility of experimental peri-implant mucositis compared with experimental gingivitis in humans. *Clin Oral Implants Res* 2012;23:182-190.
23. Heitz-Mayfield LJ, Needleman I, Salvi GE, Pjetursson BE. Consensus statements and clinical recommendations for prevention and management of biologic and technical implant complications. *Int J Oral Maxillofac Implants* 2014;29 Suppl:346-350.
24. Cha JK, Paeng K, Jung UW, Choi SH, Sanz M, Sanz-Martin I. The effect of five mechanical instrumentation protocols on implant surface topography and roughness: A scanning electron microscope and confocal laser scanning microscope analysis. *Clin Oral Implants Res* 2019;30:578-587.
25. Albrektsson T, Wennerberg A. On osseointegration in relation to implant surfaces. *Clin Implant Dent Relat Res* 2019;21 Suppl 1:4-7.
26. Brånemark PI, Adell R, Albrektsson T, Lekholm U, Lindström J, Rockler B. An experimental and clinical study of osseointegrated implants penetrating the nasal cavity and maxillary sinus. *J Oral Maxillofac Surg* 1984;42:497-505.

27. Albrektsson T, Jacobsson M. Bone-metal interface in osseointegration. *J Prosthet Dent* 1987;57:597-607.
28. Brånemark PI. Osseointegration and its experimental background. *J Prosthet Dent* 1983;50:399-410.
29. Albrektsson TO, Johansson CB, Sennerby L. Biological aspects of implant dentistry: osseointegration. *Periodontol 2000* 1994;4:58-73.
30. Moon IS, Berglundh T, Abrahamsson I, Linder E, Lindhe J. The barrier between the keratinized mucosa and the dental implant. An experimental study in the dog. *J Clin Periodontol* 1999;26:658-663.
31. Berglundh T, Lindhe J, Ericsson I, Marinello CP, Liljenberg B, Thomsen P. The soft tissue barrier at implants and teeth. *Clin Oral Implants Res* 1991;2:81-90.
32. Berglundh T, Abrahamsson I, Welander M, Lang NP, Lindhe J. Morphogenesis of the peri-implant mucosa: an experimental study in dogs. *Clin Oral Implants Res* 2007;18:1-8.
33. Tomasi C, Tessarolo F, Caola I, Wennström J, Nollo G, Berglundh T. Morphogenesis of peri-implant mucosa revisited: an experimental study in humans. *Clin Oral Implants Res* 2014;25:997-1003.
34. Tonetti MS, Imboden M, Gerber L, Lang NP. Compartmentalization of inflammatory cell phenotypes in normal gingiva and peri-implant keratinized mucosa. *J Clin Periodontol* 1995;22:735-742.
35. Renvert S, Polyzois I. Treatment of pathologic peri-implant pockets. *Periodontol 2000* 2018;76:180-190.
36. McCracken M. Dental implant materials: commercially pure titanium and titanium alloys. *J Prosthodont* 1999;8:40-43.
37. Shah FA, Trobos M, Thomsen P, Palmquist A. Commercially pure titanium (cp-Ti) versus titanium alloy (Ti6Al4V) materials as bone anchored implants - Is one truly better than the other? *Mater Sci Eng C Mater Biol Appl* 2016;62:960-966.
38. Bosshardt DD, Chappuis V, Buser D. Osseointegration of titanium, titanium alloy and zirconia dental implants: current knowledge and open questions. *Periodontol 2000* 2017;73:22-40.
39. Cionca N, Hashim D, Mombelli A. Zirconia dental implants: where are we now, and where are we heading? *Periodontol 2000* 2017;73:241-258.
40. Altuna P, Lucas-Taulé E, Gargallo-Albiol J, Figueras-Álvarez O, Hernández-Alfaro F, Nart J. Clinical evidence on titanium-zirconium dental implants: a systematic review and meta-analysis. *Int J Oral Maxillofac Surg* 2016;45:842-850.
41. Sivaswamy V, Bahl V. Surface Modifications of Commercial Dental Implant Systems: An Overview. *J Long Term Eff Med Implants* 2023;33:71-77.
42. Jemat A, Ghazali MJ, Razali M, Otsuka Y. Surface Modifications and Their Effects on Titanium Dental Implants. *Biomed Res Int* 2015;2015:791725.
43. Quirynen M, Vogels R, Peeters W, van Steenberghe D, Naert I, Haffajee A. Dynamics of initial subgingival colonization of 'pristine' peri-implant pockets. *Clin Oral Implants Res* 2006;17:25-37.
44. Fürst MM, Salvi GE, Lang NP, Persson GR. Bacterial colonization immediately after installation on oral titanium implants. *Clin Oral Implants Res* 2007;18:501-508.
45. Quirynen M, Bollen CM. The influence of surface roughness and surface-free energy on supra- and subgingival plaque formation in man. A review of the literature. *J Clin Periodontol* 1995;22:1-14.
46. Abrahamsson I, Berglundh T, Lindhe J. Soft tissue response to plaque formation at different implant systems. A comparative study in the dog. *Clin Oral Implants Res* 1998;9:73-79.
47. Costa FO, Takenaka-Martinez S, Cota LO, Ferreira SD, Silva GL, Costa JE. Peri-implant disease in subjects with and without preventive maintenance: a 5-year follow-up. *J Clin Periodontol* 2012;39:173-181.
48. Schwarz F, Derks J, Monje A, Wang HL. Peri-implantitis. *J Periodontol* 2018;89 Suppl 1:S267-S290.
49. Koyanagi T, Sakamoto M, Takeuchi Y, Ohkuma M, Izumi Y. Analysis of microbiota associated with peri-implantitis using 16S rRNA gene clone library. *J Oral Microbiol* 2010;2.
50. Mombelli A, Decaillet F. The characteristics of biofilms in peri-implant disease. *J Clin Periodontol* 2011;38 Suppl 11:203-213.
51. Persson GR, Renvert S. Cluster of bacteria associated with peri-implantitis. *Clin Implant Dent Relat Res* 2014;16:783-793.
52. Heitz-Mayfield LJ, Lang NP. Comparative biology of chronic and aggressive periodontitis vs. peri-implantitis. *Periodontol 2000* 2010;53:167-181.
53. Kumar PS, Mason MR, Brooker MR, O'Brien K. Pyrosequencing reveals unique microbial signatures associated with healthy and failing dental implants. *J Clin Periodontol* 2012;39:425-433.
54. Berglundh T, Armitage G, Araujo MG, et al. Peri-implant diseases and conditions: Consensus report of workgroup 4 of the 2017 World Workshop on the Classification of Periodontal and Peri-Implant Diseases and Conditions. *J Periodontol* 2018;89 Suppl 1:S313-S318.
55. Heitz-Mayfield LJA, Salvi GE. Peri-implant mucositis. *J Clin Periodontol* 2018;45 Suppl 20:S237-s245.
56. Pontoriero R, Tonelli MP, Carnevale G, Mombelli A, Nyman SR, Lang NP. Experimentally induced peri-implant mucositis. A clinical study in humans. *Clin Oral Implants Res* 1994;5:254-259.
57. Meyer S, Giannopoulou C, Courvoisier D, Schimmel M, Müller F, Mombelli A. Experimental mucositis and experimental gingivitis in persons aged 70 or over. Clinical and biological responses. *Clin Oral Implants Res* 2017;28:1005-1012.

58. Jepsen S, Berglundh T, Genco R, et al. Primary prevention of peri-implantitis: managing peri-implant mucositis. *J Clin Periodontol* 2015;42 Suppl 16:S152-157.
59. Lang NP, Karring T. *Proceedings of the 1st European Workshop on Periodontology, Charter House at Ittingen, Thurgau, Switzerland, February 1-4, 1993*: Quintessence; 1994.
60. Lindhe J, Berglundh T, Ericsson I, Liljenberg B, Marinello C. Experimental breakdown of peri-implant and periodontal tissues. A study in the beagle dog. *Clin Oral Implants Res* 1992;3:9-16.
61. Renvert S, Hirooka H, Polyzois I, Kelekis-Cholakis A, Wang HL, 3 aWG. Diagnosis and non-surgical treatment of peri-implant diseases and maintenance care of patients with dental implants - Consensus report of working group 3. *Int Dent J* 2019;69 Suppl 2:12-17.
62. Derks J, Schaller D, Hakansson J, Wennstrom JL, Tomasi C, Berglundh T. Effectiveness of Implant Therapy Analyzed in a Swedish Population: Prevalence of Peri-implantitis. *J Dent Res* 2016;95:43-49.
63. Mombelli A, Lang NP. Clinical parameters for the evaluation of dental implants. *Periodontol 2000* 1994;4:81-86.
64. Romandini M, Berglundh J, Derks J, Sanz M, Berglundh T. Diagnosis of peri-implantitis in the absence of baseline data: A diagnostic accuracy study. *Clin Oral Implants Res* 2021;32:297-313.
65. Lindquist LW, Rockler B, Carlsson GE. Bone resorption around fixtures in edentulous patients treated with mandibular fixed tissue-integrated prostheses. *J Prosthet Dent* 1988;59:59-63.
66. Mombelli A, van Oosten MA, Schurch E, Jr., Land NP. The microbiota associated with successful or failing osseointegrated titanium implants. *Oral Microbiol Immunol* 1987;2:145-151.
67. Renvert S, Persson GR, Pirih FQ, Camargo PM. Peri-implant health, peri-implant mucositis, and peri-implantitis: Case definitions and diagnostic considerations. *J Periodontol* 2018;89 Suppl 1:S304-S312.
68. Lindhe J, Meyle J, Group DoEWoP. Peri-implant diseases: Consensus Report of the Sixth European Workshop on Periodontology. *J Clin Periodontol* 2008;35:282-285.
69. Romandini M, Lima C, Pedrinaci I, Araoz A, Soldini MC, Sanz M. Prevalence and risk/protective indicators of peri-implant diseases: A university-representative cross-sectional study. *Clin Oral Implants Res* 2021;32:112-122.
70. Lang NP, Wetzel AC, Stich H, Caffesse RG. Histologic probe penetration in healthy and inflamed peri-implant tissues. *Clin Oral Implants Res* 1994;5:191-201.
71. Monje A, Amerio E, Farina R, et al. Significance of probing for monitoring peri-implant diseases. *Int J Oral Implantol (Berl)* 2021;14:385-399.
72. Rakic M, Struillou X, Petkovic-Curcin A, et al. Estimation of bone loss biomarkers as a diagnostic tool for peri-implantitis. *J Periodontol* 2014;85:1566-1574.
73. Salvi GE, Lang NP. Diagnostic parameters for monitoring peri-implant conditions. *Int J Oral Maxillofac Implants* 2004;19 Suppl:116-127.
74. Heitz-Mayfield LJ. Peri-implant diseases: diagnosis and risk indicators. *J Clin Periodontol* 2008;35:292-304.
75. Herrera D, Berglundh T, Schwarz F, et al. Prevention and treatment of peri-implant diseases-The EFP S3 level clinical practice guideline. *J Clin Periodontol* 2023:4-76.
76. Ericsson I, Berglundh T, Marinello C, Liljenberg B, Lindhe J. Long-standing plaque and gingivitis at implants and teeth in the dog. *Clin Oral Implants Res* 1992;3:99-103.
77. Renvert S, Polyzois I. Risk indicators for peri-implant mucositis: a systematic literature review. *J Clin Periodontol* 2015;42 Suppl 16:S172-186.
78. Levignac J. Periimplantation osteolysis- periimplantosis - periimplantitis. *Rev Fr Odontostomatol* 1965;12:1251-1260.
79. Tarnow DP. Increasing Prevalence of Peri-implantitis: How Will We Manage? *J Dent Res* 2016;95:7-8.
80. Koldsland OC, Scheie AA, Aass AM. Prevalence of peri-implantitis related to severity of the disease with different degrees of bone loss. *J Periodontol* 2010;81:231-238.
81. Roos-Jansaker AM, Lindahl C, Renvert H, Renvert S. Nine- to fourteen-year follow-up of implant treatment. Part II: presence of peri-implant lesions. *J Clin Periodontol* 2006;33:290-295.
82. Kordbacheh Changi K, Finkelstein J, Papapanou PN. Peri-implantitis prevalence, incidence rate, and risk factors: A study of electronic health records at a U.S. dental school. *Clin Oral Implants Res* 2019;30:306-314.
83. Vignoletti F, Di Domenico GL, Di Martino M, Montero E, de Sanctis M. Prevalence and risk indicators of peri-implantitis in a sample of university-based dental patients in Italy: A cross-sectional study. *J Clin Periodontol* 2019;46:597-605.
84. Rakic M, Galindo-Moreno P, Monje A, et al. How frequent does peri-implantitis occur? A systematic review and meta-analysis. *Clin Oral Investig* 2018;22:1805-1816.
85. Derks J, Tomasi C. Peri-implant health and disease. A systematic review of current epidemiology. *J Clin Periodontol* 2015;42 Suppl 16:S158-171.
86. Klinge B, Meyle J. Peri-implant tissue destruction. The Third EAO Consensus Conference 2012. *Clin Oral Implants Res* 2012;23 Suppl 6:108-110.
87. Atieh MA, Alsabeeha NH, Faggion CM, Jr., Duncan WJ. The frequency of peri-implant diseases: a systematic review and meta-analysis. *J Periodontol* 2013;84:1586-1598.

88. Mombelli A, Muller N, Cionca N. The epidemiology of peri-implantitis. *Clin Oral Implants Res* 2012;23 Suppl 6:67-76.
89. Karoussis IK, Brägger U, Salvi GE, Bürgin W, Lang NP. Effect of implant design on survival and success rates of titanium oral implants: a 10-year prospective cohort study of the ITI Dental Implant System. *Clin Oral Implants Res* 2004;15:8-17.
90. Lang NP, Berglundh T, Working Group 4 of Seventh European Workshop on P. Periimplant diseases: where are we now?--Consensus of the Seventh European Workshop on Periodontology. *J Clin Periodontol* 2011;38 Suppl 11:178-181.
91. Bullon P, Fioroni M, Goteri G, Rubini C, Battino M. Immunohistochemical analysis of soft tissues in implants with healthy and peri-implantitis condition, and aggressive periodontitis. *Clin Oral Implants Res* 2004;15:553-559.
92. Berglundh T, Donati M. Aspects of adaptive host response in periodontitis. *J Clin Periodontol* 2005;32 Suppl 6:87-107.
93. Harris LG, Mead L, Müller-Oberländer E, Richards RG. Bacteria and cell cytocompatibility studies on coated medical grade titanium surfaces. *J Biomed Mater Res A* 2006;78:50-58.
94. Leonhardt A, Renvert S, Dahlen G. Microbial findings at failing implants. *Clin Oral Implants Res* 1999;10:339-345.
95. Ata-Ali J, Candel-Marti ME, Flichy-Fernández AJ, Peñarrocha-Oltra D, Balaguer-Martinez JF, Peñarrocha Diago M. Peri-implantitis: associated microbiota and treatment. *Med Oral Patol Oral Cir Bucal* 2011;16:e937-943.
96. Shibli JA, Melo L, Ferrari DS, Figueiredo LC, Faveri M, Feres M. Composition of supra- and subgingival biofilm of subjects with healthy and diseased implants. *Clin Oral Implants Res* 2008;19:975-982.
97. Zhuang LF, Watt RM, Mattheos N, Si MS, Lai HC, Lang NP. Periodontal and peri-implant microbiota in patients with healthy and inflamed periodontal and peri-implant tissues. *Clin Oral Implants Res* 2016;27:13-21.
98. Staubli N, Walter C, Schmidt JC, Weiger R, Zitzmann NU. Excess cement and the risk of peri-implant disease - a systematic review. *Clin Oral Implants Res* 2017;28:1278-1290.
99. Karoussis IK, Salvi GE, Heitz-Mayfield LJ, Brägger U, Hämmerle CH, Lang NP. Long-term implant prognosis in patients with and without a history of chronic periodontitis: a 10-year prospective cohort study of the ITI Dental Implant System. *Clin Oral Implants Res* 2003;14:329-339.
100. Cho-Yan Lee J, Mattheos N, Nixon KC, Ivanovski S. Residual periodontal pockets are a risk indicator for peri-implantitis in patients treated for periodontitis. *Clin Oral Implants Res* 2012;23:325-333.
101. Ryder MI, Fujitaki R, Johnson G, Hyun W. Alterations of neutrophil oxidative burst by in vitro smoke exposure: implications for oral and systemic diseases. *Ann Periodontol* 1998;3:76-87.
102. Tipton DA, Dabbous MK. Effects of nicotine on proliferation and extracellular matrix production of human gingival fibroblasts in vitro. *J Periodontol* 1995;66:1056-1064.
103. César-Neto JB, Duarte PM, de Oliveira MC, et al. Smoking modulates interferon-gamma expression in the gingival tissue of patients with chronic periodontitis. *Eur J Oral Sci* 2006;114:403-408.
104. Graswinckel JE, van der Velden U, van Winkelhoff AJ, Hoek FJ, Loos BG. Plasma antibody levels in periodontitis patients and controls. *J Clin Periodontol* 2004;31:562-568.
105. Sopori ML, Kozak W. Immunomodulatory effects of cigarette smoke. *J Neuroimmunol* 1998;83:148-156.
106. Chrcanovic BR, Albrektsson T, Wennerberg A. Smoking and dental implants: A systematic review and meta-analysis. *J Dent* 2015;43:487-498.
107. Keenan JR, Veitz-Keenan A. The impact of smoking on failure rates, postoperative infection and marginal bone loss of dental implants. *Evid Based Dent* 2016;17:4-5.
108. Sgolastra F, Petrucci A, Severino M, Gatto R, Monaco A. Smoking and the risk of peri-implantitis. A systematic review and meta-analysis. *Clin Oral Implants Res* 2015;26:e62-e67.
109. Rinke S, Ohl S, Ziebolz D, Lange K, Eickholz P. Prevalence of periimplant disease in partially edentulous patients: a practice-based cross-sectional study. *Clin Oral Implants Res* 2011;22:826-833.
110. Carra MC, Blanc-Sylvestre N, Courtet A, Bouchard P. Primordial and primary prevention of peri-implant diseases: A systematic review and meta-analysis. *J Clin Periodontol* 2023;50 Suppl 26:77-112.
111. Stene LC, Ruiz PL, Åsvold BO, et al. How many people have diabetes in Norway in 2020? *Tidsskr Nor Laegeforen* 2020;140.
112. Brem H, Tomic-Canic M. Cellular and molecular basis of wound healing in diabetes. *J Clin Invest* 2007;117:1219-1222.
113. Rayfield EJ, Ault MJ, Keusch GT, Brothers MJ, Nechemias C, Smith H. Infection and diabetes: the case for glucose control. *Am J Med* 1982;72:439-450.
114. Klein R, Klein BE, Moss SE. Relation of glycemic control to diabetic microvascular complications in diabetes mellitus. *Ann Intern Med* 1996;124:90-96.
115. Stettler C, Allemann S, Jüni P, et al. Glycemic control and macrovascular disease in types 1 and 2 diabetes mellitus: Meta-analysis of randomized trials. *Am Heart J* 2006;152:27-38.
116. Wagner J, Spille JH, Wiltfang J, Naujokat H. Systematic review on diabetes mellitus and dental implants: an update. *Int J Implant Dent* 2022;8:1.

117. Monje A, Catena A, Borgnakke WS. Association between diabetes mellitus/hyperglycaemia and peri-implant diseases: Systematic review and meta-analysis. *J Clin Periodontol* 2017;44:636-648.
118. Chrcanovic BR, Albrektsson T, Wennerberg A. Diabetes and oral implant failure: a systematic review. *J Dent Res* 2014;93:859-867.
119. Pjetursson BE, Helbling C, Weber HP, et al. Peri-implantitis susceptibility as it relates to periodontal therapy and supportive care. *Clin Oral Implants Res* 2012;23:888-894.
120. Monje A, Wang HL, Nart J. Association of Preventive Maintenance Therapy Compliance and Peri-Implant Diseases: A Cross-Sectional Study. *J Periodontol* 2017;88:1030-1041.
121. Gay IC, Tran DT, Weltman R, et al. Role of supportive maintenance therapy on implant survival: a university-based 17 years retrospective analysis. *Int J Dent Hyg* 2016;14:267-271.
122. Serino G, Hultin K. Periimplant Disease and Prosthetic Risk Indicators: A Literature Review. *Implant Dent* 2019;28:125-137.
123. Ferreira SD, Silva GL, Cortelli JR, Costa JE, Costa FO. Prevalence and risk variables for peri-implant disease in Brazilian subjects. *J Clin Periodontol* 2006;33:929-935.
124. Souza AB, Tormena M, Matarazzo F, Araujo MG. The influence of peri-implant keratinized mucosa on brushing discomfort and peri-implant tissue health. *Clin Oral Implants Res* 2016;27:650-655.
125. Monje A, Blasi G. Significance of keratinized mucosa/gingiva on peri-implant and adjacent periodontal conditions in erratic maintenance compliers. *J Periodontol* 2019;90:445-453.
126. Ramanauskaite A, Schwarz F, Sader R. Influence of width of keratinized tissue on the prevalence of peri-implant diseases: A systematic review and meta-analysis. *Clin Oral Implants Res* 2022;33 Suppl 23:8-31.
127. Sanz-Martín I, Regidor E, Navarro J, Sanz-Sánchez I, Sanz M, Ortiz-Vigón A. Factors associated with the presence of peri-implant buccal soft tissue dehiscences: A case-control study. *J Periodontol* 2020; 91: 1003-1010.
128. Sanz M, Schwarz F, Herrera D, et al. Importance of keratinized mucosa around dental implants: Consensus report of group 1 of the DGI/SEPA/Osteology Workshop. *Clin Oral Implants Res* 2022;33 Suppl 23:47-55.
129. Chrcanovic BR, Albrektsson T, Wennerberg A. Reasons for failures of oral implants. *J Oral Rehabil* 2014;41:443-476.
130. Lee SJ, Alamri O, Cao H, Wang Y, Gallucci GO, Lee JD. Occlusion as a predisposing factor for peri-implant disease: A review article. *Clin Implant Dent Relat Res* 2022; 25: 734-742.
131. Hammerle CHF, Tarnow D. The etiology of hard- and soft-tissue deficiencies at dental implants: A narrative review. *J Periodontol* 2018;89 Suppl 1:S291-S303.
132. Monje A, Pons R, Insua A, Nart J, Wang HL, Schwarz F. Morphology and severity of peri-implantitis bone defects. *Clin Implant Dent Relat Res* 2019;21:635-643.
133. Serino G, Ström C. Peri-implantitis in partially edentulous patients: association with inadequate plaque control. *Clin Oral Implants Res* 2009;20:169-174.
134. Blanco C, Liñares A, Dopico J, et al. Peri-implantitis, systemic inflammation, and dyslipidemia: a cross-sectional biochemical study. *J Periodontal Implant Sci* 2021;51:342-351.
135. Chaushu L, Tal H, Sculean A, Fernández-Tomé B, Chaushu G. Effects of peri-implant infection on serum biochemical analysis. *J Periodontol* 2021;92:436-445.
136. Schwarz F, Herten M, Sager M, Bieling K, Sculean A, Becker J. Comparison of naturally occurring and ligature-induced peri-implantitis bone defects in humans and dogs. *Clin Oral Implants Res* 2007;18:161-170.
137. Schwarz F, Sahm N, Schwarz K, Becker J. Impact of defect configuration on the clinical outcome following surgical regenerative therapy of peri-implantitis. *J Clin Periodontol* 2010;37:449-455.
138. Monje A, Pons R, Sculean A, Nart J, Wang HL. Defect angle as prognostic indicator in the reconstructive therapy of peri-implantitis. *Clin Implant Dent Relat Res* 2023; 25: 992-999.
139. Sanz M, Chapple IL, Working Group 4 of the VEWoP. Clinical research on peri-implant diseases: consensus report of Working Group 4. *J Clin Periodontol* 2012;39 Suppl 12:202-206.
140. Heitz-Mayfield LJ, Mombelli A. The therapy of peri-implantitis: a systematic review. *Int J Oral Maxillofac Implants* 2014;29 Suppl:325-345.
141. Polyzois I. Treatment Planning for Periimplant Mucositis and Periimplantitis. *Implant Dent* 2019;28:150-154.
142. Schwarz F, Schmucker A, Becker J. Efficacy of alternative or adjunctive measures to conventional treatment of peri-implant mucositis and peri-implantitis: a systematic review and meta-analysis. *Int J Implant Dent* 2015;1:22.
143. Figuero E, Graziani F, Sanz I, Herrera D, Sanz M. Management of peri-implant mucositis and peri-implantitis. *Periodontol 2000* 2014;66:255-273.
144. Carcuac O, Derks J, Abrahamsson I, Wennstrom JL, Berglundh T. Risk for recurrence of disease following surgical therapy of peri-implantitis-A prospective longitudinal study. *Clin Oral Implants Res* 2020;31:1072-1077.
145. Renvert S, Roos-Jansaker AM, Claffey N. Non-surgical treatment of peri-implant mucositis and peri-implantitis: a literature review. *J Clin Periodontol* 2008;35:305-315.
146. Wagner TP, Pires PR, Rios FS, et al. Surgical and non-surgical debridement for the treatment of peri-implantitis: a two-center 12-month randomized trial. *Clin Oral Investig* 2021; 25: 5723-5733.

147. Lang NP, Salvi GE, Sculean A. Nonsurgical therapy for teeth and implants-When and why? *Periodontol 2000* 2019;79:15-21.
148. Berglundh T, Wennstrom JL, Lindhe J. Long-term outcome of surgical treatment of peri-implantitis. A 2-11-year retrospective study. *Clin Oral Implants Res* 2018;29:404-410.
149. Renvert S, Polyzois I, Claffey N. Surgical therapy for the control of peri-implantitis. *Clin Oral Implants Res* 2012;23 Suppl 6:84-94.
150. Koldsland OC, Aass AM. Supportive treatment following peri-implantitis surgery: An RCT using titanium curettes or chitosan brushes. *J Clin Periodontol* 2020;47:1259-1267.
151. Serino G, Turri A, Lang NP. Maintenance therapy in patients following the surgical treatment of peri-implantitis: a 5-year follow-up study. *Clin Oral Implants Res* 2015;26:950-956.
152. Liñares A, Sanz-Sánchez I, Dopico J, Molina A, Blanco J, Montero E. Efficacy of adjunctive measures in the non-surgical treatment of peri-implantitis: A systematic review. *J Clin Periodontol* 2023;50 Suppl 26:224-243.
153. De Waal YCM, Vangsted TE, Van Winkelhoff AJ. Systemic antibiotic therapy as an adjunct to non-surgical peri-implantitis treatment: A single-blind RCT. *Journal of Clinical Periodontology* 2021;48:996-1006.
154. Blanco C, Pico A, Dopico J, Gándara P, Blanco J, Liñares A. Adjunctive benefits of systemic metronidazole on non-surgical treatment of peri-implantitis. A randomized placebo-controlled clinical trial. *J Clin Periodontol* 2022;49:15-27.
155. Carcuac O, Derks J, Charalampakis G, Abrahamsson I, Wennstrom J, Berglundh T. Adjunctive Systemic and Local Antimicrobial Therapy in the Surgical Treatment of Peri-implantitis: A Randomized Controlled Clinical Trial. *J Dent Res* 2016;95:50-57.
156. Carcuac O, Derks J, Abrahamsson I, Wennstrom JL, Petzold M, Berglundh T. Surgical treatment of peri-implantitis: 3-year results from a randomized controlled clinical trial. *J Clin Periodontol* 2017;44:1294-1303.
157. Ramanauskaite A, Fretwurst T, Schwarz F. Efficacy of alternative or adjunctive measures to conventional non-surgical and surgical treatment of peri-implant mucositis and peri-implantitis: a systematic review and meta-analysis. *Int J Implant Dent* 2021;7:112.
158. Schmage P, Thielemann J, Nergiz I, Scorziello TM, Pfeiffer P. Effects of 10 cleaning instruments on four different implant surfaces. *Int J Oral Maxillofac Implants* 2012;27:308-317.
159. Schwarz F, Sculean A, Romanos G, et al. Influence of different treatment approaches on the removal of early plaque biofilms and the viability of SAOS2 osteoblasts grown on titanium implants. *Clin Oral Investig* 2005;9:111-117.
160. Stein JM, Conrads G, Abdelbary MMH, et al. Antimicrobial efficiency and cytocompatibility of different decontamination methods on titanium and zirconium surfaces. *Clin Oral Implants Res* 2023;34:20-32.
161. Luengo F, Sanz-Esporrín J, Noguerol F, Sanz-Martín I, Sanz-Sánchez I, Sanz M. In vitro effect of different implant decontamination methods in three intraosseous defect configurations. *Clin Oral Implants Res* 2022.
162. Ichioka Y, Derks J, Dahlén G, Berglundh T, Larsson L. Mechanical removal of biofilm on titanium discs: An in vitro study. *J Biomed Mater Res B Appl Biomater* 2022;110:1044-1055.
163. Kreisler M, Kohnen W, Christoffers AB, et al. In vitro evaluation of the biocompatibility of contaminated implant surfaces treated with an Er : YAG laser and an air powder system. *Clin Oral Implants Res* 2005;16:36-43.
164. Larsen OI, Enersen M, Kristoffersen AK, et al. Antimicrobial Effects of Three Different Treatment Modalities on Dental Implant Surfaces. *J Oral Implantol* 2017;43:429-436.
165. John G, Becker J, Schwarz F. Rotating titanium brush for plaque removal from rough titanium surfaces--an in vitro study. *Clin Oral Implants Res* 2014;25:838-842.
166. Sanchez MC, Llama-Palacios A, Blanc V, Leon R, Herrera D, Sanz M. Structure, viability and bacterial kinetics of an in vitro biofilm model using six bacteria from the subgingival microbiota. *J Periodontal Res* 2011;46:252-260.
167. Pereira da Silva CH, Vidigal GM, Jr., de Uzeda M, de Almeida Soares G. Influence of titanium surface roughness on attachment of Streptococcus sanguis: an in vitro study. *Implant Dent* 2005;14:88-93.
168. Louropoulou A, Slot DE, Van der Weijden F. The effects of mechanical instruments on contaminated titanium dental implant surfaces: a systematic review. *Clin Oral Implants Res* 2014;25:1149-1160.
169. Bermejo P, Sánchez MC, Llama-Palacios A, Figuero E, Herrera D, Sanz M. Topographic characterization of multispecies biofilms growing on dental implant surfaces: An in vitro model. *Clin Oral Implants Res* 2019;30:229-241.
170. Nemer Vieira LF, Lopes de Chaves e Mello Dias EC, Cardoso ES, Machado SJ, Pereira da Silva C, Vidigal GM, Jr. Effectiveness of implant surface decontamination using a high-pressure sodium bicarbonate protocol: an in vitro study. *Implant Dent* 2012;21:390-393.
171. Keim D, Nickles K, Dannewitz B, Ratka C, Eickholz P, Petsos H. In vitro efficacy of three different implant surface decontamination methods in three different defect configurations. *Clin Oral Implants Res* 2019;30:550-558.
172. Korello K, Eickholz P, Zuhr O, Ratka C, Petsos H. In vitro efficacy of non-surgical and surgical implant surface decontamination methods in three different defect configurations in the presence or absence of a suprastructure. *Clin Implant Dent Relat Res* 2023.

173. Sahrmann P, Ronay V, Hofer D, Attin T, Jung RE, Schmidlin PR. In vitro cleaning potential of three different implant debridement methods. *Clin Oral Implants Res* 2015;26:314-319.
174. Sahrmann P, Winkler S, Gubler A, Attin T. Assessment of implant surface and instrument insert changes due to instrumentation with different tips for ultrasonic-driven debridement. *BMC Oral Health* 2021;21:25.
175. Muxika A, Etxabide A, Uranga J, Guerrero P, de la Caba K. Chitosan as a bioactive polymer: Processing, properties and applications. *Int J Biol Macromol* 2017;105:1358-1368.
176. Fakhri E, Eslami H, Maroufi P, et al. Chitosan biomaterials application in dentistry. *Int J Biol Macromol* 2020;162:956-974.
177. Sahariah P, Másson M. Antimicrobial Chitosan and Chitosan Derivatives: A Review of the Structure-Activity Relationship. *Biomacromolecules* 2017;18:3846-3868.
178. Costa EM, Silva S, Costa MR, et al. Chitosan mouthwash: toxicity and in vivo validation. *Carbohydr Polym* 2014;111:385-392.
179. Wohlfahrt JC, Evensen BJ, Zeza B, et al. A novel non-surgical method for mild peri-implantitis- a multicenter consecutive case series. *Int J Implant Dent* 2017;3:38.
180. Wohlfahrt JC, Aass AM, Koldsland OC. Treatment of peri-implant mucositis with a chitosan brush-A pilot randomized clinical trial. *Int J Dent Hyg* 2019;17:170-176.
181. Cosgarea R, Roccuzzo A, Jepsen K, Sculean A, Jepsen S, Salvi GE. Efficacy of mechanical/physical approaches for implant surface decontamination in non-surgical submarginal instrumentation of peri-implantitis. A systematic review. *J Clin Periodontol* 2023;50 Suppl 26:188-211.
182. Abduljabbar T, Javed F, Kellesarian SV, Vohra F, Romanos GE. Effect of Nd:YAG laser-assisted non-surgical mechanical debridement on clinical and radiographic peri-implant inflammatory parameters in patients with peri-implant disease. *J Photochem Photobiol B* 2017;168:16-19.
183. Arısan V, Karabuda ZC, Arıcı SV, Topçuoğlu N, Külekçi G. A randomized clinical trial of an adjunct diode laser application for the nonsurgical treatment of peri-implantitis. *Photomed Laser Surg* 2015;33:547-554.
184. Renvert S, Samuelsson E, Lindahl C, Persson GR. Mechanical non-surgical treatment of peri-implantitis: a double-blind randomized longitudinal clinical study. I: clinical results. *J Clin Periodontol* 2009;36:604-609.
185. Roccuzzo A, Klossner S, Stähli A, et al. Non-surgical mechanical therapy of peri-implantitis with or without repeated adjunctive diode laser application. A 6-month double-blinded randomized clinical trial. *Clin Oral Implants Res* 2022;33:900-912.
186. Hentenaar DFM, De Waal YCM, Stewart RE, Van Winkelhoff AJ, Meijer HJA, Raghoebar GM. Erythritol airpolishing in the non-surgical treatment of peri-implantitis: A randomized controlled trial. *Clin Oral Implants Res* 2021;32:840-852.
187. Selimović A, Bunæs DF, Lie SA, Lobekk MA, Leknes KN. Non-surgical treatment of peri-implantitis with and without erythritol air-polishing a 12-month randomized controlled trial. *BMC Oral Health* 2023;23:240.
188. Tong Z, Fu R, Zhu W, Shi J, Yu M, Si M. Changes in the surface topography and element proportion of clinically failed SLA implants after in vitro debridement by different methods. *Clin Oral Implants Res* 2021;32:263-273.
189. Bertoldi C, Lusuardi D, Battarra F, Sassatelli P, Spinato S, Zaffe D. The maintenance of inserted titanium implants: in-vitro evaluation of exposed surfaces cleaned with three different instruments. *Clin Oral Implants Res* 2017;28:57-63.
190. Roccuzzo A, De Ry SP, Sculean A, Roccuzzo M, Salvi GE. Current Approaches for the Non-surgical Management of Peri-implant Diseases. *Current Oral Health Reports* 2020;7:274-282.
191. Tian Y, Chen C, Xu X, et al. A Review of 3D Printing in Dentistry: Technologies, Affecting Factors, and Applications. *Scanning* 2021;2021:9950131.
192. Sidhom M, Zaghloul H, Mosleh IE, Eldwakhly E. Effect of Different CAD/CAM Milling and 3D Printing Digital Fabrication Techniques on the Accuracy of PMMA Working Models and Vertical Marginal Fit of PMMA Provisional Dental Prosthesis: An In Vitro Study. *Polymers (Basel)* 2022;14.
193. Tuna T, Kuhlmann L, Bishti S, Sirazitdinova E, Deserno T, Wolfart S. Removal of simulated biofilm at different implant crown designs with interproximal oral hygiene aids: An in vitro study. *Clin Oral Implants Res* 2019;30:627-636.
194. Luengo F, Sanz-Esporrín J, Noguerol F, Sanz-Martín I, Sanz-Sánchez I, Sanz M. In vitro effect of different implant decontamination methods in three intraosseous defect configurations. *Clin Oral Implants Res* 2022;33:1087-1097.
195. Matsubara VH, Leong BW, Leong MJL, Lawrence Z, Becker T, Quaranta A. Cleaning potential of different air abrasive powders and their impact on implant surface roughness. *Clin Implant Dent Relat Res* 2020;22:96-104.
196. Virto L, Odeh V, Garcia-Quismondo E, et al. Electrochemical decontamination of titanium dental implants. An in vitro biofilm model study. *Clin Oral Implants Res* 2023;34:486-497.
197. Charalampakis G, Ramberg P, Dahlén G, Berglundh T, Abrahamsson I. Effect of cleansing of biofilm formed on titanium discs. *Clin Oral Implants Res* 2015;26:931-936.
198. Fransson C, Wennström J, Berglundh T. Clinical characteristics at implants with a history of progressive bone loss. *Clin Oral Implants Res* 2008;19:142-147.

199. Monje A, Vera M, Muñoz-Sanz A, Wang HL, Nart J. Suppuration as diagnostic criterium of peri-implantitis. *J Periodontol* 2021;92:216-224.
200. Persson GR, Roos-Jansaker AM, Lindahl C, Renvert S. Microbiologic results after non-surgical erbium-doped:yttrium, aluminum, and garnet laser or air-abrasive treatment of peri-implantitis: a randomized clinical trial. *J Periodontol* 2011;82:1267-1278.
201. Alpaslan Yayli NZ, Talmac AC, Keskin Tunc S, Akbal D, Altindal D, Ertugrul AS. Erbium, chromium-doped: yttrium, scandium, gallium, garnet and diode lasers in the treatment of peri-implantitis: clinical and biochemical outcomes in a randomized-controlled clinical trial. *Lasers Med Sci* 2022;37:665-674.
202. Shibli JA, Ferrari DS, Siroma RS, de Figueiredo LC, de Faveri M, Feres M. Microbiological and clinical effects of adjunctive systemic metronidazole and amoxicillin in the non-surgical treatment of peri-implantitis: 1 year follow-up. *Braz. Oral Res.* 2019;33.
203. Renvert S, Lessem J, Dahlen G, Renvert H, Lindahl C. Mechanical and repeated antimicrobial therapy using a local drug delivery system in the treatment of peri-implantitis: a randomized clinical trial. *J Periodontol* 2008;79:836-844.
204. Renvert S, Lessem J, Dahlén G, Lindahl C, Svensson M. Topical minocycline microspheres versus topical chlorhexidine gel as an adjunct to mechanical debridement of incipient peri-implant infections: a randomized clinical trial. *J Clin Periodontol* 2006;33:362-369.
205. Machtei EE, Romanos G, Kang P, et al. Repeated delivery of chlorhexidine chips for the treatment of peri-implantitis: A multicenter, randomized, comparative clinical trial. *J Periodontol* 2021;92:11-20.
206. Bassetti M, Schär D, Wicki B, et al. Anti-infective therapy of peri-implantitis with adjunctive local drug delivery or photodynamic therapy: 12-month outcomes of a randomized controlled clinical trial. *Clin Oral Implants Res* 2014;25:279-287.
207. Esposito M, Grusovin MG, Worthington HV. Treatment of peri-implantitis: what interventions are effective? A Cochrane systematic review. *Eur J Oral Implantol* 2012;5 Suppl:S21-41.
208. Hashim D, Cionca N, Combescure C, Mombelli A. The diagnosis of peri-implantitis: A systematic review on the predictive value of bleeding on probing. *Clin Oral Implants Res* 2018;29 Suppl 16:276-293.
209. Persson GR, Samuelsson E, Lindahl C, Renvert S. Mechanical non-surgical treatment of peri-implantitis: a single-blinded randomized longitudinal clinical study. II. Microbiological results. *J Clin Periodontol* 2010;37:563-573.
210. Karring ES, Stavropoulos A, Ellegaard B, Karring T. Treatment of peri-implantitis by the Vector system. *Clin Oral Implants Res* 2005;16:288-293.
211. Derks J, Ichioka Y, Dionigi C, et al. Prevention and management of peri-implant mucositis and peri-implantitis: A systematic review of outcome measures used in clinical studies in the last 10 years. *J Clin Periodontol* 2023;50 Suppl 25:55-66.
212. Almohandes A, Lund H, Carcuac O, Petzold M, Berglundh T, Abrahamsson I. Accuracy of bone-level assessments following reconstructive surgical treatment of experimental peri-implantitis. *Clin Oral Implants Res* 2022;33:433-440.
213. Norum TA, Aass AM, Koldsland OC. Pain and morbidity after non-surgical and surgical treatment of peri-implantitis. *Acta Odontol Scand* 2019;77:624-629.
214. Edwards P, Roberts I, Clarke M, et al. Increasing response rates to postal questionnaires: systematic review. *BMJ* 2002;324:1183.
215. Freiman JA, Chalmers TC, Smith H, Jr., Kuebler RR. The importance of beta, the type II error and sample size in the design and interpretation of the randomized control trial. Survey of 71 "negative" trials. *N Engl J Med* 1978;299:690-694.
216. Hansen E, Fonager K, Freund KS, Lous J. The impact of non-responders on health and lifestyle outcomes in an intervention study. *BMC Res Notes* 2014;7:632.
217. Merli M, Bernardelli F, Giulianelli E, et al. Short-term comparison of two non-surgical treatment modalities of peri-implantitis: Clinical and microbiological outcomes in a two-factorial randomized controlled trial. *J Clin Periodontol* 2020;47:1268-1280.
218. Rodríguez Del Águila M, González-Ramírez A. Sample size calculation. *Allergol Immunopathol (Madr)* 2014;42:485-492.
219. Serdar CC, Cihan M, Yücel D, Serdar MA. Sample size, power and effect size revisited: simplified and practical approaches in pre-clinical, clinical and laboratory studies. *Biochem Med (Zagreb)* 2021;31:010502.
220. Hait S, Valentín JL, Jiménez AG, et al. Poly(acrylonitrile-co-butadiene) as polymeric crosslinking accelerator for sulphur network formation. *Heliyon* 2020;6:e04659.
221. Acar SB, Tasdelen MA, Karaagac B. Methacrylate-functionalized POSS influence on cross-linking and mechanical properties of styrene-butadiene rubber. *Iran. Polym. J.* 2021;30:697-705.
222. Virto L, Simões-Martins D, Sánchez MC, Encinas A, Sanz M, Herrera D. Antimicrobial effects of a new brushing solution concept on a multispecies in vitro biofilm model growing on titanium surfaces. *Clin Oral Implants Res* 2022;33:209-220.
223. Charalampakis G, Rabe P, Leonhardt A, Dahlen G. A follow-up study of peri-implantitis cases after treatment. *J Clin Periodontol* 2011;38:864-871.

224. Horn H, Lackner S. Modeling of biofilm systems: a review. *Adv Biochem Eng Biotechnol* 2014;146:53-76.
225. Aguirre-Zorzano LA, Estefania-Fresco R, Telletxea O, Bravo M. Prevalence of peri-implant inflammatory disease in patients with a history of periodontal disease who receive supportive periodontal therapy. *Clin Oral Implants Res* 2015;26:1338-1344.
226. Sanchez MC, Llama-Palacios A, Fernandez E, et al. An in vitro biofilm model associated to dental implants: structural and quantitative analysis of in vitro biofilm formation on different dental implant surfaces. *Dent Mater* 2014;30:1161-1171.
227. Blanc V, Isabal S, Sanchez MC, et al. Characterization and application of a flow system for in vitro multispecies oral biofilm formation. *J Periodontal Res* 2014;49:323-332.
228. Sanchez MC, Marin MJ, Figuero E, et al. Quantitative real-time PCR combined with propidium monoazide for the selective quantification of viable periodontal pathogens in an in vitro subgingival biofilm model. *J Periodontal Res* 2014;49:20-28.
229. Renvert S, Lindahl C, Roos Jansaker AM, Persson GR. Treatment of peri-implantitis using an Er:YAG laser or an air-abrasive device: a randomized clinical trial. *J Clin Periodontol* 2011;38:65-73.
230. Sahm N, Becker J, Santel T, Schwarz F. Non-surgical treatment of peri-implantitis using an air-abrasive device or mechanical debridement and local application of chlorhexidine: a prospective, randomized, controlled clinical study. *J Clin Periodontol* 2011;38:872-878.
231. Roos-Jansaker AM, Almhojd US, Jansson H. Treatment of peri-implantitis: clinical outcome of chloramine as an adjunctive to non-surgical therapy, a randomized clinical trial. *Clin Oral Implants Res* 2017;28:43-48.
232. Nart J, Pons R, Valles C, Esmatges A, Sanz-Martin I, Monje A. Non-surgical therapeutic outcomes of peri-implantitis: 12-month results. *Clin Oral Investig* 2020;24:675-682.
233. Sedgwick P. Bias in clinical trials. *BMJ* 2011;343:d4176.
234. Easterbrook PJ, Berlin JA, Gopalan R, Matthews DR. Publication bias in clinical research. *Lancet* 1991;337:867-872.
235. Fransson C, Lekholm U, Jemt T, Berglundh T. Prevalence of subjects with progressive bone loss at implants. *Clin Oral Implants Res* 2005;16:440-446.
236. Maximo MB, de Mendonca AC, Renata Santos V, Figueiredo LC, Feres M, Duarte PM. Short-term clinical and microbiological evaluations of peri-implant diseases before and after mechanical anti-infective therapies. *Clin Oral Implants Res* 2009;20:99-108.
237. Simonis P, Dufour T, Tenenbaum H. Long-term implant survival and success: a 10-16-year follow-up of non-submerged dental implants. *Clin Oral Implants Res* 2010;21:772-777.
238. Cecchinato D, Parpaiola A, Lindhe J. Mucosal inflammation and incidence of crestal bone loss among implant patients: a 10-year study. *Clin Oral Implants Res* 2014;25:791-796.
239. Zetterqvist L, Feldman S, Rotter B, et al. A prospective, multicenter, randomized-controlled 5-year study of hybrid and fully etched implants for the incidence of peri-implantitis. *J Periodontol* 2010;81:493-501.
240. Dalago HR, Schuldt Filho G, Rodrigues MA, Renvert S, Bianchini MA. Risk indicators for Peri-implantitis. A cross-sectional study with 916 implants. *Clin Oral Implants Res* 2017;28:144-150.
241. Schuldt Filho G, Dalago HR, Oliveira de Souza JG, Stanley K, Jovanovic S, Bianchini MA. Prevalence of peri-implantitis in patients with implant-supported fixed prostheses. *Quintessence Int* 2014;45:861-868.
242. Konstantinidis IK, Kotsakis GA, Gerdes S, Walter MH. Cross-sectional study on the prevalence and risk indicators of peri-implant diseases. *Eur J Oral Implantol* 2015;8:75-88.
243. Daubert DM, Weinstein BF, Bordin S, Leroux BG, Flemming TF. Prevalence and predictive factors for peri-implant disease and implant failure: a cross-sectional analysis. *J Periodontol* 2015;86:337-347.
244. Mir-Mari J, Mir-Orfila P, Figueiredo R, Valmaseda-Castellon E, Gay-Escoda C. Prevalence of peri-implant diseases. A cross-sectional study based on a private practice environment. *J Clin Periodontol* 2012;39:490-494.
245. Fardal Ø, Grytten J. A comparison of teeth and implants during maintenance therapy in terms of the number of disease-free years and costs -- an in vivo internal control study. *J Clin Periodontol* 2013;40:645-651.
246. Kelley K, Rausch JR. Sample size planning for longitudinal models: accuracy in parameter estimation for polynomial change parameters. *Psychol Methods* 2011;16:391-405.
247. John G, Sahm N, Becker J, Schwarz F. Nonsurgical treatment of peri-implantitis using an air-abrasive device or mechanical debridement and local application of chlorhexidine. Twelve-month follow-up of a prospective, randomized, controlled clinical study. *Clin Oral Investig* 2015;19:1807-1814.
248. Salmasi V, Lii TR, Humphreys K, Reddy V, Mackey SC. A literature review of the impact of exclusion criteria on generalizability of clinical trial findings to patients with chronic pain. *Pain Rep* 2022;7:e1050.
249. Schulz KF, Altman DG, Moher D, Group C. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *BMJ* 2010;340:c332.
250. Chung KC, Song JW, Group WS. A guide to organizing a multicenter clinical trial. *Plast Reconstr Surg* 2010;126:515-523.
251. Jo D. The interpretation bias and trap of multicenter clinical research. *Korean J Pain* 2020;33:199-200.
252. Roccuzzo M, De Angelis N, Bonino L, Aglietta M. Ten-year results of a three-arm prospective cohort study on implants in periodontally compromised patients. Part 1: implant loss and radiographic bone loss. *Clin Oral Implants Res* 2010;21:490-496.

253. Schwarz F, Sculean A, Rothamel D, Schwenzer K, Georg T, Becker J. Clinical evaluation of an Er:YAG laser for nonsurgical treatment of peri-implantitis: a pilot study. *Clin Oral Implants Res* 2005;16:44-52.
254. Wang CW, Ashnagar S, Gianfilippo RD, Arnett M, Kinney J, Wang HL. Laser-assisted regenerative surgical therapy for peri-implantitis: A randomized controlled clinical trial. *J Periodontol* 2021;92:378-388.
255. Riben-Grundstrom C, Norderyd O, Andre U, Renvert S. Treatment of peri-implant mucositis using a glycine powder air-polishing or ultrasonic device: a randomized clinical trial. *J Clin Periodontol* 2015;42:462-469.
256. Schwendicke F, Tu YK, Stolpe M. Preventing and Treating Peri-Implantitis: A Cost-Effectiveness Analysis. *J Periodontol* 2015;86:1020-1029.
257. Mdala I, Olsen I, Haffajee AD, Socransky SS, Thoresen M, de Blasio BF. Comparing clinical attachment level and pocket depth for predicting periodontal disease progression in healthy sites of patients with chronic periodontitis using multi-state Markov models. *J Clin Periodontol* 2014;41:837-845.
258. Kullman L, Al-Asfour A, Zetterqvist L, Andersson L. Comparison of radiographic bone height assessments in panoramic and intraoral radiographs of implant patients. *Int J Oral Maxillofac Implants* 2007;22:96-100.
259. Preus HR, Torgersen GR, Koldsland OC, et al. A new digital tool for radiographic bone level measurements in longitudinal studies. *BMC Oral Health* 2015;15:107.

Appendices

Appendix 1. Non-surgical and ***adjunctive laser therapies*** in peri-implantitis treatment: selected randomized clinical trials with ***6-month follow-up***

Author, (year)	Treatment group (number of implants)	Clinical outcomes (baseline)	Clinical outcomes (baseline vs 6 months)	Findings
Abduljabbar et al., (2017)	**Test**: Plastic curettes, Nd:YAG (n=35)	BoP (%): 10.5 PPD (mm): 2.5 MBL (mm): 2.2	BoP (%): -39.8 PPD (mm): -2.8 MBL (mm): +0.1	The test method did **NOT** lead to statistically significant results
	Control: Plastic curettes (n=39)	BoP (%): 8.8 PPD (mm): 4 MBL (mm): 1.7	BoP (%): -39.8% PPD (mm): -1.6 MBL (mm): -0.1	
Arisan et al., (2015)	**Test**: Scaling, diode laser (n=24)	BoP (%): 100 Mean PPD (mm): 4.7 MBL (mm): 2.13 Microbiological outcome	BoP (%): -4.2 Mean PPD (mm): -0.17 MBL (mm): 0.66 Microbiological outcome	The test method did **NOT** lead to statistically significant results
	Control: Scaling (n=24)	BoP (%): 100 Mean PPD (mm): 4.4 MBL (mm):2.35 Microbiological outcome	BoP (%): 0 Mean PPD (mm): -0.21 MBL (mm): 0.28 Microbiological outcome	
Persson et al., (2010)	**Test**: Titanium curettes (n=17)	BoP (n, implants): 17 Mean PPD (mm): 5.1 Bacterial counts	BoP (n, implants): 3 Mean PPD (mm): -0.2 Bacterial counts	**No** group differences were observed. Both treatments **failed** to reduce the bacterial counts
	Control: Ultrasonic device (n=14)	BoP (n, implants): 14 Mean PPD (mm): 5.2 Bacterial counts	BoP (n, implants): 2 Mean PPD (mm): -0.3 Bacterial counts	
Persson et al., (2011)	**Test**: Er:YAG laser (n=55)	BoP (%): 100 / 100 Mean PPD (mm): 6.9 / 6.7 Bacterial counts *(improved / not improved)*[#]	BoP (%): 37.5 / 77.8 Mean PPD (mm): 5.8 / 5.9 Bacterial counts *(improved / not improved)*[#]	**NO** statistically significant differences between the groups. Both treatments **failed** to reduce the bacterial counts.
	Control: Air-abrasive (Perioflow) (n=45)	BoP (%): 100 / 100 Mean PPD (mm): 6.5 /6.1 Bacterial counts *(improved / not improved)*[#]	BoP (%): 66.7 / 75 Mean PPD (mm): 5.2 / 5.3 Bacterial counts *(improved / not improved)*[#]	
Renvert et al., (2009)	**Test**: Titanium curettes (n=17)	Mean BoP: 1.7 Mean PPD (mm): 4.0 Total bacteria count	Mean BoP: -0.3 Mean PPD (mm): 0 Total bacteria count	The test method did **NOT** lead to statistically significant results
	Control: Ultrasonic device (Vector system) (n=14)	Mean BoP: 1.7 Mean PPD (mm): 4.3 Total bacteria count	Mean BoP: -0.5 Mean PPD (mm): -0.4 Total bacteria count	

Renvert et al., (2011)	**Test**: Er:YAG laser (n=55)	No bleeding (%): 0.0 SoP (%): 30.9	No bleeding (%): 30.9 SoP (%): 10.9 PPD decrease 1.1-2.0mm (%): 12.1 PPD decrease 2.1-3.0 mm (%): 4.2	The test method did **NOT** lead to statistically significant results
	Control: Air-abrasive (Perioflow) (n=45)	No bleeding (%): 2.0 SoP (%): 31.1	No bleeding (%): 25.0 SoP (%): 11.1 PPD decrease 1.1-2.0 mm (%): 14.0% PPD decrease 2.1-3.0 mm (%): 7.9	
Roccuzzo et al., (2022)	**Test**: Titanium curettes, stainless steel curettes, sterile saline, diode laser (n=15)	BoP (%): 62.5 PPD (mm): 5.4 MBL (mm): 2.6	BoP (%): -15.3 PPD (mm): 1.3 MBL (mm): -0.1	The test method did **NOT** lead to statistically significant results
	Control: Titanium curettes, stainless steel curettes, sterile saline, non-activated diode laser (n=15)	BoP (%): 62.8 PPD (mm): 5.3 MBL (mm): 2.3	BoP (%): -15.4 PPD (mm): -1.5 MBL (mm): -0.1	
Yayli et al., (2021)	**Test 1**: Diode laser (n=19)	BoP (%): 88.1 PPD (mm): 4.1	BoP (%): -26.2 PPD (mm): -0.9	**Er,Cr:YSGG** laser-assisted therapy significantly reduced **PPD** scores compared to diode laser and control treatment
	Test 2: Er,Cr:YSGG laser (n=17)	BoP (%): 100 PPD (mm): 4.5	BoP (%): -48.8 PPD (mm): -1.2	
	Control: (n=18)	BoP (%): 72.1 PPD (mm): 4.1	BoP (%): -11.3 PPD (mm): -0.5	

BoP: bleeding on probing; **MBL**: marginal bone loss; **PPD**: probing pocket depth; **SoP:** suppuration
[#]**improved**: no bone loss, PPD reduction ≥0.5 mm; **not improved**: bone loss

Appendix 2. Non-surgical and ***adjunctive laser therapies*** in peri-implantitis treatment: selected randomized clinical trials with ***12-month follow-up***

Author, (year)	Treatment group (number of implants)	Clinical outcomes (baseline)	Clinical outcomes (baseline vs 12 months)	Findings
Hentenaar et al., (2021)	**Test**: Air-polishing with erythritol powder (n=66)	Mean PPD (mm): 4.8 BoP (%): 93.9 SoP (%): 54.5 PI (%): 45.5 VAS: 2.1	Mean PPD (mm): - BoP (%): - SoP (%): - PI (%): - VAS: -	Limited clinical benefits at 12 months. The test method did **NOT** lead to statistically significant results.
	Control: Ultrasonic device with PEEK tip (n=73)	Mean PPD (mm): 5.0 BoP (%): 91.8 SoP (%): 42.5 PI (%): 43.8 VAS: -2.6	Mean PPD (mm): - BoP (%): - SoP (%): - PI (%): - VAS: -	
Selimovic et al., (2023)	**Test**: Ultrasonic/curette instrumentation, erythritol polishing (n=20)	Mean PPD (mm): 4.5 BoP (%): 59.7 SoP (%): 32.3 Mean CBL (mm): 3.6 VAS 1: 8.8 VAS 2: 7.4	Mean PPD (mm): -0.3 BoP (%): -23.2 SoP (%): -24.6 Mean CBL (mm): -0.1 VAS 1: +0.8 VAS 2: +1.4	Limited clinical benefits at 12 months. The test method did **NOT** lead to statistically significant results.
	Control: Ultrasonic device, curette instrumentation (n=20)	Mean PPD (mm): 4.4 BoP (%): 58.1 SoP (%): 16.7 Mean CBL (mm): 3.1 VAS 1: 9.4 VAS 2: 9.1	Mean PPD (mm): -0.6 BoP (%): -25.8 SoP (%): -10.2 Mean CBL (mm): +0.3 VAS 1: +0.1 VAS 2: 0.0	

BoP: bleeding on probing; **CBL**: crestal bone loss; **PI**: plaque index; **PPD**: probing pocket depth; **SoP:** suppuration; **VAS**: visual analogue scale

Appendix 3. Non-surgical and ***chemical adjunctive therapies*** in peri-implantitis treatment: selected randomized clinical trials with ***6-month follow-up***

Author, (year)	Treatment group (number of implants)	Clinical outcomes (baseline)	Clinical outcomes (baseline vs 6 months)	Findings
Alhumaidan et al., (2022)	**Test**: Plastic curettes, minocycline hydrochloride (n=12)	mGI: 1.1 mPI: 3.0 CBL, mesial (mm): 4.8 CBL, distal (mm): 5.1	mGI: -0.3 mPI: -0.4 CBL, mesial (mm): -0.5 CBL, distal (mm): -0.7	The test method did **NOT** lead to statistically significant results between smokers
	Control: Plastic curettes (n=12)	mGI: 1.1 mPI: 2.7 CBL, mesial: 5.1 CBL, distal: 5.1	mGI: -0.2 mPI: +0.1 CBL, mesial: -0.6 CBL, distal: -0.8	
Karring et al., (2005)	**Test**: Vector system (n=11)	Plaque (%): 23.7 BoP: (%): 63.6 PPD (mm): 5.8	Plaque (%): -14.6 BoP: (%): -27.2 PPD (mm): 0	The test method did **NOT** lead to statistically significant results
	Control: Carbon fiber curette (n=11)	Plaque (%): 23.7 BoP (%): 72.7 PPD (mm): 6.2	Plaque (%): -5.5 BoP (%): +9.1 PPD (mm): +0.6	
Laleman et al., (2019)	**Test**: Titanium curette, Air-N-Go Easy air polisher, probiotic drops (n=9)	BoP (%): 87 mBI: 1.92 PPD (mm): 5.2 PI (%): 15 Microbiological outcomes	BoP (%): -28 mBI: -1.0 PPD (mm): -1.0 PI (%): -13 Microbiological outcomes	The test method did **NOT** lead to statistically significant results
	Control: Titanium curette, Air-N-Go Easy air polisher, placebo drops (n=10)	BoP (%): 87 mBI: 1.96 PPD (mm): 5.5 PI (%): 8 Microbiological outcomes	BoP (%): -34 mBI: -0.74 PPD (mm): -1.3 PI (%): -1 Microbiological outcomes	
Merli et al., (2020)	**Test 1**: Nonsurgical debridement (n=15)	RxMeanBD (mm): 3.1 Mean PPD (mm): 4.2 Mean CAL (mm): 4.3 BoP sites: 2.9	RxMeanBD (mm): 0.2 Mean PPD (mm): 0.2 Mean CAL (mm): 0.1 BoP sites: 0.4 VAS during treatment: 2.1	The test method did **NOT** lead to statistically significant results. VAS scores for patients treated with glycine powder were significantly higher ($p < 0.05$)
	Test 2: Non-surgical debridement, desiccant (n=16)	RxMeanBD (mm): 4.0 Mean PPD (mm): 4.5 Mean CAL (mm): 4.9 BoP sites: 2.5	RxMeanBD (mm): -0.1 Mean PPD (mm): 0.5 Mean CAL (mm): 0.6 BoP sites: 0.2 VAS during treatment: 3.3	
	Test 3: Non-surgical debridement, glycine powder (n=12)	RxMeanBD (mm): 4.0 Mean PPD (mm): 4.8 Mean CAL (mm): 5.2 BoP sites: 2.8	RxMeanBD (mm): -0.2 Mean PPD (mm): 0.1 Mean CAL (mm): 0.1 BoP sites: 0.7 VAS during treatment: 3.9	
	Test 4: Non-surgical debridement, desiccant, glycine powder (n=14)	RxMeanBD (mm): 3.5 Mean PPD (mm): 4.0 Mean CAL (mm): 4.2 BoP sites: 2.7	RxMeanBD (mm): -0.1 Mean PPD (mm): 0.8 Mean CAL (mm): 0.7 BoP sites: 0.8 VAS during treatment: 5.0	

Sahm et al., (2011)	**Test**: Oral hygiene program, Perioflow with glycine powder (n=15)	PI: 1.2 BoP (%): 94.6 PPD (mm): 3.8 CAL (mm): 4.8	PI: -0.1 BoP (%): -43.5 PPD (mm): -0.6 CAL (mm): -0.4	Test group showed **significantly higher changes** in mean BoP ($p < 0.05$)
	Control: Oral hygiene program, carbon curettes (n=15)	PI: 1.0 BoP (%): 95.3 PPD (mm): 4.5 CAL (mm): 4.8	PI: -0.2 BoP (%): -11.0 PPD (mm): 0.5 CAL (mm): -0.5	
Schär et al., (2013)	**Test**: Titanium curettes, glycine-based powder air-polishing, photodynamic therapy, irrigation with 3% H_2O_2 (n=20)	Mean PPD (mm): 4.2 Mean CAL (mm): 2.7 BoP positive sites: 4.0 Mean mPI: 0.1	Mean PPD (mm): -0.4 Mean CAL (mm): -0.2 BoP positive sites: -2.5 Mean mPI: -0.1	The test method did **NOT** lead to statistically significant results, except for mPI.
	Control: Titanium curettes, glycine-based powder air-polishing, minocycline HCl microspheres, irrigation with 3% H_2O_2 (n=20)	Mean PPD (mm): 4.4 Mean CAL (mm): 2.7 BoP positive sites: 4.4 Mean mPI: 0.2	Mean PPD (mm): -0.5 Mean CAL (mm): -0.2 BoP positive sites: -2.3 Mean mPI: -0.2	
Schwarz et al., (2005)	**Test**: ERL laser treatment (n=16)	PI: 1.1 BoP (%): 83 PPD (mm): 5.4 CAL (mm): 5.8	PI: 0.0 BoP (%): -52 PPD (mm): -0.8 CAL (mm): -0.7	Test method led to **statistically significant higher reduction** of BoP
	Control: Laser treatment, 0.2% chlorhexidine digluconate solution irrigation, 0.2% chlorhexidine gel (n=16)	PI: 1.0 BoP (%): 80 PPD (mm): 5.5 CAL (mm): 6.2	PI: 0.0 BoP (%): -22 PPD (mm): -0.7 CAL (mm): -0.6	
Tada et al., (2017)	**Test**: Azithromycin 500 mg (1×3 days), 1 probiotic tablet 1 pr day (n=15)	PPD (mm): 3.9 BoP (0-6): 3.7 mPI (0-3): 1.7 mBI (0-3): 1.9	PPD (mm): -0.7 BoP (0-6): -1.7 mPI (0-3): -0.1 mBI (0-3): -0.9	**No** significant difference in **BoP** or **mPI** between test and control groups. The **mBI** scores were significantly lower.
	Control: Azithromycin 500 mg (1×3 days), 1 placebo tablet pr day (n=15)	PPD (mm): 4.0 BoP (0-6): 3.7 mPI (0-3): 1.5 mBI (0-3): 2.0	PPD (mm): -0.5 BoP (0-6): -1.4 mPI (0-3): 0.3 mBI (0-3): -0.5	

BoP: bleeding on probing; **CAL**: clinical attachment level; **CBL**: crestal bone loss; **mBI**: modified bleeding index; **mGI**: modified gingival index; **mPI**: modified plaque index; **PI**: plaque index; **PPD**: probing pocket depth; **RxMeanBD**: radiographic mean bone defect; **VAS**: visual analogue scale

Appendix 4. Non-surgical and ***chemical adjunctive therapies*** in peri-implantitis treatment: selected randomized clinical trials with ***12-month follow-up***

Author, (year)	Treatment group (number of implants)	Clinical outcomes (baseline)	Clinical outcomes (baseline vs 12 months)	Findings
Basetti et al., (2014)	**Test**: Titanium curette, glycine-based powder air-polishing, photodynamic therapy (n=20)	Mean BoP (of 6 sites): 4.0 Mean PPD (mm): 4.2 Mean CAL (mm): 2.7	Mean BoP (of 6 sites): -2.3 Mean PPD (mm): -0.1 Mean CAL (mm): -0.1	The test method did **NOT** lead to statistically significant results
	Control: Titanium curette, glycine-based powder air-polishing, minocycline microsphere (n=20)	Mean BoP (of 6 sites): 4.4 PPD (mm): 4.4 Mean CAL (mm): 2.7	Mean BoP (of 6 sites): -2.8 PPD (mm): -0.6 Mean CAL (mm): +0.3	
Blanco et al., (2022)	**Test**: Non-surgical debridement, systemic metronidazole (n=16)	PPD (mm): 6.7 CAL (mm): 7.3 FMBS (%): 39.9 FMPS (%): 20.9 RBL (mm): 6.3	PPD (mm): -2.4 CAL (mm): -2.2 FMBS (%): -19.1 FMPS (%): -9.8 RBL (mm): -2.3	Significant **improvements** in clinical, radiographic, and microbiological parameters were observed when systemic metronidazole was prescribed.
	Control: Non-surgical debridement, placebo (n=16)	PPD (mm): 5.9 CAL (mm): 6.1 FMBS (%): 50.5 FMPS (%): 21.2 RBL (mm): 5.5	PPD (mm): -1.0 CAL (mm): -0.6 FMBS (%): -21.0 FMPS (%): -12.4 RBL (mm): -1.1	
Gomi et al., (2015)	**Test**: Full-mouth scaling and root planing, azithromycin 3 days before treatment (n=10)	PPD (mm): 4.3 BoP (%): 27.9	PPD (mm): -1.0 BoP (%): -23.5	The reduction in **PPD** and **BoP** positive sites was **statistically significant** at 12 months ($p < 0.05$)
	Control: Full-mouth scaling ant root planing (n=10)	PPD (mm): 4.4 BoP (%): 25.7	PPD (mm): -0.2 BoP (%): -5.9	
John et al., (2015)	**Treatment 1**: Airflow, glycine powder (n=12)	PI: 1.2 BoP (%): 99.0 PPD (mm): 3.7 Mucosal recession (mm): 1.5 CAL (mm): 5.2	PI: +0.6 BoP (%): -41.2 PPD (mm): -0.5 Mucosal recession (mm): -0.1 CAL (mm): -0.6	The **Airflow group** showed significantly higher decrease in **BoP** scores ($p < 0.05$)
	Treatment 2: carbon curettes, chlorhexidine digluconate (n=13)	PI: 1.2 BoP (%): 94.7 PPD (mm): 3.9 Mucosal recession (mm): 1.0 CAL (mm): 5.0	PI: -0.3 BoP (%): -16.6 PPD (mm): -0.4 Mucosal recession (mm): -0.1 CAL (mm): -0.5	
Machtei et al., (2012)	**Test**: Supragingival plaque removal, chlorhexidine chip insertion (Periochip®) (n=146)	IPD (mm): 6.2 RAL (mm): 6.7 R (mm): 0.5 BoP (%): 100	IPD (mm): -1.7 RAL (mm): -1.5 R (mm): +0.3 BoP (%): 50.3	Changes in **R** and **RAL** were significantly larger for the **TEST** method
	Control: Supragingival plaque removal (n=144)	IPD (mm): 6.1 RAL (mm): 6.3 R (mm): 0.3 BoP (%): 100	IPD (mm): -1.5 RAL (mm): -1.4 R (mm): +0.2 BoP (%): 55.2	

Renvert et al., (2006)	**Test**: Oral hygiene instructions, mechanical treatment, minocycline microspheres (n=16)	Mean plaque score (%): 50 PPD (%): 3.9 Mean BoP (%): 88	Mean plaque score (%): -23 PPD (%): -0.3 Mean BoP (%): -17	The test method did **NOT** lead to statistically significant results
	Control: Oral hygiene instructions, mechanical treatment, chlorhexidine gel (n=14)	Mean plaque score (%): 45 PPD (%): 3.9 Mean BoP (%): 86	Mean plaque score (%): -24 PPD (%): 0 Mean BoP (%): -8	
Renvert et al., (2008)	**Test**: Scaling, root planing, minocycline microspheres (n=57)	PPD, four sites (mm): 3.9 LPS (%): 50 Enteric rods (%): 0.9 Enterococci (%): 0.1 *P. gingivalis**: 0.5 *P. intermedia**: 1.0 *T. forsythia**: 0.33 *A. actinomycetemcomitans**: 0.52 *F. nucleatum**: 0.38 *T. denticola**: 0.91	PPD, four sites (mm): -0.3 LPS (%): -28 Enteric rods (%): -2.4 Enterococci (%): -0.02 *P. gingivalis**: -0.45 *P. intermedia**: -0.48 *T. forsythia**: 0.17 *A. actinomycetemcomitans**: -0.45 *F. nucleatum**: +0.34 *T. denticola**: -0.34	A **significant difference** between the groups was observed for **PPD** reduction at six months. The reduction was **NOT** significant at 12 months. The reduction in **bleeding** sites was **statistically significant** at 12 months ($p < 0.05$).
	Control: Scaling, root planing, 1% chlorhexidine gel (n= 38)	PPD, four sites (mm): 3.9 LPS (%): 60 Enteric rods (%): 0.02 Enterococci (%): 0.12 *P. gingivalis**: 0.73 *P. intermedia**: 0.78 *T. forsythia**: 0.49 *A. actinomycetemcomitans**: 0.54 *F. nucleatum**: 0.41 *T. denticola**: 1.11	PPD, four sites (mm): -0.2 LPS (%): -33 Enteric rods (%): +1.95 Enterococci (%): +0.11 *P. gingivalis**: -0.62 *P. intermedia**: -0.26 *T. forsythia**: -0.22 *A. actinomycetemcomitans**: -0.54 *F. nucleatum**: -0.31 *T. denticola**: -0.52	
Shibli et al., (2019)	**Test**: Non-surgical debridement, metronidazole (400 mg), and amoxicillin (500 mg) (n=20)	PI (%): 56.6 GI (%): 10.4 PPD (mm): 7.0 CAL (mm): 7.2 BoP (%): 86.6 SoP (%): 8.8	PI (%): -7.2 GI (%): -6.8 PPD (mm): -3.1 CAL (mm): -3.0 BoP (%): -51.0 SoP (%): -8.8	The test method did **NOT** lead to statistically significant difference in PPD and CAL between the groups ($p > 0.05$)
	Control: Non-surgical treatment, placebo (n=20)	PI (%): 61.6 GI (%): 10.0 PPD (mm): 5.5 CAL (mm): 5.9 BoP (%): 85.0 SoP (%): 5.0	PI (%): +0.2 GI (%): -5.3 PPD (mm): -1.7 CAL (mm): -1.5 BoP (%): 44.7 SoP (%): +0.3	

BoP: bleeding on probing; **CAL**: clinical attachment level; **FMBS**: full-mouth bleeding score; **FMPS**: full-mouth plaque score; **GI**: gingival index: **IPD**: implant pocket depth; **LPS**: local plaque score; **PI**: plaque index; **PPD**: probing pocket depth; **R**: recession; **RAL**: relative attachment level; **RBL**: radiographic bone level; **SoP:** suppuration
***Based on mean score of 0 to 5

I

Received: 26 July 2023 | Revised: 23 December 2023 | Accepted: 30 December 2023

DOI: 10.1002/cre2.841

ORIGINAL ARTICLE

Clinical and Experimental Dental Research Open Access WILEY

Anatomical three-dimensional model with peri-implant defect for in vitro assessment of dental implant decontamination

Sadia Nazir Khan[1] | Odd Carsten Koldsland[2] | Hanna Tiainen[3] | Carl Hjortsjö[1]

[1]Department of Prosthetics and Oral Function, Faculty of Dentistry, University of Oslo, Oslo, Norway

[2]Department of Periodontology, Faculty of Dentistry, University of Oslo, Oslo, Norway

[3]Department of Biomaterials, Faculty of Dentistry, University of Oslo, Oslo, Norway

Correspondence

Sadia Nazir Khan, Department of Prosthetics and Oral Function, Faculty of Dentistry, Institute for Clinical Dentistry, University of Oslo, Oslo, Norway.
Email: sadiak@odont.uio.no

Funding information

Institute for Clinical Dentistry, Faculty of Dentistry, University of Oslo, Norway

Abstract

Objectives: Access to the implant surface plays a significant role in effective mechanical biofilm removal in peri-implantitis treatment. Mechanical decontamination may also alter the surface topography of the implant, potentially increasing susceptibility to bacterial recolonization. This in vitro study aimed to evaluate a newly developed, anatomically realistic, and adaptable three-dimensional (3D)printed model with a peri-implant bone defect to evaluate the accessibility and changes of dental implant surfaces after mechanical decontamination treatment.

Material and Methods: A split model of an advanced peri-implant bone defect was prepared using 3D printing. The function of the model was tested by mechanical decontamination of the exposed surface of dental implants (Standard Implant Straumann AG) coated with a thin layer of colored occlusion spray. Two different instruments for mechanical decontamination were used. Following decontamination, the implants were removed from the split model and photographed. Image analysis and fluorescence spectroscopy were used to quantify the remaining occlusion spray both in terms of area and total amount, while scanning electron microscopy and optical profilometry were used to analyze alteration in the implant surface morphology.

Results: The 3D model allowed easy placement and removal of the dental implants without disturbing the implant surfaces. Qualitative and quantitative assessment of removal of the occlusion spray revealed differences in the mechanism of action and access to the implant surface between tested instruments. The model permitted surface topography analysis following the decontamination procedure.

Conclusion: The developed 3D model allowed a realistic simulation of decontamination of implant surfaces with colored occlusion spray in an advanced peri-implant defect. 3D printing allows easy adaptation of the model in terms of the shape and location of the defect. The model presents a valuable tool for in vitro investigation of the accessibility and changes of the implant surface after mechanical and chemical decontamination.

KEYWORDS

CADCAM, dental implants, mechanical decontamination, peri-implantitis

1 | INTRODUCTION

Despite high survival rates, dental implants encounter biological complications, including peri-implant bone loss. Peri-implantitis affects 20%–35% of implant patients and 10%–20% of implants, varying with disease classification (Atieh et al., 2013; Kordbacheh Changi et al., 2019; Romandini et al., 2021). Effective decontamination of implant surfaces and complete biofilm removal is essential in treating peri-implantitis and sustaining healthy peri-implant tissues (Costa et al., 2018).

Several chemical, mechanical, and laser treatments have been suggested for disrupting bacterial plaque on implant surfaces (Verket et al., 2023). The most proposed devices for mechanical decontamination include titanium and plastic curettes, air abrasives, and ultrasound devices. These instruments can be used alone or combined and may also act as carriers for topical antimicrobial agents. However, the synergistic effect of mechanical nonsurgical or surgical treatments combined with adjunctive therapies has demonstrated limited efficacy (Ramanauskaite et al., 2021). Access to all implant surfaces is crucial for reducing the bacterial load, as residual biofilm and hard deposits on the implant surface may sustain inflammation in the peri-implant soft and hard tissues.

The hard tissue morphology of the bone defect and other local factors impact the clinical outcome of decontamination treatment (Monje, Pons, et al., 2019; Schwarz et al., 2010). Chronic inflammation may cause anatomical changes resulting in soft tissue recession, bone configuration changes, and intraorally exposed implant surface. Since peri-implant bone defects vary in configuration and severity (Schwarz et al., 2010), it is uncertain whether the tools for mechanical decontamination can access all areas of the implant surface for effective biofilm removal.

The decontamination capability of commercially available devices has been investigated in several in vitro and in vivo studies. In vitro decontamination studies have been performed on titanium objects such as discs, coins, sheets, healing abutments, and various other objects simulating the dental implant surface (John et al., 2014; Louropoulou et al., 2014; Pereira da Silva et al., 2005; Sánchez et al., 2011). Studies evaluating decontamination effectiveness on dental implants have been conducted on implants alone or implants inserted in blocks of different materials (Bermejo et al., 2019; Cha et al., 2019; Keim et al., 2019; Nemer Vieira et al., 2012). In recent publications, three-dimensional (3D) blocks holding dental implants have been designed with different bone defect configurations to evaluate the decontamination effectiveness in the presence of varied bone defect anatomies and teeth (Keim et al., 2019; Korello et al., 2023; Luengo et al., 2022; Ronay et al., 2017; Steiger-Ronay et al., 2017). While these models allow for decontamination assessment in the presence of various defect angulations and configurations, they may not fully capture the complexity of real-life clinical situations, where access to one implant surface may be more challenging than others due to differences in defect morphology on site level and the presence of adjacent teeth. Anatomically realistic in vitro 3D models provide a valuable tool for evaluating decontamination effectiveness at the site level. Furthermore, these models may serve as educational tools, providing students and professionals with an enhanced understanding of decontamination and its associated challenges.

Computer-aided design and manufacturing (CADCAM) combined with 3D printing enables the fabrication of accurate models of complex objects (van Noort, 2012). CADCAM facilitates the modification of peri-implant defects, tooth anatomy, implant angulation, and implant-retained supraconstructions, simulating various clinical scenarios. Models with anatomical design, nonsymmetrical peri-implant defects, and access to remove the implant without disturbing the implant surface have not been manufactured or evaluated in the published literature.

This study aimed to assess the feasibility of an anatomically realistic in vitro 3D-printed model with an advanced peri-implant defect to evaluate access and mechanical damage to implant surfaces caused by different tools for mechanical decontamination.

2 | MATERIALS AND METHODS

2.1 | In vitro 3D model design and manufacturing

A phantom dental model (N222; Colombia Dentoform Corporation) was scanned using a workbench scanner (S600 ARTI, Software Zirkonzahn.Scan, Version 5051; Zirkonzahn®). One premolar was removed digitally with a computer-aided design (CAD) program (Zirkonzahn.Archive, Version 7058; Zirkonzahn®). A 3b bone defect was designed in the edentulous premolar area as described by Monje, Pons et al. (2019). A titanium dental implant (Standard Implant, Ø 4.1 mm RN, SLA® 12 mm; Straumann AG) was scanned. The phantom dental model scan and the dental implant scan were superimposed in a planned position (Zirkonzahn.Modellier, Version 6173_6958_x64, Zirkonzahn®). The phantom model was split into two at the center of the occlusal surface of the teeth (Meshmixer™, Version 3.5.474; Autodesk®). A volume according to the scanned dental implant was stamped out digitally (Zirkonzahn.Modifier, version v.21.3_6.25448; Zirkonzahn®). STL files were forwarded to a slicer (CHITUBOX V1.8.1) and to a 3D printer (Phrozen Sonic XL 4K; Phrozen Tech Co., Ltd). The 3D models were printed in a resin material (Fotodent® model, 385/405 nm, Ref. D35400, Dreve [$n = 12$]). The printed models were rinsed with 95% ethanol (Antibac®) and a liquid for solvent cleanser (IMPRIMO® Cleaning Liquid; SCHEUGROUP) in an ultrasonic bath for 40 min (Model no. UCI-230, SERIAL NO 931236817; Colténe/Whaledent Inc.). After cleansing, the models were light cured (Photopol. 230 V-50/60 Hz 300 W; Dentalfarm) with light-emitting diode (LED)/UV cure replacement bulbs (LED 9 W 405 nm; FEPshop BV). The cured models were assembled (Figure 1a) and scanned using the same workbench scanner as previously introduced ($n = 5$). Scans were superimposed to assess the similarity of the printed 3D models using the same CAD program as used for designing the 3D model. The dimensions of the peri-implant defect surrounding the inserted implant were digitally measured using the same software (Figure 2).

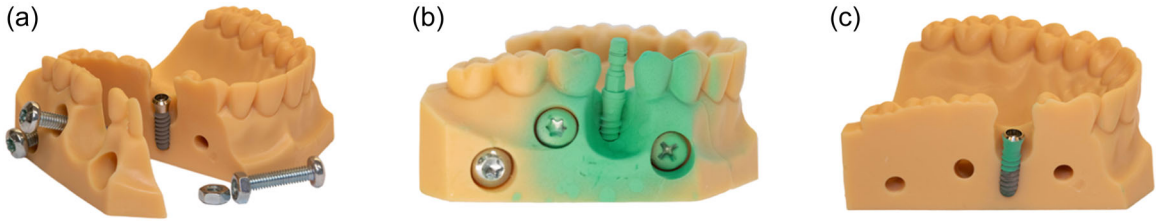


FIGURE 1 Anatomically realistic, three-dimensional, printed resin model with an advanced peri-implant defect. The model was designed with a split design for easy access to implant removal. (a) The dental implant was inserted into the split model and the parts were fixed with screws. (b) The exposed implant was sprayed with the colored occlusion spray after the model had been assembled. (c) The front panel of the model was unmounted for easy removal of the implant. The colored spray was not spread beyond the exposed area.

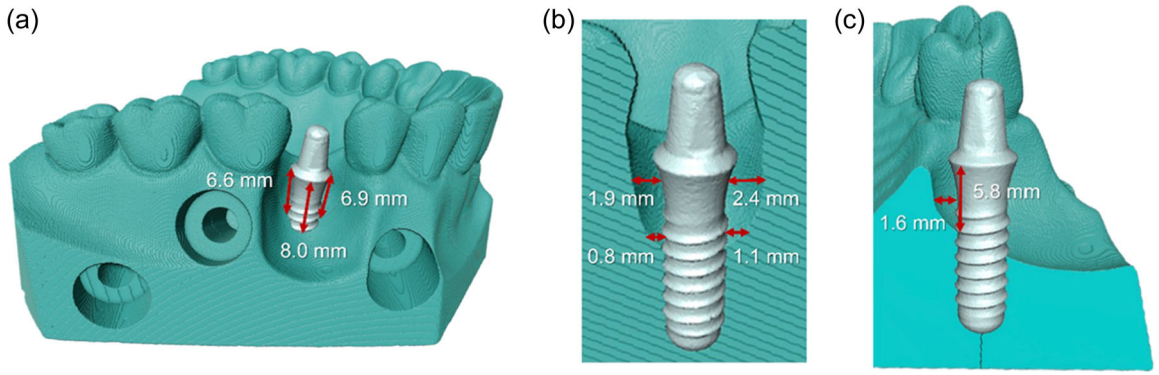


FIGURE 2 Peri-implant defect dimensions measured with the computer-aided design software (mm). (a) Vertical height at the mesial, buccal, and distal implant surface. (b) Horizontal defect dimensions at the second thread and the implant neck. (c) Vertical and horizontal defect dimensions measured at the palatal implant surface.

Sterile titanium dental implants (Standard Implant, Ø 4.1 mm RN, SLA® 12 mm; Straumann AG) ($n = 36$). The implants were carefully mounted to the split models, and the parts were put together with nuts and bolts (DIN 934 M4 G 340004&M4X20; Arvid Nilsson). The exposed implant surface was coated with a thin even layer of colored occlusion spray 1 h before decontamination treatment (Okklufine Premium; FINO GmbH) (Figure 1b,c). The film thickness was assessed by using optical profilometry (S neox, Sensofar). For this purpose, a small area on the machined implant collar was masked using polyvinyl siloxane impression material (Provil® novo; Heraeus) during the application of the occlusion spray layer on the implant surface ($n = 3$). The mask was carefully removed, and three nonoverlapping areas ($0.87 \times 0.65\ mm^2$) of the boundary region between the sprayed and unsprayed implant surface were imaged on each sample in confocal mode using red light and EPI ×20 objective. The average step height between the spray film and the masked area was then measured from each recorded image ($n = 9$) (Sensomap Standard 7.4; Sensofar).

2.2 | Decontamination strategy

To test the ability of the model to reveal differences in decontamination outcomes following various decontamination protocols, the exposed implant surface was mechanically decontaminated using decontamination methods with different modes of action operated by one experienced clinician using the following methods with implants receiving no decontamination treatment serving as controls ($n = 6$):

1. Oscillating chitosan brush (OCB; Bioclean®; Labrida).
2. Polyetheretherketone tip for an ultrasonic unit (US-PEEK; PI Instrument®; E.M.S. Electro Medical Systems) with water irrigation.
3. Irrigation with water spray using a three-way dental syringe (water).

To assess the capacity of the model to show differences between treatment outcomes of additional chemical debridement treatments in combination with OCB, four additional groups were tested ($n = 3$):

4. OCB with water irrigation (OCB + water);
5. OCB with chlorhexidine gel (CG; Corsodyl®; 1% Dental Gel) (OCB + CG);
6. OCB with blank gel (BG; 5% (wt/vol) methylcellulose; Sigma-Aldrich) dissolved in deionized water (OCB + BG).
7. CG (Corsodyl®; 1% Dental Gel).

All OCBs were soaked in 0.9% sodium chloride solution for 2 min before being attached to an oscillating handpiece (NSK) at 1000 revolutions per minute.

Decontamination was performed on the exposed part of the implant for 2 min for all treatment groups. Following decontamination, the 3D models were photographed before the implants were carefully dismounted from the open models without disturbing the implant surface. Remaining moisture and loose particles were removed from the implant surface using clean, pressured air before photographing the samples. The implants were numbered groupwise before decontamination assessment, and the operator performing the implant characterization was therefore blinded for treatment allocation.

2.3 | Decontamination assessments

2.3.1 | Photographing image analysis

The implants were fixed in a custom-made holder by a transfer piece and photographed from the buccal, distal, palatal, and mesial sites with a single-reflex camera (Nikon D3200 equipped with a Nikon 105 mm f/2.8D AF Micro Nikkor macro lens). For this purpose, the transfer piece was marked on its buccal side with a marker pen before removing the implants from the model. Imaging was performed at a fixed angle perpendicular to the height axis of the implant using fixed imaging settings and lighting conditions. Images were calibrated for white balance and contrast (Adobe Lightroom Classic) and binarized based on color histograms (ImageJ®; Figure S1). Implant diameter was used to calibrate the scale of the images. Based on the recorded images, the area still covered with the occlusion spray was determined on site level using ImageJ (Schneider et al., 2012). To compensate for potential minor alterations in implant orientation and imaging angle between experiments and test groups, all quantitative results of the image analysis are presented as a ratio between the area covered with green occlusion spray and the total projected implant area for each implant in percentage. The analyzer was blinded to decontamination allocation. Image analysis was repeated three times for each group.

2.3.2 | Fluorescence spectroscopy

To assess the total amount of occlusion spray remaining on the implant surface, the implants were then unmounted from the holder and placed in 1.7 mL microcentrifuge tubes after carefully removing the transfer piece from the implants. The remaining occlusion spray film was removed by bath sonicating the implants in 1 mL isopropanol for 20 min at 60°C. To reduce solvent evaporation, the closed microtubes were sealed with Parafilm® M laboratory film (Bemis Company) for the duration of the sonication. The solvent containing the dispersed occlusion spray particles was collected and used to estimate the amount of remaining occlusion spray film using a fluorometer (Qubit 4; Invitrogen). Fluorescence values were read using blue LED excitation at 470 nm and recording emission at far red (665–720 nm) wavelengths and converted to wt/vol concentration via a standard curve recorded for serial dilutions of known concentration of the used fluorescent occlusion spray particles dispersed in the same solvent (Figure S2).

2.3.3 | Evaluation of surface topography

Following removal of the occlusion spray film from the implant surface, the implants were dried using clean pressured air and imaged using a tabletop scanning electron microscope (SEM; TM-3030; Hitachi) and optical profilometer (S neox; Sensofar) to assess the extent of potential mechanical damage caused on the machined and SLA implant surface ($n = 3$). For SEM imaging, the samples were fixed on an aluminum holder using conductive carbon and adhesive copper tape and imaged by detecting backscattered electrons generated at 15 kV acceleration voltage. The surface topography of the decontaminated implants was further visualized and quantified using optical profilometry. An area of $0.87 \times 0.65\ mm^2$ at three randomly chosen nonoverlapping positions on the buccal surface of both the machined implant collar and the sandblasted and acid-etched implant screw was imaged in confocal mode using EPI ×20 objective ($n = 3$). Image processing (form removal and Gaussian filter: nesting index 50 µm) and quantification of surface parameters were performed using SensoMap Standard 7.4 (Sensofar).

2.4 | Statistical analysis

The implant area covered with residual occlusion spray versus the total implant surface was described using percentage. All image measurements were performed three times, and the means and standard deviations were calculated. The means and SD of the fluorescence measurements for each decontamination group and the control implants were calculated. For the optical profilometry data, the means and SD for the machined implant collar and the sandblasted acid-etched implant screw were calculated. Differences between the decontamination groups and implant sites were tested using one-way analysis of variance. Statistical significance was considered at $p < .05$. Statistical analyses were performed using SPSS (IBM Statistics, Version 28.0.1.1.14; IBM).

3 | RESULTS

3.1 | Peri-implant defect model

The printed 3D models showed minimal variation in model parts and the defect area (Figure 3). All models showed the exact precise fit between the components. Implants were inserted identically, exposing the same number of threads at the respective surfaces. Buccally, the defect led to the exposure of four threads (8.0 mm). Distally and mesially, two threads (6.6 and 6.9 mm, respectively) and palatally one thread (5.8 mm) were exposed as illustrated in Figure 2, which also

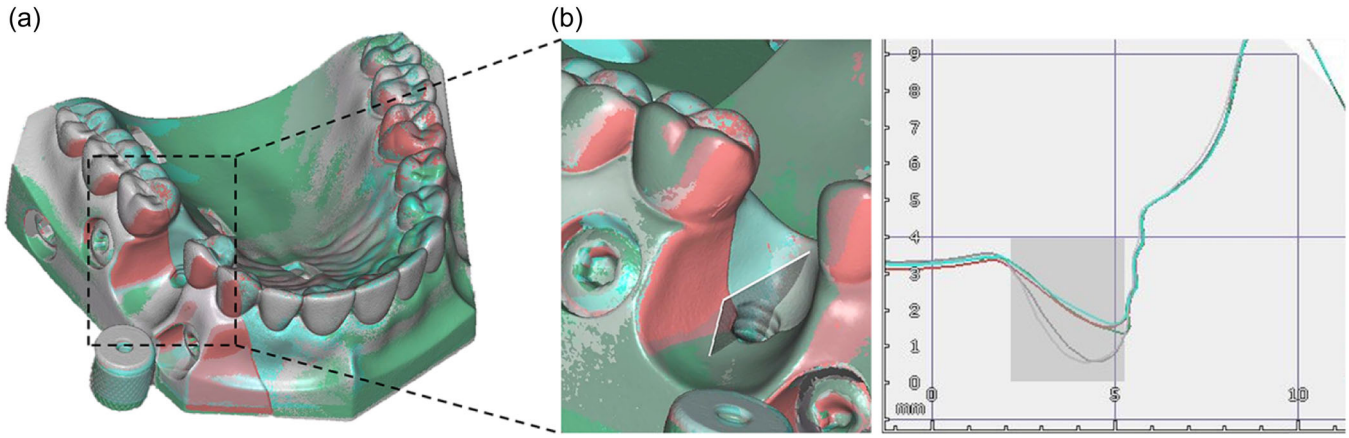


FIGURE 3 Evaluation of printed three-dimensional (3D) model similarity using computer-aided design software. (a) Five printed models were scanned and assigned five distinct color codes before they were superimposed. The superimposed models exhibited minimal variation in model parts or defect configurations. (b) Dimensional variations within digitally cut cross-sections of the superimposed 3D scans of the printed model were limited to a few micrometers and are considered insignificant for the function of the model. These minimal variations are caused by shrinkage during final curing of the model and are smaller than the resolution of the 3D printing used to produce the models. Area highlighted in gray corresponds to the volume occupied by the implant and cannot therefore be scanned accurately due to restricted access to light within this area of the model during scanning.

illustrates the horizontal dimensions of the peri-implant defect. The amount of occlusion spray on the unexposed implant surface was minimal (Figure 1c). The film thickness of the occlusion spray layer on the implant surface was 9.6 ± 1.4 µm, as measured by optical profilometry and illustrated with SEM images in Figure S3. The occlusion spray was composed of solid inorganic particles embedded in a polymeric matrix and morphologically resembled a multispecies biofilm grown on titanium implant surface (Figure S3A–D).

3.2 | Visual inspection and image analysis of the implant surface decontaminated mechanically

Visual inspection of the implant surfaces in the control group demonstrated completely intact occlusion spray with a pattern analogous to the defect morphology (Figure 4a and Figure S4). A similar intact layer of green occlusion spray was observed on implant areas not reached by the decontamination instruments in the OCB and US-PEEK groups, particularly in areas adjacent to the defect margins and the valley areas immediately below the threads in US-PEEK samples.

Implants decontaminated with the OCB showed efficient removal of the green occlusion spray from the machined implant collar. However, the occlusion spray appeared visibly darker green for the threaded implant area compared to the control implants, indicating that the OCB accessed the entire surface but was not able to decontaminate the surface fully. In contrast, only partial removal of the colored occlusion spray with visible areas of intact spray layer at all four implant sites for both the machined collar and the threaded implant area was observed for the US-PEEK group (Figure S4). Although the US-PEEK instrument did not reach the entire surface, a

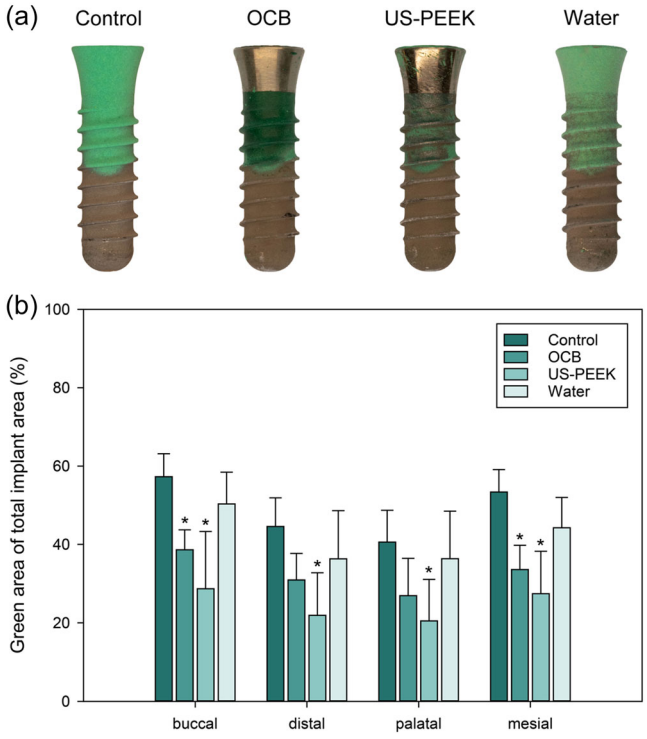


FIGURE 4 Image analysis of the area covered with residual occlusion spray versus total implant area. (a) The extent of occlusion spray coverage on the buccal surface of the control implant and changes in the occlusion spray coverage after mechanical decontamination treatment. (b) The ratio of area covered with remnant occlusion spray versus total implant area was calculated for control implants and implants treated with mechanical decontamination ($n = 6$, mean ± SD, $^*p < .05$ compared to control at respective implant site). OCB, oscillating chitosan brush; US-PEEK, ultrasonic polyetheretherketone tip.

significantly larger amount of green occlusion spray was removed from the threaded area compared to the OCB group (Figure 4b).

No decontamination method resulted in complete removal of the occlusion spray, as shown in Figure 4b. At the buccal and mesial implant surfaces, the removal of the occlusion spray showed statistical significance in the OCB and US-PEEK groups when compared to the control group ($p < .05$). Notably, the occlusion spray removal was significantly higher in the US-PEEK group compared to the control group at all four sites ($p < .05$), whereas there was no statistically significant difference between the water and control group at any implant site.

Overall, the implants in the water group demonstrated the least removal of the occlusion spray, with results resembling the control group. The green occlusion spray color in the water spray group was slightly lighter at both the implant neck and the threaded implant area compared to the control implants. Nevertheless, no implant areas were displayed without the occlusion spray in the water group, indicating that the occlusion spray was affected by the water but not removed.

3.3 | Visual inspection and image analysis of combined chemical and mechanical decontamination

Equivalent to the water group, the occlusion spray was minimally affected by the CG, as shown in Figure 4 and Figure S5. Furthermore, OCB combined with water irrigation showed results similar to OCB without water irrigation, with a clean implant neck and a darker green occlusion spray layer at the threaded implant area. OCB combined with CG was the group that differed from all other decontamination groups as minimal to no occlusion spray remained at the machined implant neck and the threaded area after decontamination (Figure 5a and Figure S5). A clean implant neck and darker green occlusion spray on the threaded part were displayed when OCB was combined with BG. However, the darker green occlusion spray layer seemed thinner, and more of the implant surface was shown through the occlusion spray compared to the OCB group. The image analysis showed no statistically significant difference in the removal of the occlusion spray between the OCB combined with water or BG compared to OCB without irrigation at any of the four implant sites. For OCB combined with CG, the removal of occlusion spray was significantly higher compared to all other OCB groups as well as the CG and the control group for all implant sites ($p < .05$; Figure 5b).

3.4 | Assessment of residual occlusion spray

Residual occlusion spray was removed from the implant surface with sonication in isopropanol. Fluorescence spectroscopy results showing the amount of residual occlusion spray on the implant surface after decontamination are displayed in Figure 6.

Implants decontaminated with OCB showed higher amount of residual occlusion spray than the water spray, control, and US-PEEK group. The high fluorescence values were also demonstrated when OCB was combined with water due to difficulty of dispersing condensed residual occlusion spray detached from the implant surface. A decrease in the values was shown when OCB was combined with the BG. The lowest values were obtained for OCB combined with CG, corroborating the results from the image analysis.

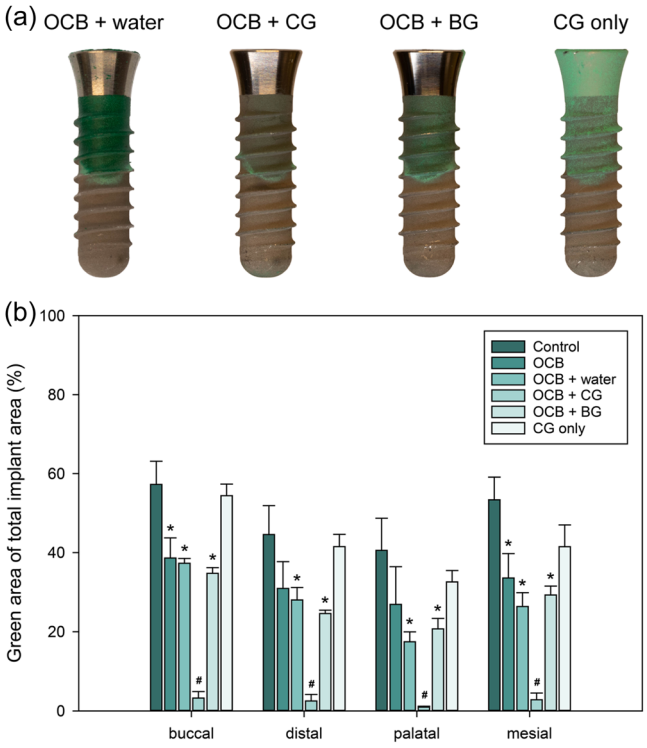


FIGURE 5 Qualitative and quantitative analysis of residual spray on implant surfaces. (a) Changes in the colored occlusion spray on the buccal implant surface after chemical or combined mechanical and chemical decontamination treatment. (b) Area with remnant occlusion spray versus total implant area for the control implant and implants decontaminated with chemical or combined chemical and mechanical decontamination ($n = 3$, mean ± SD, $^{*}p < .05$ compared to control at respective implant site, $^{\#}p < .05$ compared to all other groups at respective implant site). BG, blank gel; CG, chlorhexidine gel; OCB, oscillating chitosan brush.

3.5 | Surface topography

Figure 7a presents the SEM images for the control and the decontaminated machined and SLA surfaces. The images showed notable changes in the machined and threaded implant area in the US-PEEK group, displayed as scratching and disruptions. The scratches were irregular in size and shape and distributed throughout the machined implant neck. For the threaded implant area, pronounced flattening of the microscale peaks on the sandblasted surface was observed, especially on the apex of the implant threads. OCB and water spray groups revealed no visible alterations in the surface texture of the machined implant neck or the SLA-treated implant body.

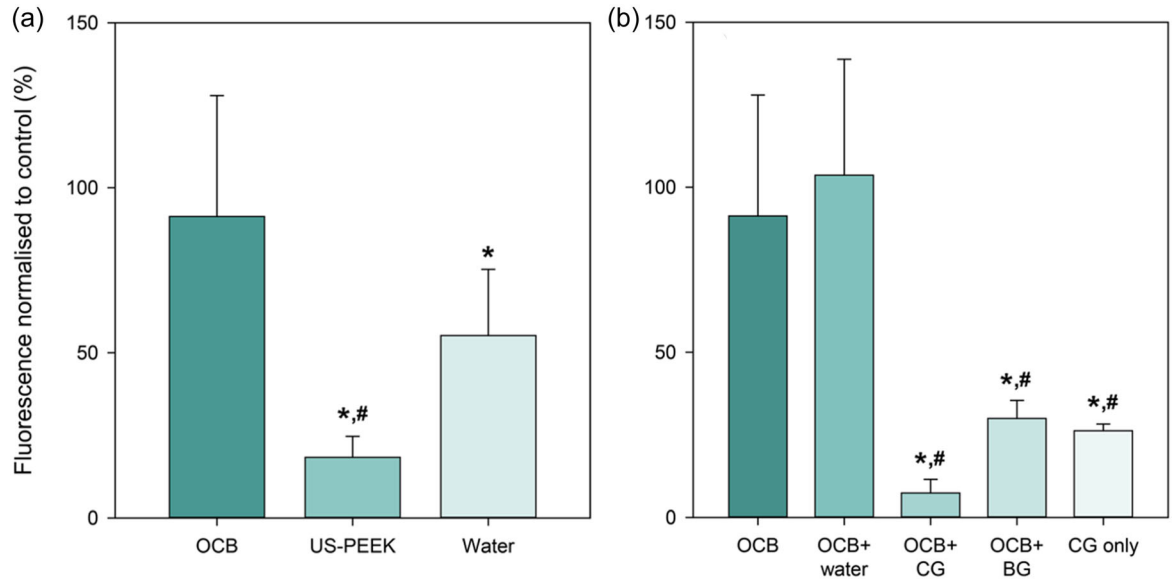


FIGURE 6 The mean fluorescence intensity of remnant occlusion spray indicating the total amount of unremoved colored occlusion spray on the decontaminated implant surface normalized to control (mean ± SD, (a): $n = 6$, (b) $n = 3$, $^{*}p < .05$ compared to control, $^{\#}p < .05$ compared to OCB). BG, blank gel; CG, chlorhexidine gel; OCB, oscillating chitosan brush; US-PEEK, ultrasonic polyetheretherketone tip.

The results obtained from the optical profilometry are displayed in Figure 7b. No statistically significant difference in the average surface roughness (Sa), surface skewness (Ssk), kurtosis (Sku), or core fluid retention (Sci) was observed between the groups for the machined or the SLA surface.

4 | DISCUSSION

This study aimed to evaluate a CADCAM-produced anatomical 3D model to investigate the efficacy of mechanical and chemical decontamination of dental implants. CADCAM production was selected because it allows easy modification of peri-implant defect configuration and dimensions. Furthermore, the method facilitates the production of models with various nonsymmetrical defects, potentially within the same model. It also allows the design of bone defects based on data from cone beam computed tomography or intraoral scans obtained during surgical intervention. The precision and reproducibility of the models are assured by machine fabrication. The model can be designed for integration in a phantom head for educational purposes and hands-on clinical training.

The effectiveness of tools for mechanical decontamination of dental implants has previously been investigated using 3Dprinted models with different standardized defect configurations as carriers for dental implants (Korello et al., 2023; Luengo et al., 2022; Matsubara et al., 2020). Contrary to the present study, the models were not designed anatomically with a split design. While evaluating decontamination using nonanatomical models with symmetrical defects is valuable, incorporating nonsymmetrical defects with realistic contours offers a more clinically relevant assessment of decontamination methods in vitro.

The bone defect shape is another factor influencing access to contaminated implant surfaces. Understanding the application of treatment methods in varying defect configurations may provide relevant knowledge for managing peri-implant diseases (Keim et al., 2019; Luengo et al., 2022). Moreover, the accessibility of implant surfaces may differ at the site level, particularly in nonsymmetrical defects and the presence of teeth. The use of a model with a split design facilitates the evaluation of the implant on specific sites.

In addition to addressing bone defect morphology, replicating biofilms poses additional challenges, as simulations do not fully replicate the clinical situation and behave differently compared to natural biofilm. A common approach in in vitro studies to evaluate the efficacy of decontamination methods is using ink staining as a biofilm mimic (Luengo et al., 2022; Matsubara et al., 2020; Mensi et al., 2020; Ronay et al., 2017; Sahrmann et al., 2015). The simulated biofilms do not adhere to the implant surface in the same manner as natural biofilms. Additionally, using sprays and inks introduces a treatment bias as they are visible to the operator. However, this bias could be circumvented using colored filters that absorb light at the specific wavelengths of the ink or spray.

In the present study, *colored* occlusion spray, simulating biofilm, was applied to implant surfaces. In the control group, implants demonstrated consistent spray application to the exposed implant surface. The implants were removed, preserving intact occlusion spray and indicating no spray application on parts embedded in the model, as demonstrated in Figure 1. The model had a split design to facilitate the convenient insertion and removal of dental implants without disturbing the biofilm or coatings. This feature enables the reuse of the model components in multiple experiments, particularly with autoclavable resin.

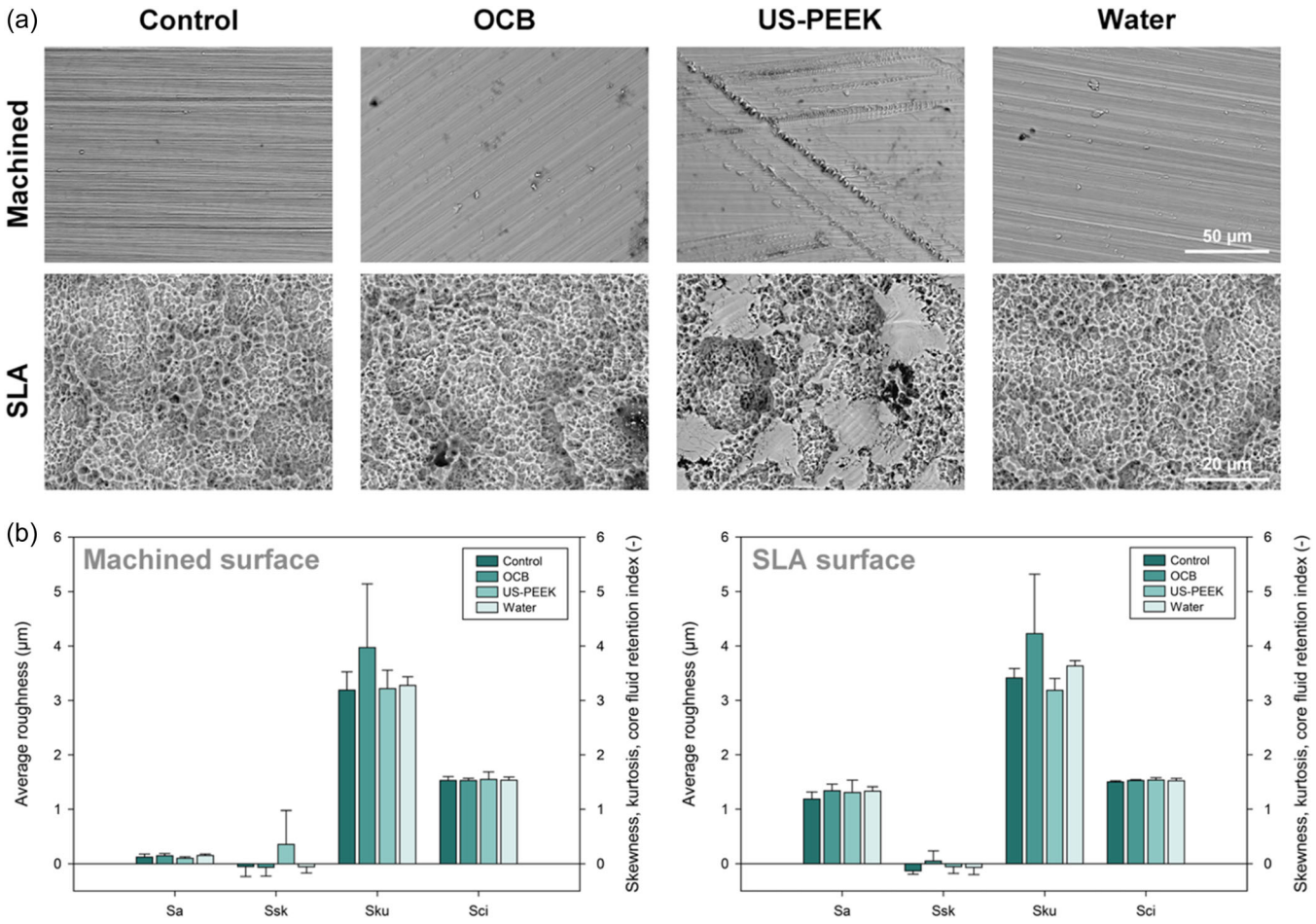


FIGURE 7 (a) Micromorphologic surface changes analyzed on scanning electron microscopy images (×2000 magnification) of an untreated control implant and implants after decontamination. Visible changes of surface topography were only observed in the US-PEEK group. (b) Average surface texture parameters of the decontaminated machined implant neck and the SLA surface showed no remarkable changes in the overall surface topography. ($n = 3$, mean ± SD). OCB, oscillating chitosan brush; US-PEEK, ultrasonic polyetheretherketone tip.

Colored occlusion spray, containing microscale particles embedded in a polymeric matrix, was used to mimic biofilm (Figure S3). In composition, the occlusion spray is assembled as biofilm and may provide a better simulation of biofilm than ink (Figure S3,E,F). The use of colored occlusion spray to mimic biofilm was previously reported by Tuna et al. (2019) in an in vitro study with the aim of studying the removal of simulated biofilm on implant-retained restorations.

Visual inspection of implants after decontamination showed various distinct differences in the residual occlusion spray layer for each tested decontamination method, demonstrating the capacity of this model to reveal differences in not only the access to the implant surface but also the mechanism of action and the efficacy of the different tested decontamination strategies in removing the occlusion spray from the surface. Implant areas that mechanical devices could not reach displayed an intact green shade similar to the spray layer on the control implants. In contrast, areas with complete decontamination had no residual occlusion spray. A dark, green, glossy surface of the residual occlusion spray layer was found on the SLA surface, regardless of whether or not water irrigation was used in the OCB groups (Figures 4 and 5). In the absence of any residual occlusion spray on the machined implant collar, this demonstrated that OCB reached the entire implant surface but was incapable of removing the spray in the micro-rough surface of the implant body. Furthermore, the reduced thickness of the residual spray layer (Figure 6b) when combined with a gel (CHX or BG) was visually observed as a pale shade of green (Figures 5 and Figure S5), indicating that the gel was capable of assissting the mechanical decontamination process using this device. These results illustrates that the presented 3D model, combined with occlusion spray as biofilm mimic, allows to differentiate between restricted accessibility and limited decontamination capacity.

In vitro decontamination studies using ink as biofilm simulation report quantitative comparisons between the methods (Luengo et al., 2022; Matsubara et al., 2020). In the present study, residual green occlusion spray assessment following decontamination involved visual inspection, image analysis, and fluorescence spectroscopy. Furthermore, the present study demonstrates the implementation of fluorescence spectroscopy to analyze the total amount of occlusion spray remaining on implant surfaces, extending beyond assessments of the residual spray area analyzed through image

analysis. Fluorescence evaluation distinctly differentiated the instruments' efficiency in removing or compressing the occlusion spray more tightly, as observed in the OCB group. Following the OCB decontamination process, the colored occlusion spray exhibited densely packed and adhered to the SLA surface, posing challenges during the sonication removal. The sonication procedure did not effectively break up the larger, condensed occlusion spray particles. Consequently, this caused an overestimation of the spray content, which was observed as elevated fluorescence levels in the OCB groups.

In the present study, two machine-driven decontamination tools with distinct mechanisms of action were selected to assess the novel, anatomically realistic, 3D-printed model. The defect configuration was designed to differ in depth and width across implant surfaces to evaluate the instruments' reach. In line with comparative studies utilizing ink (Luengo et al., 2022; Matsubara et al., 2020), the colored occlusion spray was not completely removed in any decontamination group. However, the decontamination groups demonstrated significantly better spray removal at the buccal and mesial implant sites than the control group ($p < .05$). For the decontamination methods, the US-PEEK group performed significantly better in occlusion spray removal compared to the control groups at all four sites ($p < .05$), demonstrating the functionality of the model.

In contrast, decontamination with OCB exhibited greater efficiency in removing occlusion spray from the machined implant collar compared to the rough SLA implant surface. The residual occlusion spray for OCB remained prominent on all sites within the SLA area, however, with a changed green shade. The change in the green color indicated that the OCB reached all implant areas: threads, valleys, and flanks, but was unable to remove the green occlusion spray from the roughened SLA surface of the implant. This observation was further supported by the high fluorescence values, indicating aggregated spray material. These results show that the developed 3D model allowed for the evaluation of the implants at the site level and the comparison of machined and threaded implant surfaces.

Additional elements such as water irrigation and CG were incorporated to further assess the model. The impact of water irrigation on the results for the US-PEEK group was investigated through the incorporation of a decontamination group with water-only. Implants in the water group showed no removal of occlusion spray. Additionally, the mechanism of action for OCB was further investigated by adding the following decontamination groups to the study: OCB with water, OCB with CG, and OCB with BG. Decontamination groups of OCBs with or without water showed similar results with darker green remnant occlusion spray. These findings suggested that the results for the US-PEEK were not attributed to sonication and not simultaneous water use. The inclusion of chlorhexidine gel was motivated by its frequent clinical use. A BG group was established to determine whether the efficacy in spray removal was due to the gel viscosity or its specific content. When OCB was combined with BG or CG, all OCB groups showed effective removal of the occlusion spray at all four sites compared to the control group ($p < .05$). The fluorescence analysis revealed that OCB combined with CG was more effective in removing occlusion spray than CG alone, both quantitatively and qualitatively. Effective removal of the occlusion spray using the OCB combined with CG can be explained by the isopropanol content in CG and its capacity to dissolve the colored occlusion spray. However, CG alone was inadequate to dissolve a substantial amount of the occlusion spray from the implant surface, as shown when CG was used without OCB. Thus, the combined mechanical action of OCB and the chemical properties of chlorhexidine enhanced the removal of the occlusion spray. This outcome indicates the utility of the 3D model in assessing the efficacy of combined mechanical and chemical decontamination methods.

Efficient decontamination of the threaded implant area was demonstrated for the US-PEEK group. However, the SEM images showed scratching of the machined implant collar and flattening porosities in the threaded SLA implant area. In contrast, the OCB group showed efficient decontamination of the machined implant neck while preserving intact surface topography. Bacterial colonization on implants is influenced by surface roughness (Louropoulou et al., 2012). The roughness and threaded design make the implant surface more prone to biofilm buildup than natural teeth (Quirynen & van Steenberghe, 1993). The risk is also present on smooth implant surfaces (Quirynen & Bollen, 1995). Scratching of the machined implant area may facilitate biofilm accumulation, potentially leading to inflammation and bone loss in susceptible patients. However, no direct evidence links instrument-induced roughness to biofilm accumulation (Louropoulou et al., 2012; Monje, Insua, et al., 2019). Furthermore, micromechanical damage to the implant surface may cause corrosion and release of nanoscale particles into surrounding tissues, potentially triggering adverse reactions in peri-implant tissues (Kotsakis & Olmedo, 2021).

Data from optical profilometry showed no statistically significant differences between the decontamination groups or the implant surfaces. However, the SEM images revealed scratching of the machined implant neck and the threaded area in the US-PEEK group. The variation between profilometry and SEM results may be attributed to differences in magnification. SEM images were obtained at ×2000, whereas profilometry used a ×20 objective, resulting in larger region of interest containing only few scratches on otherwise intact implant surface that was analyzed by the profilometer. Additionally, the limited number of implants scanned for profilometry is a study limitation.

It is important to acknowledge the limitations of the present study. The absence of model installation in the phantom head before decontamination and the exclusion of supracontructions may impact the applicability of the findings. The incorporation of soft tissue imitation in the model, as suggested by Korello et al. (2023), was not included in the present study. Future research is warranted to incorporate these elements. Nevertheless, the easy removal of implants after decontamination enabled quantitative and qualitative analysis of the implant surface, providing a detailed assessment of the decontamination efficacy.

5 | CONCLUSION

The present in vitro study describes a promising 3D-printed model designed for evaluating devices used in the mechanical decontamination of the implant surfaces in advanced peri-implant defects. The model is anatomically realistic and can easily be modified to mimic different bone defect topographies with high accuracy and reproducibility, providing a standardized method to assess accessibility and implant surface damage. Although no decontamination group achieved complete removal of the colored occlusion spray, the results in the present study showed that the 3D model allowed postdecontamination analysis of implants at the site level. Additionally, changes in the occlusion spray can be used to detect areas with inaccessible and incomplete decontamination. Further studies should explore different peri-implant defect configurations and incorporate true, dynamic biofilm. Results from in vitro investigations using colored coatings to simulate biofilm should be interpreted cautiously as they present a treatment bias.

AUTHOR CONTRIBUTIONS

Sadia Nazir Khan, Odd Carsten Koldsland, Hanna Tiainen, and Carl Hjortsjö contributed to the conception, design, data acquisition, and interpretation and drafted and critically revised the manuscript. Sadia Nazir Khan and Hanna Tiainen performed the statistical analyses.

ACKNOWLEDGMENTS

The authors would like to thank dental technician, Mateusz Roch Chamszyk, for computer-aided design and three-dimensional printing of the resin models with peri-implant bone defects and senior engineer, Alejandro Barrantes, for optical profilometry. Implants for this study were donated by Institut Straumann AG and the chitosan brushes by Labrida AS (Labrida BioClean®; Labrida AS). The study was funded by the Institute for Clinical Dentistry, Faculty of Dentistry, University of Oslo, Norway

CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID

Sadia Nazir Khan http://orcid.org/0000-0003-3925-326X
Odd Carsten Koldsland http://orcid.org/0000-0001-6842-8255
Carl Hjortsjö http://orcid.org/0000-0003-3873-0490

REFERENCES

Atieh, M. A., Alsabeeha, N. H. M., Faggion, Jr. C. M., & Duncan, W. J. (2013). The frequency of peri-implant diseases: A systematic review and meta-analysis. *Journal of Periodontology*, *84*(11), 1586–1598.

Bermejo, P., Sánchez, M. C., Llama-Palacios, A., Figuero, E., Herrera, D., & Sanz, M. (2019). Topographic characterization of multispecies biofilms growing on dental implant surfaces: An in vitro model. *Clinical Oral Implants Research*, *30*(3), 229–241.

Cha, J. K., Paeng, K., Jung, U. W., Choi, S. H., Sanz, M., & Sanz-Martín, I. (2019). The effect of five mechanical instrumentation protocols on implant surface topography and roughness: A scanning electron microscope and confocal laser scanning microscope analysis. *Clinical Oral Implants Research*, *30*(6), 578–587.

Costa, F. O., Ferreira, S. D., Cortelli, J. R., Lima, R. P. E., Cortelli, S. C., & Cota, L. O. M. (2018). Microbiological profile associated with peri-implant diseases in individuals with and without preventive maintenance therapy: A 5-year follow-up. *Clinical Oral Investigations*, *23*, 3161–3171.

John, G., Becker, J., & Schwarz, F. (2014). Rotating titanium brush for plaque removal from rough titanium surfaces--an in vitro study. *Clinical Oral Implants Research*, *25*(7), 838–842.

Keim, D., Nickles, K., Dannewitz, B., Ratka, C., Eickholz, P., & Petsos, H. (2019). In vitro efficacy of three different implant surface decontamination methods in three different defect configurations. *Clinical Oral Implants Research*, *30*(6), 550–558.

Kordbacheh Changi, K., Finkelstein, J., & Papapanou, P. N. (2019). Peri-implantitis prevalence, incidence rate, and risk factors: A study of electronic health records at a U.S. dental school. *Clinical Oral Implants Research*, *30*(4), 306–314.

Korello, K., Eickholz, P., Zuhr, O., Ratka, C., & Petsos, H. (2023). In vitro efficacy of non-surgical and surgical implant surface decontamination methods in three different defect configurations in the presence or absence of a suprastructure. *Clinical Implant Dentistry and Related Research*, *25*(3), 549–563.

Kotsakis, G. A., & Olmedo, D. G. (2021). Peri-implantitis is not periodontitis: Scientific discoveries shed light on microbiome-biomaterial interactions that may determine disease phenotype. *Periodontology 2000*, *86*(1), 231–240.

Louropoulou, A., Slot, D. E., & Van der Weijden, F. A. (2012). Titanium surface alterations following the use of different mechanical instruments: A systematic review. *Clinical Oral Implants Research*, *23*(6), 643–658.

Louropoulou, A., Slot, D. E., & Van der Weijden, F. (2014). The effects of mechanical instruments on contaminated titanium dental implant surfaces: a systematic review. *Clinical Oral Implants Research*, *25*(10), 1149–1160.

Luengo, F., Sanz-Esporrín, J., Noguerol, F., Sanz-Martín, I., Sanz-Sánchez, I., & Sanz, M. (2022). In vitro effect of different implant decontamination methods in three intraosseous defect configurations. *Clinical Oral Implants Research*, *33*, 1087–1097.

Matsubara, V. H., Leong, B. W., Leong, M. J. L., Lawrence, Z., Becker, T., & Quaranta, A. (2020). Cleaning potential of different air abrasive powders and their impact on implant surface roughness. *Clinical Implant Dentistry and Related Research*, *22*(1), 96–104.

Mensi, M., Viviani, L., Agosti, R., Scotti, E., Garzetti, G., & Calza, S. (2020). Comparison between four different implant surface debridement methods: an in-vitro experimental study. *Minerva Stomatologica*, *69*(5), 286–294.

Monje, A., Insua, A., & Wang, H. L. (2019). Understanding peri-implantitis as a plaque-associated and site-specific entity: On the local predisposing factors. *Journal of Clinical Medicine*, *8*(2), 279.

Monje, A., Pons, R., Insua, A., Nart, J., Wang, H. L., & Schwarz, F. (2019). Morphology and severity of peri-implantitis bone defects. *Clinical Implant Dentistry and Related Research*, *21*(4), 635–643.

Nemer Vieira, L. F., Lopes de Chaves e Mello Dias, E. C., Cardoso, E. S., Machado, S. J., Pereira da Silva, C., & Vidigal, G. M. (2012). Effectiveness of implant surface decontamination using a high-pressure sodium bicarbonate protocol: an in vitro study. *Implant Dentistry*, *21*(5), 390–393.

van Noort, R. (2012). The future of dental devices is digital. *Dental Materials*, *28*(1), 3–12.

Pereira da Silva, C. H. F., Vidigal, Jr. G. M., de Uzeda, M., & de Almeida Soares, G. (2005). Influence of titanium surface

roughness on attachment of *Streptococcus sanguis*: An in vitro study. *Implant Dentistry*, *14*(1), 88–93.

Quirynen, M., & Bollen, C. M. L. (1995). The influence of surface roughness and surface-free energy on supra- and subgingival plaque formation in man. A review of the literature. *Journal of Clinical Periodontology*, *22*(1), 1–14.

Quirynen, M., & van Steenberghe, D. (1993). Bacterial colonization of the internal part of two-stage implants. An in vivo study. *Clinical Oral Implants Research*, *4*(3), 158–161.

Ramanauskaite, A., Fretwurst, T., & Schwarz, F. (2021). Efficacy of alternative or adjunctive measures to conventional non-surgical and surgical treatment of peri-implant mucositis and peri-implantitis: A systematic review and meta-analysis. *International Journal of Implant Dentistry*, *7*(1), 112.

Romandini, M., Berglundh, J., Derks, J., Sanz, M., & Berglundh, T. (2021). Diagnosis of peri-implantitis in the absence of baseline data: A diagnostic accuracy study. *Clinical Oral Implants Research*, *32*(3), 297–313.

Ronay, V., Merlini, A., Attin, T., Schmidlin, P. R., & Sahrmann, P. (2017). In vitro cleaning potential of three implant debridement methods. Simulation of the non-surgical approach. *Clinical Oral Implants Research*, *28*(2), 151–155.

Sahrmann, P., Ronay, V., Hofer, D., Attin, T., Jung, R. E., & Schmidlin, P. R. (2015). In vitro cleaning potential of three different implant debridement methods. *Clinical Oral Implants Research*, *26*(3), 314–319.

Sánchez, M. C., Llama-Palacios, A., Blanc, V., León, R., Herrera, D., & Sanz, M. (2011). Structure, viability and bacterial kinetics of an in vitro biofilm model using six bacteria from the subgingival microbiota. *Journal of Periodontal Research*, *46*(2), 252–260.

Schneider, C. A., Rasband, W. S., & Eliceiri, K. W. (2012). NIH Image to ImageJ: 25 years of image analysis. *Nature Methods*, *9*(7), 671–675.

Schwarz, F., Sahm, N., Schwarz, K., & Becker, J. (2010). Impact of defect configuration on the clinical outcome following surgical regenerative therapy of peri-implantitis. *Journal of Clinical Periodontology*, *37*(5), 449–455.

Steiger-Ronay, V., Merlini, A., Wiedemeier, D. B., Schmidlin, P. R., Attin, T., & Sahrmann, P. (2017). Location of unaccessible implant surface areas during debridement in simulated peri-implantitis therapy. *BMC Oral Health*, *17*(1), 137.

Tuna, T., Kuhlmann, L., Bishti, S., Sirazitdinova, E., Deserno, T., & Wolfart, S. (2019). Removal of simulated biofilm at different implant crown designs with interproximal oral hygiene aids: An in vitro study. *Clinical Oral Implants Research*, *30*(7), 627–636.

Verket, A., Koldsland, O. C., Bunaes, D., Lie, S. A., & Romandini, M. (2023). Non-surgical therapy of peri-implant mucositis-mechanical/physical approaches: A systematic review. *Journal of Clinical Periodontology*, *50*, 135–145.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Khan, S. N., Koldsland, O. C., Tiainen, H., & Hjortsjö, C. (2024). Anatomical three-dimensional model with peri-implant defect for in vitro assessment of dental implant decontamination. *Clinical and Experimental Dental Research*, *10*, e841. https://doi.org/10.1002/cre2.841

II

Title page

The efficacy in decontaminating dental implants of an oscillating chitosan brush compared with an ultrasonic PEEK-tip. An in vitro study using a dynamic biofilm model

Correspondence: Honorato Ribeiro-Vidal, ETEP (Etiology and Therapy of Periodontal and Peri-Implant Diseases) Research Group, Faculty of Odontology, University Complutense of Madrid, Madrid, Spain

E-mail: honorato.vidal@gmail.com

Data availability statement: The data that support the findings of this study are available on request from the corresponding author.

Funding statement: This study was funded by Institute for Clinical Dentistry, Faculty of Dentistry, University of Oslo, Norway, and ETEP (Etiology and Therapy of Periodontal and Peri-Implant Diseases) Research Group Faculty of Odontology, University Complutense, Madrid, Spain. Implants for this study were donated by Institut Straumann AG.

III

Received: 5 November 2021 | Revised: 16 August 2022 | Accepted: 6 September 2022

DOI: 10.1111/clr.14007

ORIGINAL ARTICLE

CLINICAL ORAL IMPLANTS RESEARCH WILEY

Non-surgical treatment of mild to moderate peri-implantitis using an oscillating chitosan brush or a titanium curette—A randomized multicentre controlled clinical trial

Sadia N. Khan[1] | Odd Carsten Koldsland[2] | Ann-Marie Roos-Jansåker[3,4] | Johan Caspar Wohlfahrt[2] | Anders Verket[2] | Ibrahimu Mdala[5] | Anna Magnusson[6] | Eirik Salvesen[7] | Carl Hjortsjö[1]

[1]Department of Prosthetics and Oral Function, Institute of Clinical Dentistry, Faculty of Dentistry, University of Oslo, Oslo, Norway

[2]Department of Periodontology, Institute of Clinical Dentistry, Faculty of Dentistry, University of Oslo, Oslo, Norway

[3]Department of Periodontology, Public Dental Health Services, Kristianstad, Sweden

[4]Department of Periodontology, Blekinge Hospital, Karlskrona, Sweden

[5]Department of General Practice, University of Oslo, Oslo, Norway

[6]Department of Periodontology, Public Dental Health Services, Örebro, Sweden

[7]Oris Dental, Stavanger, Norway

Correspondence
Sadia N. Khan, Specialist in Prosthodontics, Department of Prosthetics and Oral Function, Institute for Clinical Dentistry, Faculty of Dentistry, University of Oslo, Oslo, Norway.
Email: sadiak@odont.uio.no

Funding information
Norges Forskningsråd, Grant/Award Number: Project number 256756

Abstract

Objectives: This prospective, parallel-group, examiner-blinded, multicentre, randomized, controlled clinical trial aimed to assess the efficacy of an oscillating chitosan brush (OCB) versus titanium curettes (TC) on clinical parameters in the non-surgical treatment of peri-implantitis.

Material and Methods: In five dental specialist clinics, 39 patients with one implant with mild to moderate peri-implantitis, defined as 2–4 mm radiographic reduced bone level, bleeding index (BI) ≥ 2, and probing pocket depth (PPD) ≥ 4 mm were randomly allocated to test and control groups, receiving OCB or TC debridement, respectively. Treatment was performed at baseline and three months. PPD, BI, and Plaque index (PI) were measured at six sites per implant and recorded by five blinded examiners at baseline, one, three, and six month(s). Pus was recorded as present/not present. Changes in PPD and BI were compared between groups and analysed using multilevel partial ordinal and linear regression.

Results: Thirty-eight patients completed the study. Both groups showed significant reductions in PPD and BI at six months compared with baseline ($p < .05$). There was no statistically significant difference in PPD and BI changes between the groups. Eradication of peri-implant disease as defined was observed in 9.5% of cases in the OCB group and 5.9% in the TC group.

Conclusions: Within the limitations of this six-month multicentre clinical trial, non-surgical treatment of peri-implantitis with OCB and TC showed no difference between the interventions. Eradication of disease was not predictable for any of the groups.

KEYWORDS
dental implants, multilevel analysis, peri-implantitis, randomized controlled trial, single-blind method

1 | INTRODUCTION

The formation of dysbiotic biofilm on dental implants is associated with peri-implant inflammation and peri-implant bone loss (Costa et al., 2019). Peri-implant mucositis presents when mucosa surrounding dental implants shows clinical signs of bleeding on probing (BoP), erythema, swelling, and/or purulent exudate (Berglundh et al., 2018). Peri-implant mucositis often involves an increase in probing pocket depth (PPD). Inflammation in the peri-implant mucosa, together with progressive peri-implant bone loss, is termed peri-implantitis. Various case definitions have been applied in intervention studies in the absence of an unequivocal grading system (Sanz, Chapple, & Working Group 4 of the VIII European Workshop on Periodontology, 2012).

Peri-implantitis has become a concern, and data suggest the prevalence to be more than 30% of all patients treated with dental implants and exceeding 20% of all dental implants (Kordbacheh Changi et al., 2019). Even though this is a common complication, to date, there is no treatment protocol showing predictable outcomes.

Several devices for the non-surgical treatment of peri-implantitis have been presented in the literature. It has been reported that mechanical debridement is difficult to achieve in the irregularities on the implant surface and is thus ineffective in the treatment of peri-implantitis (Renvert & Polyzois, 2018).

In an animal study, the outcome of non-surgical versus surgical treatment of peri-implantitis was tested with a better outcome for new bone-to-implant contact after surgical treatment (Schwarz et al., 2006). Nevertheless, it has been suggested that non-surgical intervention should be considered prior to surgical treatment, as the results may be beneficial and make surgery more efficient due to reduced inflammation at the surgical site (Schwarz et al., 2015). Improvement of clinical inflammatory parameters has been demonstrated after non-surgical treatment of peri-implantitis (Schwarz et al., 2015), but an arrest of peri-implant bone loss over time is dubious (Berglundh et al., 2018). Recent data suggest that implants treated by surgical means are at risk of recurrent peri-implantitis (Carcuac et al., 2020). However, complete disease resolution is rare in severe cases (Renvert et al., 2019; Roccuzzo et al., 2020). Regardless of treatment strategy, debridement devices that do not adversely affect the implant surface are preferred (Cha et al., 2019). Moreover, it is important that the instrument used for implant debridement does not leave non-biocompatible remnants that aggravate bone destruction through foreign-body reactions (van Velzen et al., 2016).

An oscillating chitosan brush (OCB) aimed at the removal of biofilm in less accessible surfaces around dental implants and teeth is commercially available (Labrida BioClean®, Labrida AS, Oslo, Norway). Chitosan is a natural polysaccharide derived from chitin. It is biocompatible, biodegradable, and has antibacterial activity (Muxika et al., 2017). The effectiveness of this novel instrument for debridement of implant surfaces in the treatment of peri-implantitis has been demonstrated (Larsen et al., 2017; Wohlfahrt et al., 2017, 2019). In a case study with a six-month follow-up, patients treated with an OCB showed a significant reduction in inflammation parameters (Wohlfahrt et al., 2017). The efficacy of the OCB in supportive treatment following peri-implantitis surgery was questioned in a recent randomized clinical trial (RCT) (Koldsland & Aass, 2020). RCTs that assess the efficacy of OCBs in the non-surgical treatment of peri-implantitis are still lacking. Non-surgical treatment of mucositis and peri-implantitis using titanium curettes (TC) with and without adjunctive antibiotics has been evaluated in several RCTs (Hallstrom et al., 2012; Renvert et al., 2009; Wohlfahrt et al., 2017). Partial reduction in peri-implant inflammation was observed for the treatment modalities at six months (Hallstrom et al., 2012; Renvert et al., 2009). Non-surgical treatment with TC combined with photodynamic therapy or minocycline microspheres showed a statistically significant ($p < .05$) reduction in peri-implant inflammation three months after initial treatment (Schär et al., 2013). There is no evidence that one non-surgical approach is more effective than the other when treating peri-implant disease (Roccuzzo et al., 2020; Wang et al., 2019).

This multicentre randomized controlled clinical trial aimed to evaluate the following outcomes: PPD, bleeding index (BI), presence of pus, radiographic bone level following non-surgical mechanical treatment of peri-implantitis with an OCB (test) and TC (control) at one, three, and six month(s) after initial therapy. In addition, patient-reported pain during intervention was evaluated at three months.

2 | MATERIALS AND METHODS

2.1 | Study design

A two-arm parallel-group, multicentre, randomized, examiner-blinded, controlled study was designed to test the efficacy of two treatment modalities in the management of mild to moderate peri-implantitis, defined as PPD ≥ 4 mm, BI score of at least 2, and a peri-implant radiographic bone level of 2–4 mm measured from the most coronal intraosseous part of the implant (Sanz et al., 2012). The patients were randomly allocated to either the test group and treated with an OCB or the control group treated with TC. One single implant was treated in each patient. When several implants with treatment needs in the same patients were observed, one implant was randomly selected by drawing lots to assure unbiased inclusion.

The null hypothesis was that there would be no difference in the reduction of evaluated parameters between the two groups.

The study was performed in compliance with Good Clinical Practice, and the Declaration of Helsinki (Fortaleza, Brazil, 2013). The study was registered at ClinicalTrials.gov (12/08/2017, NCT03373448) and approved by the Regional Committee for Medical and Health Research Ethics, South-East Norway (REK sør-øst 2017/710) and by Swedish Ethical Review Authority, Linköping (EPN 2017/36-31).

2.2 | Sample size assessment and power

The study was designed to have 80% power to detect a PPD change of 1 mm between the test and the control group. With a

standard deviation of 0.8 mm (Aljateeli et al., 2014) and an alpha level of 0.05, the appropriate number of patients per group was calculated to be 17.

2.3 | Study population

Consecutive patients presenting for supportive dental care or patients referred for treatment of peri-implantitis between April 2018 and October 2019 were invited to participate in this study. In total, 45 patients were assessed for study eligibility in five dental specialist practices in Norway and Sweden.

Absence of visual plaque around the included implant at baseline was a prerequisite for inclusion. All included patients underwent oral hygiene instructions until this criterion was met. Periodontal and/or endodontic diseases were treated prior to the start of the study.

Inclusion criteria encompassed: (1) Peri-implantitis as defined above on an implant in function for more than 12 months; (2) age above 18 years; (3) eligible for treatment in an outpatient dental clinic (ASA I and II); (4) full-mouth plaque scores ≤20% prior to final inclusion; (5) signed informed consent; and (6) consent to complete all follow-up visits.

Exclusion criteria included: patients/implants registered with (1) peri-implant bone loss >4 mm; (2) supraconstructions that for technical reasons made it impossible to access the implant for clinical measurements; (3) technical complications which, according to the examiners' judgment, had contributed to the disease state and were not possible to resolve prior to final inclusion; (4) mobile implant; (5) diagnosed active periodontal disease; (6) implants previously treated for peri-implantitis with grafting materials; (7) receiving medications known to induce mucosal hyperplasia; (8) receiving systemic antibiotics < three months prior to inclusion; (9) acute or chronic medical conditions that constituted an unwarranted risk and that would limit the patients' ability to participate in the study; (10) unwillingness to undergo treatment; (11) advanced, untreated, and uncontrolled peri-implantitis on neighbouring implants; (12) patients presented with poorly designed prosthetic constructions resulting in non-balanced traumatic occlusion; (13) ongoing or previous radiotherapy to the head–neck region; (14) ongoing chemotherapy; and (15) ongoing corticosteroid treatment.

2.4 | Randomization and allocation concealment

All patients were appointed a patient number and randomly assigned to treatment in blocks of 10. Computer-generated block randomization was performed by the study administrator (RANDOM.ORG, Randomness and Integrity Services Ltd., Dublin, Ireland). Clinicians who performed treatment were provided with lists consisting of patient numbers and treatment assignments. The examiners were blinded to the treatment allocation.

2.5 | Clinical and radiographic outcomes

The patients were clinically examined by five specialists in periodontology. Calibration meeting was held to discuss the study protocol.

Examinations were performed at baseline prior to treatment and at one-, three-, and six- month(s) post treatment. The evaluated parameters for the included implants were PPD, BI, plaque index (PI), pus, and height of keratinized mucosa (KM). PPD, BI, and PI registrations were performed at six sites per implant (mesiobuccal, buccal, distobuccal, distopalatal, palatal, and mesiopalatal).

Plaque was assessed by running a probe along the implant neck. Plaque was scored using the PI and dichotomized as present or not present (O'Leary et al., 1972). BI was used to classify the degree of inflammation as 0–3, according to Roos-Jansaker et al. (2007). Degrees were noted as 0 = no bleeding, 1 = isolated minimal bleeding spots, 2 = blood forming a confluent red line on the margin, and 3 = heavy or profound bleeding. PPD was measured in millimetres. Pus was registered as present or not present. Keratinized mucosa was assessed midbuccaly with a periodontal probe.

Clinical examinations were performed using a manual 0.20 N defined force periodontal probe (University of North Carolina, DB764R, AESCULAR B Braun, Germany). The implant-retained fixed dental prosthesis was in place during the entire study period.

Peri-apical radiographs were obtained using the long-cone paralleling technique with digital X-rays. ImageJ®, image processing and analysis software program was used to measure peri-implant radiographic bone level (RBL) at baseline (Preus et al., 2015). The RBL was analysed by one blinded examiner as the distance from the implant neck to bone-to-implant contact. The size of intraoral phosphor plates and sensors were used to calibrate the radiographs. Baseline and 6-month RBL were measured three times. The intra-examiner agreement test resulted in an intraclass correlation coefficient (ICC) of 0.98.

Digital intraoral radiographs were taken at six months to observe potential adverse effects or further bone loss.

PPD, BI, PI, and pus were registered at baseline and one, three and six month(s) following baseline. PPD and BI were registered for the same sites throughout the study. For PPD, the mean of the sites ≥4 mm was calculated for each time point for each implant.

PPD change was used as primary outcome variable. The secondary outcome variables were BI/pus, change in radiographic bone level, and patient-reported pain during intervention.

A composite outcome of disease eradication was based on frequency analysis of implants with absence of peri-implant sites with pocket depth PPD ≥ 4 mm, no bleeding (BI 0)/suppuration, and no radiographic bone loss between baseline and 6 months. Inflammation control was defined as BI = 0 at any implant site.

Following treatment at three months, patients were asked to record pain associated with the treatment via a visual analogue scale (VAS). No pain was categorized as 0 and worst possible pain as 10.

2.6 | Treatment procedures

The implants in the test group were debrided with an OCB. The brush was soaked in sterile saline for two minutes before it was seated on an oscillating dental handpiece (NSK ER10, TEQ-Y, Nakanishi International Inc., Tochigi, Japan) as recommended in the instructions for use by the manufacturer. The implants in the control group were debrided utilizing TC (Langer and Langer, Rønvig, Denmark). Infiltration anaesthesia was used if requested by the patient. The included implants were treated for two minutes and irrigated with sterile saline after mechanical debridement for both the test and control groups. Treatment was performed at baseline and repeated at three months after initial therapy, at implants with PPD ≥ 4 mm and BI > 0 by five registered dental hygienists. In cases with multiple implants with treatment need, all implants were treated in the same session with the same assigned treatment.

2.7 | Data management and statistical analysis

All clinical recordings and patient data were registered in a web-based clinical report form (Nettskjema Version 2.0, University of Oslo, Norway). Per-protocol (PP) analysis was performed on patients who were assessed at all time-points. Calculations and analyses were performed using Stata Statistical Software, Version 16.1 (StataCorp.2001. Statistical Software: Release 7.0. College Station, TX: Stata Corporation). The significance level was set at $\alpha = 0.05$.

Characteristics of the patients and the implants in the test and control groups were described using frequencies and percentages for categorical variables and means with standard deviations (*SDs*) for continuous data.

Data on PPD and BI were obtained at baseline and at one-, three-, and six month(s) for one implant in each individual. The included patients attended five different clinics. Therefore, to account for possible

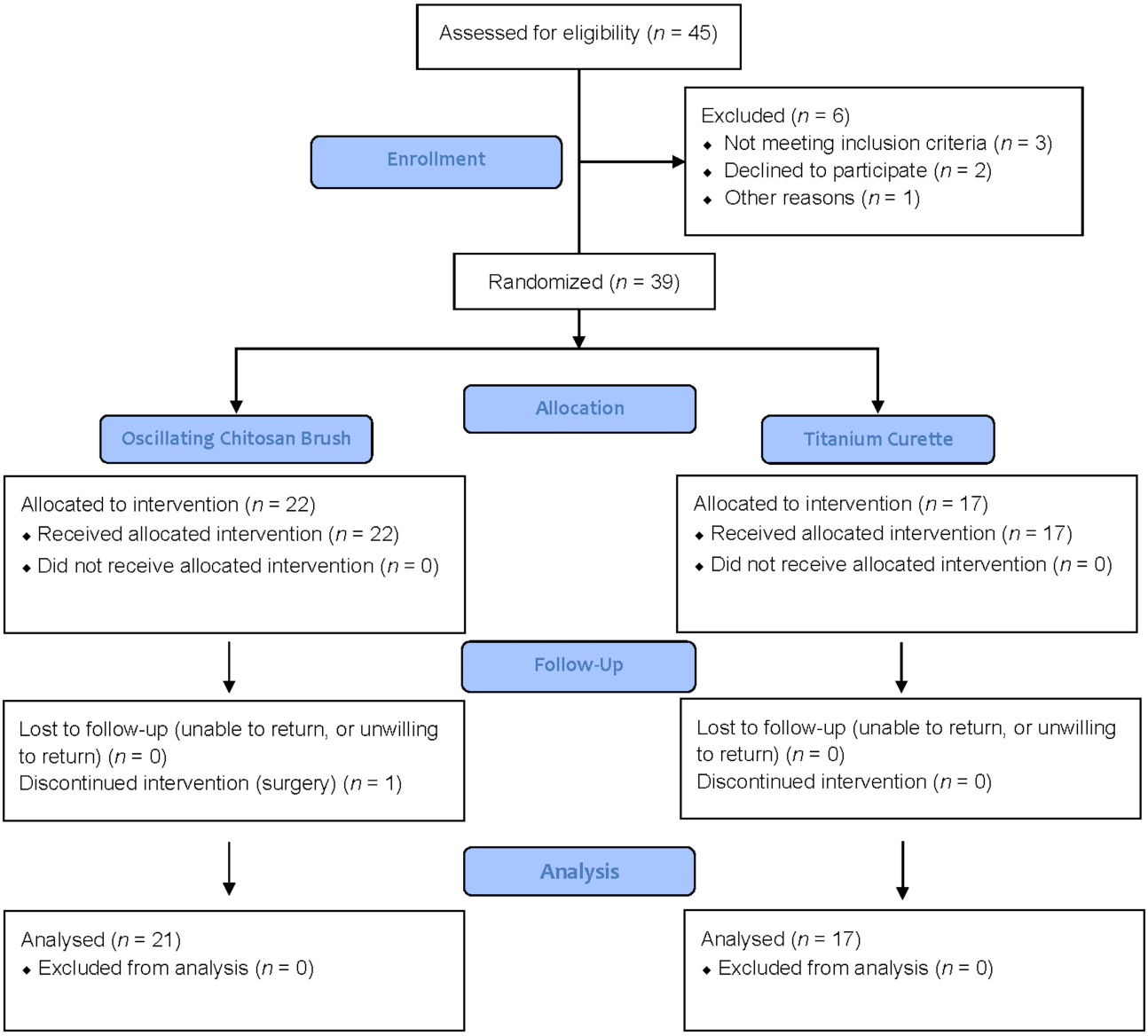


FIGURE 1 A CONSORT flowchart of enrolment, allocation, follow-up, and analysis

dependences of the data within participants who were nested within clinics, three-level linear and partial ordinal multilevel models with random intercept and random effect of time (level 3) on patients (level 2) and on clinics at level 1 for PPD and BI were performed, respectively. The assumption of proportionality between categories of the ordered variable was violated for some independent variables. A multilevel partial ordinal logistic model using gologit2 was fitted to the BI data. The differences in PPD and BI between the groups at each study time point were obtained from the two-way interaction of time with the groups. Estimates of ICC, which described the amount of variability in both PPD and BI that could be attributed to differences between patients and clinicians, were obtained.

Consolidation Standards of Reporting Trials (CONSORT) Statement guidelines were followed (Schulz et al., 2010).

3 | RESULTS

After verification of inclusion and exclusion criteria, 39 patients gave informed consent and were enroled in the present study. The flow diagram of the study is presented in Figure 1. No adverse events were reported. Complete observations were available for 38 patients. There were no statistically significant baseline differences in patient and implant characteristics between the groups (Table 1). No group differences in PI were detected throughout the study period.

3.1 | Changes in PPD between groups

Table 2 show the changes in PPD between the groups at each time point. Figure 2 shows the changes in PPD from baseline for the groups at each study time point. There were no statistically significant differences in PPD changes between the groups at baseline or after one, three, and six month(s).

3.2 | Changes in BI and presence of pus between groups

There were no statistically significant differences in BI between the groups at any time point (Table 2). At six months, pus was registered at 33.3% and 64.7% of the implants in the test and control group, respectively ($p > .05$). Figure 3 shows the probability of BI 0–3 between and within both groups.

3.3 | Radiographic bone level

At six months, the mean radiographic bone level was 2.5 mm (±0.5) for the test group and 2.6 mm (±0.7) for the control group. The between group RBL change from baseline to six months was not statistically significant.

3.4 | Composite outcome

At six months, two implants in the test group (9.5%) and one implant in the control group (5.9%) presented disease eradication with PPD < 4 mm, no bleeding (BI 0), and no change in radiographic bone level compared with baseline. The difference between the groups was not statistically significant.

3.5 | Keratinized mucosa

The mean KM at baseline is presented in Table 1. At baseline, 36.8% ($n = 7$) implants in the test group and 54.5% ($n = 6$) implants in the control group had KM ≤ 2 mm.

3.6 | Withdrawal

One implant in the OCB-group showed progression of peri-implant disease at the 3 months screening and was excluded from the study for surgical intervention.

3.7 | Intraclass correlation coefficient

Estimates of ICC for both the variance component and adjusted models are presented in Table S1. For PPD, 7.7% of the variability was explained by the differences between the clinics, while 51.2% of the variability was nested in patients. For BI, 10.3% of the variability was explained by the differences between the clinics, while patients nested in clinics explained 22.2% of the variability.

3.8 | Visual analogue scale for pain

VAS information for pain during treatment was recorded for 22 patients (OCB (n) = 14, TC (n) = 8), leading to a response rate of 57.9%. Seven patients received anaesthesia before treatment and were therefore not asked to fill out the VAS form (OCB (n) = 4, TC (n) = 3). No data were reported for nine patients because screeners did not collect VAS forms (OCB (n) = 4, TC (n) = 6). The mean VAS scores (±SD) for the test and control groups were 2.9 (±1.93) and 3.4 (±2.09), respectively. There was no statistically significant difference in VAS between the groups ($p > .05$).

4 | DISCUSSION

In the present prospective intervention study comparing non-surgical treatment with OCB and TC showed no significant difference between the treatment groups. However, both interventions significantly reduced the investigated inflammation parameters at implants affected by peri-implantitis. This might be explained by

TABLE 1 Baseline characteristics of the study patients and implants

Variable	Total	(%)	Test group	(%)	Control group	(%)
Patients/Implants (*n*)	38	100	21	55.3	17	44.7
Mean age (±*SD*)	61.99		62.86 (±12.19)		61.12 (±3.67)	
Gender						
Male	14	36.8	5	23.8	9	52.9
Female	24	63.2	16	76.2	8	47.1
Daily smoker	5	13.2	4	19.0	1	5.9
Diabetes	7	18.4	4	19.0	3	17.6
Tooth loss due to periodontitis	11	28.2	6	27.3	5	29.4
Front	17	44.7	8	38.1	9	52.9
Premolar	18	47.4	11	52.4	7	41.2
Molar	3	7.9	2	9.5	1	5.9
Non-modified implant surface	5	13.2	3	14.3	2	11.8
Screw-retained	31	81.6	16	76.2	15	88.2
Cement-retained	6	15.9	5	23.8	1	5.9
Not reported	1	2.6	0	0	1	5.9
Implant-retained crown	15	39.5	10	66.7	5	33.3
Implant-retained fixed dental prosthesis	23	60.5	11	47.8	12	52.2
Keratinized mucosa (mm)	30	78.9	2.8	90	2.5	64.7
Implants with suppuration	22	57.9	11	52.4	11	64.7
Radiographic bone level (±*SD*)	38	100	2.43 (±0.51)	55.3	2.58 (±0.58)	44.7
BI ≥ 2 (±*SD*)	38	100	2.33 (±0.48)	55.3	2.24 (±0.44)	44.7
PPD mean ≥ 4 mm (±*SD*)	38	100	5.3 (±0.16)	55.3	5.5 (±0.29)	44.7
PPD ≥ 4 mm (±*SD*)	38	100	6.8 (±1.6)	55.3	6.5 (±1.7)	44.7
PPD ≥ 6 mm (±*SD*)	29	76.3	7.6 (±1.1)	39.5	7.0 (±1.5)	36.8

Abbreviations: BI, bleeding index; PPD, probing pocket depth.

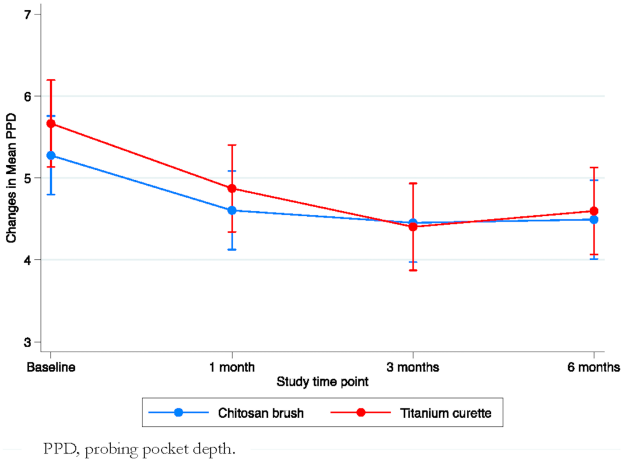
the sample size. A larger sample might have resulted in a different outcome. The included patients underwent an initial hygiene phase and two active treatments: at baseline and three months. PPD and the BI score significantly reduced between baseline and six months in both groups. In a pilot RCT assessing treatment of peri-implant mucositis with an OCB and TC, a similar decrease in inflammation parameters was reported (Wohlfahrt et al., 2019). In contrast to the study by Wohlfahrt et al. (2019) and the present study, Koldsland and Aass (2020) showed that supportive treatment with OCB or TC failed to further reduce PPD and BoP among patients in maintenance following recent surgical treatment of peri-implantitis. Both studies were RCTs comparing treatment with an OCB and TC, but the included patients had different disease entities and treatment strategies. A possible explanation for the different outcomes in the study performed by Wohlfahrt et al. (2019), Koldsland and Aass (2020), and the present study may be the treatment of mucositis, post-surgical maintenance, and treatment of peri-implantitis, respectively. Moreover, supportive treatment after peri-implantitis surgery involves debridement of a complex intraorally exposed implant surface. The moderately rough implant surface may complicate home care maintenance and professional removal of biofilm, which was indicated by increasing plaque scores throughout the study period compared with baseline (Koldsland & Aass, 2020).

The results of the present study agree with the findings from a RCT where non-surgical, mechanical treatment with TC and an ultrasonic device was compared (Renvert et al., 2009). The studies are also comparable in terms of the number of included patients, follow-up period, and a hand instrument that was compared with a machine-driven device. In contrast to the present study, Renvert et al. (2009) included patients with high plaque scores at baseline. The plaque scores diminished significantly at the end of the study (from 73% to 53%). Significantly reduced BoP score within the groups and no difference between the groups was reported from

TABLE 2 Changes in mean PPD and BI between the groups at each time point obtained from partial ordinal and linear multilevel regression model with clinic and patient random effects

	Baseline	1 month		3 months		6 months	
	β (95% CI)	*p*-value	*β* (95% CI)	*p*-value	*β* (95% CI)	*p*-value	*β* (95% CI)	*p*-value
Group (ref: TC)								
PPD Test group	0.4 (−1.01, 0.2)	.22	−0.3 (−0.9, 0.4)	.40	−0.1 (−0.6, 0.7)	.88	−0.1 (−0.7, 0.5)	.74
	OR (95% CI)	***p*-value**	**OR (95% CI)**	***p*-value**	**OR (95% CI)**	***p*-value**	**OR (95% CI)**	***p*-value**
Between the groups (ref: Control)								
Profuse versus no bleeding (BI 3 vs BI 0)	0.8 (0.2, 3.6)	.77	4.3 (0.5, 35.0)	.17	0.4 (0.1, 1.6)	.21	0.8 (0.1, 12.9)	.89
Line versus no bleeding (BI 2 vs BI 0)	2.5 (0.6, 10.4)	.20	1.5 (0.6, 4.2)	.43	1.4 (0.5, 4.1)	.49	0.8 (0.3, 2.4)	.72
Spot versus no bleeding (BI 1 vs BI 0)	0.7 (0.3, 2.1)	.53	1.3 (0.5, 3.3)	.62	0.3 (0.1, 1.0)	.06	1.2 (0.5, 3.4)	.68

Abbreviations: BI, bleeding index; CI, confidence interval; PPD, probing pocket depth; TC, titanium curettes.



PPD, probing pocket depth.

FIGURE 2 Changes in mean PPD between and within the groups

baseline to six months, as in the present study. A longer follow-up period and frequently repeated treatment may be required to demonstrate significant differences between non-surgical mechanical treatment modalities (Bertoldi et al., 2017). Mechanical debridement alone may be inadequate in the absence of an ideal treatment frequency (Karring et al., 2005).

To date, some RCTs have compared different non-surgical modalities for the treatment of peri-implantitis (Faggion et al., 2014). The studies vary with regard to disease severity among the included patients and what is considered a successful endpoint (Faggion et al., 2014). In the present multicentre study, radiographs and PPD registration from the time of first prosthetic loading were not mandatory to be included. The reduced bone level was measured on digital radiographs taken at the time of study recruitment. An assumed initial bone level was used as a reference for the bone loss measurements. In a consensus report, a minimum of 2 mm of assumed bone loss in addition to inflammation was suggested as criteria for peri-implantitis for clinical studies when the baseline bone level is not known (Sanz et al., 2012). Thus, cases with 2–4 mm assumed bone level reduction were included in the current trial. Renvert et al. included implants with 1.5 mm mean bone loss (Renvert et al., 2009). Furthermore, the peri-implant tissues of the selected implants in the present study demonstrated. In a recent consensus report, RBLs ≥ 3 mm apical to the most intraosseous part of the implant, along with BoP and PPD ≥ 6 mm, were suggested as case definitions of peri-implantitis when data from the time of prosthetic loading are unavailable (Berglundh et al., 2018). In the present study, PPD ≥ 4 mm at the time of study enrolment was decided as an inclusion criterion and later used as a reference for changes in PPD throughout the study. Nevertheless, 76.3% of all implants in the present study had PPD ≥ 6 mm and could be classified as peri-implantitis according to Berglundh et al. (2018).

In the present study, patients were recruited from specialist clinics in Norway and Sweden. The multicentre study design is considered to have several benefits: the participation of a compound group of investigators, the participants being included and treated by different centres may improve the validity of the results, and the main advantage is the recruitment of patients from a wider population. To achieve the benefits of a multicentre study, it is an absolute prerequisite that the data are collected in the same reliable way throughout the project period. Calibration of operators can be challenging because of the number of operators and geographical distance. In the present study, calibration meeting was held to assure study quality. However, practical calibration training was not performed, and ICC was not calculated.

BI, pus, and PPD are surrogate parameters for inflammation, and reduction in these parameters may indicate a positive but potentially transient outcome of an intervention and may not be an indication of permanent resolution of peri-implantitis (Faggion et al., 2014). This emphasizes the importance of regular evaluation of treatment and preventive maintenance therapy to control

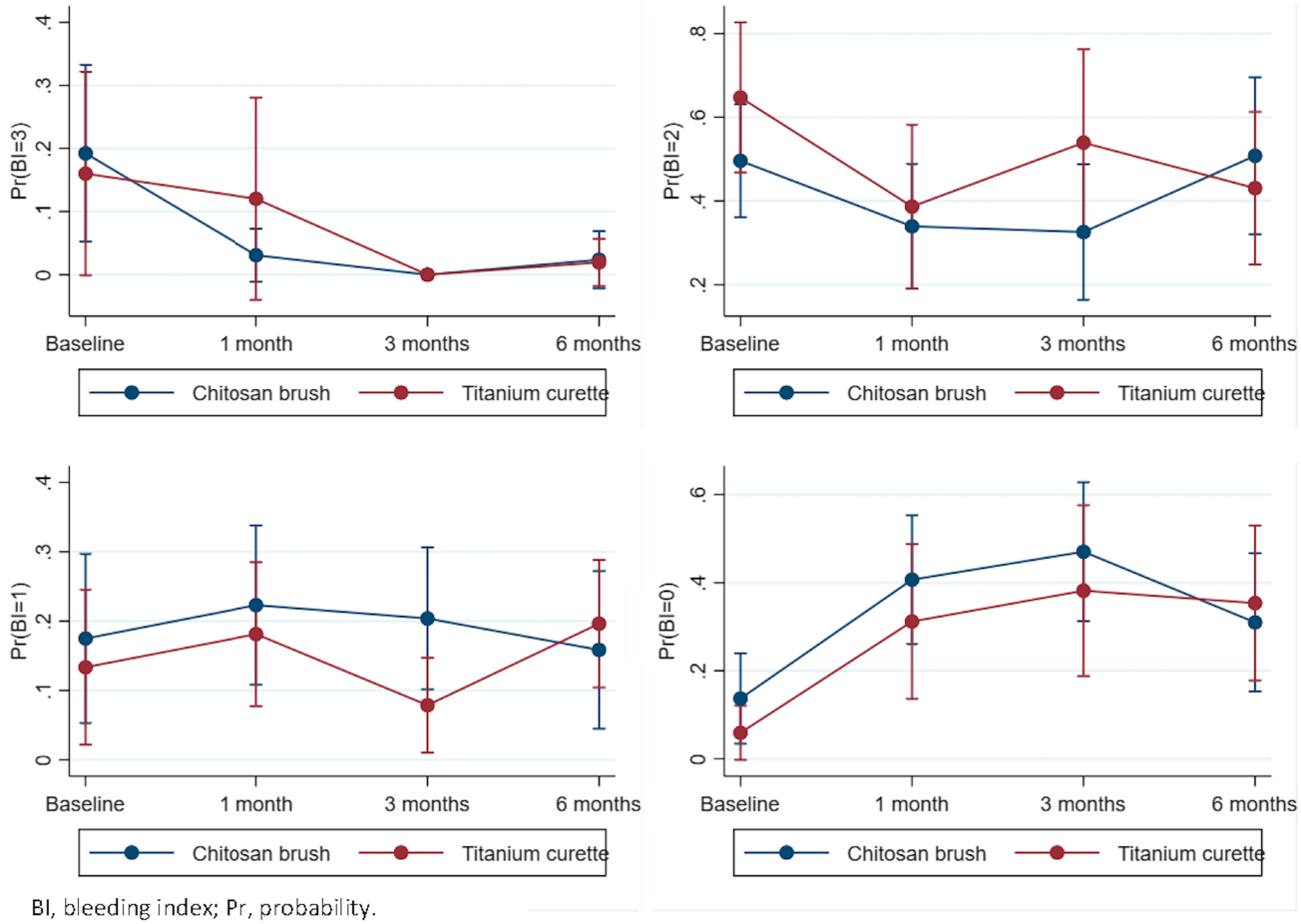


FIGURE 3 The probability of BI 0–3 between and within the groups at each time point

peri-implant disease. Further investigations may focus on how long it is possible to keep the implant free of inflammation.

The efficacy of non-surgical treatment with TCs, air-polishing with glycine-based powder, ultrasonic curettes, or photodynamic therapy has been assessed and reported in a range of studies, but it remains inconclusive as to what treatment the exhibits superior outcome of peri-implant inflammation (2019). Several of these studies emphasize the importance of non-surgical intervention as the preferred treatment (Faggion et al., 2014). Although no golden standard for treatment of peri-implantitis has been defined (Graziani et al., 2012), TC was chosen as the control treatment in the present study based on what seems to be a common method among many implant clinics and in the literature. For non-surgical treatment, data suggest that greater PPD reduction can be achieved when a combination of therapies is applied (Faggion et al., 2014). In the present study, mechanical debridement alone was performed.

In an in vitro study comparing instrumentation with an OCB, TC, and Er:YAG laser, shallow alterations of the implant surface for the TC group were observed when debridement was performed for three minutes (Larsen et al., 2017). In vivo, the release of titanium particles from the damaged/scratched implant surface to the peri-implant tissues may trigger the immune system by a foreign-body reaction and result in osteolysis of the peri-implant bone (Purdue et al., 2007). Furthermore, an altered implant surface may affect the recolonization of microbial biofilm and the proliferation of soft and hard tissue cells (Cao et al., 2018).

In the present study, the control group had a higher incidence of implants with KM ≤ 2 mm than the test group. Although more brushing discomfort and plaque accumulation is reported for implants with KM ≤ 2 mm (Souza et al., 2016), lower height of KM is not clearly associated with higher risk of peri-implantitis (Schwarz et al., 2018). And thus, it might be speculated that insufficient KM might affect the outcome of treatment. In the present study, the PI remained equally low in both groups.

Peri-implant health is characterized by the absence of all inflammation signs (Berglundh et al., 2018). In the present study, pus was registered at 52.4% and 64.7% of the implants at baseline in the test and control groups, respectively. At six months, the pus score decreased to 33.3% for the test group. No change in the presence of pus was registered from baseline to six months for the control group. In a recent RCT, pus scores varied from 19% to 37% at baseline and decreased to 0% to 15% at six months (Merli et al., 2020). The study patients were allocated to four intervention groups and

treated either non-surgically alone or in combination with chemical agents (Merli et al., 2020). No pus was registered at six months when implants were treated with two chemical agents in addition to mechanical debridement (Merli et al., 2020). Contrary to our study, the implant-retained restorations were removed before the mechanical debridement (Merli et al., 2020). In addition, the study patients were instructed to rinse with a 0.12% chlorhexidine solution twice per day for the first 15 days (2020).

In the present study, no statistically significant difference between treatments with an OCB or TC was detected. Positive changes in PPD and BI were registered, but this may be a short, transient stage. BI results should be interpreted with caution due to the large confidence intervals. A decrease in the presence of pus was observed in the test group. The presence of pus in both groups indicates active disease and the need for further intervention. Studies with longer follow-up with an assessment of radiographic bone loss and a larger sample size may be interesting to pursue.

The current study, while limited in size has sought to assess the efficacy of an oscillating chitosan brush (OCB) versus titanium curettes (TC). A major limitation in estimating the sample size of the current clustered study was the unavailability from literature of statistical measures such as the intra-cluster correlation (ICC), hence we relied on a reasonable educated guess. However, the richness of the data generated in this study can be used in formulating hypotheses of much bigger studies.

Within the limitations of this six-month multicentre clinical trial, it can be concluded that non-surgical treatment of peri-implantitis with OCB or TC demonstrated equal efficacy and no difference between the interventions. Eradication of disease was not predictable for any of the groups.

AUTHOR CONTRIBUTIONS

Sadia N. Khan: Conceptualization (lead); data curation (lead); formal analysis (equal); investigation (supporting); project administration (lead); software (lead); supervision (equal); writing – original draft (lead); writing – review and editing (lead). **Odd Carsten Koldsland:** Conceptualization (lead); methodology (lead); supervision (lead); writing – review and editing (lead). **Ann-Marie Roos-Jansåker:** Conceptualization (equal); investigation (lead); methodology (equal); validation (equal); writing – review and editing (equal). **Johan Caspar Wohlfahrt:** Conceptualization (lead); methodology (lead); writing – review and editing (equal). **Anders Verket:** Investigation (lead); writing – review and editing (equal). **Carl Hjortsjö:** Methodology (lead); project administration (lead); supervision (lead); validation (lead); writing – review and editing (lead).

ACKNOWLEDGMENTS

We acknowledge and thank the dental hygienists Åsa Thelin, Dagrun Ernø, Ane-Mari Igland Naustdal, Monica Palmér Tvedt, Sofia Hald, Menaka Seevaratnam, and Helena Olsson for their participation in the study, head engineer Gerald Torgersen for blinding, calibrating the digital radiographs, and Anne Christophersen for administration and moderating the study.

FUNDING INFORMATION

The study was funded by the Research Council of Norway and university grants.

CONFLICT OF INTEREST

Caspar Wohlfahrt is the inventor and patentholder of the chitosan brush (Labrida Bioclean®, Labrida AS Oslo, Norway). Ann Marie Roos-Jansåker and Caspar Wohlfahrt are shareholders in Labrida AS. Drs Khan, Koldsland, Verket, Mdala, Magnusson, Salvesen and Hjortsjö report no conflicts of interest related to this study.

DATA AVAILABILITY STATEMENT

Data are stored at a central university facility and are available upon request from the corresponding author, S.K.

ETHICAL APPROVAL

The study was approved by the Regional Committee for Medical and Health Research Ethics, South-East Norway (REK sør-øst 2017/710) and by Swedish Ethical Review Authority, Linköping (EPN 2017/36-31).

PATIENT CONSENT STATEMENT

Informed consent was signed by the study patients prior to study start.

PERMISSION TO REPRODUCE MATERIAL FROM OTHER SOURCES

Not applicable.

CLINICAL TRIALS REGISTRATION

The study was registered at ClinicalTrials.gov (12/08/2017, NCT03373448).

ORCID

Sadia N. Khan https://orcid.org/0000-0003-3925-326X

REFERENCES

Aljateeli, M., Koticha, T., Bashutski, J., Sugai, J. V., Braun, T. M., Giannobile, W. V., & Wang, H. L. (2014). Surgical periodontal therapy with and without initial scaling and root planing in the management of chronic periodontitis: A randomized clinical trial. *Journal of Clinical Periodontology*, *41*(7), 693–700. https://doi.org/10.1111/jcpe.12259

Berglundh, T., Armitage, G., Araujo, M. G., Avila-Ortiz, G., Blanco, J., Camargo, P. M., Chen, S., Cochran, D., Derks, J., Figuero, E., Hämmerle, C. H. F., Heitz-Mayfield, L. J. A., Huynh-Ba, G., Iacono, V., Koo, K. T., Lambert, F., McCauley, L., Quirynen, M., Renvert, S., … Zitzmann, N. (2018). Peri-implant diseases and conditions: Consensus report of workgroup 4 of the 2017 world workshop on the classification of periodontal and peri-implant diseases and conditions. *Journal of Periodontology*, *89 Suppl 1*(Suppl 1), S313–S318. https://doi.org/10.1002/JPER.17-0739

Bertoldi, C., Lusuardi, D., Battarra, F., Sassatelli, P., Spinato, S., & Zaffe, D. (2017). The maintenance of inserted titanium implants: In-vitro evaluation of exposed surfaces cleaned with three different instruments. *Clinical Oral Implants Research*, *28*(1), 57–63. https://doi.org/10.1111/clr.12759

Cao, J., Wang, T., Pu, Y., Tang, Z., & Meng, H. (2018). Influence on proliferation and adhesion of human gingival fibroblasts from different titanium surface decontamination treatments: An in vitro study. *Archives of Oral Biology*, *87*, 204–210. https://doi.org/10.1016/j.archoralbio.2017.12.013

Carcuac, O., Derks, J., Abrahamsson, I., Wennstrom, J. L., & Berglundh, T. (2020). Risk for recurrence of disease following surgical therapy of peri-implantitis—A prospective longitudinal study. *Clinical Oral Implants Research*, *31*(11), 1072–1077. https://doi.org/10.1111/clr.13653

Cha, J. K., Paeng, K., Jung, U. W., Choi, S. H., Sanz, M., & Sanz-Martin, I. (2019). The effect of five mechanical instrumentation protocols on implant surface topography and roughness: A scanning electron microscope and confocal laser scanning microscope analysis. *Clinical Oral Implants Research*, *30*(6), 578–587. https://doi.org/10.1111/clr.13446

Costa, F. O., Ferreira, S. D., Cortelli, J. R., Lima, R. P. E., Cortelli, S. C., & Cota, L. O. M. (2019). Microbiological profile associated with peri-implant diseases in individuals with and without preventive maintenance therapy: A 5-year follow-up. *Clinical Oral Investigations*, *23*(8), 3161–3171. https://doi.org/10.1007/s00784-018-2737-y

Faggion, C. M., Jr., Listl, S., Fruhauf, N., Chang, H. J., & Tu, Y. K. (2014). A systematic review and Bayesian network meta-analysis of randomized clinical trials on non-surgical treatments for peri-implantitis. *Journal of Clinical Periodontology*, *41*(10), 1015–1025. https://doi.org/10.1111/jcpe.12292

Graziani, F., Figuero, E., & Herrera, D. (2012). Systematic review of quality of reporting, outcome measurements and methods to study efficacy of preventive and therapeutic approaches to peri-implant diseases. *Journal of Clinical Periodontology*, *39*(Suppl 12), 224–244. https://doi.org/10.1111/j.1600-051X.2011.01832.x

Hallstrom, H., Persson, G. R., Lindgren, S., Olofsson, M., & Renvert, S. (2012). Systemic antibiotics and debridement of peri-implant mucositis. A randomized clinical trial. *Journal of Clinical Periodontology*, *39*(6), 574–581. https://doi.org/10.1111/j.1600-051X.2012.01884.x

Karring, E. S., Stavropoulos, A., Ellegaard, B., & Karring, T. (2005). Treatment of peri-implantitis by the vector system. *Clinical Oral Implants Research*, *16*(3), 288–293. https://doi.org/10.1111/j.1600-0501.2005.01141.x

Koldsland, O. C., & Aass, A. M. (2020). Supportive treatment following peri-implantitis surgery: An RCT using titanium curettes or chitosan brushes. *Journal of Clinical Periodontology*, *47*(10), 1259–1267. https://doi.org/10.1111/jcpe.13357

Kordbacheh Changi, K., Finkelstein, J., & Papapanou, P. N. (2019). Peri-implantitis prevalence, incidence rate, and risk factors: A study of electronic health records at a U.S. dental school. *Clinical Oral Implants Research*, *30*(4), 306–314. https://doi.org/10.1111/clr.13416

Larsen, O. I., Enersen, M., Kristoffersen, A. K., Wennerberg, A., Bunaes, D. F., Lie, S. A., & Leknes, K. N. (2017). Antimicrobial effects of three different treatment modalities on dental implant surfaces. *The Journal of Oral Implantology*, *43*(6), 429–436. https://doi.org/10.1563/aaid-joi-D-16-00147

Merli, M., Bernardelli, F., Giulianelli, E., Carinci, F., Mariotti, G., Merli, M., Pini-Prato, G., & Nieri, M. (2020). Short-term comparison of two non-surgical treatment modalities of peri-implantitis: Clinical and microbiological outcomes in a two-factorial randomized controlled trial. *Journal of Clinical Periodontology*, *47*(10), 1268–1280. https://doi.org/10.1111/jcpe.13345

Muxika, A., Etxabide, A., Uranga, J., Guerrero, P., & de la Caba, K. (2017). Chitosan as a bioactive polymer: Processing, properties and applications. *International Journal of Biological Macromolecules*, *105*(Pt 2), 1358–1368. https://doi.org/10.1016/j.ijbiomac.2017.07.087

O'Leary, T. J., Drake, R. B., & Naylor, J. E. (1972). The plaque control record. *Journal of Periodontology*, *43*(1), 38. https://doi.org/10.1902/jop.1972.43.1.38

Preus, H. R., Torgersen, G. R., Koldsland, O. C., Hansen, B. F., Aass, A. M., Larheim, T. A., & Sandvik, L. (2015). A new digital tool for radiographic bone level measurements in longitudinal studies. *BMC Oral Health*, *15*, 107. https://doi.org/10.1186/s12903-015-0092-9

Purdue, P. E., Koulouvaris, P., Potter, H. G., Nestor, B. J., & Sculco, T. P. (2007). The cellular and molecular biology of periprosthetic osteolysis. *Clinical Orthopaedics and Related Research*, *454*, 251–261. https://doi.org/10.1097/01.blo.0000238813.95035.1b

Renvert, S., Hirooka, H., Polyzois, I., Kelekis-Cholakis, A., Wang, H. L., & Working, G. (2019). Diagnosis and non-surgical treatment of peri-implant diseases and maintenance care of patients with dental implants - consensus report of working group 3. *International Dental Journal*, *69*(Suppl 2), 12–17. https://doi.org/10.1111/idj.12490

Renvert, S., & Polyzois, I. (2018). Treatment of pathologic peri-implant pockets. *Periodontology 2000*, *76*(1), 180–190. https://doi.org/10.1111/prd.12149

Renvert, S., Samuelsson, E., Lindahl, C., & Persson, G. R. (2009). Mechanical non-surgical treatment of peri-implantitis: A double-blind randomized longitudinal clinical study. I: Clinical results. *Journal of Clinical Periodontology*, *36*(7), 604–609. https://doi.org/10.1111/j.1600-051X.2009.01421.x

Roccuzzo, A., De Ry, S. P., Sculean, A., Roccuzzo, M., & Salvi, G. E. (2020). Current approaches for the non-surgical management of peri-implant diseases. *Current Oral Health Reports*, *7*(3), 274–282. https://doi.org/10.1007/s40496-020-00279-x

Roos-Jansaker, A. M., Renvert, H., Lindahl, C., & Renvert, S. (2007). Surgical treatment of peri-implantitis using a bone substitute with or without a resorbable membrane: A prospective cohort study. *Journal of Clinical Periodontology*, *34*(7), 625–632. https://doi.org/10.1111/j.1600-051X.2007.01102.x

Sanz, M., Chapple, I. L., & Working Group 4 of the VIII European Workshop on Periodontology. (2012). Clinical research on peri-implant diseases: Consensus report of Working group 4. *Journal of Clinical Periodontology*, *39*(Suppl 12), 202–206. https://doi.org/10.1111/j.1600-051X.2011.01837.x

Schär, D., Ramseier, C. A., Eick, S., Arweiler, N. B., Sculean, A., & Salvi, G. E. (2013). Anti-infective therapy of peri-implantitis with adjunctive local drug delivery or photodynamic therapy: Six-month outcomes of a prospective randomized clinical trial. *Clinical Oral Implants Research*, *24*(1), 104–110. https://doi.org/10.1111/j.1600-0501.2012.02494.x

Schulz, K. F., Altman, D. G., Moher, D., & CONSORT Group (2010). CONSORT 2010 statement: Updated guidelines for reporting parallel group randomised trials. *BMJ*, *340*, c332. https://doi.org/10.1136/bmj.c332

Schwarz, F., Becker, K., & Renvert, S. (2015). Efficacy of air polishing for the non-surgical treatment of peri-implant diseases: A systematic review. *Journal of Clinical Periodontology*, *42*(10), 951–959. https://doi.org/10.1111/jcpe.12454

Schwarz, F., Derks, J., Monje, A., & Wang, H. L. (2018). Peri-implantitis. *Journal of Periodontology*, *89*(Suppl 1), S267–S290. https://doi.org/10.1002/JPER.16-0350

Schwarz, F., Jepsen, S., Herten, M., Sager, M., Rothamel, D., & Becker, J. (2006). Influence of different treatment approaches on non-submerged and submerged healing of ligature induced peri-implantitis lesions: An experimental study in dogs. *Journal of Clinical Periodontology*, *33*(8), 584–595. https://doi.org/10.1111/j.1600-051X.2006.00956.x

Souza, A. B., Tormena, M., Matarazzo, F., & Araujo, M. G. (2016). The influence of peri-implant keratinized mucosa on brushing discomfort

and peri-implant tissue health. *Clinical Oral Implants Research*, *27*(6), 650–655. https://doi.org/10.1111/clr.12703

van Velzen, F. J., Lang, N. P., Schulten, E. A., & Ten Bruggenkate, C. M. (2016). Dental floss as a possible risk for the development of peri-implant disease: An observational study of 10 cases. *Clinical Oral Implants Research*, *27*(5), 618–621. https://doi.org/10.1111/clr.12650

Wang, C. W., Renvert, S., & Wang, H. L. (2019). Nonsurgical treatment of periimplantitis. *Implant Dentistry*, *28*(2), 155–160. https://doi.org/10.1097/ID.0000000000000846

Wohlfahrt, J. C., Aass, A. M., & Koldsland, O. C. (2019). Treatment of peri-implant mucositis with a chitosan brush-a pilot randomized clinical trial. *International Journal of Dental Hygiene*, *17*(2), 170–176. https://doi.org/10.1111/idh.12381

Wohlfahrt, J. C., Evensen, B. J., Zeza, B., Jansson, H., Pilloni, A., Roos-Jansåker, A. M., di Tanna, G. L., Aass, A. M., Klepp, M., & Koldsland, O. C. (2017). A novel non-surgical method for mild peri-implantitis—A multicenter consecutive case series. *International Journal of Implant Dentistry*, *3*(1), 38. https://doi.org/10.1186/s40729-017-0098-y

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Khan, S. N., Koldsland, O. C., Roos-Jansåker, A.-M., Wohlfahrt, J. C., Verket, A., Mdala, I., Magnusson, A., Salvesen, E., & Hjortsjö, C. (2022). Non-surgical treatment of mild to moderate peri-implantitis using an oscillating chitosan brush or a titanium curette—A randomized multicentre controlled clinical trial. *Clinical Oral Implants Research*, *33*, 1254–1264. https://doi.org/10.1111/clr.14007

IV

Received: 27 November 2022 | Revised: 22 March 2023 | Accepted: 15 April 2023

DOI: 10.1111/clr.14078

ORIGINAL ARTICLE

CLINICAL ORAL IMPLANTS RESEARCH WILEY

Non-surgical treatment of mild to moderate peri-implantitis with an oscillating chitosan brush or a titanium curette—12-month follow-up of a multicenter randomized clinical trial

Sadia N. Khan[1] | Odd Carsten Koldsland[2] | Ann-Marie Roos-Jansåker[3,4] | Johan Caspar Wohlfahrt[2] | Anders Verket[2] | Ibrahimu Mdala[5] | Anna Magnusson[6] | Eirik Salvesen[7] | Carl Hjortsjö[1]

[1]Department of Prosthetics and Oral Function, Institute of Clinical Dentistry, Faculty of Dentistry, University of Oslo, Oslo, Norway

[2]Department of Periodontology, Institute of Clinical Dentistry, Faculty of Dentistry, University of Oslo, Oslo, Norway

[3]Department of Periodontology, Faculty of Odontology, Malmö University, Malmö, Sweden

[4]Department of Periodontology, Blekinge Hospital, Karlskrona, Sweden

[5]Department of General Practice, University of Oslo, Oslo, Norway

[6]Department of Periodontology, Faculty of Medicine and Health, School of Medical Sciences, Orebro University, Orebro, Sweden

[7]Private Practice, Stavanger, Norway

Correspondence
Sadia N. Khan, Department of Prosthetics and Oral Function, Institute for Clinical Dentistry, Faculty of Dentistry, University of Oslo, PO. Box 1109, Oslo 0317, Norway.
Email: sadiak@odont.uio.no

Funding information
Norges Forskningsråd

Abstract

Objectives: To study clinical and radiographic outcomes after non-surgical treatment of peri-implantitis using either an oscillating chitosan brush (OCB) or titanium curette (TC) and to observe changes in clinical signs of inflammation after repeated treatment.

Methods: Thirty-nine patients with dental implants (n = 39) presented with radiographic bone level (RBL) of 2–4 mm, bleeding index (BI) ≥ 2, and probing pocket depth (PPD) ≥ 4 mm were randomly assigned to mechanical debridement with OCB (test) or TC (control). Treatment was performed at baseline and repeated at 3, 6, and 9 months in cases with > 1 implant site with BI ≥ 1 and PPD≥4 mm. Blinded examiners recorded PPD, BI, pus, and plaque. The radiographic bone level change between baseline and 12 months was calculated. A multistate model was used to calculate transitions of BI.

Results: Thirty-one patients completed the study. Both groups exhibited a significant reduction in PPD, BI, and pus at 12 months compared to baseline. Radiographic analysis showed stable mean RBL in both groups at 12 months. There was no statistically significant difference in any of the parameters between the groups.

Conclusions: Within the limitations of this 12-month multicenter randomized clinical trial, non-surgical treatment of peri-implantitis with OCB or TC showed no statistically significant differences between the groups. Clinical improvements and, in some cases, disease resolution, was observed in both groups. However, persistent inflammation was a common finding which further puts emphasis on the need for further treatment.

KEYWORDS
clinical trial, dental implants, peri-implantitis, titanium

Key findings: Clinical signs of inflammation were reduced in both groups at 12 months compared to baseline, but no statistically significant intergroup differences were observed.

Clinical trials registration: The study was registered at ClinicalTrials.gov (12/08/2017, NCT03373448).

1 | INTRODUCTION

Peri-implantitis, defined as biofilm-associated inflammation in peri-implant mucosa and progressive peri-implant bone loss (Berglundh et al., 1992, 2018), affects approximately 30% of all dental implants (Romandini et al., 2021). It is a widespread but false understanding that 'implants are for life' and that implants are better than teeth. Many patients have exaggerated expectations of rehabilitation with dental implants. Thus, biological complications such as peri-implantitis can be challenging for patients and clinicians (Abrahamsson et al., 2017; Insua et al., 2017).

The prevalence of peri-implantitis has been reported to vary between 1% and 47% on the patient level. Various disease definitions explain the significant variations in reported prevalence numbers (Derks & Tomasi, 2015). This issue was addressed at the World Workshop on the Classification of Periodontal and Peri-implant Diseases and Conditions (Berglundh et al., 2018). A case definition was proposed based on three criteria; (1) the presence of peri-implant signs of inflammation, (2) radiographic evidence of bone loss following initial bone remodeling, and (3) increased probing pocket depth (PPD) compared to probing depth measurements after prosthetic loading of the implant. Since baseline radiographs and probing depths are not always available, it was proposed that peri-implantitis diagnosis be based on radiographic bone level ≥3 mm combined with bleeding on probing (BoP) and PPD ≥ 6 mm. Studies employing similar definitions reported prevalence numbers of approximately 15% on the patient level after 9–14 years of function (Derks et al., 2016; Roos-Jansaker et al., 2006).

Biofilm accumulation is considered the main etiological factor for the inflammatory response in peri-implant soft- and hard tissues (Berglundh et al., 1992, 2018). The treatment focuses on controlling the inflammation by reducing the bacterial load around the infected implant (Renvert et al., 2019). A comparison of surgical versus non-surgical treatment has demonstrated superior outcomes for surgical approaches in cases with deeper peri-implant pockets (Polyzois, 2019). Although the non-surgical treatment of peri-implantitis is unpredictable, studies have also reported efficacious non-surgical protocols (Machtei et al., 2021). Early intervention, at a bone loss of 2–4 mm, is preferable as the outcome of surgical intervention largely depends on the amount of bone loss at the implant (Koldsland et al., 2018; Serino & Turri, 2011). Given the prevalence of peri-implantitis, non-surgical methods may reduce the treatment burden on specialist teams as general practitioners and dental hygienists may perform the treatment.

Furthermore, non-surgical methods generally require fewer resources from the dental team. Non-surgical treatment may be performed before surgical treatment, allowing the clinician to assess the peri-implant tissues´ response to treatment (Polyzois, 2019) and reduce the microbial load before surgery. Non-surgical intervention may also reduce the degree of inflammation and thereby facilitate surgical treatment (Schwarz et al., 2015). Developing effective non-surgical treatment methods is essential for treating patients where surgical treatment is contraindicated or for patients unwilling to undergo surgery. Surgical treatment may lead to soft tissue recession and influence the esthetic outcome in cases with high smile lines (Montero et al., 2022).

The affected implant's short- and long-term re-evaluation is indicated due to constant changes in plaque and inflammation (Polyzois, 2019). Outcomes of a recent multicenter randomized controlled clinical trial (RCT) demonstrated reductions in inflammatory parameters but rarely disease resolution when treatment was performed non-surgically with an oscillating chitosan brush (OCB) or titanium curettes (TC; Khan et al., 2022). Similar findings with a reduction in bleeding sites but no reduction in PPD and stable RBL were observed when implants were non-surgically treated with carbon fiber curettes or a Vector® system (Karring et al., 2005). Karring et al. (2005) defined peri-implantitis as BOP, PPD ≥5 mm, 1.5 mm radiographic bone loss, and exposed implant threads. Renvert et al. (2009) reported equivalent findings, with incomplete resolution of peri-implant inflammation 6 months after initial non-surgical treatment with titanium curettes or an ultrasonic device. Although eradication of the disease is rare, a decrease in inflammation seems to be a common feature in RCTs with shorter follow-up times and repeated non-surgical intervention (Karring et al., 2005; Sahm et al., 2011). The efficacy of repeated therapy over time has been evaluated for peri-implant mucositis, peri-implantitis, and after peri-implantitis surgery (Bassetti et al., 2014; Koldsland & Aass, 2020; Riben-Grundstrom et al., 2015). Despite post-surgical follow-up and repeated treatments every third month, peri-implant bleeding was observed 18 months after the first follow-up (Koldsland & Aass, 2020). For mucositis and peri-implantitis, a decrease in diseased sites was observed after repeated treatments and follow-up for 12 months. Clinical follow-ups and retreatments seem crucial considering the non-linear and progressive bone loss pattern in peri-implantitis (Berglundh et al., 2018).

Using a graded bleeding score may be beneficial in evaluating patients´ risk of destructive disease (Newbrun, 1996). Bleeding Index (BI; Roos-Jansåker et al., 2007) allows for identifying sites at risk of further bone destruction. Because BI includes four degrees of bleeding scores (0 = no bleeding, 1 = bleeding spot, 2 = bleeding line, 3 = profuse bleeding), it may be used to estimate the probability of transitions from one state to another. The likelihood of disease progression for periodontitis has been estimated using multistate Markov models (Mdala et al., 2014). Markov models are helpful when a condition involves a persistent risk. Markov models estimate the transition from one state to another for chronic diseases with a staged progression. There is a need to understand disease development in healthy and diseased peri-implant sites to reduce mortality and choose the proper treatment intervention and frequency. To our knowledge, transition analysis for clinical inflammation parameters for peri-implantitis has not been performed per se.

The present multicenter RCT aimed to evaluate repeated non-surgical treatment of peri-implantitis with an OCB or a TC. This study assessed changes in the following parameters: PPD, BI, presence of pus, and RBL 12 months after initial treatment (Sanz & Chapple, 2012). Furthermore, implant sites with and without inflammation were evaluated by assessing the transitions for BI scores during the study period.

2 | MATERIALS AND METHODS

2.1 | Study design

This randomized, prospective, two-arm, multicenter, controlled clinical trial including five specialist dental practices. The study was registered at clinicaltrials.gov (NCT03373448). Research ethical boards approved the trial in Norway and Sweden (REK south-east 2017/710, Linköping (EPN 2017/36–31). The study was conducted according to the principles in the Declaration of Helsinki (Fortaleza, Brazil). Good clinical practice (GCP) for medical devices and the Consolidation Standards of Reporting Trials (CONSORT) guidelines for clinical trials were followed (Schulz et al., 2010). A calibration meeting was held to discuss the study protocol prior to the study start. The detailed clinical protocol and study design have been published (Khan et al., 2022).

2.2 | Primary and secondary outcome variables

The primary outcome was a change in PPD. Secondary outcome variables included changes in BI, pus, and RBL.

2.3 | Sample size assessment and power

The calculation of the required sample size was based on the primary outcome; PPD. Alpha was set as 5%. To detect a difference of 1 mm for PPD between the groups, 17 patients per group were required to provide 80% statistical power (ß = 0.2).

2.4 | Study population

Patients diagnosed with peri-implantitis (mild/moderate) in dental specialist practices between April 2018 and February 2020 were invited to participate in the study. Mild to moderate peri-implantitis was defined as 2–4 mm radiographic reduction in peri-implant bone level, PPD ≥4 mm, and BI ≥2. One implant per patient was included in the study. Once patients had given written informed consent, they were randomly allocated to the test or control group.

Patients were included based on the following criteria:

1. Peri-implantitis as defined on an implant in function for more than 12 months.
2. Age ≥ 18 years.
3. Eligible for treatment in a dental clinic (ASA I and II, American Society of Anesthesiologists).
4. Full-mouth plaque scores ≤20% at the study start.
5. No plaque at the included implant.
6. Informed consent.
7. Consent to complete all follow-ups.

Patients were excluded based on the following criteria:

1. Supraconstructions that made it impossible to access the implant for clinical measurements.
2. Technical complications which had contributed to peri-implantitis and were not possible to resolve before final inclusion.
3. Mobile implant.
4. Active periodontal disease.
5. Implants treated for peri-implantitis with grafting materials.
6. Mucosal hyperplasia-inducing medications.
7. Systemic antibiotics ≤3 months prior to inclusion.
8. Acute or chronic medical conditions that would limit the patients´ ability to participate in the trial.
9. Advanced and uncontrolled peri-implantitis on proximate implants.
10. Patients presented with severely overloaded implants.
11. Previous or current radiotherapy to the head–neck region.
12. Current chemotherapy.
13. Current corticosteroid treatment.

Complementary inclusion and exclusion criteria were published in a previous publication (Khan et al., 2022).

2.5 | Null hypothesis

The null hypothesis was no statistically significant difference in the reduction of peri-implant inflammation (PPD, BI, and pus) 12 months after initial debridement between the two intervention groups.

2.6 | Randomization and allocation concealment

The allocation concealment between the two groups was conducted by the study administrator using computer-generated block randomization (RANDOM.ORG., Randomness and Integrity Services Ltd., Dublin, Ireland). Patients were randomly assigned to treatment in blocks of 10.

2.7 | Clinical and radiographic assessment

Probing pocket depth, BI, pus, Plaque Index (PI), and height of keratinized mucosa (KM) were registered at baseline before treatment and at 1, 3, 6, and 12 months after initial treatment.

Probing pocket depth, BI, and PI (Plaque Index) were measured at six sites per implant (mesiobuccal, buccal, distobuccal, mesiolingual, lingual, and distolingual) using a manual 0.20 N defined force periodontal probe (University of North Carolina, DB764R, AESCULAP, B Braun, Germany). Specialists in periodontology performed the assessment. The examiners were blinded to treatment allocation. The implant-retained supra-constructions were not removed for clinical examination or treatment.

Radiographic examinations were performed at baseline, 6- and 12 months post-treatment. Periapical radiographs were obtained using the long-cone paralleling technique with digital X-rays. ImageJ® image processing and analysis software program was used to measure changes in peri-implant RBL (Preus et al., 2015). Intraoral phosphor plates and sensors were used to calibrate the radiographs. Three examiners assessed the RBL twice for radiographs taken at baseline and 12 months. Information on patient data, time of examination, and clinic affiliation was removed from the radiographs before the analysis. The size of the sensor and phosphor plates were used to calibrate the radiographs. ImageJ® roentgenological attachment analyzer plugin converted markings on the radiographs to numeric data. The RBL was calculated as the distance from the implant neck to the first bone-to-implant contact.

2.8 | Clinical outcomes

The outcome variables were assessed at baseline before treatment and 3, 6, and 12 months after initial treatment.

The following clinical variables were assessed at the affected implant:

1. PI—presence or absence of plaque (O'Leary et al., 1972). Registered by running the probe along the marginal surface of the implant (Mombelli et al., 1987).
2. Pus—presence or absence of pus/suppuration.
3. BI—registered 30 s after probing. The bleeding scores were categorized into four categories; score 0 = no bleeding, score 1 = isolated bleeding spot, score 2 = blood forming a red line, and score 3 = profuse bleeding (Roos-Jansåker et al., 2007).
4. PPD registered in millimeters.
5. Height of keratinized mucosa (KM) was assessed midbuccaly with a periodontal probe.

2.9 | Treatment interventions and protocol

There were two parallel treatment arms. Prior to inclusion, all study patients underwent an initial hygiene phase with oral hygiene instructions. At the baseline registration and intervention, the PI was 0 for all implant surfaces. Implants in the test group were treated with an OCB for 2 min. The OCB was soaked in sterile saline prior to treatment. The control group treatment was performed using TC for 2 min (Langer and Langer, Rønvig, Denmark). Peri-implant pockets were irrigated with saline after mechanical treatment in both groups. Treatment was performed at baseline and repeated at 3, 6, and 9 months in cases with >1 implant site with BI ≥ 1 and (PPD) ≥ 4 mm (Figure 1). Local infiltration anesthesia was administrated when required by the patients. Both treatment modalities were performed non-surgically. Treatment was performed by five authorized dental hygienists.

2.10 | Treatment outcomes

Disease eradication: PPD <4 mm, BI 0, and no reduction in RBL compared to baseline.Treatment success: ≤1 implant site with BI ≤1, absence of pus, PPD ≤5 mm, and absence of progressive bone loss.Resolution of inflammation: BI 0.Disease improvement: BI = 1. Peri-implantitis recurrence/progression: RBL increase, and/or PPD increase, and/or BI ≥2.

2.11 | Data management and statistical analysis

Calculations and analysis were performed using Stata Statistical Software, Version 16.1 (StataCorp.2001. Statistical Software: Release 7.0. Stata Corporation). A *p*-value less than .05 was considered statistically significant.

Data were analyzed by per-protocol (PP) analysis on assessed patients at all time points. In addition, the intention-to-treat (ITT) principle was used, meaning that all randomized patients were included in the analysis using multiple imputations generated in R (R app 4.0.3 GUI Mac OS, R Foundation for Statistical Computing, Vienna, Austria). Implant and patient characteristics were described with percentages for categorical variables and means with standard deviations (SD) for continuous data.

Probing pocket depth, BI, pus, and PI data were obtained at baseline, 1, 3, 6, and 12 months for one implant in each patient. Patients were included in five different dental practices. The mean of PPD sites ≥4 mm was calculated for all implants at each study time point. A three-level linear regression model for PPD and a logistic regression model for BI with random intercept and random effect of time (level 3) were used to account for possible

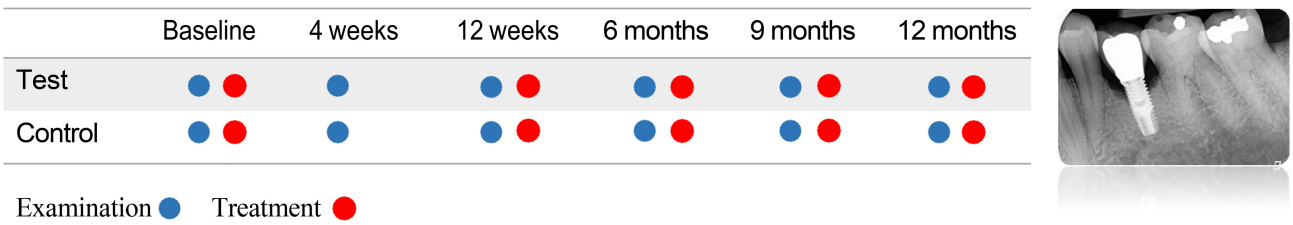


FIGURE 1 Patients were examined at baseline, 1, 3, 6, 9 and 12 months. Treatment was performed at baseline and retreated every third month in cases with PPD ≥ 4 mm and BI ≥ 1.

dependences of the data within the patients (level 2) who were nested within the clinics (level 1). Within and between the group changes in PPD and BI at each study time point were obtained from the two-way interaction of time with the groups. Variability in PPD and BI attributed to differences in patients, and clinics were described using estimates of ICC.

The transitions between BI states were modeled using a three-state Markov model. For the Markov analysis, BI 0 was considered healthy, and BI 1 was a state between health and disease. BI 2 and BI 3 were merged into one state and categorized as sick. All states were considered transient. The analysis was performed at the site level for both groups. Each implant was presented with six site measurements (mesiobuccal, buccal, distobuccal, distopalatal, palatal, and mesiopalatal). In addition, BI transitions at the implant level were performed based on the highest BI at baseline and 12 months. Markov analysis was performed with the msm package in R (R app 4.0.3 GUI Mac OS, R Foundation for Statistical Computing).

3 | RESULTS

A total of 31 patients with peri-implantitis, as defined above, completed the scheduled 12 months examination appointment. The study flow chart is presented in Figure 2. The baseline implant and patient characteristics for both groups and the dropouts are presented in Table 1. No adverse reactions related to the treatments were reported.

3.1 | Clinical withdrawal

A total of eight patients were withdrawn from the study by the clinicians (Figure 2). In the test group, one patient was excluded at the follow-up between 3 and 6 months, and three patients between 6 and 12 months. All withdrawals in the control group were at the follow-ups between 6 and 12 months. Radiographic and clinical data at baseline for the dropouts are presented in Table 2.

3.2 | Clinical and radiographic changes between the study groups (per-protocol; $n = 31$)

The changes in PPD, BI, PI, pus, plaque, KM, and RBL from baseline to 6 and 12 months are presented in Table 3.

3.2.1 | Probing pocket depth

Changes in the mean PPD at 6 and 12 months are reported in Table 3. Differences in PPD between the groups are presented in Table 4. Changes in mean PPD at each study point from baseline to 12 months between and within the groups are demonstrated in Figure 3. Both treatments resulted in a statistically significant reduction in PPD at 3, 6, and 12 months compared to baseline ($p < .05$). No statistically significant differences between the groups were registered at any time point. Reduction in PPD was statistically significant between 6 and 12 months for the test group ($p < .05$).

3.2.2 | Bleeding index

The results from the ordinal logistic regression model with the following comparisons: no bleeding (BI 0) and spot bleeding (BI 1) combined vs line and profuse bleeding combined, demonstrated a statistically significant decrease in BI 2 and BI 3 at the implant level in the test and the control group from baseline to 12 months (Figure 3b). The differences between the groups were not a statistically significant at 1, 3, 6, or 12 months ($p > .05$; Table 4).

3.2.3 | Pus

The number of implants with pus decreased significantly in both study groups between baseline to 12 months (Table 3). However, there was no statistically significant difference between the groups ($p > .05$).

3.2.4 | Radiographic bone level

At baseline, three patients in the test group and six patients in the control group had RBL ≥ 3 mm. The mean RBL for both study groups at baseline, 6, and 12 months are presented in Table 3. The radiographic bone level was stable in all patients, and the change in bone levels between baseline and 12 months was not statistically significant for any of the groups. The intraclass correlation coefficient (ICC) describing the intra-examiner agreement was 0.98.

3.2.5 | Composite outcome

At 12 months, one implant in the test group and none in the control group presented disease eradication according to the criteria: PPD <4 mm, no bleeding (BI 0), and no changes in RBL compared to baseline. Treatment success was defined as ≤1 implant site with BI ≤ 1, absence of pus, PPD ≤ 5 mm, and no progressive bone loss was achieved for three implants in the test group and one implant in the control group. The differences between the groups were not statistically significant ($p > .05$).

3.2.6 | Height of keratinized mucosa

The mean KM at baseline, 6 and 12 months for both groups is presented in Table 3.

At baseline, about 70% of implants in the test group and 60% in the control group had KM ≥ 2 mm. At 12 months, the number of

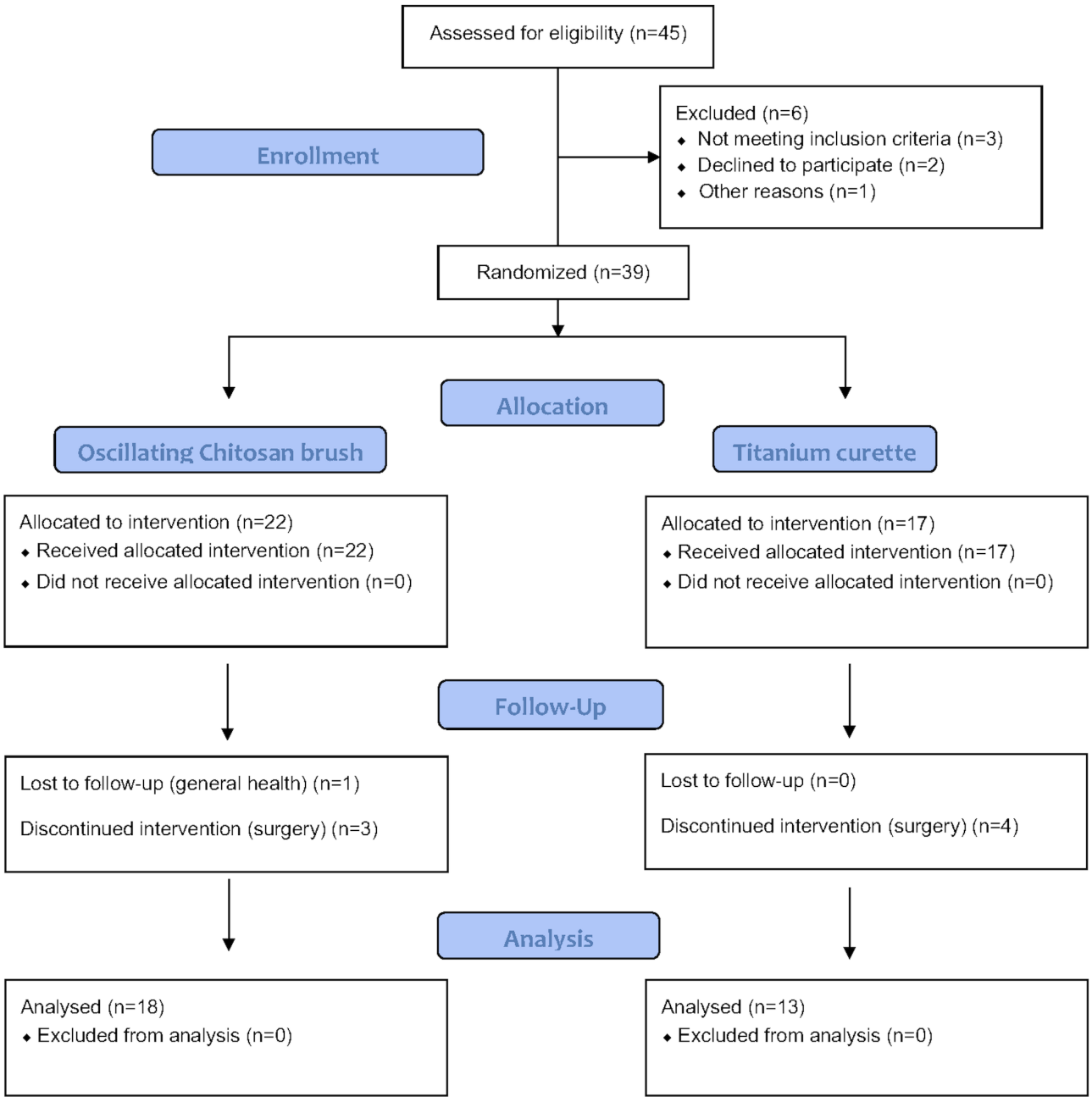


FIGURE 2 A CONSORT flowchart of enrollment, allocation, follow-up, and analysis.

implants with KM ≥ 2 mm decreased to approximately 60% in the test group and to 40% in the control group.

3.2.7 | Plaque index

Plaque scores at the included implants changed throughout the study period, without significant differences between the study time points ($p > .05$) Figure 4. At the site level, an association between bleeding on probing and the presence of plaque was not observed.

3.3 | Clinical and radiographic changes between the study groups (intention-to-treat; $n = 39$)

Study group differences derived from the intention-to-treat (ITT) analysis are presented in Tables 5 and 6. In both study groups, the radiographic bone level remained stable throughout the study period. No statistically significant differences between the groups were observed for PPD, BI, presence of pus, or RBL. PPD, BI, and plaque changes are presented in included implants changed throughout S1-S3.

TABLE 1 Baseline characteristics of all randomized study patients ($n=39$) and dropouts ($n=8$).

	Patients randomized to the study		Dropouts		*p*-values	
Variable	**OCB**	**TC**	**OCB**	**TC**	**OCB**	**TC**
Subjects/Implants (*n*)	22	17	4	4		
Mean age (± SD)	62.86 (±12.2)	61.12 (±3.7)	61.5 (±9.0)	65.5 (±11.4)	.84	.18
Gender						
Male (M), *n* patients (%)	5 (22.7)	9 (52.9)	0 (0.0)	1 (25.0)	.29	.31
Female (F), *n* patients (%)	17 (77.3)	8 (47.1)	4 (100.0)	3 (75.0)	.01	.31
Daily smoker; *n* patients (%)	4 (18.2)	1 (5.9)	0 (0.0)	0 (0.0)	.35	.62
Diabetes; *n* patients (%)	5 (22.7)	3 (17.6)	2 (75.0)	0 (0.0)	.04	.36
Tooth loss due to periodontitis; *n* patients (%)	6 (27.3)	5 (29.4)	1 (25.0)	0 (0.0)	.92	.21
Front; *n* implants (%)	8 (36.4)	9 (52.9)	1 (25.0)	2 (50.0)	.66	.92
Premolar; *n* implants (%)	11 (50.0)	7 (41.2)	2 (50.0)	2 (50.0)	1.0	.74
Molar; *n* implants (%)	3 (13.6)	1 (5.9)	1 (25.0)	0 (0.0)	.56	.62
Screw-retained; *n* implants (%)	17 (77.3)	15 (88.2)	4 (100.0)	4 (100.0)	.01	.01
Cement-retained; *n* implants (%)	5 (22.7)	1 (5.9)	–	–		
Not reported; *n* implants (%)	0 (0.0)	1 (5.9)	–	–		
Implant-retained crown; *n* implants (%)	10 (45.5)	5 (33.3)	1 (25.0)	2 (50.0)	.87	.53
Implant-retained fixed dental prosthesis; *n* implants (%)	12 (54.5)	12 (52.2)	3 (75.0)	2 (50.0)	.44	.94

Abbreviations: OCB, oscillating chitosan brush; TC, titanium curettes.

TABLE 2 Radiographic and clinical data of the complete cases and dropout patients at baseline ($n=8$).

	Complete cases ($n=31$)		Dropouts ($n=8$)		*p* values	
Variable	**OCB**	**TC**	**OCB**	**TC**	**OCB**	**TC**
Subjects/Implants (*n*)	18	13	4	4		
Radiographic bone level (±SD)	2.4 (±0.7)	2.9 (±0.5)	2.6 (±0.2)	2.5 (±0.5)	.58	.29
PPD[§] mean (mm)[a]	5.0 (±0.8)	5.3 (±1.4)	5.0 (±0.3)	5.3 (±0.6)	1.00	1.00
Mean (PPD ≥ 4 mm)[a] (±SD)	5.2 (±0.9)	5.6 (±0.1)	5.4 (±0.5)	5.3 (±0.6)	.73	.08
Mean (PPD ≥ 6 mm)[a] (±SD)	6.7 (±0.5)	6.5 (±0.9)	8.0 (±1.7)	6.2 (±0.3)	.01	.53
BI ≥ 2 (%)	100.0	100.0	100.0	100.0	1.00	1.00
BI 0 (%)	0.0	0.0	0.0	0.0	-	-
BI 1 (%)	0.0	0.0	0.0	0.0	-	-
BI 2 (%)	61.1	76.9	100.0	75.0	.06	.93
BI 3 (%)	38.9	23.1	0.0	25.0	.13	.93
Pus (%)	50	53.8	25.0	100.0	.36	.12
Plaque (%)	2.8	9.0	0.0	0.0	.73	.01
Keratinized mucosa ≥ 2 mm (%)	72.2	61.5	25.0	75.0	.08	.62

Abbreviations: BI, bleeding index; OCB, oscillating chitosan brush; PPD, probing pocket depth; TC, titanium curettes.

[a]PPD mean = mean of 6 measurements at selected sites, whereas mean (PPD ≥ 4 mm) = mean of measurements ≥4 mm, and mean (PPD ≥ 6 mm) is the mean of measurements 6 ≥ mm.

3.4 | Markov models

During the 12-month interval, a large number of healthy sites (BI0), remained healthy. Comparably, a large number of inflamed sites (BI2 + BI3) remained inflamed. The transitions between the BI states for both groups at the site level are demonstrated in Figure 5. BI transitions at the implant level between baseline and 12 months are presented in Figure 6.

TABLE 3 Changes in mean PPD and BI between the groups at each time point obtained from the linear and ordinal logistic multilevel regression model with clinic (level 1), patient random effects (level 2), and time (level 3), based on per-protocol analysis ($n = 31$).

	Baseline		6 months		12 months			
	OCB ($n = 18$)	TC ($n = 13$)	OCB ($n = 18$)	TC ($n = 13$)	OCB ($n = 18$)	TC ($n = 13$)	*p* value[a]	*p* value[b]
Clinical parameters								
Radiographic bone level (±SD)	2.4 (±0.7)	2.9 (±0.5)	2.5 (±0.5)	2.7(±0.7)	2.5 (±0.5)	3.1 (±0.7)	.62	.41
PPD[§] mean (mm)[c]	5.0 (±0.8)	5.3 (±1.4)	4.5 (±1.1)	4.7 (±0.1)	3.9(±1.2)	3.9 (±1.1)	.01	.01
Mean (PPD ≥ 4 mm) (±SD)[c]	5.2 (±0.9)	5.6 (±0.1)	4.5 (±1.1)	4.3 (±0.9)	4.0 (± 1.2)	4.0 (± 1.1)	.01	.01
Mean (PPD ≥ 6 mm) (±SD)[c]	6.7 (±0.5)	6.5 (±0.9)	6.8 (±0.7)	6.3(±0.6)	6.7 (±0.7)	6.6 (±0.7)		
BI ≥ 2 (%)	100.0	100.0	77.8	61.5	44.4	76.9	.02	.01
BI 0 (%)	–	–	–	15.4	11.1	0	–	–
BI 1 (%)	–	–	22.2	23.1	44.4	23.1	–	–
BI 2 (%)	61.1	76.9	77.8	61.5	38.9	76.9		
BI 3 (%)	38.9	23.1	-	-	5.6	0	.01	.07
Pus (%)	50	53.8	33.3	53.8	16.7	0	.05	.01
Plaque (%)	2.8	9.0	4.6	19.2	10.2	5.1	.4	.7
Keratinized mucosa ≥ 2 mm (%)	72.2	61.5	77.8	69.2	77.8	38.5	.70	.24

Abbreviations: BI, bleeding index; OCB, oscillating chitosan brush; PPD, probing pocket depth; TC, titanium curettes.

[a]Difference between baseline and 12 months for OCB.

[b]Difference between baseline and 12 months for TC.

[c]PPD mean = mean of 6 measurements at selected sites, whereas mean (PPD ≥ 4 mm) = mean of measurements ≥4 mm, and mean (PPD ≥ 6 mm) is the mean of measurements 6 ≥ mm.

TABLE 4 Changes in mean PPD and BI between the groups at each time point obtained from the linear and ordinal logistic multilevel regression model with clinic (level 1), patient random effects (level 2), and time (level 3), based on per-protocol analysis ($n = 31$).

	Baseline		1 month		3 months		6 months		12 months	
	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value
Group (ref.: TC)										
PPD OCB	0.5 (−0.3, 1.2)	.4	0.3 (−0.4, 1.0)	.4	−0.1 (−0.8, 0.6)	.8	−0.2 (−0.9, 0.5)	.6	0.1(0.7, 0.8)	.9
	OR (95% CI)	***p*-value**	**OR (95% CI)**	***p*-value**	**OR (95% CI)**	***p*-value**	**OR (95% CI)**	***p*-value**	**OR (95% CI)**	
Between the groups (ref.: TC)										
BI 0–1 vs. BI 2–3	0.8(0.2, 3.3)	.7	0.8(0.2, 3.5)	.8	0.7(0.2, 3.0)	.6	2.9(0.7, 12.4)	.1	0.3(0.1, 1.3)	.1

Abbreviations: BI, bleeding index; CI, confidence interval; OCB, oscillating chitosan brush; PPD, probing pocket depth; TC, titanium curettes.

4 | DISCUSSION

This 12-month multicenter, single-blinded RCT aimed to evaluate the efficacy of repeated non-surgical mechanical treatment of peri-implantitis performed with an OCB or TC after an initial hygiene phase. Implants in both groups demonstrated a statistically significant reduction in BI and PPD at 3, 6, and 12 months after initial treatment. Pus was significantly reduced in both groups at 12 months compared to baseline. The null hypothesis was not rejected as the difference in the reduction of inflammation parameters between the groups was not statistically significant at any time point.

It is demonstrated that disease resolution is unpredictable after non-surgical peri-implantitis treatment; thus, novel methods should be developed and tested (Roccuzzo et al., 2020). Non-surgical treatment of mucositis and peri-implantitis with OCB compared to TC has been evaluated in clinical studies with equal efficacy for both treatment modalities (Khan et al., 2022; Koldsland & Aass, 2020; Wohlfahrt et al., 2017) The presence of pus was resolved in 19.1% of the implants in the OCB group and with no reduction in the TC group when the treatments were compared in an RCT with a 6-month follow-up (2022). Further reduction in the presence of pus following repeated treatments over 12 months was observed in the present study (2022).

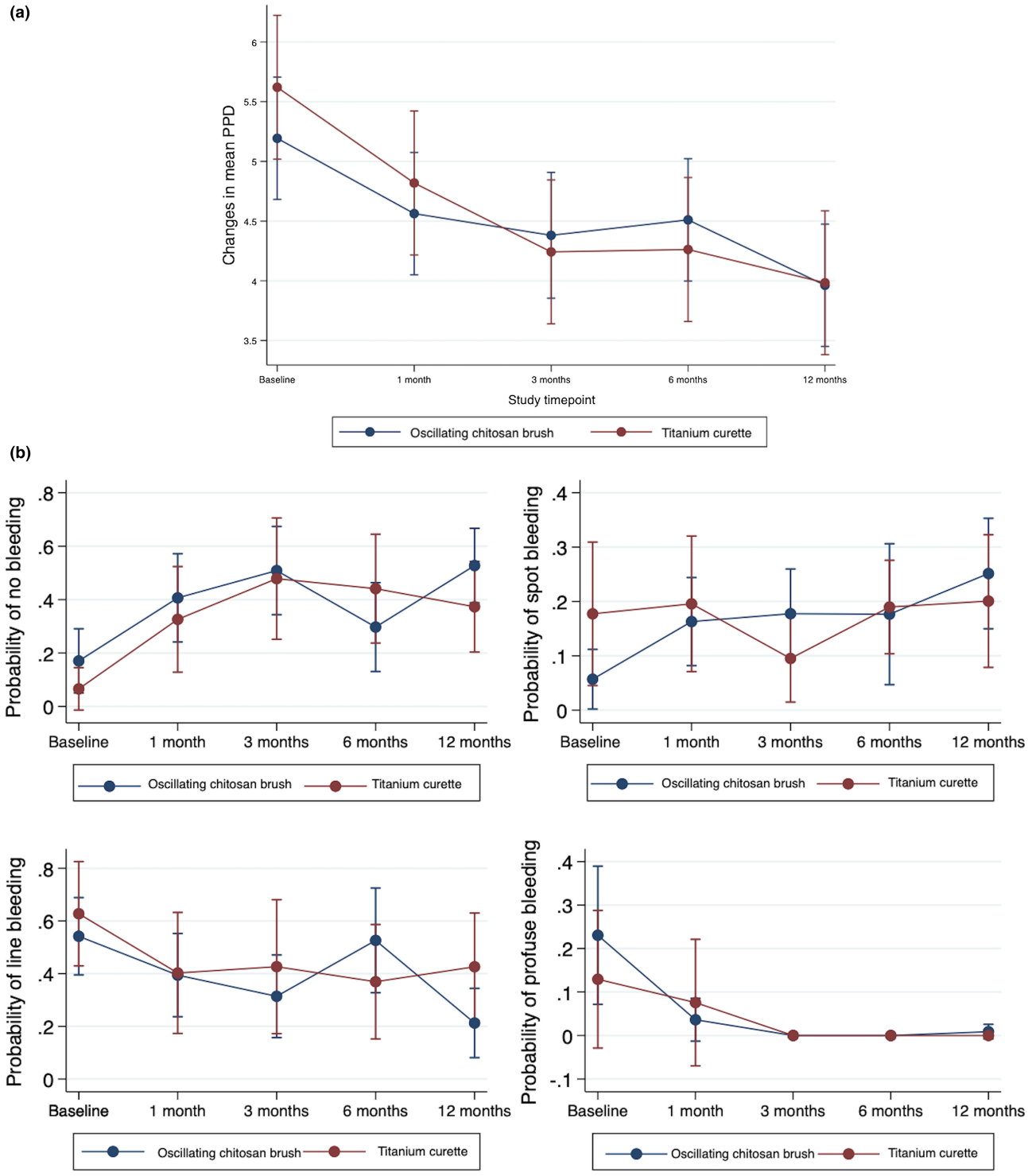


FIGURE 3 (a) Changes in mean PPD ≥ 4 mm, from baseline to 12 months between and within the groups (per-protocol; $n = 31$). (b) The probability of BI 0–3 at implant level between and within the groups at each time point between baseline and 12 months (per-protocol; $n = 31$).

Peri-implantitis has been reported to progress in a non-linear, accelerating pattern if no treatment is performed (Fransson et al., 2010). Patients with peri-implant mucositis who are not provided adequate preventive maintenance care show an increase in total bacterial load and a higher prevalence of peri-implantitis after 5 years (Costa et al., 2019). As of today, there has yet to be a consensus on a protocol for peri-implant maintenance or supportive therapy, both concerning instruments that should be applied and the frequency of care.

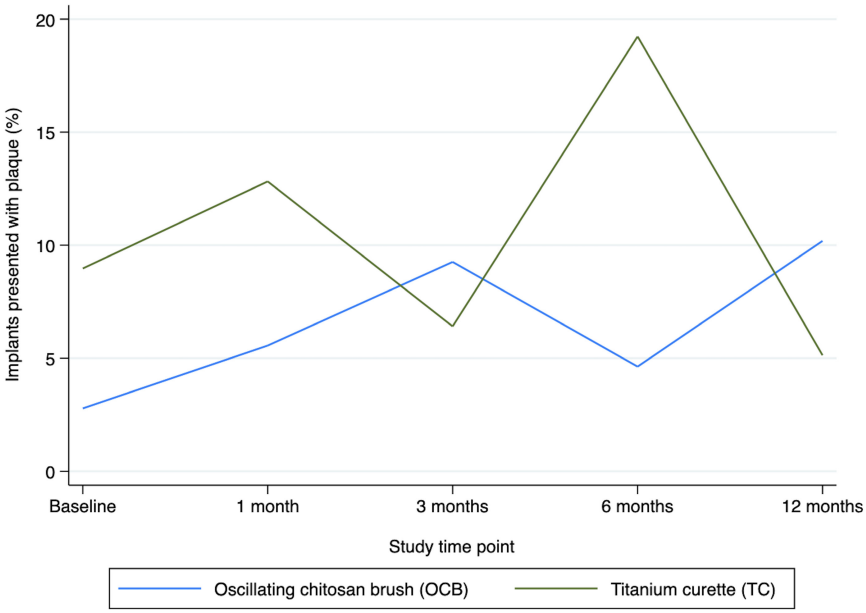


FIGURE 4 Implants with plaque at all study timepoints (per-protocol; $n = 31$).

TABLE 5 Radiographic and clinical data based on *intention-to-treat* analysis at baseline, 6 months and 12 months for both study groups ($n = 39$).

	Baseline		6 months		12 months			
	OCB ($n = 22$)	TC ($n = 17$)	OCB ($n = 22$)	TC ($n = 17$)	OCB ($n = 22$)	TC ($n = 17$)	*p* value[a]	*p* value[b]
Clinical parameters								
Radiographic bone level (±SD)	2.4 (±0.1)	2.8 (±0.1)	2.5 (±0.5)	2.6 (±0.7)	2.5 (±0.7)	3.0 (±0.5)	.51	.12
PPD mean (mm)[c]	5.1 (±0.9)	5.3 (±1.6)	4.5 (±1.1)	4.4 (±1.0)	3.9 (±1.2)	3.9 (±1.1)	.01	.01
Mean (PPD ≥ 4 mm), (±SD)[c]	5.3 (±0.7)	5.6 (±1.4)	4.9 (±0.1)	5.0 (±0.1)	4.7 (±0.6)	4.6 (±0.1)	.01	.01
Mean (PPD ≥ 6 mm), (±SD) [c]	6.8 (±0.6)	6.5 (±0.9)	6.7 (±0.7)	6.3 (±0.6)	6.5 (±0.7)	6.5 (±0.5)	.16	1.00
BI ≥ 2 (%)	88.7	93.3	51.6	40.9	22.0	38.3	.01	.01
BI 0 (%)	2.3	1.3	17.2	24.5	50.6	31.9	.01	.01
BI 1 (%)	9.0	5.4	31.2	34.6	27.4	29.8	.11	.06
BI 2 (%)	85.3	87.4	51.2	40.3	21.6	37.4	.01	.01
BI 3 (%)	3.4	5.9	0.4	0.6	0.4	0.9	.52	.42
Pus (%)	47.5	64.7	33.3	64.7	17.5	8.4	.03	.01
Plaque (%)	2.4	6.7	4.2	20.6	9.9	5.7	.27	.9
Keratinized mucosa ≥ 2 (mm) (%)	71.6	68.6	66.9	58.9	75.3	71.9	.78	.83

Abbreviations: BI, bleeding index; OCB, oscillating chitosan brush; TC, titanium curettes.

[a]Difference between baseline and 12 months for OCB.

[b]Difference between baseline and 12 months for TC.

[c]PPD mean = mean of 6 measurements at selected sites, whereas mean (PPD ≥ 4 mm) = mean of measurements ≥4 mm, and mean (PPD ≥ 6 mm) is the mean of measurements ≥6 mm.

Treatment of both peri-implant mucositis and peri-implantitis aims to reduce the bacterial load and control the inflammation. In the present study, FMPS < 20% and no plaque at the included implants at baseline were prerequisites. At baseline, the test group had the lowest plaque scores, with an increasing trend throughout the study period. The control group had a higher number of implants with plaque at baseline compared to the test group and showed a reduction in plaque at 3 months. The number of implants with plaque

TABLE 6 Changes in mean PPD and BI between the groups at each time point obtained from the linear and ordinal logistic multilevel regression model with clinic (level 1), patient random effects (level 2), and time (level 3) based on imputed data ($n = 39$).

	Baseline		1 month		3 months		6 months		12 months	
	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value	ß (95% CI)	*p*-value
Group (ref.: TC)										
PPD OCB	- 0.3 (−1.2, 0.5)	.48	0.1 (−0.7, 1.0)	.80	0.1 (−1.0, 0.8)	.82	0.1 (−0.8, 1.0)	.78	0.2 (−1.4, 0.9)	.71
	OR (95% CI)	*p*-value	OR (95% CI)	*p*-value	OR (95% CI)	*p*-value	OR (95% CI)	*p*-value	OR (95% CI)	
Between the groups (ref.: TC)										
BI 0–1 vs. BI 2–3	0.2 (0.1, 0.9)	.03	0.5 (0.2, 1.5)	.24	0.5 (0.2, 1.4)	.20	1.0 (0.4, 3.0)	.97	0.5 (0.1, 1.4)	.16

Abbreviations: BI, bleeding index; CI, confidence interval; OCB, oscillating chitosan brush; PPD, probing pocket depth; TC, titanium curettes.

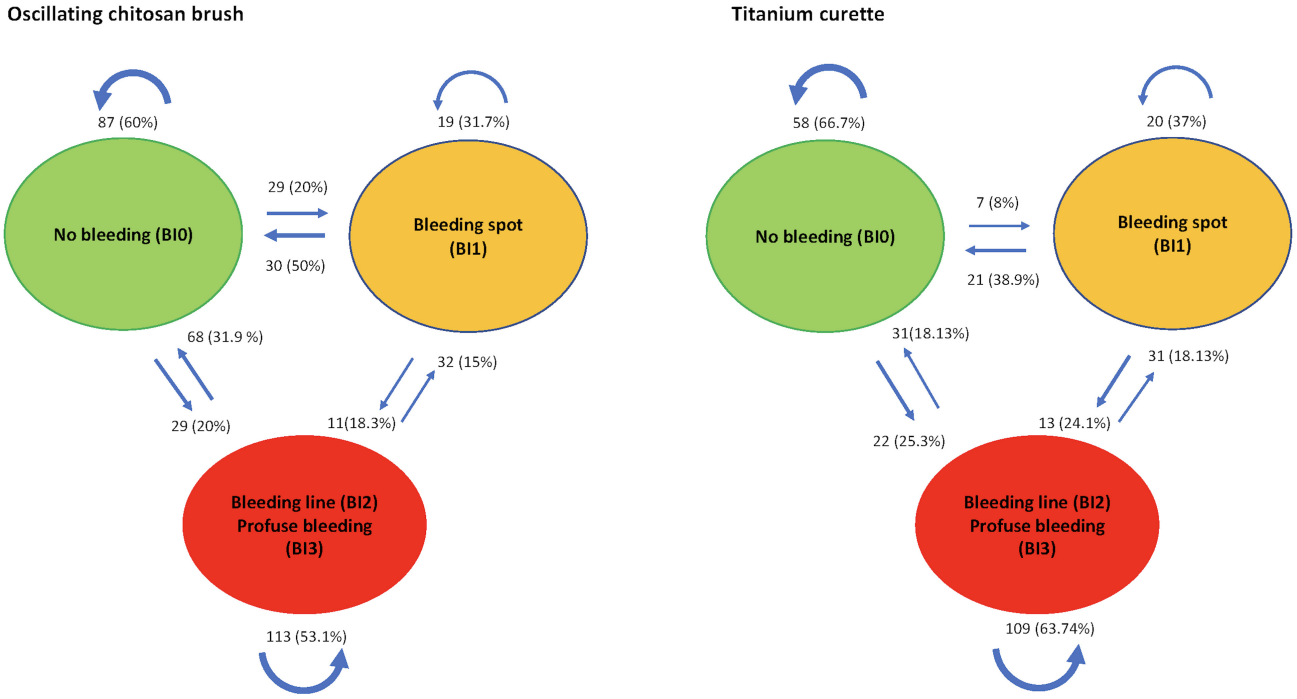


FIGURE 5 A multi-state Markov model for peri-implantitis. BI 0 correspond to health, BI 1 to a state between health and disease and BI 2 and 3 as disease. The number and percentage of site transitions between the different states are represented by the arrows. The model shows all transitions through the study period of 12 months ($n = 31$).

in the control group increased significantly from 3 to 6 months. At 12 months, the implants in the control group showed a significant reduction in the presence of plaque, with values lower than at baseline. A causal relationship between plaque and peri-implant inflammation has been reported in previous studies (Pontoriero et al., 1994; Serino & Ström, 2009). The difference in plaque levels may have affected the results in the present study.

In the present study, 53.1% of the implant sites in the test group and 63.7% of the sites in the control group remained at BI 2–3 through the study period despite four active treatments. Furthermore, transition of healthy implant sites (BI 0) to diseased sites (B1 2–3) was observed in both groups. Contrary to the within-group results from the regression analysis, results from the Markov model indicated active disease. The transition from health to disease (BI 0 to BI 2–3) and infrequent improvement of sites with BI 2–3 is an important finding in the present study, as complete disease improvement was not achieved according to the BI transitions. In comparison, the regression analysis showed a statistically significant reduction in BI and PPD at 6 and 12 months within both groups. Transitions analysis at the implant level showed that most implants had BI2-3 at 12 months. Bleeding improvement from BI2-3 to BI1 was a common finding in both study groups. While transitions from BI2-3 to BI0 were observed in 11.2% of the implants in the test group, no implants in the control group showed improvement from BI2-3 to BI0. Transitions between the different BI scores are not reported for either peri-implant mucositis or peri-implantitis in the literature. The present study is the first attempt to estimate the disease initiation of healthy sites. Although, after the examinations

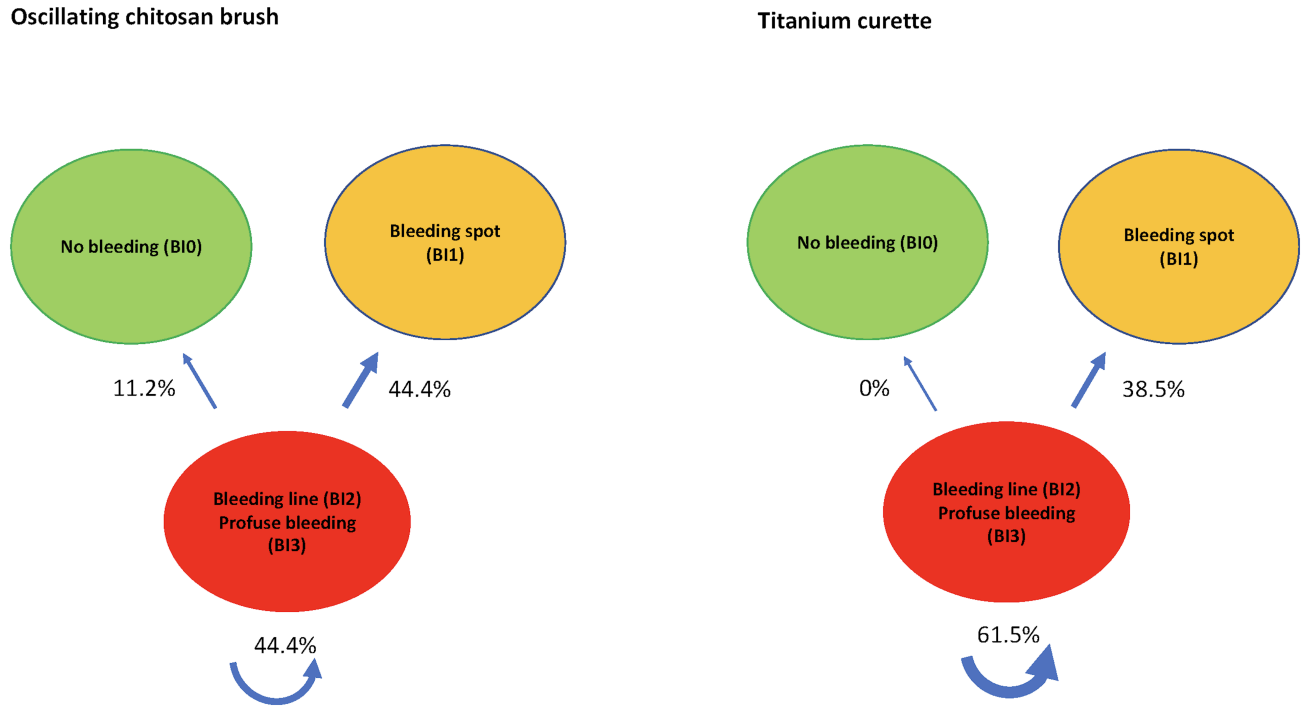


FIGURE 6 Transitions between BI states reported on implant level between baseline and 12 months. Each implant is presented with the highest BI score at each study timepoint ($n = 31$).

at 3 and 6 months, only sites presented with bleeding on probing were retreated, it is conceivable that the patients´ hygiene routines were positively influenced by the fact that they participated in a study with repeated follow-ups, known as the Hawthorne effect (Sedgwick & Greenwood, 2015).

The PPD reduction in the test group was statistically significant from 6 to 12 months in the present study. Contrary to the control group, the test group showed an increase in PPD between 3 to 6 months. The statistically significant PPD decrease in the test group between 6 to 12 months could be related to the PPD increase between 3 and 6 months.

In the present study, the baseline RBL was 2.4 (±0.7) mm and 2.9 (±0.5) mm for the test and the control group, respectively, leading to a difference in baseline characteristics among the groups. However, the CONSORT guidelines do not encourage significance testing of the baseline characteristics and describe the differences as 'results by chance' and not bias (Schulz et al., 2010). At 12 months, the RBL increased to 2.5 (±0.5) mm and 3.1 (±0.7) mm for the test and control groups, respectively. The registered RBL may have been influenced by the inter-examiner difference in radiographic technique and various digital x-ray equipment in all five clinics participating in the present multicenter study. Radiographs from the time of prosthetic loading were not available. The first radiographs were obtained at the time of study recruitment. Thus, a different threshold for peri-implantitis should have been used since baseline radiographic data were lacking, namely 3 mm bone loss and 6 mm PPD.

The required sample size to achieve 80% study power was calculated to be 17 patients in each group. At baseline, 21 patients were included in the test group and 17 in the control group (Khan et al., 2022). At 12 months, the test group consisted of 18 patients and the control group 13 patients. The multicenter randomization process leading to differential attrition rates between the test and control group was caused by separate randomizations at the five clinics. The skewing is a limitation of the present study, and significant differences may have been present with a high number of patients.

Intention-to-treat analysis with imputed data was compared to the results derived from the per-protocol analysis of complete cases. Analyses based on the per-protocol method run the risk of attrition bias as dropout patients may differ from those who remain. In the present study, one patient dropped out, and seven were excluded during the study period. The reason for exclusion was mainly recurrence or worsening of the disease and indication for surgical intervention, but the dropout rates were not significantly different in the two groups (4/22 vs. 4/17). Systematic differences in baseline data of complete cases and the dropouts were compared using regression analysis. For the radiographic and clinical data at baseline, a statistically significant difference was observed in the presence of plaque and the odds of being in BI0-1 vs. BI2-3 for the control group.

Within the limitations of this 12 months multicenter randomized clinical trial, non-surgical treatment of peri-implantitis with OCB and TC demonstrated no statistically significant difference between the treatment groups. Although this finding does not demonstrate an equivalence between the treatment methods, in view of the small sample sizes, it should be noted from the figures showing the time development of various features that none of the treatments seems

superior to each other. Clinical improvements in both groups and some cases disease resolution were achieved. Differences between test and control groups in changes in inflammation were not statistically significant but due to withdrawals, power was low. However, persistent inflammation was a common finding that further puts emphasis on the need for further treatment. Studies with larger sample sizes are important in the future.

AUTHOR CONTRIBUTIONS

J.C.W. conceived the idea. A.V., A.M.R.J., A.M., E.S., and S.N.K collected the data. I.M. and S.N.K. analyzed the data. S.N.K. led the writing. All co-authors have approved the manuscript.

FUNDING INFORMATION

The study was funded by the Research Council of Norway and University grants.

CONFLICT OF INTEREST STATEMENT

Caspar Wohlfahrt is the inventor and patent holder of the chitosan brush (Labrida Bioclean®, Labrida AS Oslo, Norway). Ann Marie Roos-Jansåker and Caspar Wohlfahrt are shareholders in Labrida AS. Drs Khan, Koldsland, Verket, Mdala, Magnusson, Salvesen and Hjortsjö report no conflicts of interest related to this study.

DATA AVAILABILITY STATEMENT

Data are stored at a central university facility and are available upon request from the corresponding author, S.K.

ETHICS STATEMENT

The study was approved by the Regional Committee for Medical and Health Research Ethics, South-East Norway (REK sør-øst 2017/710) and by the Swedish Ethical Review Authority, Linköping (EPN 2017/36–31).

PATIENT CONSENT STATEMENT

Informed consent forms were signed by participants prior to the study start.

PERMISSION TO REPRODUCE MATERIAL FROM OTHER SOURCES

Not applicable.

ORCID

Sadia N. Khan https://orcid.org/0000-0003-3925-326X
Odd Carsten Koldsland https://orcid.org/0000-0001-6842-8255

REFERENCES

Abrahamsson, K. H., Wennstrom, J. L., Berglundh, T., & Abrahamsson, I. (2017). Altered expectations on dental implant therapy; views of patients referred for treatment of peri-implantitis. *Clinical Oral Implants Research*, *28*(4), 437–442. https://doi.org/10.1111/clr.12817

Bassetti, M., Schar, D., Wicki, B., Eick, S., Ramseier, C. A., Arweiler, N. B., Sculean, A., & Salvi, G. E. (2014). Anti-infective therapy of peri-implantitis with adjunctive local drug delivery or photodynamic therapy: 12-month outcomes of a randomized controlled clinical trial. *Clinical Oral Implants Research*, *25*(3), 279–287. https://doi.org/10.1111/clr.12155

Berglundh, T., Armitage, G., Araujo, M. G., Avila-Ortiz, G., Blanco, J., Camargo, P. M., Chen, S., Cochran, D., Derks, J., Figuero, E., Hämmerle, C. H. F., Heitz-Mayfield, L. J. A., Huynh-Ba, G., Iacono, V., Koo, K. T., Lambert, F., McCauley, L., Quirynen, M., Renvert, S., … Zitzmann, N. (2018). Peri-implant diseases and conditions: Consensus report of workgroup 4 of the 2017 world workshop on the classification of periodontal and Peri-implant diseases and conditions. *Journal of Periodontology*, *89*(Suppl 1), S313–S318. https://doi.org/10.1002/JPER.17-0739

Berglundh, T., Lindhe, J., Marinello, C., Ericsson, I., & Liljenberg, B. (1992). Soft tissue reaction to de novo plaque formation on implants and teeth. An experimental study in the dog. *Clinical Oral Implants Research*, *3*(1), 1–8. https://doi.org/10.1034/j.1600-0501.1992.030101.x

Costa, F. O., Ferreira, S. D., Cortelli, J. R., Lima, R. P. E., Cortelli, S. C., & Cota, L. O. M. (2019). Microbiological profile associated with peri-implant diseases in individuals with and without preventive maintenance therapy: A 5-year follow-up. *Clinical Oral Investigations*, *23*(8), 3161–3171. https://doi.org/10.1007/s00784-018-2737-y

Derks, J., Schaller, D., Hakansson, J., Wennstrom, J. L., Tomasi, C., & Berglundh, T. (2016). Effectiveness of implant therapy analyzed in a Swedish population: Prevalence of peri-implantitis. *Journal of Dental Research*, *95*(1), 43–49. https://doi.org/10.1177/0022034515608832

Derks, J., & Tomasi, C. (2015). Peri-implant health and disease. A systematic review of current epidemiology. *Journal of Clinical Periodontology*, *42*(Suppl 16), S158–S171. https://doi.org/10.1111/jcpe.12334

Fransson, C., Tomasi, C., Pikner, S. S., Gröndahl, K., Wennström, J. L., Leyland, A. H., & Berglundh, T. (2010). Severity and pattern of peri-implantitis-associated bone loss. *Journal of Clinical Periodontology*, *37*(5), 442–448. https://doi.org/10.1111/j.1600-051X.2010.01537.x

Insua, A., Monje, A., Wang, H. L., & Inglehart, M. (2017). Patient-centered perspectives and understanding of peri-implantitis. *Journal of Periodontology*, *88*(11), 1153–1162. https://doi.org/10.1902/jop.2017.160796

Karring, E. S., Stavropoulos, A., Ellegaard, B., & Karring, T. (2005). Treatment of peri-implantitis by the vector system. *Clinical Oral Implants Research*, *16*(3), 288–293. https://doi.org/10.1111/j.1600-0501.2005.01141.x

Khan, S. N., Koldsland, O. C., Roos-Jansåker, A. M., Wohlfahrt, J. C., Verket, A., Mdala, I., Magnusson, A., Salvesen, E., & Hjortsjö, C. (2022). Non-surgical treatment of mild to moderate peri-implantitis using an oscillating chitosan brush or a titanium curette - a randomized multicentre controlled clinical trial. *Clinical Oral Implants Research*, *33*, 1254–1264. https://doi.org/10.1111/clr.14007

Koldsland, O. C., & Aass, A. M. (2020). Supportive treatment following peri-implantitis surgery: An RCT using titanium curettes or chitosan brushes. *Journal of Clinical Periodontology*, *47*(10), 1259–1267. https://doi.org/10.1111/jcpe.13357

Koldsland, O. C., Wohlfahrt, J. C., & Aass, A. M. (2018). Surgical treatment of peri-implantitis: Prognostic indicators of short-term results. *Journal of Clinical Periodontology*, *45*(1), 100–113. https://doi.org/10.1111/jcpe.12816

Machtei, E. E., Romanos, G., Kang, P., Travan, S., Schmidt, S., Papathanasiou, E., Tatarakis, N., Tandlich, M., Liberman, L. H., Horwitz, J., Bassir, S. H., Myneni, S., Shiau, H. J., Shapira, L., Donos, N., Papas, A., Meyle, J., Giannobile, W. V., Papapanou, P. N., & Kim, D. M. (2021). Repeated delivery of chlorhexidine chips for the treatment of peri-implantitis: A multicenter, randomized, comparative

clinical trial. *Journal of Periodontology*, *92*(1), 11–20. https://doi.org/10.1002/jper.20-0353

Mdala, I., Olsen, I., Haffajee, A. D., Socransky, S. S., Thoresen, M., & de Blasio, B. F. (2014). Comparing clinical attachment level and pocket depth for predicting periodontal disease progression in healthy sites of patients with chronic periodontitis using multi-state Markov models. *Journal of Clinical Periodontology*, *41*(9), 837–845. https://doi.org/10.1111/jcpe.12278

Mombelli, A., van Oosten, M. A., Schurch, E., Jr., & Land, N. P. (1987). The microbiota associated with successful or failing osseointegrated titanium implants. *Oral Microbiology and Immunology*, *2*(4), 145–151. https://doi.org/10.1111/j.1399-302x.1987.tb00298.x

Montero, E., Roccuzzo, A., Molina, A., Monje, A., Herrera, D., & Roccuzzo, M. (2022). Minimal invasiveness in the reconstructive treatment of peri-implantitis defects. *Periodontology 2000*, 1–18. https://doi.org/10.1111/prd.12460

Newbrun, E. (1996). Indices to measure gingival bleeding. *Journal of Periodontology*, *67*(6), 555–561. https://doi.org/10.1902/jop.1996.67.6.555

O'Leary, T. J., Drake, R. B., & Naylor, J. E. (1972). The plaque control record. *Journal of Periodontology*, *43*(1), 38. https://doi.org/10.1902/jop.1972.43.1.38

Polyzois, I. (2019). Treatment planning for periimplant mucositis and periimplantitis. *Implant Dentistry*, *28*(2), 150–154. https://doi.org/10.1097/ID.0000000000000869

Pontoriero, R., Tonelli, M. P., Carnevale, G., Mombelli, A., Nyman, S. R., & Lang, N. P. (1994). Experimentally induced peri-implant mucositis. A clinical study in humans. *Clinical Oral Implants Research*, *5*(4), 254–259. https://doi.org/10.1034/j.1600-0501.1994.050409.x

Preus, H. R., Torgersen, G. R., Koldsland, O. C., Hansen, B. F., Aass, A. M., Larheim, T. A., & Sandvik, L. (2015). A new digital tool for radiographic bone level measurements in longitudinal studies. *BMC Oral Health*, *15*, 107. https://doi.org/10.1186/s12903-015-0092-9

Renvert, S., Hirooka, H., Polyzois, I., Kelekis-Cholakis, A., Wang, H. L., & Working, G. (2019). Diagnosis and non-surgical treatment of peri-implant diseases and maintenance care of patients with dental implants - consensus report of working group 3. *International Dental Journal*, *69*(Suppl 2), 12–17. https://doi.org/10.1111/idj.12490

Renvert, S., Samuelsson, E., Lindahl, C., & Persson, G. R. (2009). Mechanical non-surgical treatment of peri-implantitis: A double-blind randomized longitudinal clinical study. I: Clinical results. *Journal of Clinical Periodontology*, *36*(7), 604–609. https://doi.org/10.1111/j.1600-051X.2009.01421.x

Riben-Grundstrom, C., Norderyd, O., Andre, U., & Renvert, S. (2015). Treatment of peri-implant mucositis using a glycine powder air-polishing or ultrasonic device: A randomized clinical trial. *Journal of Clinical Periodontology*, *42*(5), 462–469. https://doi.org/10.1111/jcpe.12395

Roccuzzo, A., De Ry, S. P., Sculean, A., Roccuzzo, M., & Salvi, G. E. (2020). Current approaches for the non-surgical management of peri-implant diseases. *Current Oral Health Reports*, *7*(3), 274–282. https://doi.org/10.1007/s40496-020-00279-x

Romandini, M., Lima, C., Pedrinaci, I., Araoz, A., Soldini, M. C., & Sanz, M. (2021). Prevalence and risk/protective indicators of peri-implant diseases: A university-representative cross-sectional study. *Clinical Oral Implants Research*, *32*(1), 112–122. https://doi.org/10.1111/clr.13684

Roos-Jansaker, A. M., Lindahl, C., Renvert, H., & Renvert, S. (2006). Nine- to fourteen-year follow-up of implant treatment. Part I: Implant loss and associations to various factors. *Journal of Clinical Periodontology*, *33*(4), 283–289. https://doi.org/10.1111/j.1600-051X.2006.00907.x

Roos-Jansåker, A. M., Renvert, H., Lindahl, C., & Renvert, S. (2007). Surgical treatment of peri-implantitis using a bone substitute with or without a resorbable membrane: A prospective cohort study. *Journal of Clinical Periodontology*, *34*(7), 625–632. https://doi.org/10.1111/j.1600-051X.2007.01102.x

Sahm, N., Becker, J., Santel, T., & Schwarz, F. (2011). Non-surgical treatment of peri-implantitis using an air-abrasive device or mechanical debridement and local application of chlorhexidine: A prospective, randomized, controlled clinical study. *Journal of Clinical Periodontology*, *38*(9), 872–878. https://doi.org/10.1111/j.1600-051X.2011.01762.x

Sanz, M., & Chapple, I. L. (2012). Clinical research on peri-implant diseases: Consensus report of Working group 4. *Journal of Clinical Periodontology*, *39*(Suppl 12), 202–206. https://doi.org/10.1111/j.1600-051X.2011.01837.x

Schulz, K. F., Altman, D. G., Moher, D., & CONSORT Group. (2010). CONSORT 2010 statement: Updated guidelines for reporting parallel group randomised trials. *BMJ*, *340*, c332. https://doi.org/10.1136/bmj.c332

Schwarz, F., Becker, K., & Renvert, S. (2015). Efficacy of air polishing for the non-surgical treatment of peri-implant diseases: A systematic review. *Journal of Clinical Periodontology*, *42*(10), 951–959. https://doi.org/10.1111/jcpe.12454

Sedgwick, P., & Greenwood, N. (2015). Understanding the Hawthorne Effect. *BMJ*, *351*, h4672. https://doi.org/10.1136/bmj.h4672

Serino, G., & Ström, C. (2009). Peri-implantitis in partially edentulous patients: Association with inadequate plaque control. *Clinical Oral Implants Research*, *20*(2), 169–174. https://doi.org/10.1111/j.1600-0501.2008.01627.x

Serino, G., & Turri, A. (2011). Outcome of surgical treatment of peri-implantitis: Results from a 2-year prospective clinical study in humans. *Clinical Oral Implants Research*, *22*(11), 1214–1220. https://doi.org/10.1111/j.1600-0501.2010.02098.x

Wohlfahrt, J. C., Evensen, B. J., Zeza, B., Jansson, H., Pilloni, A., Roos-Jansaker, A. M., Di Tanna, G. L., Aass, A. M., Klepp, M., & Koldsland, O. C. (2017). A novel non-surgical method for mild peri-implantitis- a multicenter consecutive case series. *International Journal of Implant Dentistry*, *3*(1), 38. https://doi.org/10.1186/s40729-017-0098-y

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Khan, S. N., Koldsland, O. C., Roos-Jansåker, A.-M., Wohlfahrt, J. C., Verket, A., Mdala, I., Magnusson, A., Salvesen, E., & Hjortsjö, C. (2023). Non-surgical treatment of mild to moderate peri-implantitis with an oscillating chitosan brush or a titanium curette—12-month follow-up of a multicenter randomized clinical trial. *Clinical Oral Implants Research*, *34*, 684–697. https://doi.org/10.1111/clr.14078