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Short-term comparison of two non-surgical treatment modalities of peri-implantitis: Clinical and microbiological outcomes in a two-factorial randomized controlled trial

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Abstract

Aim: To compare the efficacy of two different therapies (amino acid glycine abrasive powder and a desiccant material) and their combination in the non-surgical treatment of peri-implantitis.

Materials and Methods: This was an examiner-blind randomized clinical trial, with 2-factorial design with a follow-up of 6 months. The combination of the two factors resulted in four interventions: (a) non-surgical debridement alone (C); (b) non-surgical debridement and a desiccant material (H); (c) non-surgical debridement and glycine powder (G); and (d) non-surgical debridement, desiccant material and glycine powder (HG).

Results: Sixty-four patients with peri-implantitis were randomized, 16 for each intervention. After six months, two implants failed in the G intervention. Mean pocket depth reduction was higher in patients treated with the desiccant material (estimated difference: 0.5 mm; 95% CI from 0.1 to 0.9 mm, $p = .0229$) while there was no difference in the patients treated with glycine powder (estimated difference: 0.1 mm; 95% CI from −0.3 to 0.5 mm, $p = .7333$). VAS for pain during intervention and VAS for pain after one week were higher for patients treated with glycine powder ($p = .0056$ and $p = .0339$, respectively). The success criteria and other variables did not reveal differences between interventions.

Conclusions: In this 6-month follow-up study, pocket reduction was more pronounced in patients using the desiccant material. Pain was higher in patients using glycine. All the interventions resulted in low success rate.

KEYWORDS

dental disinfectants, dental implant, peri-implantitis, randomized controlled trial

Clinical trial registration number: ISRCTN Registry, reference number: ISRCTN13724605.

1 | INTRODUCTION

The consensus report of the 2017 World Workshop of the classification of periodontal and peri-implant diseases defined peri-implantitis as a plaque-associated pathological condition occurring in tissues around dental implants, characterized by inflammation in the peri-implant mucosa and subsequent progressive loss of supporting bone (Berglundh et al., 2018).

At the moment, there is no reliable evidence suggesting the most effective interventions for treating peri-implantitis (Esposito, Grusovin, & Worthington, 2012). Non-surgical treatments usually include debridement with curettes or via air abrasion; these can be supplemented with local antibiotic or anti-infective therapy (Liñares, Pico, Blanco, & Blanco, 2019; Muthukuru, Zainvi, Esplugues, & Flemmig, 2012; Nart et al., 2020). On the basis of currently available RCTs, there is insufficient evidence to support that any particular non-surgical treatment for peri-implantitis showed better performance than debridement alone (Faggion, Listl, Frühauf, Chang, & Tu, 2014).

Comparing alternative peri-implant treatments is not only relevant for the identification of the most effective therapeutic approach, but also to compare treatment cost (Listl & Birch, 2013). In cases where surgery is not required, debridement alone, abrasive powder using amino acid glycine, debridement combined with PerioChip® and debridement combined with local antibiotics were identified as treatment strategies with comparably better value for money than Er:YAG laser monotherapy, Vector™ System, debridement combined with chlorhexidine and photodynamic therapy (Listl et al., 2015).

Abrasive powder using amino acid glycine has been shown effective in cleaning the contaminated implant surfaces (Tastepe, van Waas, Liu, & Wismeijer, 2012). Similar results were obtained using the abrasive powder amino acid glycine in a randomized controlled trial (John, Sahm, Becker, & Schwarz, 2015; Renvert, Lindahl, Roos Jansåker, & Persson, 2011).

Recently, clinical cases showing peri-implant mucositis and peri-implantitis were treated using an oral tissue decontaminant agent containing a concentrated aqueous mixture of hydroxybenzenesulphonic and hydroxymethoxybenzene acids and sulphuric acid (HybenX) (Lombardo et al., 2015; Pini-Prato, Magnani, & Rotundo, 2016). This desiccant agent has showed greater reduction in clinical, microbial and inflammatory mediators compared to scaling and root planing alone in patients with chronic periodontitis (Isola et al., 2018). Its efficacy has not yet been tested in a randomized trial in the treatment of peri-implantitis.

Currently, there is great uncertainty regarding the most cost-efficient approach to peri-implantitis therapy (Figuero, Graziani, Sanz, Herrera, & Sanz, 2014). In addition, no literature has been published regarding the added values and interactions of combining therapies.

The evaluation of more than one treatment in the same randomized controlled trial can be achieved using a parallel-group design. However, this requires increased sample size and can be inefficient, especially if there is also interest in considering combinations of the treatments (Montgomery, Peters, & Little, 2003). An alternative may be a factorial trial, where for two interventions participants are allocated to receive neither intervention, one or the other, or both (Montgomery et al., 2003). The effect of the glycine powder and the effect of the desiccant agent on peri-implantitis could be simultaneously assessed using a factorial randomized trial.

The aim of this factorial randomized controlled trial was to compare the efficacy of two different therapies (abrasive powder amino acid glycine and a desiccant material) and their combinations in the treatment of peri-implantitis. The null hypothesis was that there is no difference in change probing depth between the use or not of the glycine power and the use or not of the desiccant material in the non-surgical treatment of peri-implantitis.

Clinical Relevance

Scientific rationale for the study: Currently, there is great uncertainty regarding the most efficient approach for non-surgical peri-implantitis therapy. This factorial randomized controlled trial compared the efficacy of abrasive powder amino acid glycine, a desiccant material and their combinations in the treatment of peri-implantitis.

Principal finding: For many clinical and microbiological variables, there were no differences between treatments. Pain was higher in patients using glycine. Pocket reduction was more pronounced in patients using desiccant material.

Practical implications: Non-surgical treatment alone or associated with the use of a desiccant agent could be preferred to more complicate non-surgical therapies.

2 | MATERIALS AND METHODS

2.1 | Setting, locations and ethics committee

The study took place at a private centre in Rimini (Italy). The dental office obtained the approval of the local authorities to conduct clinical studies (protocol number 221722/P).

The patients read and asked questions inherent to the study prior to signing the informed consent. The study was conducted in accordance with the ethical principles that have their origins in the Declaration of Helsinki 2013. An independent Ethics Committee (Ethical Committee IRST-IRCCS—Area Vasta Romagna) approved this clinical study (protocol 450/2016 I.5/282, date 27-01-2016). The principal investigator has 10 years of experience in dental implant surgery and dental implant prosthesis rehabilitation.

2.2 | Trial design

This study was written in accordance with the Consort 2010 explanation and elaboration guidelines for reporting parallel-group randomized controlled trials (Moher et al., 2010) and with the design, analysis and presentation of factorial randomized controlled trials (Montgomery et al., 2003).

This was a monocentred, examiner-blind randomized clinical trial, with balanced randomization and 2-factorial design. All of the patients underwent debridement with ultrasonic dedicated scalers. The two factors at two levels each were as follows:

1. Factor glycine powder: abrasive powder amino acid glycine (AirFlow®, EMS);
2. Factor desiccant material: application of a gel of concentrated aqueous mixture of hydroxybenzenesulphonic and hydroxymethoxybenzene acids and sulphuric acid (HybenX®, Epien Medical Inc.).

The four interventions were as follows:

1. Non-surgical debridement alone (C);
2. Non-surgical debridement and desiccant material (H);
3. Non-surgical debridement and glycine powder (G);
4. Non-surgical debridement, glycine powder and desiccant material (HG).

2.3 | Eligibility criteria for participants

Eligible participants were adults, aged 18 or older, suffering from peri-implantitis at an implant site. The eligible criteria are reported in Table 1. The severity of peri-implantitis was registered using a published definition of severity (Karlsson et al., 2019). With this method, an implant was identified affected by mucositis/mild peri-implantitis in the presence of BoP and bone loss ≤2 mm (group B) or

TABLE 1 Eligible criteria

Inclusion criteria
Presence of at least one screw-type titanium implant exhibiting signs of peri-implantitis: maximum probing depth from 5 to 8 mm, bleeding on probing or suppuration, and radiographic bone loss beyond crestal bone-level changes resulting from initial bone remodelling
Radiographic infra-osseous component of the defect ≤ 5 mm
Radiographic suprabony component of the defect ≤ 4mm
18 years old or older (completed skeletal growth)
Presence of at least 2 mm of keratinized mucosa
Implant loading of at least 6 months
Exclusion criteria
Patients incapable of giving informed consent
Mobile implant
Head- and neck-irradiated patients
Patients undergoing chemo- or immunosuppressive therapy over the previous 5 years
Patients treated or undergoing treatment with intravenous amino-bisphosphonates
Patients with poor oral hygiene and motivation
Untreated periodontitis
Uncontrolled diabetes
Pregnancy and lactating period
Substance abusers
Allergy to chlorhexidine or phenolic or sulphur compounds
Smoking more than 20 cigarettes per day or the equivalent
Patients unable to attend the 3-year follow-up

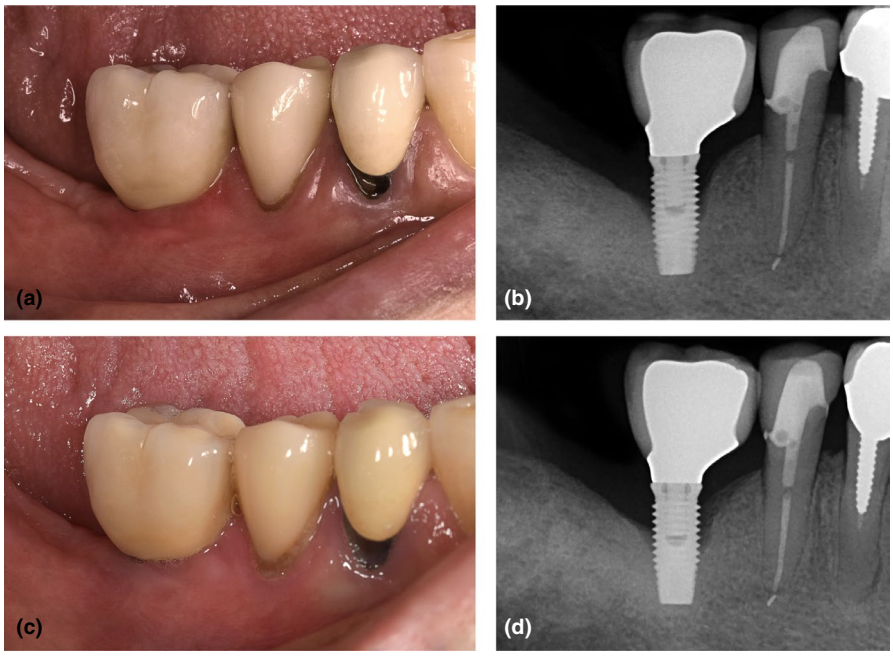

FIGURE 1 (a) Clinical baseline of a patient treated with HG. (b) Radiographic baseline of a patient treated with HG. (c) Clinical follow-up of a patient treated with HG. (d) Radiographic follow-up of a patient treated with HG

affected by moderate/severe peri-implantitis in the presence of BoP and bone loss >2 mm (group C) (Karlsson et al., 2019).

2.4 | Interventions

Before treatment, all patients were enrolled in an oral hygiene programme and received supramucosal/gingival professional implant/tooth cleaning using a rubber cup and polishing paste as well as oral hygiene instructions according to individual needs. Patients rinsed with chlorhexidine mouthwash (0.2%) for 1 min prior to the intervention.

The procedures were performed following prosthetic removal. The envelope with the randomization code was opened after the removal of the prosthetic reconstructions.

Only one implant was selected in each patient for this study. The operator was FB.

Anaesthesia was not performed. The implant surface was debrided with ultrasonic dedicated scalers until the clinician deemed the area properly cleaned. If both the glycine powder and the desiccant material should have been used, they were applied in sequence (first glycine powder) after the debridement procedure.

As regards the gel of the desiccant material, it was positioned inside the peri-implant pocket and it was left in situ for 30 s; then, the material was removed by thorough physiological saline solution of the treated area. The physiological saline solution was also used after the procedure of the glycine powder.

After the treatment, patients were instructed to refrain from mechanical plaque removal in the area of intervention for 1 week and to use chlorhexidine mouthrinse (0.12%) twice a day for 15 days. Gentle wiping with a soft brush was started after the first week. Patients were advised to avoid smoking during the prescribed recovery period.

Patients were seen at 1 week, 1 month, 3 months and 6 months for maintenance with supragingival prophylaxis (Figure 1).

2.5 | Outcome measures

Primary outcome variable was the reduction in probing depth (PD reduction).

Secondary outcome variables were as follows: implant failure, complications (including re-treatments), change in radiographic bone level, VAS for pain, VAS for satisfaction, Oral Health Impact Profile (OHIP-14), variation in keratinized tissue (KT), variation in recession (Rec reduction), bleeding on probing reduction (BoP reduction), variation in clinical attachment level, a composite index of success and microbiological sampling.

Outcome measurements were registered by an assessor blinded to the treatment selected (EG). The examiner was calibrated and subjected to an intra-rater agreement test for probing depth, resulting in an intra-class correlation coefficient of 0.84, considered excellent (Fleiss, 1986, Merli et al., 2014). The analysis was carried out before the start of the study by performing repeated measurements after two hours on 420 sites. Systematic error between replicate measurements using Dahlberg estimator resulted in 0.5 mm (Springate, 2012).

Probing depth, recession depth and bleeding on probing were measured at four sites of the implant (vestibular, mesial, distal and lingual) using a PCP-15 periodontal probe (Hu-Friedy) at baseline and after 6 months. Variation in clinical attachment level (CAL variation) was registered adding the values of probing depth and recession depth. KT was assessed at mid-buccally with a PCP-15 periodontal probe.

Peri-implant marginal bone-level change was assessed on periapical intra-oral radiographs taken with the parallel technique at baseline and at 6-month follow-up. Reference points for the linear measurements were the coronal margin of the implant collar and the most coronal point of bone-to-implant contact. The mesial and distal sites were averaged for each implant. The radiographic measurements were relativized considering the length of the implant. The radiographs were examined by one examiner (Marco M). The examiner was calibrated and subjected to an intra-rater agreement test, resulting in an intra-class correlation coefficient of 0.98, considered excellent (Fleiss, 1986). The analysis was carried out by performing repeated measurements after two weeks on 42 sites of implants not included in the study. Systematic error between replicate measurements using Dahlberg estimator resulted in 0.16 mm (Springate, 2012).

A composite index of success criteria was based on implant survival, absence of a peri-implant site with PD ≥ 5 mm with concomitant BoP or suppuration and no bone loss between baseline and 6-month radiograph (Heitz-Mayfield et al., 2018).

VAS for patient pain was registered immediately after the procedure, after 1 week and after 6 months. The VAS was from 0 (no pain) to 10 (the worst possible pain). VAS for patient satisfaction was registered after 6 months. The VAS was from 0 (completely dissatisfied) to 10 (completely satisfied).

Patient oral health was based on the questionnaire of the Oral Health Impact Profile (OHIP-14), which was compiled prior to treatment and 6 months after treatment (Slade, 1997). The Italian version of OHIP-14 was used (Franchignoni et al., 2010).

2.6 | Microbiological sampling

Subgingival bacterial samples were taken immediately before debridement and at 1 and 6 months after debridement. The procedure was carried out before the probing assessment.

The area to be sampled was isolated with cotton rolls to prevent contamination from saliva. Samples were taken with two sterile endodontic paper points of size 70 at the site of the treated titanium implant with the deepest probing depth. The paper points were inserted at the bottom of the pocket and kept in situ for 20 s (Persson, Samuelsson, Lindahl, & Renvert, 2010). The microbiological procedures were performed in a masked mode at a specialized laboratory (LAB Srl) and are reported in Appendix 1.

2.7 | Sample size

To detect a difference between treatments of 0.5 mm in pocket depth change between baseline and 6-month follow-up, standard deviation of 0.6 mm in agreement with the study of Sahm, Becker, Santel, & Schwarz, 2011, with a two-side 5% significance level and power of 80%, a sample size of 64 patients was necessary, given an anticipated drop-out rate of 20%. This sample size determination follows the common procedure in factorial trials based on target effect size for each of the factors with their respective controls (Montgomery et al., 2003). The sample size is based on the assumption that there is no interaction between the factors (Montgomery et al., 2003).

2.8 | Random sequence

For allocation of the participants, two computer-generated lists of random numbers were used. Two lists were generated, one for the factor glycine powder and one for the factor desiccant material. A blocked randomization was applied to include 32 patients in each level of each factor, resulting in 16 patients in each intervention (C, H, G and HG).

2.9 | Allocation concealment

The allocation sequence was concealed from the researcher (MN) enrolling and assessing participants in sequentially numbered, opaque, sealed and stapled envelopes. The envelopes were opened by the operator (FB) only after the removal of the prosthetic reconstructions.

2.10 | Blinding

Whereas the operator (FB) and the patient were aware of the allocated arm, outcome assessors were kept blinded to the allocation. The complications were registered and treated by the operator (FB) in a non-blinded mode.

2.11 | Statistical methods

Descriptive statistics were performed (MN) using mean and standard deviation for quantitative data and frequency and percentage for qualitative data.

The statistical unit of the analysis was the patient. For radiographic bone level and for pocket depth, the mean among the sites was considered and presented at patient level.

Analysis of covariance (ANCOVA) was performed for change in radiographic bone level, change in pocket depth, recession reduction and change in clinical attachment level. Value at baseline was the covariate, and a two-factorial model was constructed. The two factors were the glycine and the desiccant material (HybenX). A secondary analysis was performed to test the interaction term that was added to the model only if significant (Montgomery et al., 2003). If the interaction term was significant, the Tukey HSD post hoc test was used to confirm where the difference occurred between interventions.

The qualitative variables (implant failure and complications) were subjected to the logistic regression model using the two-factorial model. The interaction term was added to the model only if significant. Odds ratio (OR) was used to estimate the results.

General linear models using the two-factorial model were performed for chair time, VAS pain immediately after intervention, VAS pain after 1 week and VAS satisfaction. The interaction term was added to the model only if significant.

ANCOVA models with previous logarithmic transformation were used for the microbiological analyses.

Graphical residual analyses were performed to test the assumption of homoscedasticity and normality.

The intention-to-treat method of analysis was utilized.

In the light of the scarce literature on non-surgical therapy in the management of peri-implantitis, an additional analysis was performed pooling the values of the four treatments. Differences between baseline and 6 months were tested for clinical variables with *t* tests for paired data and for microbiological variables with Wilcoxon paired test. The significant level was set at 0.05. The statistical software was JMP v. 13 (SAS Institute Inc.).

3 | RESULTS

3.1 | Participant flow and recruitment

Patients were recruited and treated in a private clinic (Rimini, Italy) from March 2016 to July 2018. The last 6-month follow-up was done in January 2019.

Sixty-four patients were consecutively enrolled in the trial and randomized in a factorial two-factor two-level non-surgical study: 32 patients were randomized to glycine, and 32 patients were randomized to the desiccant material so that four interventions of 16 patients each were established.

For explicative reasons, the descriptive results are reported considering the factors and their combinations in the four cells of the study. Nevertheless, the inferential analysis considers only the main factors (glycine powder and the desiccant material), while the interaction was investigated as a secondary analysis.

Two patients belonging to the HG intervention dropped out after debridement, one of them moved away, and the other declined to return to the follow-up visit. One patient belonging to the H intervention dropped out at 3 months for general health problems, and one patient belonging to the G intervention dropped out at 5 months and declined to return to the follow-up visit.

There were five deviations of the protocol: one patient with 0 mm of keratinized mucosa was included and treated in the H intervention; two patients with maximum pocket depth of 9 mm were

included and treated in the G intervention; two patients with a suprabony component of the defect of 5 mm were included (one in the H and one in the HG intervention). In addition, four patients refused to take the radiographs at 6-month follow-up (one patient of the G intervention and three patients of the H intervention).

3.2 | Baseline data

The main baseline patient characteristics are shown in Table 2. There were no apparent baseline imbalances between the four interventions.

3.3 | Outcomes and estimation

The main patient characteristics after treatment are shown in Table 3.

Two implants failed in the G intervention due to the progression of their peri-implantitis, and no implant failed in the C, H or HG intervention.

Pocket depth reduction was higher in patients treated with desiccant material (estimated difference: 0.5 mm; 95% CI from 0.1 to 0.9 mm, $p = .0229$), while there was no difference for the patients treated with glycine powder (estimated difference: 0.1 mm; 95% CI from −0.3 to 0.5 mm, $p = .7333$) (Table 4). The covariate (pocket depth at baseline) was not significant ($p = .17$).

One complication was observed in the C intervention (swelling after one month treated with chlorhexidine gel and then hesitated in vestibular gingival hypertrophy), three complications were observed in the H intervention (two patients had swelling after one week and were untreated; another patient had swelling and was treated with antibiotics), four complications were observed in the G intervention (two patients had swelling at 3 months, treated with antibiotics and then hesitated to peri-implantitis and implant failure, one patient had inflammation and profuse bleeding at the treated site at 3 months treated with chlorhexidine gel and the other patient had peri-implantitis at 6 months treated with a surgical approach), and two complications occurred in the HG intervention (two patients had swelling, one of them one week after intervention treated with chlorhexidine gel and the other one month after intervention treated with antibiotics).

TABLE 2 Patient characteristics at baseline

Variable	C *N* = 16	H *N* = 16	G *N* = 16	HG *N* = 16
Age [years]	64.5 (8.3)	60.3 (10.7)	66.4 (9.4)	60.3 (8.5)
Gender [female]	9 (56%)	12 (75%)	9 (56%)	10 (62%)
Site maxillae	7 (44%)	4 (25%)	9 (56%)	11 (69%)
Site molars	9 (56%)	9 (56%)	5 (31%)	7 (44%)
Smokers	3 (19%)	4 (25%)	2 (12%)	4 (25%)
Edentulous	1 (6%)	1 (6%)	3 (19%)	6 (38%)
OHIP-14	6.2 (6.5)	4.4 (3.6)	6.1 (5.4)	3.3 (3.3)
Implant age [years]	10.1 (5.0)	9.1 (5.3)	9.7 (4.3)	8.1 (3.3)
Nobel implant	9 (56%)	7 (44%)	7 (44%)	8 (50%)
Thommen implant	3 (19%)	4 (25%)	4 (25%)	6 (37%)
RxMeanBD [mm]	3.3 (1.2)	3.9 (1.2)	3.6 (1.7)	3.6 (0.9)
KT [mm]	3.2 (1.1)	2.6 (1.0)	2.9 (1.4)	3.1 (1.9)
PDMean [mm]	4.4 (1.1)	5.0 (1.2)	5.1 (1.5)	4.9 (1.1)
RecMean [mm]	0.1 (0.1)	0.4 (0.5)	0.2 (0.9)	0.1 (0.2)
CALMean [mm]	4.4 (1.0)	5.4 (1.2)	5.4 (1.6)	5.0 (0.9)
BoP Sites	3.3 (0.8)	2.9 (1.3)	3.6 (0.8)	3.6 (0.8)
Presence of suppuration	4 (25%)	3 (19%)	4 (25%)	6 (37%)
Severity (Group B)	2 (13%)	1 (6%)	3 (19%)	1 (6%)
Severity (Group C)	14 (87%)	15 (94%)	13 (81%)	15 (94%)

Note: C: only non-surgical debridement; H: non-surgical debridement and desiccant material; G: non-surgical debridement and glycine powder; HG: non-surgical debridement, desiccant material and glycine powder. Between parenthesis standard deviations for quantitative variables and percentage for qualitative variables. RxMeanBD: radiographic mean bone defect. KT: keratinized tissue. PD: pocket depth. CAL: clinical attachment level. BoP Sites: number of sites per implant with bleeding on probing. Group B: presence of BoP and bone loss ≤ 2 mm. Group C: presence of BoP and bone loss > 2 mm.

TABLE 3 Patient characteristics after randomization and at 6 months of follow-up

Variable	C $N = 16$	H $N = 15$	G $N = 13$	HG $N = 14$	Total $N = 58$
Failures	0 (0%)	0 (0%)	2 (13%)[a]	0 (0%)	2/60 (3%)
Complications	1 (6%)	3 (20%)	4 (27%)[a]	2 (14%)	10/60 (17%)
VAS pain (during treatment)	2.1 (2.1)	3.3 (2.7)[b]	3.9 (2.7)[b]	5.0 (2.5)[b]	3.6 (2.7) $N = 64$
VAS pain (after 1 week)	0.6 (1.0)	0.9 (1.6)[b]	1.8 (2.5)[b]	1.7 (2.2)[b]	1.2 (1.9) $N = 64$
RxMeanBD [mm]	3.1 (1.5)	4.0 (1.8)[c]	4.0 (1.8)[c]	3.5 (1.0)	3.6 (1.5)
RxMeanBD reduction [mm]	0.2 (0.8)	−0.1 (0.9)[c]	−0.2 (1.0)[c]	−0.1 (0.7)	−0.0 (0.8)
KT [mm]	3.2 (1.4)	2.1 (1.3)	2.4 (1.8)	3.1 (1.9)	2.7 (1.6)
KT difference [mm]	0.0 (0.9)	−0.5 (0.9)	−0.7 (1.3)	−0.2 (0.7)	−0.3 (1.0)
PDMean [mm]	4.2 (1.3)	4.5 (1.2)	4.8 (1.3)	4.0 (1.2)	4.4 (1.3)
PD reduction [mm]	0.2 (0.7)	0.5 (0.9)	0.1 (0.8)	0.8 (0.8)	0.4 (0.8)
RecMean [mm]	0.1 (0.2)	0.3 (0.5)	0.3 (0.7)	0.2 (0.4)	0.2 (0.5)
Rec reduction [mm]	−0.0 (0.2)	0.1 (0.3)	0.0 (0.4)	−0.1 (0.3)	−0.0 (0.3)
CALMean [mm]	4.3 (1.3)	4.9 (1.3)	5.2 (1.5)	4.2 (1.0)	4.6 (1.3)
CAL reduction [mm]	0.1 (0.6)	0.6 (0.9)	0.1 (0.9)	0.7 (0.8)	0.4 (0.8)
BoP sites	2.9 (0.8)	2.5 (1.7)	2.8 (1.3)	2.7 (1.3)	2.8 (1.3)
BoP site reduction	0.4 (0.9)	0.5 (1.8)	0.7 (1.3)	0.8 (1.2)	0.6 (1.3)
Presence of suppuration	2 (12%)	2 (13%)	2 (15%)	0 (0%)	6/58 (10%)
Success criteria	6 (37%)	3 (25%)[c]	2 (14%)[d]	6 (43%)	17/56 (30%)
VAS pain (after 6 months)	0.7 (1.5)	0.5 (2.1)	0.2 (0.4)	0.4 (0.9)	0.5 (1.4)
OHIP-14	4.4 (5.7)	4.6 (6.5)	2.6 (3.8)	2.9 (6.3)	3.7 (5.6)
OHIP-14 reduction	1.8 (6.1)	0.1 (4.2)	4.0 (6.4)	0.0 (5.6)	1.4 (5.7)
VAS satisfaction	6.9 (2.6)	7.8 (2.6)	7.5 (3.0)	8.2 (2.5)	7.6 (2.6)

Note: C: only non-surgical debridement; H: non-surgical debridement and desiccant material; G: non-surgical debridement and glycine powder; HG: non-surgical debridement, desiccant material and glycine powder. Between parenthesis standard deviations for quantitative variables and percentage for qualitative variables. RxMeanBD: radiographic mean bone defect. KT: keratinized tissue. PD: pocket depth. CAL: clinical attachment level. BoP Sites: number of sites per implant with bleeding on probing.

[a]15 patients.

[b]16 patients.

[c]12 patients.

[d]14 patients.

The complications were not significantly different between the treatments. For complications, the glycine factor had an OR = 1.8 (95% CI from 0.4 to 7.0; $p = .4177$) and the desiccant factor had an OR = 1.1 (95% CI from 0.3 to 4.3; $p = .9068$).

VAS for pain during intervention was higher for patients treated with glycine powder (difference: 1.8; 95% CI from 0.5 to 3.0; $p = .0056$), while it was not significant for patients treated with desiccant material (difference: 1.2; 95% CI from −0.1 to 2.4; $p = .0602$).

VAS for pain after 1 week was higher for patients treated with glycine powder (difference: 1.1; 95% CI from 0.1 to 2.0; $p = .0339$), while it was not significant for patients treated with the desiccant material (difference: 0.1; 95% CI from −0.8 to 1.1; $p = .7635$).

No differences were found in radiographic bone-level reduction at 6 months for patients treated with the desiccant material (estimated difference: −0.1 mm; 95% CI from −0.6 to 0.4 mm, $p = .6934$) and for patients treated with glycine powder (estimated difference:

TABLE 4 Detailed results of the primary variable, PD reduction (mm)

		Desiccant	Factor		Difference 95% CI p-value
		No	Yes	Total	
Glycine	No	0.2 (0.7) *N* = 16	0.5 (0.9) *N* = 15	0.3 (0.8) *N* = 31	0.1 From −0.3 to 0.5 *p* = .7333
Factor	Yes	0.1 (0.8) *N* = 13	0.8 (0.8) *N* = 14	0.4 (0.8) *N* = 27	
Total		0.1 (0.7) *N* = 29	0.6 (0.8) *N* = 29		
Difference 95% CI p-value		0.5 From 0.1 to 0.9 *p* = .0229			

Note: Interaction is not significant ($p = .25$). Standard deviation in parentheses.

−0.2 mm; 95% CI from −0.6 to 0.3 mm, $p = .4408$). The covariate (bone level at baseline) was not significant ($p = .54$).

As regards KT modifications, there were no differences at 6 months for patients treated with desiccant material (estimated difference: −0.1 mm; 95% CI from −0.6 to 0.4 mm, $p = .7630$) and for patients treated with glycine powder (estimated difference: −0.2 mm; 95% CI from −0.7 to 0.4 mm, $p = .5202$).

There were no differences in recession reduction at 6 months for patients treated with desiccant material (estimated difference: −0.0 mm; 95% CI from −0.2 to 0.1 mm, $p = .8003$) and for patients treated with glycine powder (estimated difference: −0.1 mm; 95% CI from −0.2 to 0.1 mm, $p = .3474$). The covariate (recession at baseline) was significant ($p < .0001$), indicating a small increase in recession for implants that had no baseline recessions.

CAL reduction was higher in patients treated with the desiccant material (estimated difference: 0.5 mm; 95% CI from 0.1 to 0.9 mm, $p = .0245$), while there was no difference for the patients treated with glycine powder (estimated difference: 0.0 mm; 95% CI from −0.4 to 0.4 mm, $p = .9988$). The covariate (CAL at baseline) was not significant ($p = .19$).

For BoP reduction, there were no differences at 6 months for patients treated with the desiccant material (estimated difference: 0.2; 95% CI from −0.5 to 0.8 mm, $p = .5701$) and for patients treated with glycine powder (estimated difference: 0.1 mm; 95% CI from −0.5 to 0.8 mm, $p = .6917$).

The percentage of success criteria was low in all the interventions ranging from 14% to 43% (Table 3). The implants with success criteria were not significantly different between the treatments. For the success criteria, the glycine factor had an OR = 0.8 (95% CI from 0.3 to 2.2; $p = .73$) and the desiccant factor had an OR = 1.5 (95% CI from 0.5 to 4.7; $p = .50$).

VAS for pain after 6 months was not significant for patients treated with desiccant material (difference: −0.0; 95% CI from −0.7 to 0.7; $p = .9846$) and for patients treated with glycine powder (difference: −0.3; 95% CI from −1.0 to 0.4; $p = .4225$).

For OHIP-14 reduction, there were no differences at 6 months for patients treated with desiccant material (estimated difference: −1.5; 95% CI from −4.3 to 1.2 mm, $p = .2655$) and for patients treated with glycine powder (estimated difference: 1.4 mm; 95% CI from −1.3 to 4.1 mm, $p = .3151$).

VAS for satisfaction after 6 months was not significant for patients treated with desiccant material (difference: 0.7; 95% CI from −0.6 to 2.2; $p = .2479$) and for patients treated with glycine powder (difference: 0.5; 95% CI from −0.9 to 1.9; $p = .5036$).

The interaction test was not significant for all the analyses.

3.4 | Ancillary analysis (bacterial counts)

Means and standard deviations for bacterial counts are reported in Tables 5–7. Microbiological counts were not different between interventions (all p-values on the logarithmic transformation were >0.05).

The interaction test was not significant for all the analyses.

3.5 | Additional analysis on pooled data

Means and standard deviation for pooled data are reported in Table 3. Differences between baseline and 6 months were find for KT ($p = .009$), PD ($p = .0005$), CAL ($p = .001$) and BoP sites ($p = .002$), while there were no differences for BD ($p = .74$), Rec ($p = .52$) and OHIP ($p = .06$). Regarding the microbiological data, a reduction was found only for *Porphyromonas gingivalis* count ($p = .02$).

4 | DISCUSSION

The aim of this 6-month factorial randomized controlled trial was to compare the efficacy of two different non-surgical therapies (abrasive powder amino acid glycine and a desiccant material) and their combinations in the treatment of peri-implantitis. The two therapies were simultaneously evaluated using a factorial design (Montgomery et al., 2003,). This design is more efficient in respect to a parallel trial, assuming the absence of interaction between the two treatments (Esposito & Nieri, 2016; Pandis, Walsh, Polychronopoulou, Katsaros, & Eliades, 2014).

TABLE 5 Bacteria count at baseline. Mean (standard deviation)

Bacteria	C *N* = 16	H *N* = 16	G *N* = 16	HG *N* = 16
Total bacteria	111,042 (141,310)	236,812 (288,326)	319,594 (313,900)	233,650 (396,180)
Median [IQR]	40,941 [10,319; 148,729]	104,726 [17,202; 453,568]	286,416 [23,467; 564,604]	86,988 [19,225; 295,280]
Aggregatibacter actinomycetemcomitans	0 (0)	1,353 (5,411)	309 (1,077)	0 (0)
Median [IQR]	0 [0; 0]	0 [0; 0]	0 [0; 0]	0 [0; 0]
Porphyromonas gingivalis	7,697 (30,361)	12,047 (35,098)	17,596 (51,343)	393 (1,107)
Median [IQR]	0 [0; 201]	0 [0; 198]	0 [0; 968]	0 [0; 193]
Tannerella forsythia	2,248 (6,054)	710 (1,428)	1,449 (2,966)	244 (525)
Median [IQR]	30 [0; 840]	0 [0; 839]	185 [0; 828]	0 [0; 212]
Treponema denticola	1,515 (4,949)	6,670 (17,135)	5,027 (18,364)	1,113 (4,289)
Median [IQR]	0 [0; 181]	0 [0; 1,595]	0 [0; 197]	0 [0; 59]
Fusobacterium nucleatum	8,853 (26,644)	22,356 (44,295)	70,970 (155,190)	20,412 (67,091)
Median [IQR]	411 [68; 2,795]	1,261 [181; 1,977]	1,902 [565; 91,623]	365 [0; 3,344]
Campylobacter rectus	355 (822)	3,232 (11,110)	3,240 (8,500)	4,889 (18,917)
Median [IQR]	75 [0; 245]	0 [0; 521]	0 [0; 535]	0 [0; 514]

Note: C: only non-surgical debridement; H: non-surgical debridement and desiccant material; G: non-surgical debridement and glycine powder; HG: non-surgical debridement, desiccant material and glycine powder; IQR: inter-quartile range.

The null hypothesis was rejected since pocket depth reduction was greater in patients treated with desiccant material than in those not treated with the desiccant material; nevertheless, there was no difference between patients treated with glycine powder and those treated without the use of glycine powder. There were only two failures in the G intervention and no failures in the other interventions. The difference between therapies was not significant, but for this variable, the study is underdimensioned at least at 6-month follow-up. On the basis of currently available RCTs, there is insufficient evidence to support that any particular non-surgical treatment for peri-implantitis has shown better performance than others in term of failures or complications (Esposito et al., 2012; Muthukuru et al., 2012).

In this study, the patients were treated without the use of local anaesthesia. Light pain during treatment was reported by the patients. It was more pronounced when the abrasive powder amino acid glycine was used. The pain VAS was more pronounced when the abrasive powder amino acid glycine was used even in the subsequent days, as reported at the one-week follow-up.

On average, the change in radiographic bone level was near to zero for each intervention. The period of 6 months is too short to highlight considerable modification of the bone levels. Consequently, there were no differences between the investigated treatments.

Pocket depth reduction and clinical attachment reduction were higher in patients treated with desiccant material (estimated difference: 0.5 mm), while there was no difference for the patients treated with glycine powder. The difference obtained for the patients treated with desiccant material is precisely that which had considered the minimal important difference in the "a priori" sample size determination. The use of desiccant material would seem promising to treat peri-implantitis; nevertheless, for the other clinical variables, such as change in bone level, recession, keratinized tissue or bleeding on probing, no differences were found. This desiccant agent has showed greater reduction in probing depth and bleeding on probing compared to scaling and root planing alone in patients with chronic periodontitis (Isola et al., 2018). At the moment, there are no other RCTs that used the desiccant material on peri-implantitis: for these reasons, it is unclear the value of this desiccant agent in the clinical outcome of non-surgical treatment of peri-implantitis.

In the present study, the glycine powder (air polishing) did not reveal any major improvement in clinical variables, as probing depth reduction or bleeding on probing reduction. These results are partially discordant with respect to a meta-analysis that showed an improved efficacy of glycine powder in reducing BOP score compared with control treatments after non-surgical treatment of peri-implantitis, while PD reduction failed to reach statistical significance (Schwarz, Becker, & Renvert, 2015).

The difference in mucosal recession between baseline and 6 months was near null for each treatment. This result was similar to that of other studies (Sahm et al., 2011).

The present study was also designed to acquire information about the combination of the glycine powder and the desiccant material. Therefore, a factorial design was implemented. The interaction term was insignificant for all the clinical variables. No clinical advantage of the glycine powder and the desiccant material combination was highlighted. For this reason and considering also the cost, combining different therapies in non-surgical peri-implantitis would not seem appropriate. From a cost point of view, the most

TABLE 6 Bacteria count at 1-month follow-up. Mean (standard deviation)

Bacteria	C *N* = 16	H *N* = 16	G *N* = 16	HG *N* = 14
Total bacteria	178,716 (282,550)	627,646 (1,934,812)	223,726 (335,421)	210,057 (337,342)
Median [IQR]	26,185 [2,963; 172,167]	108,260 [10,540; 328,104]	89,350 [15,143; 274,087]	73,172 [14,499; 213,701]
Aggregatibacter actinomycetemcomitans	0 (0)	299 (1,198)	0 (0)	0 (0)
Median [IQR]	0 [0; 0]	0 [0; 0]	0 [0; 0]	0 [0; 0]
Porphyromonas gingivalis	31 (82)	48,644 (194,521)	1,848 (7,281)	2,460 (8,787)
Median [IQR]	0 [0; 0]	0 [0; 48]	0 [0; 0]	0 [0; 124]
Tannerella forsythia	517 (1,308)	3,173 (10,554)	653 (1,366)	3,148 (10,689)
Median [IQR]	0 [0; 79]	0 [0; 368]	0 [0; 657]	87 [0; 513]
Treponema denticola	254 (874)	9,127 (33,545)	1,151 (3,155)	787 (2,806)
Median [IQR]	0 [0; 0]	0 [0; 74]	0 [0; 52]	0 [0; 23]
Fusobacterium nucleatum	10,587 (30,148)	36,989 (85,306)	28,798 (51,692)	10,964 (25,201)
Median [IQR]	408 [0; 1,483]	1,084 [19; 28,423]	1,923 [0; 35,387]	737 [141; 7,962]
Campylobacter rectus	175 (325)	5,743 (22,169)	759 (2,324)	1,994 (4,242)
Median [IQR]	0 [0; 212]	0 [0; 180]	0 [0; 534]	0 [0; 1,633]

Note: C: only non-surgical debridement; H: non-surgical debridement and desiccant material; G: non-surgical debridement and glycine powder; HG: non-surgical debridement, desiccant material and glycine powder; IQR: inter-quartile range.

TABLE 7 Bacteria count at 6-month follow-up. Mean (standard deviation)

Bacteria	C *N* = 16	H *N* = 15	G *N* = 13	HG *N* = 14
Total bacteria	316,164 (479,042)	436,721 (606,755)	793,718 (2,084,166)	270,615 (267,015)
Median [IQR]	119,686 [27,168; 364,486]	148,882 [13,763; 810,167]	41,819 [7,341; 507,471]	202,022 [16,662; 482,064]
Aggregatibacter actinomycetemcomitans	0 (0)	0 (0)	0 (0)	0 (0)
Median [IQR]	0 [0; 0]	0 [0; 0]	0 [0; 0]	0 [0; 0]
Porphyromonas gingivalis	3,546 (12,545)	6,942 (25,044)	229 (755)	469 (1,567)
Median [IQR]	0 [0; 42]	0 [0; 0]	0 [0; 46]	0 [0; 113]
Tannerella forsythia	1,931 (4,495)	1,899 (3,588)	568 (1,337)	401 (1,295)
Median [IQR]	0 [0; 1,663]	0 [0; 1,352]	66 [0; 203]	0 [0; 119]
Treponema denticola	3,034 (8,720)	3,637 (10,785)	721 (2,240)	103 (189)
Median [IQR]	0 [0; 472]	0 [0; 0]	0 [0; 36]	0 [0; 146]
Fusobacterium nucleatum	20,016 (40,356)	12,932 (24,437)	150,713 (522,176)	14,556 (35,020)
Median [IQR]	153 [0; 9,388]	2,998 [0; 21,843]	947 [210; 10,126]	890 [0; 2,175]
Campylobacter rectus	457 (969)	1,958 (4,926)	5,836 (19,123)	6,199 (22,867)
Median [IQR]	0 [0; 334]	0 [0; 622]	0 [0; 241]	0 [0; 157]

Note: C: only non-surgical debridement; H: non-surgical debridement and desiccant material; G: non-surgical debridement and glycine powder; HG: non-surgical debridement, desiccant material and glycine powder; IQR: inter-quartile range.

appropriate treatments proposed in this RCT are non-surgical treatment alone or the use of the desiccant agent.

Currently, there are few studies that report on the microbiological outcome following non-surgical treatment of peri-implantitis (Persson, Roos-Jansåker, Lindahl, & Renvert, 2011; Persson et al., 2010; Schwarz et al., 2015). No microbiological differences between baseline and 6-month samples were found for any species or between treatment study methods in peri-implantitis (Persson et al., 2010). Similarly, in the present study no differences were detected between interventions in total bacterial load and for the counts of the species investigated. Only a small decrease in count of *P. gingivalis* was observed in the pooled analysis. This may be due to a limitation of both devices to effectively remove hard deposits such as calculus from implant surfaces (Schwarz et al., 2015).

This study started in March 2016 and could not take into consideration the new case definition published in the recent World Workshop on the classification of peri-implant disease (Berglundh et al., 2018). Nevertheless, the presence of bleeding and/or suppuration on gentle probing and the presence of bone loss beyond crestal bone-level changes resulting from initial bone remodelling are equally considered in our case definition and in the World Workshop classification (Berglundh et al., 2018). Regarding probing depth, for the diagnosis of peri-implantitis the World Workshop classification considers the increased probing depth compared to a previous examination, while in our case definition, we considered at least one peri-implant site with probing depth ≥5 mm. Furthermore, in the inclusion criteria, we placed maximum limits for PD, infrabony and supra-osseous components of the defects because a surgical treatment was proposed to patients who had major defects.

In the light of the scarce literature on non-surgical therapy in the management of peri-implantitis, a further focus was performed pooling the values for the clinical, radiographic and microbiological outcomes of the four different interventions. In general, a reduction of KT, PD, CAL, BoP and *P. gingivalis* count was observed between baseline and 6 months. The pooled composite index of success was 30%.

A limit of this study should be the limited sample size to detect interactions between the two treatments. Nevertheless, the sample size was adequate to highlight differences in the main factors. Another limit is the short follow-up. Important variables such as implant failure and radiographic changes could be assessed in the medium or long follow-up. A follow-up of 2 years was scheduled for this study.

In conclusion, in this 6-month follow-up study, pocket reduction was more pronounced in patients using the desiccant material. Pain was higher in patients using glycine. For many clinical and microbiological variables, there were no differences between treatments. All the interventions resulted in low success rate.

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CONFLICT OF INTEREST

The authors report no conflict of interest.

AUTHOR CONTRIBUTIONS

Mauro Merli and Giovanpaolo Pini-Prato conceived the idea of the study and led the writing. Francesco Bernardelli performed the interventions. Erica Giulianelli and Marco Merli collected the clinical data. Francesco Carinci carried out the microbiological procedures. Giorgia Mariotti led the writing. Michele Nieri analysed the data and led the writing.

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APPENDIX 1

REAL-TIME POLYMERASE CHAIN REACTION

DNA was extracted and purified using standard protocols that include two consecutive incubations with lysozyme and proteinase K, followed by spin-column purification.

Primers and probe oligonucleotide were designed based on 16S rRNA gene sequences of the Human Oral Microbiome Database (HOMD 16S rRNA RefSeq version 10.1) counting 845 entries. All the sequences were aligned in order to find either consensus sequence or less conservate spots. Three real-time polymerase chain reaction (PCR) runs were performed for each sample. The first reaction quantified the total amount of bacteria using two degenerate primers and a single probe matching a highly conservated sequence of the 16S ribosomal RNA gene. The second reaction detected and quantified the three red complex bacteria, that is *Porphyromonas gingivalis*, *Tannerella forsythia* and *Treponema denticola*, in a multiplex PCR. The third reaction detected and quantified *Aggregatibacter actinomycetemcomitans*, *Fusobacterium nucleatum* and *Campylobacter rectus*, in a multiplex PCR. Each reaction included a total of six primers and three probes that were highly specific for each species.

Oligonucleotide concentrations and PCR conditions were optimized to ensure sensitivity, specificity and no inhibition in the case of unbalanced target amounts. Absolute quantification assays were performed using the Applied Biosystems 7,500 Sequence Detection System. The amplification profile was initiated by a 10-min incubation period at 95°C to activate polymerase, followed by a two-step amplification of 15 s at 95°C and 60 s at 57°C for 40 cycles. All these experiments were performed including non-template controls to exclude reagents contamination. Plasmids containing synthetic DNA target sequences (Eurofin MWG Operon, Ebersberg, Germany) were used as a standard for the quantitative analysis. Standard curves for each target were constructed in a triplex reaction, by using a mix of the same amount of plasmids, in serial dilutions ranging from 10^1 to 10^7 copies. There was a linear relationship between the threshold cycle values plotted against the log of the copy number over the entire range of dilutions. The copy numbers for individual plasmid preparations were estimated using the Thermo NanoDrop spectrophotometer.

The absolute quantification of total bacterial genome copies in samples allowed for the calculation of relative amount of red complex species. To prevent samples and polymerase chain reaction contamination, plasmid purification and handling were performed in a separate laboratory with dedicated pipettes.