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Peri-implant infections

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**IMPLANT DECONTAMINATION
WITH 2% CHLORHEXIDINE DURING
SURGICAL PERI-IMPLANTITIS
TREATMENT:
A RANDOMIZED, DOUBLE-BLIND,
CONTROLLED TRIAL**



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ABSTRACT

Objectives

The objective of this randomized, double-blind, controlled trial was to evaluate the clinical, radiographic and microbiological effects of implant surface decontamination with a 2% chlorhexidine (CHX) solution in comparison to a 0.12% chlorhexidine + 0.05% cetylpyridinium chloride (CPC) solution during resective surgical peri-implantitis treatment.

Material and methods

Forty-four patients (108 implants) with peri-implantitis were treated with resective surgical treatment consisting of bone re-contouring, surface debridement and chemical decontamination and apically repositioned flap. Patients were randomly allocated to decontamination with a 2% CHX solution (test group) or 0.12% CHX + 0.05% CPC (control group). Clinical and radiographic parameters were recorded before treatment (baseline), and at 3, 6 and 12 months after treatment. Microbiological parameters were recorded during surgery.

Results

Multilevel analysis showed no significant differences in bleeding, suppuration, probing pocket depth and radiographic bone loss between control and test group over three follow-up measurements (3, 6 and 12 months) from baseline. Both decontamination procedures resulted in significant reductions of anaerobic bacterial counts on the implant surface, but no significant difference was noted between control and test group (mean log 3.37 ± 2.34 versus 3.65 ± 2.87 , $p = 0.99$).

Conclusions

The use of a 2% CHX solution for implant surface decontamination during resective peri-implantitis therapy does not lead to improved clinical, radiographic or microbiological results compared to a 0.12% CHX + 0.05% CPC solution. Overall, the additional use of CHX reduces anaerobic bacterial load on the implant surface better than mechanical debridement alone, but does not seem to enhance clinical treatment outcomes.

(ClinicalTrials.gov number NCT01852253).

INTRODUCTION

Non-surgical approaches for the treatment of peri-implantitis have shown to be unpredictable and, in most cases, not effective (Lindhe et al. 2008). If non-surgical therapy does not resolve the inflammatory lesion it is recommended to perform access surgery, thus facilitating proper granulation tissue removal and debridement and decontamination of the implant surface. The removal of the biofilm on the implant surface is compromised by the screw-shaped design of the implant and the rough implant surface. Since mechanical debridement alone may not be sufficient to achieve complete biofilm removal additional measures, such as laser therapy, use of antibiotics and/or use of antiseptics such as hydrogen peroxide or chlorhexidine, have been proposed (for review see: Esposito et al. 2012).

A recent clinical trial has revealed that implant surface debridement and decontamination with 0.12% chlorhexidine (CHX) + 0.05% cetylpyridinium chloride (CPC) during resective surgical treatment of peri-implantitis results in a greater immediate suppression of anaerobic bacteria on the implant surface than a placebo solution (De Waal et al. 2013). However, this microbiological effect did not translate to improved clinical results. Possible explanations for this observation could be that 1) the clinical success of surgical peri-implantitis treatment is determined by factors other than the method of surface debridement and decontamination (Schwarz et al. 2011), 2) CHX has bactericidal capacity but does not actually remove biofilm from the implant surface (Chin et al. 2007) or 3) the 0.12% CHX concentration is too low to sufficiently reduce the number of colony forming units on the implant surface and consequently to exert a clinical effect.

It is known that the efficacy of CHX mouthrinse formulations is dependent on the dose of CHX being delivered, amongst other factors such as rinsing time and rinsing frequency (Cumming & Loe 1973, Lang & Ramseier-Grossmann 1981, Jenkins et al. 1994). Increasing the CHX dose reduces plaque formation and prevents gingival inflammation. However, increasing the CHX dose will also increase the probability and extent of possible side effects such as tooth staining, taste disturbance and mucosal erosion (Flötra et al. 1971). These side effects prevent the long-term use of CHX at high doses, but may be less of an issue when CHX is locally applied once for implant decontamination during surgical peri-implantitis therapy. Yet, CHX may have deleterious effects on human eukaryotic cells. This potential cell toxicity of CHX has been shown in several *in vitro* studies (Pucher & Daniel 1992, Cline & Layman 1992, Mariotti & Rumpf 1999). CHX-induced cell damage *in vitro* appears to be concentration and time dependent, notable in different cell types (fibroblasts, endothelial cells and osteoblasts) and manifest at concentrations far below those used in clinical practice (Giannelli et al. 2008). From *in vitro* research it has therefore been suggested that the direct application of CHX during surgical treatment of periodontal and peri-implant diseases might negatively interfere with the early healing phase of these diseases. Clinically, however, the application of CHX during oral surgical procedures does not seem to interfere with wound healing (Yengopal & Mickenautsch 2012, De Waal et al. 2013). It has even been shown that the application of CHX gel directly within the alveolus following extraction of impacted third molars may reduce the incidence of

alveolar osteitis, without increasing the occurrence of adverse events (Torres-Lagares et al. 2006). This holds true even if concentrations up to 1% CHX are being used (Rodríguez-Pérez et al. 2013).

Based on the dose-dependent effect of CHX and its good clinical acceptance, it may be hypothesized that increasing the dose of CHX delivered to the implant surface during resective surgical peri-implantitis treatment, will increase the decontaminating effect, which consequently leads to improved clinical treatment results. Therefore, the objective of the present study was to evaluate the clinical, radiographic and microbiological effects of implant surface decontamination with a 2% CHX solution in comparison to a 0.12% CHX + 0.05% CPC solution during resective surgical peri-implantitis treatment. The null-hypothesis of no differences in clinical and microbiological results between both treatment procedures was tested.

MATERIAL AND METHODS

The study protocol was essentially adopted from a previous study (De Waal et al. 2013). In brief, the material and methods are described below.

Participants

Participants were consecutively recruited from patients referred for treatment of peri-implantitis to the University Medical Center Groningen, the Netherlands. Written informed consent was obtained from all participants before entering the trial. Inclusion- and exclusion criteria are depicted in Figure 1. Peri-implantitis was defined as bleeding and/or suppuration on probing combined with a peri-implant probing pocket depth (PPD) ≥ 5 mm and bone loss ≥ 2 mm.

The study took place between September 2010 and April 2013, has been conducted in full accordance with the World Medical Association Declaration of Helsinki (version 2008) and was approved by the Institutional Review Board of the University Medical Center Groningen, the Netherlands (METc2010.028). US National Institutes of Health clinical trial registration was done at www.ClinicalTrials.gov (NCT01852253). The CONSORT guidelines for reporting a clinical trial were followed (Schulz et al. 2010, Moher et al. 2010, Cairo et al. 2012).

Trial design

The study is a randomized, double-blind, controlled trial evaluating the clinical, radiographic and microbiological outcomes of resective surgical treatment of peri-implantitis combined with decontamination of the implant surface using a 2% CHX solution (test group) or a 0.12% CHX + 0.05% CPC solution (control group). Follow-up time was 12 months. Patients were randomly assigned to either the test or control group using a one-to-one allocation ratio.

Randomisation

Twenty-two notes with the words 'solution A' and 22 notes with 'solution B' were put

Figure 1. Inclusion- and exclusion criteria

Inclusion criteria	<ul style="list-style-type: none">• Presence of ≥ 1 endosseous dental implant with clinical and radiographic signs of peri-implantitis (peri-implantitis defined as: bleeding and/or suppuration on probing, peri-implant probing pocket depth (PPD) ≥ 5 mm and bone loss ≥ 2 mm);• Implant function time ≥ 2 years.
Exclusion criteria	<ul style="list-style-type: none">• Medical and general contra-indications for the surgical procedures;• A history of radiotherapy to the head and neck region;• Pregnancy and lactation;• Insuline-dependent diabetes;• Use of antibiotics during the last 3 months;• Incapability to perform basal oral hygiene measures due to physical or mental disorders;• Active, uncontrolled periodontal infections of the natural dentition (PPD > 5 mm);• Implants with bone loss exceeding 2/3 of the length of the implant or implants with bone loss beyond any transverse openings in hollow implants;• Implant mobility;• Implants at which no position could be identified where proper probing measurements could be performed;• Previous surgical treatment of the peri-implantitis lesions.

into 44 identical, sequentially numbered, non-transparent envelopes according to a computer generated randomization list with a permuted block design (fixed block sizes of 4). No stratification was performed. All envelopes were irreversibly sealed, only to be opened by the surgical assistant during the surgical procedure. According to the information on the note, the surgical assistant prepared a transparent syringe with either solution A or solution B. The syringe was covered with a non-transparent sleeve, thus hiding the appearance of the solution, *i.e.* colour, from the surgeon. There were slight colour differences between the solutions, which were not visible during rinsing of the implant surface because the solution was continuously dispersed in small volumes with a 22-gauge needle. Except for colour, the test solution was otherwise similar to the control solution (no noticeable differences in taste, smell and viscosity) ensuring blinding of the surgeon, the patient and the investigator to treatment allocation.

Intervention

Before the surgical procedure, all patients received extensive oral hygiene instructions and mechanical debridement of implants, suprastructures and remaining dentition. Patients were all surgically treated by one experienced oral- and maxillofacial surgeon (G.R). Suprastructures were removed if necessary and reasonably possible (in all but sixteen patients). Incisions were made using a surgical blade (no. 15) under local anaesthesia. Flaps were designed to allow optimal access to the peri-implant bone defect for granulation tissue removal and debridement and decontamination of the implant surface. Vertical releasing incisions extending into the alveolar mucosa were placed at the mesial and distal aspects of the horizontal incision.

Full thickness mucoperiosteal flaps were raised buccally and lingually. Granulation tissue was removed using curettes (Gracey, Hu-Friedy®, Chicago, IL, USA). Bone recon-

touring, aimed at eliminating angular bony defects, was performed using a rotating round bur under saline irrigation. The implant surface was mechanically cleaned using surgical gauzes soaked in saline. After mechanical debridement, patients were randomly allocated to either the control or the test group. After treatment allocation, the implant surface was rinsed for 1 minute with a 2% CHX solution (alcohol-based) (test group) or with 0.12% CHX + 0.05% CPC without alcohol (PerioAid, Dentaaid SL, Cerdanyola, Spain) (control group). Test and control solutions were prepared by Dentaaid SL (Cerdanyola, Spain). Care was taken to continuously cover the implant surface with the solution. Subsequently, the implant surface was rinsed with abundant amounts of sterile saline for 1 minute. Suprastructures were repositioned and mucosal flaps were apically positioned and firmly sutured (Vicryl Plus®, Ethicon Inc., Somerville, NJ, USA). For both test and control group, surgery was followed by two weeks of mouth rinsing with 0.12% CHX + 0.05% CPC without alcohol (PerioAid, Dentaaid SL, Cerdanyola, Spain) two times daily during 30 seconds. Sutures were removed after 2 weeks. During follow-up examinations patients were re-instructed in oral hygiene measures and implants and teeth were cleaned as necessary. Follow-up visits were scheduled after 3 (T_3), 6 (T_6) and 12 (T_{12}) months.

Outcomes

Clinical and radiographical outcomes

The primary outcome variable was percentage of sites with bleeding on probing (% sites BoP). Secondary clinical outcome variables were presence of plaque (% sites plaque), suppuration on probing (% sites SoP), mean probing pocket depth (PPD) and mean radiographic marginal bone loss. Measurements were performed before treatment (baseline, T_0) and at 3, 6 and 12 months after surgery (T_3 , T_6 and T_{12}) by one and the same experienced examiner (Y.W.) who was blind to treatment allocation. Presence of plaque was assessed (present/absent) at four sites per implant (mesial, buccal, distal and lingual) by running a probe across the marginal surface of the implant/suprastructure. Peri-implant pocket probing was performed at 4 sites per implant using a pressure sensitive probe (probe force of 0.25 N, KerrHawe Click Probe®, Bioggo, Switzerland). PPD was scored to the nearest millimeter. Up to 30 seconds after pocket probing the presence or absence of bleeding and suppuration were assessed. Intra-oral radiographs were obtained using an aiming device and the long cone paralleling technique. Care was taken to position the film parallel to the long axis of the implant. Due to anatomical restrictions, in 16 fully edentulous patients (31 implants) no intra-oral radiographs could be obtained from all implants without pain or major distortion of the image. In these patients, orthopantomograms were taken. All radiographs were digital. Measurements were performed using Adobe Photoshop (version 10.0.1, Adobe Systems Incorporated, San Jose, CA, USA). The radiographs were calibrated using the known dimensions of the implant as reference values. A horizontal line was drawn through the shoulder of the implant and the distance from this line to the first bone-to-implant contact was measured at the mesial and distal site. Bone loss was assessed with regard to the position at which the bone is normally positioned, taking into account the different implant types and brands. Intra-examiner reproducibility of probing pocket depth measurements and radiographic examinations was determined

in a previous study ($\kappa = 0.82$ for probing pocket depth measurements; $\kappa = 0.99$ and $\kappa = 0.96$ for measurements on intra-oral radiographs and orthopantomograms respectively) and judged as very good (De Waal et al. 2013).

Microbiological outcomes

Secondary microbiological outcome variable was the difference in anaerobic bacterial load of the implant surface before and after mechanical and chemical debridement and decontamination. After flap deflection and granulation tissue removal a sample was obtained from the implant surface by rubbing a sterilized brush (Microbrush® International, Grafton, WI, USA) across the implant surface (T_{pre}). A second sample was obtained after mechanical debridement, rinsing of the implant surface with the test or control solution and subsequent rinsing with sterile saline (T_{post}). The top part of the brush was cut off and collected in a vial containing reduced transport fluid (RTF) (Syed & Loesche 1972). Separate samples were obtained from every implant presenting peri-implantitis. The vast majority of the microbiological samples were processed within 4 hours after sampling. Some samples were kept at 4°C (Van Steenberg et al. 1993) and processed within 24 hours. Samples were processed as described by Van Winkelhoff et al. (1985) and Van Steenberg et al. (1986). Total anaerobic bacterial load and presence and numbers of the putative periodontal pathogens *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, *Prevotella intermedia*, *Tannerella forsythia*, *Fusobacterium nucleatum*, *Parvimonas micra* and *Campylobacter rectus* were determined by laboratory technicians who were blind to treatment allocation.

Side effects

Any abnormalities in wound healing were recorded two weeks after the surgical procedure (during appointment for suture removal). Furthermore, the number of days patients had been using analgesics after the surgical procedure was scored.

Statistical methods

Sample size calculation

Sample size was determined by multilevel power analysis using MLPowSim version 1.0 (Centre for Multilevel Modeling, University of Bristol, Bristol, United Kingdom). Data from a previous study evaluating the clinical effect of implant surface decontamination with a 0.12% CHX solution versus implant surface decontamination with a placebo solution during resective peri-implantitis treatment were used to estimate effect size (De Waal et al. 2013). The effect (reduction in % of sites BoP) of rinsing the implant surface with a 2% CHX solution versus rinsing with a 0.12% CHX solution was expected to be at least twice the effect of rinsing the implant surface with a 0.12% CHX solution versus rinsing with a placebo solution (15% reduction versus 7.5% reduction). Sample size estimates showed that 40 patients with an average of 3 implants per patient and 3 observations (T_3 , T_6 and T_{12}) per implant would give a power (β) of more than 80% with a significance level (α) of 0.05 (two-sided test). To compensate for patient withdrawal and losses to follow-up a sample size of 44 patients was chosen (22 per group).

Statistical analysis

Multilevel modeling was used to determine the effect of the intervention over time (test group versus control group) on the primary outcome variable and the secondary clinical and radiographic outcome variables. A multilevel hierarchical three-level structure was chosen with three levels of analysis being timing of follow-up measurements (level 1), implant (level 2) and patient (level 3). Baseline values of BoP, SoP, PPD and marginal bone loss (continuous variables), smoking, dental status and history of periodontitis (dichotomous variables) and implant surface roughness (categorical variable) were a priori identified as potential confounders. With the crude analysis the effect of the intervention over time was determined, while controlling for baseline value and time. In the adjusted analysis the potential confounders smoking, dental status, history of periodontitis and implant surface roughness were additionally included in the model.

For analysis of the secondary microbiological outcome variable linear regression analysis was performed. Total anaerobic bacterial load at baseline (T_{pre}) was distributed normally after logarithmic transformation. The implant was taken as the statistical unit. Baseline values and implant surface roughness were included in the regression model. For comparison of within-group differences in detection frequency of single bacterial species between T_{pre} and T_{post} the McNemar's test was used. Between-group differences at T_{post} were analyzed using the Fisher's exact test.

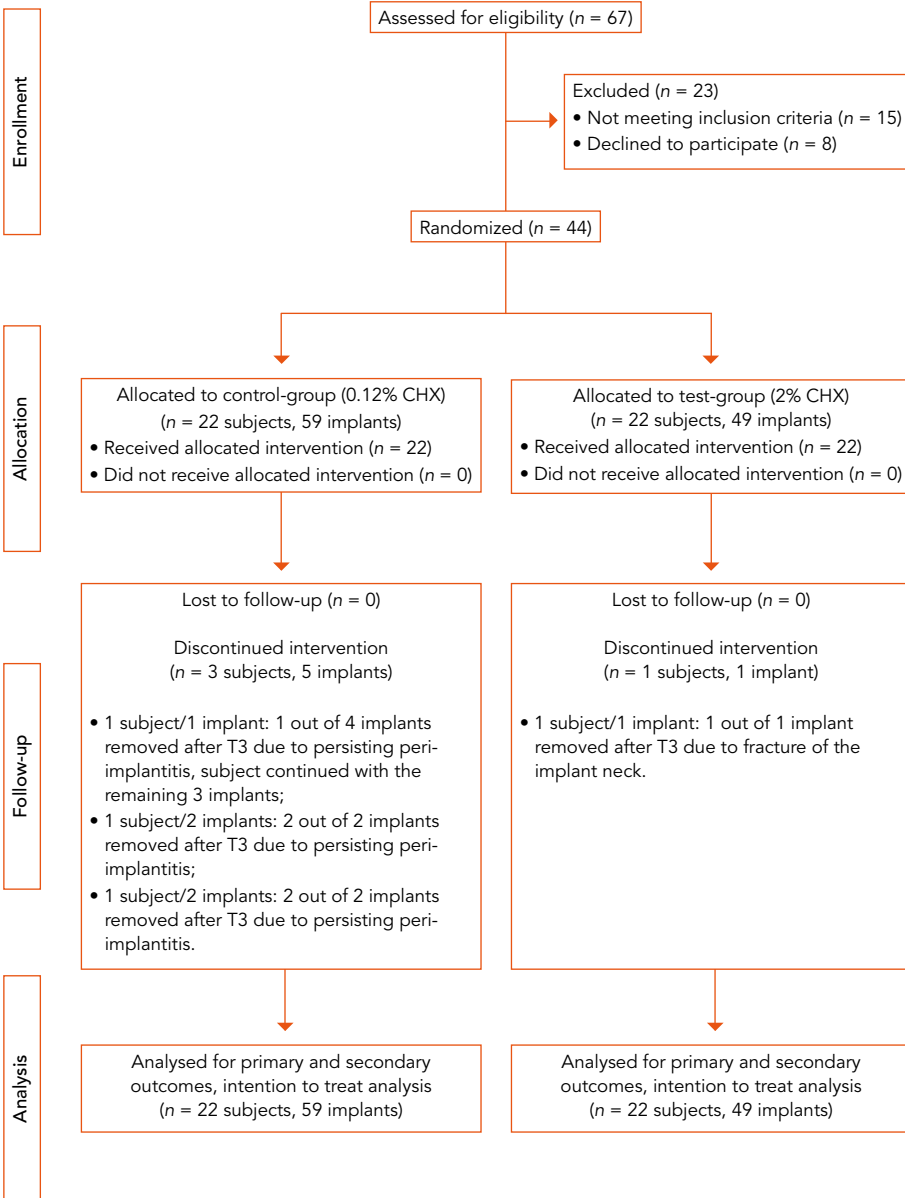
Multilevel models were analyzed using MLwiN version 2.12 (Centre for Multilevel Modeling, University of Bristol, Bristol, UK). Descriptive data and data regarding the secondary microbiological outcome variable were analyzed using IBM® SPSS® Statistics 20 (version 20.0.0.1, IMB, Armonk, NY, USA).

RESULTS

The flow of the participants throughout the different phases of the study is depicted in Figure 2. Baseline demographic patient and implant characteristics are reported in Table 1. A Total of 44 patients were included presenting 37 implants with healthy peri-implant conditions, 41 implants with peri-implant mucositis and 115 implants with peri-implantitis. Seven of the implants presenting peri-implantitis were excluded, six due to implant mobility (explantation sole treatment option) and one due to the impossibility of performing probing pocket depth measurements, leaving a total of 108 implants appropriate for inclusion.

Five implants in 3 patients from the control group had to be removed between T_3 and T_6 due to severe persisting peri-implantitis. Two patients lost both their implants (two Pitt-easy implants with Vacuum-TPS surface; two Nobel Biocare implants with Ti-Unite surface) and discontinued the study. The other patient lost 1 out of 4 treated implants (Straumann, SLA surface), but continued the study with the remaining 3 implants. One implant in the test group was removed between T_3 and T_6 due to fracture of the implant neck (Nobel Biocare, Ti-Unite surface). The patients from both groups attended all follow-up visits and no patient was lost to follow-up.

Figure 2. Flow-diagram



Clinical and radiographical outcomes

Descriptive statistics of clinical and radiographic outcomes at baseline and different follow-up visits are reported in Table 2. Results of the multilevel modeling analysis are depicted in Table 3. No significant differences in BoP, SoP, PPD and radiographic marginal bone loss were detected between control and test group over three follow-up measurements (3, 6 and 12 months) from baseline, neither in the 'crude' nor in the 'adjusted' analysis.

Microbiological outcomes

Seventy-eight out of the 108 sampled implants appeared culture positive after flap deflection and granulation tissue removal. Log-transformed mean bacterial anaerobic counts of these culture positive implants for the control and test group before and after debridement and decontamination of the implant surface are depicted in Table 4. In both groups the debridement and decontamination procedure resulted in a significant reduction of counts of anaerobic bacteria on the implant surface. However, no differences were present between control and test group (mean log 3.37 ± 2.34 and 3.65 ± 2.87 respectively, $p = 0.99$).

A. actinomycetemcomitans could not be detected on any implant surface (Table 5). The debridement and decontamination procedure resulted in reduction below detection level of *P. gingivalis*, *P. intermedia* (control group only), *T. forsythia* and *C. rectus* and significant reductions in detection frequencies of *F. nucleatum* and *P. micra*. No differences were observed between control and test group.

Side effects

No abnormalities were seen in wound healing in both groups and no differences were present in the number of days patients had been taking analgesics (3.77 ± 3.78 and 4.68 ± 4.80 in control and test group respectively, $p = 0.80$).

DISCUSSION

The present study shows that implant surface debridement and decontamination using a 2% CHX solution during resective surgical peri-implantitis treatment leads to similar clinical, radiographic and microbiological results compared to using a 0.12% CHX + 0.05% CPC solution. Thus, the null-hypothesis of no difference has to be accepted.

In the present study, the 0.12% CHX + 0.05% CPC solution was chosen as reference (control group) against which the effect of the 2% CHX solution was compared. This was done because a previous study had shown a positive effect of the 0.12% CHX solution over a placebo solution (saline) in suppressing anaerobic bacteria on the implant surface. Because the study protocol of the present study was nearly identical to the previous study, a post hoc analysis was carried out comparing the 2% CHX group from the present study with the placebo group from the previous study. These analyses revealed a marginally positive microbiological effect of the 2% CHX solution over the placebo solution ($p = 0.073$). However, no clinical effects, neither positive

Table 1. Baseline demographic characteristics of included subjects/implants

Characteristics	Control	Test
Number of patients	22	22
Age (years; mean (SD))	60.5 (11.6)	58.6 (10.2)
Gender; M (male), F (female)	M 8, F 14	M 5, F 17
Smoking; n subjects (%)		
never (or quit smoking before implant placement)	11 (50.0)	14 (63.6)
former (quit smoking after implant placement)	5 (22.7)	1 (4.5)
current	6 (27.3)	7 (31.8)
History of periodontitis; n subjects (%)	10 (45.5)	10 (45.5)
Dental status; n subjects (%)		
fully edentulous	12 (54.5)*	9 (40.9)**
partially edentulous	10 (45.5)	13 (59.1)
<i>Remaining dentition in partially edentulous subjects:</i>		
Number of teeth; mean (SD)	19.8 (6.1)	18.2 (8.7)
Number of teeth; range	8-28	2-28
Full-mouth plaque score; % (SD)	3.2 (3.8)	10.0 (10.8)
Full-mouth bleeding score; % (SD)	12.8 (12.6)	15.3 (15.2)
Full-mouth mean probing pocket depth; mm (SD)	2.1 (0.4)	2.1 (0.4)
<i>All implants:</i>		
Total number of implants (range)	113 (2-12)	80 (1-8)
Clinical diagnosis; n implants (%)		
Health (no BoP or suppuration);	30 (26.5)	7 (8.8)
Peri-implant mucositis (BoP/suppuration, but no bone loss);	23 (20.4)	18 (22.5)
Peri-implantitis;		
allocated to treatment	59 (52.2)	49 (61.3)
explantation only possible treatment option	1 (0.9)	5 (6.3)
no probing pocket depth measurements possible	0 (0)	1 (1.3)
<i>Allocated implants:</i>		
Total number of allocated implants (range)	59 (1-9)	49 (1-6)
Implant function time (years; mean (SD))	6.8 (3.9)	7.3 (5.1)
Implant surface; n implants (%)		
Nobel Biocare		
machined surface	5 (8.5)	0 (0)
porous anodized surface, TiUnite	18 (30.5)	22 (44.9)
Straumann		
titanium plasma-sprayed, TPS	0 (0)	4 (8.2)
sandblasted large grit acid-etched, SLA	15 (25.4)	9 (18.4)
sandblasted large grit acid-etched, SLActive	3 (5.1)	1 (2.0)
Astra Tech		
fluoride-modified titanium dioxide grit-blasted, Osseospeed	6 (10.2)	6 (12.2)
IMZ		
titanium plasma-sprayed, frios TPS	4 (6.8)	6 (12.2)
Pitt-easy		
vacuum titanium plasma-sprayed, V-TPS	2 (3.4)	1 (2.0)
Camlog		
abrasive-blasted acid-etched, Promote	4 (6.8)	0 (0)
Dentsply Friadent		
grit-blasted acid-etched, Friadent plus	2 (3.4)	0 (0)
Type of restoration; n implants involved (%)		
single crown	6 (10.2)	12 (24.5)
implant supported fixed partial denture	6 (10.2)	5 (10.2)
implant-teeth supported fixed partial denture	0 (0)	2 (4.1)
fixed full denture	1 (1.7)	0 (0)
overdenture	46 (78.0)	30 (61.2)
Screw or cement retained restoration; n implants involved (%)		
screw-retained	42 (71.2)	45 (91.8)
cement-retained	17 (28.8)	4 (8.2)
Implants placed in maxilla or mandible; n implants (%)		
maxilla	31 (52.5)	27 (55.1)
mandible	28 (47.5)	22 (44.9)

No significant differences were present between both groups at baseline; *3 were partially edentulous at the time of implant placement; **2 were partially edentulous at the time of implant placement.

Table 2. Descriptive statistics of clinical and radiographic parameters

	Control				Test			
	T ₀ (n = 59)	T ₃ (n = 59)	T ₆ (n = 54)	T ₁₂ (n = 54)	T ₀ (n = 49)	T ₃ (n = 49)	T ₆ (n = 48)	T ₁₂ (n = 48)
plaque								
% of sites (SD)	25.0 (30.8)	19.9 (28.5)	26.9 (34.3)	19.9 (29.7)	21.9 (33.7)	12.8 (25.1)	7.3 (14.5)	15.1 (26.2)
% of implants (n)	47.5 (28)	44.1 (26)	46.3 (25)	37.0 (20)	36.7 (18)	24.5 (12)	22.9 (11)	31.2 (15)
BoP								
% of sites (SD)	74.2 (27.8)	54.2 (30.8)	37.0 (36.9)	37.0 (35.3)	82.1 (23.9)	42.6 (31.0)	32.3 (31.4)	42.7 (34.2)
% of implants (n)	94.9 (56)	88.1 (52)	63.0 (34)	68.5 (37)	98.0 (47)	77.6 (38)	70.8 (34)	77.1 (37)
SoP								
% of sites (SD)	20.3 (25.6)	3.0 (14.0)	2.3 (8.8)	0.5 (3.4)	21.9 (24.8)	0.0 (0.0)	1.6 (8.0)	2.5 (7.7)
% of implants (n)	49.2 (29)	6.8 (4)	7.4 (4)	1.9 (1)	57.1 (28)	0.0 (0)	4.2 (2)	10.4 (5)
mean PPD	5.0 (1.2)	3.2 (1.3)	2.9 (0.8)	2.9 (0.7)	4.7 (1.0)	3.0 (0.6)	2.8 (0.6)	3.0 (0.7)
PPD ≥ 5 mm								
% of sites (SD)	60.2 (28.3)	9.7 (24.6)	7.9 (22.7)	5.6 (12.5)	57.7 (26.6)	3.1 (9.7)	3.6 (11.5)	7.3 (12.6)
% of implants (n)	100 (59)	18.6 (11)	14.8 (8)	18.5 (10)	100 (49)	10.2 (5)	10.4 (5)	27.1 (13)
PPD ≥ 6 mm								
% of sites (SD)	34.3 (31.8)	4.2 (15.5)	2.8 (14.3)	1.4 (5.8)	29.1 (31.6)	0.5 (3.6)	0.5 (3.6)	2.1 (7.0)
% of implants (n)	69.5 (41)	10.2 (6)	3.7 (2)	5.6 (3)	57.1 (28)	2.0 (1)	2.1 (1)	8.3 (4)
PPD ≥ 5 mm + BoP/SoP (same site)								
% of sites (SD)	53.4 (31.3)	8.5 (21.6)	6.5 (21.8)	4.2 (9.4)	56.1 (26.3)	2.0 (6.9)	1.0 (5.0)	6.3 (10.9)
% of implants (n)	91.5 (54)	18.6 (11)	11.1 (6)	16.7 (9)	100 (49)	8.2 (4)	4.2 (2)	25.0 (12)
PPD ≥ 6 mm + BoP/SoP (same site)								
% of sites (SD)	31.8 (31.1)	3.8 (14.5)	2.8 (14.3)	1.4 (5.8)	28.6 (31.5)	0.5 (3.6)	0.5 (3.6)	1.6 (6.1)
% of implants (n)	66.1 (39)	10.2 (6)	3.7 (2)	5.6 (3)	57.1 (28)	2.0 (1)	2.1 (1)	6.3 (3)
mean marginal bone loss	4.1 (1.6)	4.4 (1.8)	4.2 (1.7)	4.1 (1.7)	4.0 (1.5)	4.3 (1.5)	4.3 (1.6)	4.3 (1.7)

BoP = bleeding on probing; SoP = suppuration on probing; PPD = probing pocket depth; SD = standard deviation

Table 3. Mean differences in BoP, SoP, PPD and radiographic marginal bone loss between control and test group over three follow-up measurements (3, 6 and 12 months) from baseline.

Outcome variable	Crude model ¹		Adjusted model ²	
	β (95% CI)	p value	β (95% CI)	p value
% sites BoP	-3.88 (-17.74 to 9.97)	0.583	3.24 (-10.17 to 16.64)	0.636
% sites SoP	-3.00 (-7.81 to 1.81)	0.222	-4.15 (-9.07 to 0.78)	0.099
mean PPD	0.12 (-0.40 to 0.63)	0.642	-0.15 (-0.40 to 0.69)	0.601
mean marginal bone loss	-0.01 (-0.22 to 0.21)	0.950	0.02 (-0.20 to 0.23)	0.869

Note: the reference category for intervention effect is the control group. The regression coefficients (β) indicate the average differences in outcomes between control and test group over the three follow-up measurements (3, 6 and 12 months) from baseline.

¹ adjusted for baseline value and time; ² adjusted for baseline value, time, smoking, dental status, history of periodontitis and implant surface roughness; BoP = bleeding on probing; SoP = suppuration on probing; PPD = probing pocket depth; 95% CI = 95% confidence interval.

nor negative could be noted for the CHX treatment. Although initially (crude analysis) the clinical results of the 2% CHX group appeared better than the results from the placebo group, the perceived positive effects disappeared after correction for the confounding factor 'order in which patients were treated'. The influence of this confounding factor might reflect an increase in experience of the surgical team with the specific treatment protocol.

Despite the lack of clinical and radiographic differences between the control and test group, the overall treatment success of both treatment procedures was rather high compared to other studies with a more or less comparable treatment protocol (Serino & Turri 2011, De Waal et al. 2013). Combining both groups revealed an overall success at twelve months after treatment of 75% on implant level and 59% on subject level (failure defined as PPD \geq 5 mm combined with bleeding and/or suppuration on probing). Increasing the threshold for failure to PPD \geq 6 mm combined with bleeding and/or suppuration on probing revealed a success of 89% on implant level and 80% on subject level.

Interestingly, despite a more than 16-fold difference in the CHX dose being delivered to the implant surface both decontamination procedures (2% CHX versus 0.12% CHX + 0.05% CPC) resulted in significant, but similar ($p = 0.99$), reductions in counts of viable anaerobic bacteria on the implant surface. Apparently, there is a limit to the effect that can be achieved with the used procedure, *i.e.* mechanical debridement followed by continuous irrigation and refreshment of a CHX-solution during 1 minute using a syringe with a 22 gauge needle followed by 1 minute of saline rinsing. Although the 2% CHX solution did not cause any detrimental clinical effects (*e.g.* impaired wound healing or pain), it neither contributed to improved clinical or radiographic outcomes of the investigated peri-implantitis treatment.

Analysis of the bacterial samples was done using culture technique. This allowed for determination of the number of viable bacteria after the decontamination procedure. Ideally, a decontamination procedure includes both bactericidal (killing) and cleaning (removal) capacities. Unfortunately, there is no single analyzing technique available that allows for determination of, and discrimination between, both capacities.

Table 4. Log-transformed mean bacterial anaerobic counts (SD) of culture positive implants for the control and test group before (T_{pre}) and after (T_{post}) debridement and decontamination of the implant surface.

N = 78*	Total anaerobic bacterial load log-transformed mean (SD)			β (95% CI) [#]	p value
	T_{pre}	T_{post}	Difference		
control	5.25 (0.88) [44]	1.88 (2.20) [19]	3.37 (2.34)	-0.01 (-1.20 to 1.19)	0.99
test	5.63 (0.98) [34]	1.98 (2.86) [12]	3.65 (2.87)		

* Implants with baseline values of 0 excluded from analysis; SD = standard deviation; [n] = number of culture positive implants; [#] linear regression analysis, adjusted for baseline values and implant surface roughness

Several *in-vitro* studies have shown that CHX can be successful in killing bacteria in biofilms grown on titanium surfaces (Chin et al. 2007, Gosau et al. 2010). However, CHX seems only modestly effective in actually removing the biofilm (Ntrouka et al. 2011b). It seems reasonable therefore to conclude that the microbiological differences between the CHX-solutions (0.12% / 2%) and saline (placebo) are mainly caused by a killing, rather than a cleaning effect. However, it appears that this killing effect is limited, given that no concentration effect was noted in the present study, under the given exposure time of 1 minute.

The superior microbiological results of CHX over placebo did not translate to improved clinical results. One explanation for this observation could be the lack of cleaning capacity of CHX. Another explanation could be that the success of peri-implantitis treatment is determined by factors other than the method of surface debridement and decontamination (Schwarz et al. 2011). If treatment is aimed to halt further peri-implant bone loss a clean ('pristine') implant surface might indeed not be a prerequisite for achieving a successful treatment outcome. However, if re-osseointegration is the goal, a clean implant surface in addition to an unaffected implant topography seems mandatory, thus allowing for reestablishment of the initial surface atomic composition and titanium oxide layer and recreation of a surface compatible with osseointegration (Mouhyi et al. 2012).

In vitro studies have shown that other chemotherapeutic agents, such as hydrogen peroxide (H_2O_2) and citric acid (CA), might be more suitable for removal of biofilm on titanium surfaces than CHX (Ntrouka et al. 2011a, Ntrouka et al. 2011b, Mouhyi et al. 2012). However, although hydrogen peroxide and citric acid have been used for implant surface decontamination during surgical peri-implantitis treatment (Khoury & Buchmann 2001, Leonhardt et al. 2003, Roos-Jansåker et al. 2007a, Roos-Jansåker et al. 2007b), so far no randomized controlled trials have been conducted evaluating exclusively the influence of these chemotherapeutic agents on treatment outcomes. Another potentially capable chemotherapeutic agent for implant surface decontamination might be phosphoric acid. Although this agent has only been evaluated in regular implant maintenance and not yet for use during surgical peri-implantitis treatment, it appears to have a strong bactericidal and cleaning effect without being deleterious to the implant surface or surrounding tissues (Strooker et al. 1998).

Based on the present study it can be concluded that the use of a 2% CHX solution

Table 5. Number of culture positive implants of selected periodontal pathogens and mean (SD) percentage of total anaerobic bacterial load on culture positive implants for the control and test group before (T_{pre}) and after (T_{post}) debridement and decontamination of the implant surface.

N = 78*		control (n = 44)		test	
		T_{pre}	T_{post}	T_{pre}	T_{post}
Aa	n %	0	0	0	0
Pg	n %	6 27.2 (26.3)	0**	6 18.5 (10.2)	0**
Pi	n %	4 8.0 (7.4)	0	7 8.4 (9.9)	1 [1] 1.0
Tf	n %	9 4.7 (2.2)	0**	7 15.9 (19.8)	0**
Fn	n %	24 17.6 (13.1)	2** 20.5 (24.7)	15 13.6 (11.5)	3 [3]** 6.0 (6.1)
Pm	n %	26 21.8 (23.8)	5 [1]** 24.0 (18.1)	28 34.8 (26.0)	7 [1]** 55.0 (44.8)
Cr	n %	8 7.9 (4.5)	0**	4 10.5 (14.0)	0

* Implants with baseline values of 0 excluded from analysis; Aa = *A. actinomycetemcomitans*; Pg = *P. gingivalis*; Pi = *P. intermedia*; Tf = *T. forsythia*; Fn = *F. nucleatum*; Pm = *P. micra*; Cr = *C. rectus*; [n] = number of implants culture negative at baseline but culture positive after decontamination; ** significant change from baseline $p < 0.05$ (McNemar test). There were no significant differences between both groups after debridement and decontamination (Fisher's exact test).

for implant surface decontamination during resective peri-implantitis therapy does not lead to improved clinical, radiographic or microbiological results compared to using a 0.12% CHX + 0.05% CPC solution. Overall, the additional use of CHX reduces anaerobic bacterial load on the implant surface better than mechanical debridement alone, but does not seem to enhance clinical treatment outcomes. Future research should focus on other chemotherapeutic agents for implant surface decontamination. To allow discrimination between effective and ineffective (combinations of) procedures randomized controlled trials are urgently needed.

Conflict of interest and source of funding statement

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