Peri-implant infections

de Waal, Yvonne Catharina Maria

IMPLANT DECONTAMINATION DURING SURGICAL PERI-IMPLANTITIS TREATMENT: A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED TRIAL

Y.C.M. de Waal
G.M. Raghoebar
J.J.R. Huddleston Slater
H.J.A. Meijer
E.G. Winkel
A.J. van Winkelhoff

Edited version of: Journal of Clinical Periodontology 2013; 40: 186-195

ABSTRACT

Aim

The objective of this randomized, double-blind, placebo-controlled trial was to study the effect of implant surface decontamination with chlorhexidine (CHX)/cetylpyridinium chloride (CPC) on microbiological and clinical parameters.

Material and methods

Thirty patients (79 implants) with peri-implantitis were treated with resective surgical treatment consisting of apically repositioned flap, bone re-contouring and surface debridement and decontamination. Patients were randomly allocated to decontamination with 0.12% CHX + 0.05% CPC (test group) or a placebo solution (without CHX/CPC, placebo group). Microbiological parameters were recorded during surgery, clinical and radiographic parameters were recorded before (pre)treatment (baseline), and at 3, 6 and 12 months after treatment.

Results

Nine implants in two patients in the placebo group were lost due to severe persisting peri-implantitis. Both decontamination procedures resulted in significant reductions of bacterial load on the implant surface, but the test group showed a significantly greater reduction than the placebo group (log 4.21 ± 1.89 versus log 2.77 ± 2.12, $p = 0.006$). Multilevel analysis showed no differences between both groups in the effect of the intervention on bleeding, suppuration, probing pocket depth and radiographic bone loss over time.

Conclusion

Implant surface decontamination with 0.12% CHX + 0.05% CPC in resective surgical treatment of peri-implantitis leads to a greater immediate suppression of anaerobic bacteria on the implant surface than a placebo solution, but does not lead to superior clinical results. The long-term microbiological effect is unknown.
(ClinicalTrials.gov number NCT01521260).

CLINICAL RELEVANCE

Scientific rationale for study

The method of surface debridement and decontamination might influence the outcome of surgical treatment of peri-implantitis. However, limited evidence exists as to which method should be used.

Principal finding

Implant surface decontamination with chlorhexidine/cetylpyridinium chloride during resective surgical treatment of peri-implantitis leads to greater bacterial reduction, but similar clinical results compared to decontamination with a placebo solution.

Practical implications

Chlorhexidine/cetylpyridinium chloride might be useful for decontamination of the implant surface during surgical treatment of peri-implantitis, but it fails to improve clinical results significantly.

INTRODUCTION

The principal objectives for treatment of peri-implantitis are resolution of inflammation and preservation of supporting bone. If non-surgical therapy does not resolve the inflammatory lesion, access flap surgery is recommended (Lindhe et al. 2008). Surgical access to the peri-implantitis lesion allows for proper removal of granulation tissue and exposes the implant surface for debridement and decontamination.

The clinical effects of access surgery combined with surface debridement and decontamination have been investigated in only a few studies (Leonhardt et al. 2003, Máximo et al. 2009, Heitz-Mayfield et al. 2012). Three studies have evaluated resective surgical procedures, *e.g.* apically repositioned flap, bone re-contouring and/or implantoplasty, combined with debridement and decontamination of the implant surface (Romeo et al. 2005, Deppe et al. 2007, Serino & Turri 2011). Most of these studies included adjunctive systemic antibiotic therapy in their treatment protocol. Different protocols, materials and chemical compounds for decontamination of the implant surface have been used, including 10% hydrogen peroxide (Leonhardt et al. 2003), teflon curettes and abrasive sodium carbonate air-powder (Máximo et al. 2009), titanium coated curettes and surgical gauzes soaked in saline (Heitz-Mayfield et al. 2012), metronidazole gel and a tetracycline hydrochloride solution (Romeo et al. 2005), air-powder abrasive alone or in combination with CO_2 laser irradiation (Deppe et al. 2007) and an ultrasonic instrument and rotating rubber cup under chlorhexidine (CHX) irrigation (Serino & Turri 2011).

Due to the wide variation in materials and procedures that have been described for the treatment of peri-implantitis it is difficult to discriminate between effective and ineffective (components of) interventions. Therefore, it has been suggested that it may be necessary to start assessing simple interventions using a double-blind study design before gradually testing more complex treatments (Esposito et al. 2012). Future stud-

ies should compare two treatment protocols that differentiate only on one component of the intervention.
So far, only two randomized controlled trials, comparing different protocols for debridement and decontamination of the implant surface and combined surgical treatment of peri-implantitis, were published (Romeo et al. 2005, Schwarz et al. 2011). Modification of surface topography (implantoplasty) when combined with resective surgery seems to positively influence implant survival and clinical parameters such as peri-implant pocket depth, suppuration and sulcus bleeding (Romeo et al. 2005). In the second randomized controlled trial, implantoplasty was used as an adjunct to regenerative surgical procedures (Schwarz et al. 2011, 2012). The method of surface debridement and decontamination (Er:YAG laser versus plastic curettes + cotton pellets + sterile saline) did not significantly impact the clinical outcomes, neither after six months nor after two years. Unfortunately, in both studies the microbiological effects of the surface modification/decontamination procedures were not assessed.
Since peri-implantitis is an infectious disease (Lindhe et al. 2008, Zitzmann & Berglundh 2008), it seems logical to focus on anti-infective measures. The screw-shaped design of implants and the various implant surface modifications may limit the effect of mechanical debridement of implant surfaces and may advocate the use of additional therapies, such as antibiotics or antiseptics. An in vivo study showed that the antiseptics chlorhexidine, sodium hypochlorite, hydrogen peroxide, essential oils and citric acid may have some beneficial effect in reducing the bacteria load on titanium surfaces and may improve peri-implantitis therapy (Gosau et al. 2010). Chlorhexidine (CHX) has broad antibacterial activity and, for periodontal diseases, has well documented clinical efficacy and plaque-reducing capabilities (for reviews see: (Addy 1986, Jones 1997)).
The objective of the present study was to study the microbiological, clinical and radiographic effect of implant surface decontamination by a chlorhexidine (CHX)/cetylpyridinium chloride (CPC) solution in comparison to a placebo solution in resective surgical treatment of peri-implantitis. It was hypothesized that no differences would exist in reduction of counts of anaerobic bacteria on the implant surface between the two decontamination procedures.

MATERIAL AND METHODS

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 October 2009 and September 2011. The study 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 (METc2009.172). US Na-

tional Institutes of Health clinical trial registration was done at www.ClinicalTrials.gov (NCT01521260). 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 present study is a randomized, double-blind and placebo-controlled trial evaluating the microbiological, clinical and radiographic outcomes of resective surgical treatment of peri-implantitis combined with decontamination of the implant surface using 0.12% CHX + 0.05% CPC or a placebo solution. Follow-up time was 12 months. Patients were randomly assigned to either the test or placebo group using a one-to-one allocation ratio.

Randomization

Fifteen notes with the words 'solution A' and 15 notes with 'solution B' were put into 30 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 syringe with either solution A or solution B and was unaware of the composition of the solution. This information was stored and kept by an independent person not involved in the study. The placebo solution was matched to the CHX solution for taste, smell, color and viscosity, ensuring blinding of the assistant, 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 reasonably possible (in all but eight patients). Incisions were made using a surgical blade (no. 15) under local anesthesia. 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 recontouring, 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 placebo or the test group. After treatment allocation, the implant surface was rinsed for 1 minute with 0.12% CHX + 0.05% CPC without alcohol (PerioAid, Dentaid SL, Cerdanyola, Spain) (test group) or with the placebo solution (placebo group). Test and placebo solutions were prepared and distributed by Dentaid SL (Cerdanyola, Spain). The placebo solution contained the same ingredients

as the test solution, except for CHX and CPC. Care was taken to continuously cover the implant surface with the solution. This was achieved by continuous irrigation and refreshment of the solution using a syringe with a 22 gauge needle to ensure penetration into deep bony defects. 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® 3-0, Ethicon Inc., Somerville, NJ, USA). For both placebo and test group, surgery was followed by two weeks of mouth rinsing with 0.12% CHX + 0.05% CPC without alcohol (PerioAid, Dentaid 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

Primary outcome

The primary 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 and rinsing of the implant surface with the test or placebo solution and 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. All microbiological samples were processed within 24 hours as described by Van Winkelhoff et al. (1985) and Van Steenbergen 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.

Secondary outcomes

Secondary outcome parameters were presence of plaque (% sites plaque), bleeding on probing (% sites BoP), suppuration on probing (% sites SoP), mean probing pocket depth (PPD) and mean radiographic marginal bone loss. Measurements were performed before (pre)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. Reproducibility of probing pocket depth measurements was assessed by evaluating 20 implants in 8 subjects on two separate occasions, one week

Figure 1. Inclusion- and exclusion criteria

Inclusion criteria	• 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	• 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.

apart and calculating the linear weighted kappa (κ) value.
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 nine fully edentulous patients (16 implants) no intra-oral radiographs could be obtained 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. Reproducibility of radiographic examinations was assessed by evaluating radiographic images of 20 implants (10 intra-oral radiographs and 10 orthopantomograms) twice with a one-week interval. Intraclass correlation coefficients were calculated for both categories of radiographs.

Statistical methods

Sample size calculation

From the literature, no data were available for estimating the effect size. However, the microbiological effect of rinsing the dental implant surface with a CHX solution versus rinsing with a placebo solution was expected to be large.
To detect a difference of 1 standard deviation (assumed to be unknown and equal) between both groups under the null hypothesis that both group means were 0.0 with a significance level (α) of 0.05 and a power (β) of 80% using a two sided two-sample

t-test, required group sample sizes of 15 (G*Power Version 3.1.0, University of Kiel, Kiel, Germany). Since no compensation for patient withdrawal or losses to follow up was required (data regarding primary outcome variable was collected during surgical treatment) a sample size of 30 patients was chosen (15 per group).

Statistical analysis

Total anaerobic bacterial load at baseline (T_{pre}) was distributed normally after logarithmic transformation. To compare outcomes between placebo and test group linear regression analysis was performed. 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.

Because the primary outcome variable is a measure of the local effect of decontamination, the implant (and not the patient) was taken as the statistical unit, despite the fact that multiple implants were present per patient. To correct for the within patient dependency, multilevel modeling was used to determine the effect of the intervention over time (test group versus placebo group) on the secondary 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. For each outcome variable two analyses were performed (Twisk 2006). With the crude analysis the effect of the intervention over time was determined, while controlling for baseline value and time. Since follow-up was conducted at irregularly spaced time intervals and not completely similar for each patient, time was included in the crude model (Ridgers et al. 2007). In the adjusted analysis the potential confounders smoking, dental status, history of periodontitis and implant surface roughness were additionally included in the model.

Descriptive data and data regarding the primary outcome variable were analyzed using PASW® Statistics 18 (version 18.0.3, SPSS inc., Chicago, IL, USA). Multilevel models were analyzed using MLwiN version 2.12 (Centre for Multilevel Modeling, University of Bristol, Bristol, UK).

RESULTS

The flow of the participants throughout the different phases of the study is depicted in Figure 2. Eligible patients were recruited from October 2009 to September 2010 and were followed 3, 6 and 12 months after the surgical procedure. The baseline demographic patient and implant characteristics are reported in Table 1. Thirty patients with a total of 79 implants with peri-implantitis were included.

Figure 2. Flow-diagram

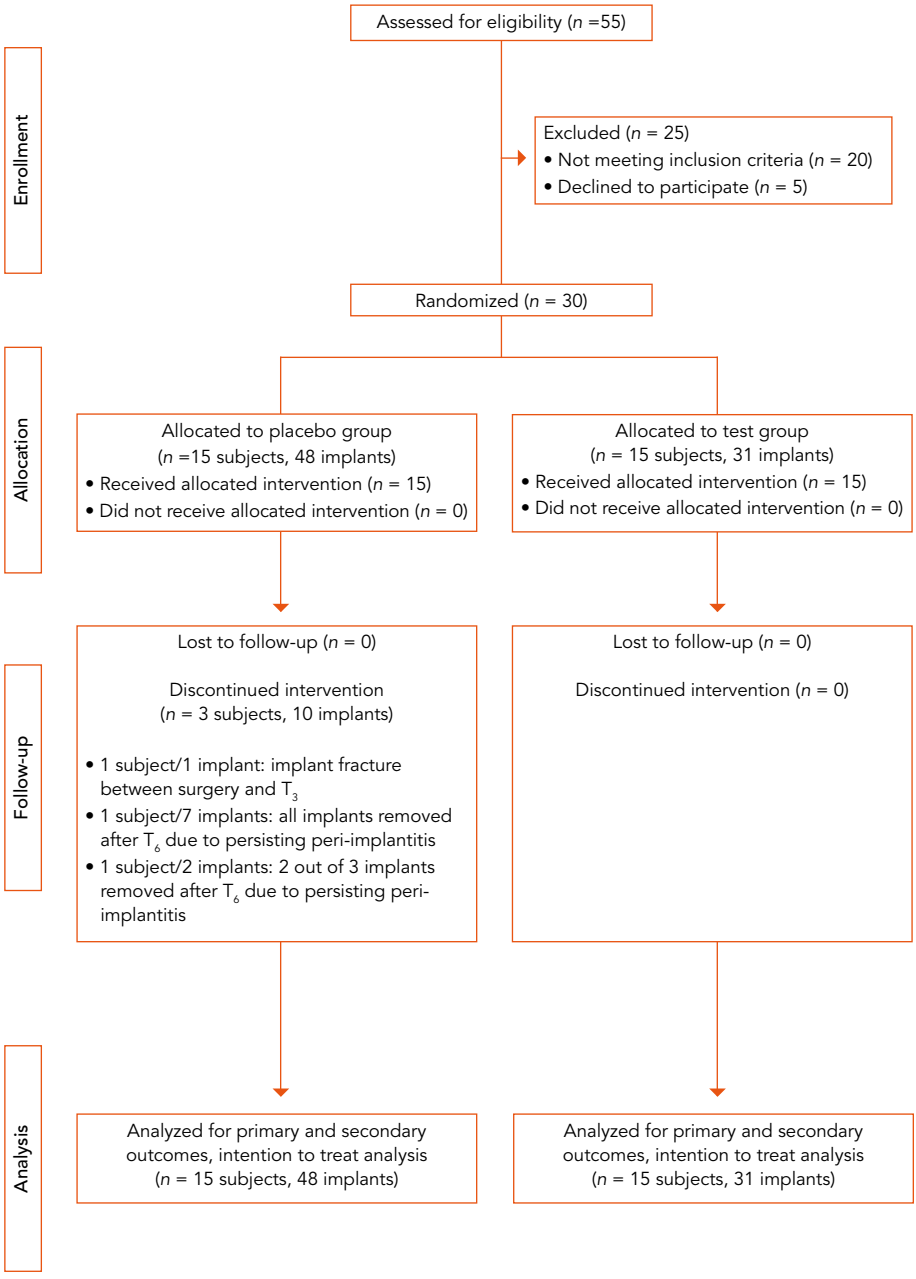

Table 1. Baseline demographic characteristics of included subjects/implants

Characteristics		Placebo	Test
Number of patients		15	15
Age (years; mean (SD))		61.5 (10.0)	59.4 (14.0)
Gender; M (male), F (female)		M 5, F 10	M 5, F 10
Smoking; *n* subjects (%)			
	never (or quit smoking before implant placement)	7 (46.7)	8 (53.3)
	former (quit smoking after implant placement)	1 (6.7)	3 (20.0)
	current	7 (46.7)	4 (26.7)
History of periodontitis; *n* subjects (%)		5 (33.3)	6 (40.0)
Dental status; *n* subjects (%)			
	fully edentulous	9 (60.0)	10* (66.7)
	partially edentulous	6 (40.0)	5 (33.3)
Total number of implants (range)		63 (2-10)	58 (1-10)
Number of implants presenting peri-implantitis (range)		48 (1-7)	31 (1-5)
Time in function (years; mean (SD))		8.6 (5.5)	9.2 (3.8)
Implant surface; *n* implants (%)			
Nobel Biocare	machined surface	1 (2.1)	4 (12.9)
	porous anodized surface, TiUnite	21 (43.8)	6 (19.4)
Straumann	titanium plasma-sprayed, TPS	5 (10.4)	0 (0)
	sandblasted large grit acid-etched, SLA	4 (8.3)	14 (45.2)
	sandblasted large grit acid-etched, SLActive	10 (20.8)	1 (3.2)
IMZ	titanium plasma-sprayed	7 (14.6)	2 (6.5)
Astra Tech	fluoride-modified titanium dioxide grit-blasted, Osseospeed	0 (0)	2 (6.5)
Dentsply Friadent	grit-blasted acid-etched, Friadent plus	0 (0)	2 (6.5)
Type of restoration; *n* implants involved (%)			
	single crown	4 (8.3)	2 (6.5)
	fixed partial denture	4 (8.3)	1 (3.2)
	fixed full denture	7 (14.5)	6 (19.4)
	overdenture	33 (68.8)	22 (71.0)
Screw or cement retained restoration; *n* implants involved (%)			
	screw-retained	44 (91.7)	28 (90.3)
	cement-retained	4 (8.3)	3 (9.7)
Implants placed in maxilla or mandible; *n* implants (%)			
	maxilla	24 (50.0)	20 (64.5)
	mandible	24 (50.0)	11 (35.5)

*3 were partially edentulous at the time of implant placement

Primary outcome

The log-transformed mean anaerobic bacterial load of the culture positive implants for the placebo and test group before and after debridement and decontamination of the implant surface are depicted in Table 2. Sixty out of the 79 sampled implant surfaces appeared culture positive after exposure and removal of granulation tissue (T_{pre}). In both groups the decontamination procedure resulted in a significant reduction of the bacterial load on the implant surface, although the test group showed a significantly greater reduction than the placebo group (4.21 ± 1.89 versus 2.77 ± 2.12, $p = 0.006$). The number of implants culture positive for the selected periodontal pathogens before and after decontamination are depicted in Table 3. *A. actinomycetemcomitans*

Table 2. Log-transformed mean bacterial anaerobic counts (SD) of culture positive implants for the placebo and test group before (T_{pre}) and after (T_{post}) debridement and decontamination of the implant surface.

N = 60*	Total anaerobic bacterial load log-transformed mean (SD)				
	T_{pre}	T_{post}	Difference	β (95% CI)#	*p* value
placebo	5.54 (1.23) [35]	2.77 (2.37) [21]	2.77 (2.12)	-1.57 (-2.68 to -0.46)	0.006**
test	5.46 (1.13) [25]	1.25 (2.11) [7]	4.21 (1.89)		

* 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; ** statistically significant difference

was not detected on any of the implant surfaces. Both decontamination procedures resulted in reduction below detection level of *P. gingivalis* and *P. intermedia* and in significant reductions in detection frequencies of *T. forsythia, F. nucleatum, P. micra* and *C. rectus*. No differences were observed between both groups.

Secondary outcomes

One implant (Nobel Biocare, machined surface) in one patient (with no other implants affected by peri-implantitis) was lost due to implant fracture between surgery and T_3. No signs of fracture were present during the surgical procedure. Nine implants in 2 patients from the placebo group had to be removed between T_6 and T_{12} due to severe persisting peri-implantitis. One patient lost all 7 treated implants (Nobel Biocare, Ti-Unite surface) and discontinued the study. The other patient lost 2 out of 3 treated implants (Straumann, TPS surface), but continued the study with the remaining implant. No implants were lost in the test group. The patients from both groups attended all follow-up visits and no patient was lost to follow-up.

Descriptive statistics of the clinical and radiographic outcomes at baseline and at different follow-up visits are depicted in Table 4. Data are reported at implant-level and, in more detail, on different sites per implant. Intra-examiner reproducibility of clinical measurements was good as indicated by a linear weighted kappa (κ) value of 0.82. Intraclass correlation coefficients were 0.99 and 0.96 for radiographic measurements on intra-oral radiographs and orthopantomograms respectively, indicating very good intra-examiner reproducibility. Clinical improvements occurred in both the test and placebo group, as indicated by the reduction in percentage of sites with BoP and/or SoP and reduction in mean PPD. However, at T_{12} almost all implants still showed at least one site with BoP (95.7%) and a substantial number of implants showed signs of suppuration (9 implants in test group, 6 of remaining implants in placebo group).

The results from the multilevel modeling analysis regarding the effect of the intervention on BoP, SoP, PPD and radiographic marginal bone loss across time are shown in Table 5. No significant differences were observed between both groups for all investigated secondary parameters, both in the 'crude' and 'adjusted' model. Smoking, dental status (fully versus partially edentulous), history of periodontitis and implant surface roughness were confounders to the 'crude' model.

Table 3. Number of culture positive implants of selected periodontal pathogens and mean (SD) percentage of total anaerobic bacterial load on culture positive implants for the placebo and test group before (T_{pre}) and after (T_{post}) debridement and decontamination of the implant surface.

N = 60*		Placebo (*n* = 35)		Test (*n* = 25)	
		T_{pre}	T_{post}	T_{pre}	T_{post}
Aa	n	0	0	0	0
	%				
Pg	n	3	0	3	0
	%	63.0 (32.2)		31.0 (36.6)	
Pi	n	2	0	1	0
	%	2.5 (0.7)		6.0	
Tf	n	13	3 [1]**	7	1 [1]
	%	9.9 (17.0)	10.0 (9.2)	4.7 (3.3)	2.0
Fn	n	20	6 [1]**	19	3 [1]**
	%	9.0 (9.9)	16.7 (19.9)	12.7 (13.1)	32.7 (30.1)
Pm	n	19	6**	18	3**
	%	12.8 (9.8)	30.8 (33.8)	19.1 (11.9)	24.0 (24.3)
Cr	n	11	2**	5	1 [1]
	%	7.5 (7.7)	22.0 (14.1)	11.8 (9.8)	4.0

* 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's test). There were no significant differences between both groups after debridement and decontamination (Fisher's exact test).

DISCUSSION

To our knowledge, this is the first randomized, double-blind, placebo-controlled trial evaluating the microbiological, clinical and radiographic effect of an implant surface decontamination procedure combined with resective surgical treatment of peri-implantitis. The results of this study indicate that decontaminating the implant surface with 0.12% CHX + 0.05% CPC leads to a greater reduction of the anaerobic bacterial load on the implant surface than using a placebo solution. Therefore, the null hypothesis of no difference can be rejected. However, this greater reduction in bacterial load did not lead to superior clinical or radiographic results over a period of 12 months. These findings are consistent with Schwarz et al. (2011, 2012) who did not find an impact of the method of surface debridement and decontamination on the clinical outcomes following combined surgical therapy of advanced peri-implantitis lesions. It was suggested that the long-term stability of the clinical outcomes may be influence by factors other than the method of surface debridement and decontamination.
CHX is considered the gold standard for oral antiseptics (Addy 1986, Jones 1997). It has been widely used and extensively tested and has a broad spectrum of antibacterial activity including gram-positive and gram-negative bacteria (Jones 1997). From the literature no comparable studies are available evaluating the immediate microbiological effect of an antiseptic agent on a genuine peri-implantitis-associated biofilm. However, an in vivo study showed that chlorhexidine, among other antiseptics such as sodium hypochlorite, hydrogen peroxide, essential oils and citric acid, may have some

Table 4. Descriptive statistics of clinical and radiographical parameters

N = 79		Placebo				Test			
		T_0 (*n* = 48)	T_3 (*n* = 47)	T_6 (*n* = 47)	T_{12} (*n* = 38)	T_0 (*n* = 31)	T_3 (*n* = 31)	T_6 (*n* = 31)	T_{12} (*n* = 31)
plaque	% of sites (SD)	17.7 (26.3)	17.6 (22.7)	28.7 (28.5)	15.8 (17.8)	9.7 (30.1)	17.7 (27.5)	18.6 (24.1)	12.9 (19.2)
	% of implants (*n*)	41.7 (20)	46.8 (22)	66.0 (31)	50.0 (19)	9.7 (3)	35.5 (11)	48.4 (15)	38.7 (12)
BoP	% of sites (SD)	79.7 (28.1)	62.6 (33.3)	60.1 (32.0)	57.2 (29.0)	80.4 (26.5)	69.4 (27.9)	54.0 (36.6)	60.5 (30.1)
	% of implants (*n*)	95.8 (46)	91.5 (43)	91.5 (43)	94.7 (36)	96.8 (30)	96.8 (30)	80.6 (25)	96.8 (30)
SoP	% of sites (SD)	14.6 (25.2)	1.6 (6.2)	4.4 (11.2)	5.9 (14.7)	31.7 (31.6)	0.8 (4.5)	5.6 (12.4)	14.5 (28.0)
	% of implants (*n*)	31.3 (15)	6.4 (3)	14.9 (7)	15.8 (6)	64.5 (20)	3.2 (1)	19.4 (6)	29.0 (9)
mean PPD	mean (SD)	5.5 (1.4)	4.0 (1.3)	4.1 (1.4)	3.7 (0.8)	6.6 (1.6)	4.2 (1.5)	4.0 (1.5)	4.3 (2.1)
PPD ≥ 5 mm	% of sites (SD)	75.2 (26.1)	26.2 (31.6)	29.6 (35.0)	17.1 (24.0)	88.2 (18.4)	39.5 (36.4)	33.1 (31.9)	33.9 (39.0)
	% of implants (*n*)	100 (48)	51.1 (24)	55.3 (26)	47.4 (17)	100 (31)	64.5 (20)	67.7 (21)	54.8 (17)
PPD ≥ 6 mm	% of sites (SD)	49.0 (33.8)	16.5 (29.0)	14.4 (30.0)	7.2 (19.2)	57.0 (33.5)	21.8 (34.0)	17.7 (30.4)	20.2 (35.0)
	% of implants (*n*)	81.3 (39)	31.9 (15)	25.6 (12)	15.8 (6)	87.1 (27)	35.5 (11)	32.3 (10)	32.3 (10)
PPD ≥ 5 mm + BoP/SoP (same site)	% of sites (SD)	70.0 (30.4)	23.6 (30.9)	25.4 (34.1)	15.8 (23.6)	80.1 (23.0)	36.3 (32.8)	23.4 (30.9)	29.0 (37.1)
	% of implants (*n*)	95.8 (46)	46.8 (22)	48.9 (23)	42.1 (16)	100 (31)	64.5 (20)	48.4 (15)	48.4 (15)
PPD ≥ 6 mm + BoP/SoP (same site)	% of sites (SD)	46.9 (33.7)	16.0 (29.1)	14.4 (30.0)	7.2 (19.2)	54.6 (33.7)	19.4 (30.1)	14.5 (27.2)	17.7 (34.3)
	% of implants (*n*)	81.3 (39)	29.8 (14)	25.5 (12)	15.8 (6)	87.1 (27)	35.5 (11)	32.3 (10)	25.8 (8)
mean marginal bone loss	mean (SD)	3.6 (1.9)	4.0 (2.0)	4.3 (2.2)	3.9 (2.0)	4.3 (2.1)	4.7 (2.2)	4.9 (2.4)	5.0 (2.5)

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

Table 5. Average differences in BoP, SoP, PPD and radiographical marginal bone loss between placebo 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	0.34 (-14.93 to 15.61)	0.965	-7.58 (-24.20 to 9.05)	0.372
% sites SoP	0.08 (-5.36 to 5.52)	0.977	-3.77 (-10.25 to 2.72)	0.255
mean PPD	-0.26 (-1.13 to 0.62)	0.563	-0.50 (-1.40 to 0.41)	0.284
mean marginal bone loss	0.01 (-0.35 to 0.38)	0.949	0.11 (-0.27 to 0.48)	0.575

Note: the reference category for intervention effect is the placebo group. The regression coefficients (β) indicate the average differences in secondary outcomes between placebo 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

beneficial effect in reducing the bacterial load on titanium surfaces and may improve peri-implantitis therapy (Gosau et al. 2010). The antibacterial mode of action is based on the ability of the cationic CHX-molecule to rapidly get attracted by the negatively charged bacterial cell surface (Rölla & Melsen 1975). Upon interaction, the integrity of the bacterial cell membrane is altered, which leads to leakage and eventually to destruction of the cell (Russell 1986). CHX does not distinguish between bacterial and non-bacterial proteins found in mature plaque (Jones 1997). Therefore, to remove extraneous protein and thereby optimize the effectiveness of the CHX-solution, we first mechanically cleaned the implant surface using gauzes soaked in saline. Furthermore, to optimize the penetration on rough implant surfaces, at sites away from application and in deep bony defects, a rinse was chosen as mode of application rather than a gel. One of the main advantages of CHX is its property of substantivity, which leads to prolonged activity (Addy 1986, Kuyyakanond & Quesnel 1992). However, *in vitro* studies have shown that CHX can be highly cytotoxic on fibroblasts, endothelial and osteoblastic cells (Babich et al. 1995, Cabral & Fernandes 2007, Giannelli et al. 2008). The cell damaged induced by CHX is concentration and time dependent and might negatively interfere with the early healing phase of oral diseases (Giannelli et al. 2008). To minimize any possible negative side effects of CHX, it was decided to rinse the implant surface and wound area with copious amounts of saline immediately after CHX rinsing. As a consequence, by washing out the wound area, the potential benefits resulting from CHX substantivity may have also diminished.
The commercially available CHX solution used in the present study also contained CPC as active ingredient. CPC is a cationic agent and has a broad antimicrobial spectrum with bactericidal effect on gram-positive pathogens and yeast in particular (Pitten & Kramer 2001). CPC has a strong immediate bactericidal effect, but lower residual effect compared to CHX (Pitten & Kramer 1999). It has been shown that the non-alcoholic formulation of 0.12% CHX + 0.05% CPC is an equally effective anti-plaque and anti-inflammatory agent as the 0.2% CHX mouthrinse with alcohol (Quirynen et al. 2001). In addition, Herrera et al. (2003) showed that the reformulation and addition

of 0.05% CPC to 0.12% CHX products may not only compensate for the absence of alcohol but may rather increase the in vitro and in vivo antimicrobial activity. Both CHX + alcohol and CHX + CPC showed high antimicrobial activity to 20 bacterial species, including periodontal pathogens.

In the present study, microbiological samples were collected using sterilized microbrushes. These were small enough to reach the areas between implant threads, but robust enough to allow rubbing of the implant surface. Because the local and immediate microbiological effect of the decontamination procedure was evaluated, data were analyzed on implant level. Microbiological parameters were not assessed over time. The clinical and radiographic data were analyzed using a multi-level model. By using multilevel modeling a correction is made for the difference in number of implants per patient and the dependency of the observations within each patient and over time. Because multilevel modeling is very flexible in handling missing data points, all longitudinal data could be used despite some incomplete patient records (*e.g.* implants that were removed during the follow-up period) (Twisk & De Vente 2002). Due to practical and anatomical limitations, radiographs could not be standardized. In addition, in many fully edentulous subjects intra-oral radiographs could not be obtained and had to be replaced by orthopantomographs. However, despite these limitations, intra-examiner reproducibility was very good both for intra-oral radiographs and orthopantomographs (intraclass correlation coefficients were 0.99 and 0.96 respectively).

No significant differences were seen between the test and placebo group over 12 months of observation in BoP, SoP, PPD and marginal bone loss. Although both groups showed improved clinical parameters as a result of treatment, complete resolution of inflammation (*i.e.* health) was almost never achieved. Sixty-six out of the 69 implants present at T_{12} showed at least one site with BoP and 15 implants additionally showed suppuration (representing either peri-implant mucositis or peri-implantitis). If the criteria for treatment failure were to be defined according to the inclusion criteria used in the present study (residual pockets ≥ 5 mm associated with bleeding and/or suppuration) treatment was only successful for 11 (38%) subjects and 38 (49%) implants. Increasing the threshold to pockets ≥ 6 mm associated with bleeding and/or suppuration results in treatment success for 17 (59%) subjects and 55 (71%) implants. These results are somewhat less than the 2-year follow-up results described by Serino & Turri (2011), who used more or less a comparable surgical approach (apically repositioned flap, bone re-contouring and mechanical cleansing of the implant surface under CHX irrigation). Two years after treatment 77% of the subjects and 75% of the implants showed no pockets ≥ 6 mm associated with bleeding/suppuration. A possible explanation for the difference between both studies is the fact that in the latter study all patients received adjunctive systemic antibiotic therapy (clindamycine). However, one could think of many other factors that may influence treatment results (*e.g.* patient factors such as smoking, plaque levels, periodontitis and dental status, implant factors, treatment factors et cetera) making a direct comparison between the present study and other studies difficult. Therefore, more randomized controlled trials are needed, each focusing on one aspect of the treatment protocol at a time.

The present study shows that implant surface decontamination with 0.12% CHX +

0.05% CPC in resective surgical treatment of peri-implantitis leads to greater suppression of anaerobic bacteria on the implant surface than a placebo solution. However, this does not translate to better clinical or radiographic outcomes of the intervention.

Conflict of interest and source of funding statement

The test and control solutions were manufactured and provided by Dentaid SL (Cerdanyola, Spain). The study was financed by the University Medical Center Groningen. Co-authors E.G. Winkel and A.J. van Winkelhoff have stock ownership in Dentaid BeNeLux B.V via their company LabOral International.

References

Addy M. (1986) Chlorhexidine compared with other locally delivered antimicrobials. A short review. *Journal of Clinical Periodontology* 13, 957-964.

Babich H., Wurzburger B.J., Rubin Y.L., Sinensky M.C. & Blau L. (1995) An in vitro study on the cytotoxicity of chlorhexidine digluconate to human gingival cells. *Cell Biology and Toxicology* 11, 79-88.

Cabral C.T. & Fernandes M.H. (2007) In vitro comparison of chlorhexidine and povidone-iodine on the long-term proliferation and functional activity of human alveolar bone cells. *Clinical Oral Investigations* 11, 155-164.

Cairo F., Sanz I., Matesanz P., Nieri M. & Pagliaro U. (2012) Quality of reporting of randomized clinical trials in implant dentistry. A systematic review on critical aspects in design, outcome assessment and clinical relevance. *Journal of Clinical Periodontology* 39 Suppl 12, 81-107.

Deppe H., Horch H.H. & Neff A. (2007) Conventional versus CO2 laser-assisted treatment of peri-implant defects with the concomitant use of pure-phase beta-tricalcium phosphate: a 5-year clinical report. *International Journal of Oral and Maxillofacial Implants* 22, 79-86.

Esposito M., Grusovin M.G. & Worthington H.V. (2012) Interventions for replacing missing teeth: treatment of peri-implantitis. *Cochrane Database of Systematic Reviews (Online) 1, CD004970.*

Giannelli M., Chellini F., Margheri M., Tonelli P. & Tani A. (2008) Effect of chlorhexidine digluconate on different cell types: a molecular and ultrastructural investigation. *Toxicology In Vitro* 22, 308-317.

Gosau M., Hahnel S., Schwarz F., Gerlach T., Reichert T.E. & Bürgers R. (2010) Effect of six different peri-implantitis disinfection methods on *in vivo* human oral biofilm. *Clinical Oral Implants Research* 21, 866-872.

Heitz-Mayfield L.J., Salvi G.E., Mombelli A., Faddy M., Lang N.P. & Implant Complication Research Group. (2012) Anti-infective surgical therapy of peri-implantitis. A 12-month prospective clinical study. *Clinical Oral Implants Research* 23, 205-210.

Herrera D., Roldán S., Santacruz I., Santos S., Masdevall M. & Sanz M. (2003) Differences in antimicrobial activity of four commercial 0.12% chlorhexidine mouthrinse formulations: an in vitro contact test and salivary bacterial counts study. *Journal of Clinical Periodontology* 30, 307-314.

Jones C.G. (1997) Chlorhexidine: is it still the gold standard? *Periodontology 2000* 15, 55-62.

Kuyyakanond T. & Quesnel L.B. (1992) The mechanism of action of chlorhexidine. *FEMS Microbiology Letters* 79, 211-215.

Leonhardt Å., Dahlén G. & Renvert S. (2003) Five-year clinical, microbiological, and radiological outcome following treatment of peri-implantitis in man. *Journal of Periodontology* 74, 1415-1422.

Lindhe J., Meyle J. & Group D of European Workshop on Periodontology. (2008) Peri-implant diseases: Consensus Report of the Sixth European Workshop on Periodontology. *Journal of Clinical Periodontology* 35, 282-285.

Máximo M.B., de Mendonça A.C., Renata Santos V., Figueiredo L.C., Feres M. & Duarte P.M. (2009) Short-term clinical and microbiological evaluations of peri-implant diseases before and after mechanical anti-infective therapies. *Clinical Oral Implants Research* 20, 99-108.

Moher D., Hopewell S., Schulz K.F., Montori V., Gotzsche P.C., Devereaux P.J., Elbourne D., Egger M., Altman D.G. & Consolidated Standards of Reporting Trials Group. (2010) CONSORT 2010 Explanation and Elaboration: Updated guidelines for reporting parallel group randomised trials. *Journal of Clinical Epidemiology* B, e1-37.

Pitten F.A. & Kramer A. (2001) Efficacy of cetylpyridinium chloride used as oropharyngeal antiseptic. *Arzneimittel-Forschung* 51, 588-595.

Pitten F.A. & Kramer A. (1999) Antimicrobial efficacy of antiseptic mouthrinse solutions. *European Journal of Clinical Pharmacology* 55, 95-100.

Quirynen M., Avontroodt P., Peeters W., Pauwels M., Coucke W. & Van Steenberghe D. (2001) Effect of different chlorhexidine formulations in mouthrinses on de novo plaque formation. *Journal of Clinical Periodontology* 28, 1127-1136.

Ridgers N.D., Stratton G., Fairclough S.J. & Twisk J.W. (2007) Long-term effects of a playground markings and physical structures on children's recess physical activity levels. *Preventive Medicine* 44, 393-397.

Rölla G. & Melsen B. (1975) On the mechanism of the plaque inhibition by chlorhexidine. *Journal of Dental Research* 54 Spec No B, B57-62.

Romeo E., Ghisolfi M., Murgolo N., Chiapasco M., Lops D. & Vogel G. (2005) Therapy of peri-implantitis with resective surgery. A 3-year clinical trial on rough

screw-shaped oral implants. Part I: clinical outcome. *Clinical Oral Implants Research* 16, 9-18.

Russell A.D. (1986) Chlorhexidine: antibacterial action and bacterial resistance. *Infection* 14, 212-215.

Schulz K.F., Altman D.G., Moher D. & CONSORT Group. (2010) CONSORT 2010 Statement: Updated guidelines for reporting parallel group randomised trials. *Journal of Clinical Epidemiology* 63, 834-840.

Schwarz F., Sahm N., Iglhaut G. & Becker J. (2011) Impact of the method of surface debridement and decontamination on the clinical outcome following combined surgical therapy of peri-implantitis: a randomized controlled clinical study. *Journal of Clinical Periodontology* 38, 276-284.

Schwarz F., John G., Mainusch S., Sahm N. & Becker J. (2012) Combined surgical therapy of peri-implantitis evaluating two methods of surface debridement and decontamination. A two-year clinical follow up report. *Journal of Clinical Periodontology* 39, 789-797

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, 1214-1220.

Slots J. (2002) Selection of antimicrobial agents in periodontal therapy. *Journal of Periodontal Research* 37, 389-398.

Syed S.A. & Loesche W.J. (1972) Survival of human dental plaque flora in various transport media. *Applied Microbiology* 24, 638-644.

Twisk J.W. & De Vente W. (2002) Attrition in longitudinal studies. How to deal with missing data. *Journal of Clinical Epidemiology* 55, 329-337.

Twisk J.W. (2006) Applied Multilevel Analysis. *Cambridge University Press*, 62-107.

Van Steenbergen M.J., Van Winkelhoff A.J., Van der Mispel L., Van der Velden U., Abbas F. & De Graaff J. (1986) Comparison of two selective media for *Actinobacillus actinomycetemcomitans*. *Journal of Clinical Microbiology* 24, 636-638.

Van Winkelhoff A.J., Van Steenbergen T.J., Kippuw N. & De Graaff J. (1985) Further characterization of *Bacteroides endodontalis*, an asaccharolytic black-pigmented *Bacteroides* species from the oral cavity. *Journal of Clinical Microbiology* 22, 75-79.

Zitzmann N.U. & Berglundh T. (2008) Definition and prevalence of peri-implant diseases. *Journal of Clinical Periodontology* 35, 286-291.

IMPLANT DECONTAMINATION 0.12% CHX