The granulation tissue preservation technique in regenerative periodontal surgery—a randomized controlled clinical trial

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Abstract

Objectives: To investigate if the application of the granulation tissue preservation technique (GTPT) in regenerative therapy of infrabony periodontal defects results in more clinical attachment level (CAL) gain and more radiographic bone gain (RBG) than the conventional resective approach 12 months after surgery.

Materials and methods: Forty patients exhibiting at least one infrabony defect with a probing pocket depth (PPD) ≥6 mm and a radiographic infrabony component (INFRAX-ray) ≥3 mm were randomly treated with the GTPT (test group) or the double-flap approach with resection of the defect-filling granulation tissue (control group). Enamel matrix derivatives were applied in both groups. Clinical and radiographic parameters were recorded at baseline (t₀), 6 months (t₁), and 12 months (t₂) after surgery. The primary outcome variable was CAL gain between t₀ and t₂.

Results: When all patients were considered, ΔCAL_t₀–t₂ did not differ significantly between the two groups (p = .160). Significant PPD reduction (test group: 4.38 ± 1.36 mm; control group: 4.06 ± 2.38 mm), CAL gain (test group: 3.75 ± 1.24 mm; control group: 2.88 ± 2.09 mm), and RBG (test group: 3.06 ± 1.74 mm; control group: 3.27 ± 2.19 mm) were achieved at t₂ in both groups. Using multivariate linear regression, PPD_t₀ and group were identified as variables with the greatest influence on ΔCAL_t₀–t₂. PPD_t₀ and INFRAX-ray were identified as variables with the greatest influence on RBG_t₀–t₂. Patients with a defect angle >22° showed significantly more CAL gain in the test group (t₀–t₁: 3.08 ± 1.38 mm; t₀–t₂: 3.62 ± 0.96 mm) than in the control group (t₀–t₁: 1.77 ± 1.54 mm; t₀–t₂: 2.18 ± 1.83 mm).

Conclusions: Regarding all patients, the study failed to show significant differences between the test and control groups. However, the GTPT appears to lead to more CAL gain in noncontaining infrabony defects.

Keywords
granulation tissue, infrabony, osseous defect, periodontitis, regeneration
1 | BACKGROUND

Minimally invasive surgical techniques have been developed to limit the extent of the surgical area, to achieve a stable primary wound closure, and thus to avoid failures in wound healing particularly in the area of the interdental papilla (Cortellini & Tonetti, 2007, 2009; Harrel, 1998; Harrel & Rees, 1995; Trombelli et al., 2009). All these flap designs have in common that the defect-filling granulation tissue is resected and discarded. However, advanced infrabony defects are often not limited to the interdental space but extend to the oral and buccal sites. In these cases, it is not sufficient to prepare only one flap, but a double flap approach must be used to ensure sufficient visibility of the root surface to be instrumented. In addition, advanced complex defects usually lack soft tissue support, which is essential for the success of the regenerative procedure. Therefore, bone substitutes are frequently used to fill the space previously occupied by granulation tissue and thus to preserve space for regeneration (Kao et al., 2015; Reynolds et al., 2003). It has been shown for different graft materials that the space created for regeneration leads to new bone formation only to a limited extent (Stavropoulos et al., 2003). Furthermore, graft materials carry the risk of microbial contamination compromising the treatment outcome.

Recently, we introduced the granulation tissue preservation technique (GTPT) for regenerative therapy of infrabony periodontal and peri-implant defects (Güney et al., 2013, 2019). Preservation of the defect-filling granulation tissue is hypothesized to serve as soft tissue support, to make the use of bone substitutes dispensable, and to enable increased wound stability particularly in the area of the interdental papilla. Moreover, the existing vascular network and the precursor cells contained in the granulation tissue can be preserved and subsequently promote wound healing. In vitro and in vivo studies have shown that cells with properties of mesenchymal stem cells reside in both inflamed periodontal and peri-implant tissues (Adam et al., 2019, 2020; Gousooupolou et al., 2021; Park et al., 2011). From this point of view, it seems to make sense to preserve the defect-filling granulation tissue in regenerative periodontal surgery. Preservation of granulation tissue was first described by Lindhe and Nyman (1985). They already stated that granulation tissue removal during access flap surgery is not a mandatory measure for creating suitable conditions for the proper healing of periodontal tissues.

The aim of the present study was to compare the GTPT with the conventional double-flap approach, in which the defect-filling granulation tissue is resected. Since soft tissue collapse was to be expected particularly in defects with missing bony support, the statistical examination focused on noncontaining defects defined by large radiographic defect angles. The hypothesis of the present study was that infrabony periodontal defects treated with the GTPT would result in more clinical attachment level (CAL) gain and more radiographic bone gain (RBG) than those treated with the conventional approach.

2 | METHODS

2.1 | Experimental design

The present clinical trial was performed in the Department of Conservative Dentistry, Periodontology, and Preventive Dentistry of Hannover Medical School (MHH) and had a prospective, randomized, controlled, and double-blinded (patients, investigator) design. In total, 40 patients with 40 deep infrabony periodontal defects received regenerative periodontal surgery using the GTPT (test group; n = 20) or the conventional double flap approach with resection of the defect-filling granulation tissue (control group; n = 20). In both groups, enamel matrix derivatives (EMDs) were used as bioactive molecules to promote periodontal regeneration. The clinical and radiographic outcomes of both groups were longitudinally followed for 12 months (Figure 1). The study was conducted in accordance with the Declaration of Helsinki as revised in 2013. The Institutional Review Board (Ethical Committee of MHH) approved the study protocol (ethics vote no. 5556) and all participants signed informed consent.

2.2 | Patient and defect eligibility

Systemically healthy individuals presenting with advanced periodontitis (Stage III or IV) and at least one isolated deep, mostly interproximal infrabony defect were considered appropriate for this trial. The inclusion criteria were

1. Probing pocket depth (PPD) ≥ 6 mm
2. Radiographic infrabony component (INFRAX-ray) ≥ 3 mm
3. Positive response to sensitivity test with refrigerant spray
4. Hygiene index (HI, see below) ≥ 40% recorded during the first step of periodontal therapy

When more than one eligible defect was available, the one with the largest INFRAX-ray (see below) was selected. The exclusion criteria comprised:

1. Heavy smokers (>10 cigarettes daily)
2. Pregnancy
3. Breastfeeding
4. Intake of antibiotics and/or nonsteroidal antirheumatic drugs within the previous 3 months
5. Systemic diseases with known impact on periodontal health

Before baseline examination, all patients received the first and second steps of periodontal therapy consisting of supragingival dental biofilm control, oral hygiene instructions, professional mechanical plaque removal, elimination of possible plaque-retentive factors, and subgingival instrumentation (Sanz et al., 2020). Six weeks after completion of the second step of periodontal therapy, clinical and radiographic baseline examinations were conducted.
2.3 | Clinical and radiographic parameters were recorded at baseline (t0) and 6-months (t1) and 12-months (t2) follow-up visits

The clinical parameters recorded at baseline (t0), 6 months (t1), and 12 months (t2) after surgery included bleeding on probing (BOP, Ainamo & Bay, 1975), full mouth bleeding score (FMBS), HI, PPD, recession depth (RED), and CAL. BOP, FMBS, PPD, RED, and CAL were assessed at six sites per tooth (mesiobuccal, buccal, distobuccal, mesio-oral, oral, disto-oral) using a WHO periodontal probe. BOP was assessed dichotomously (yes/no) and subsequently used to calculate the FMBS (Cortellini et al., 1993). The FMBS was calculated using the formula: sum of bleeding sites/sum of all sites × 100 in percent. The HI is a modification of the plaque control record (O’Leary et al., 1972) and was used to assess the quality of oral hygiene measures at home. For this purpose, all teeth were stained using a plaque revelator (Mira-2-Ton, Hager & Werken) and evaluated dichotomously (yes/no) at four sites per tooth (mesial, distal, buccal, oral). The HI was calculated using the formula: sum of plaque-free sites/sum of all sites × 100 in percent. The simplified composite outcome measure (COM) was used to evaluate the success of regenerative periodontal surgery (Trombelli et al., 2020). Briefly, the changes in PPD and CAL between t0 and t2 were assigned to the following categories in a 2 × 2-frequency table:

1. CAL gain ≥ 3 mm, residual PPD ≤ 4 mm (treatment success),
2. CAL gain ≥ 3 mm, residual PPD > 4 mm,
3. CAL gain < 3 mm, residual PPD ≤ 4 mm,
4. CAL gain < 3 mm, residual PPD > 4 mm (treatment failure).

X-rays were taken at t0, t1, and t2. X-ray film holders (Super-Bite, Kerr) were individualized using addition-curing silicone (Silagum-Putty, DMG) to warrant a reproducible beam path and best possible comparability of the radiographic images (Figure 2). The following distances were measured at sites affected by the infrabony defect using a software program for dental imaging (byzzKlinik, orangedental):

1. Distance from the cementoenamel junction to the bottom of the defect (CEJ-BD\(_{\text{X-ray}}\))
2. Distance from the cementoenamel junction to the root tip (CEJ-RT\(_{\text{X-ray}}\))

The RBG at t1 and t2 was calculated using the formula:

\[ \text{RBG}_{t0-t2} = (\text{CEJ-BD}_{\text{X-ray}})_{t0} - (\text{CEJ-RT}_{\text{X-ray}})_{t0} \times (\text{CEJ-RT}_{\text{X-ray}})_{t2} / (\text{CEJ-RT}_{\text{X-ray}})_{t2} \]

Besides this, (INFRAX\(_{\text{X-ray}}\) = distance from the bone crest to the bottom of the defect) and the radiographic defect angle were determined using the byzzKlinik software.

2.4 | Surgical procedure

Analgesia was achieved through infiltration or block anesthesia using an epinephrine-containing local anesthetic (Ultracain D-S forte, Sanofi-Aventis). Circumferential, strictly intrasulcular incisions were conducted at the defect-related teeth using a microsurgical blade.
An oblique, z-shaped incision was applied at the interdental space to connect the intrasulcular incisions of two adjacent teeth. The localization of the interdental incision was dependent on the defect morphology (residual buccal and/or oral bone wall) and the width of the interdental space. In interdental spaces with a width $\geq$ 2 mm and a preserved buccal and/or oral bone wall, the interdental incision was ideally placed on the marginal bone crest (Figure 3), which sometimes had to be localized by bone sounding. Afterward, the mucoperiosteal flap was first prepared on the side of the alveolar process, where the marginal bone crest had been preserved. This procedure allowed the granulation tissue to be separated from the root surface and the bony defect walls under direct vision. For interdental spaces with a width $<$ 2 mm or those with missing buccal and oral bone walls (one-wall defects), the interdental incision was placed centrally in the area of the proximal contact. In this procedure, the defect-filling granulation tissue was divided into a buccal and an oral portion (Video Clip S1). Generally, mobilization of the interdental soft tissues (including granulation tissue) was performed with sharp, microsurgical instruments (e.g., periotome PT1X, Goldman-Fox Knife KGF11X, Periosteal Elevator PH26M; all from Hu-Friedy) and under constant contact with the bony defect walls. The mucoperiosteal flaps were mobilized only to the extent that sufficient space was available for instrumentation of the defect-related root surface(s). This was performed with sonically driven instruments (SONICflex, KaVo Dental) and Gracey mini curettes (American Eagle Instruments, Young Innovations Europe). After conscientious removal of the microbial deposits, the clinical infrabony component (INFRA) was measured. Subsequently, the regenerative procedure was applied. The root surface was conditioned for 2 min with 24% EDTA (PrefGel®, Straumann) and irrigated with sterile isotonic sodium chloride solution. After careful air-drying, EMDs (Emdogain®, Straumann) were applied onto the root surface(s). The mucoperiosteal flaps were repositioned and fixed at the base of the interdental papilla with interrupted sutures (GORE-TEX Suture CV-6, W. L. Gore & Associates). In the test group, special focus was placed on the exact repositioning of the granulation tissue into its original position within the infrabony defect (Figure 3). In the control group, the granulation tissue was

![Figure 2](image_url)
completely resected before the interdental papillae were repositioned and fixed. Finally, the operating area was gently compressed with saline-soaked gauze for 1 min.

3 | POSTOPERATIVE CARE

Patients were instructed to spare the surgical area, to refrain from mechanical plaque control, and to use instead a mouth rinse containing 0.2% chlorhexidine digluconate twice daily. Patients attended weekly control sessions during the first 3 weeks. At each visit, the surgical area was carefully cleaned and epithelial wound healing of the interdental papilla was assessed using the early healing index (Wachtel et al., 2003). The sutures were removed 2 weeks after surgery. After this initial 3-week wound healing phase, patients were again allowed to brush their teeth with a very soft toothbrush. The use of interdental brushes was permitted depending on the progress of the papillary soft tissue healing. Supportive periodontal therapy was given at 3-monthly intervals. This included professional supra- and subgingival tooth cleaning and remotivation and re-instruction of the patients to maintain the best possible oral hygiene.

3.1 | Statistical analysis

The Institute of Biostatistics (MHH) conducted the sample size calculation (nQuery Advisor 7.0) and randomized patient allocation. The difference of CAL between t0 and t2 ($\Delta$CAL$_{t0-t2}$) was defined as the primary outcome variable. A mean difference of 1.5 mm was expected between the test and control groups. The sample size calculation assumed that there was an unrelated problem, a Type I error of 5% (two-sided), and a standard deviation of 1.5 mm. For this setting, a power of 92% was calculated for 20 patients per group. The values used for the sample size calculation were based on data obtained from studies using the modified Widman flap as resective approach (Heitz-Mayfield et al., 2002) and using the simplified papilla preservation flap as a tissue-preserving approach (Cortellini et al., 2001). The randomized patient allocation was carried out by telephone on
the day of the surgical intervention. The randomization list was based on permuted blocks with random block lengths. Patients were allocated in a 1:1 ratio. One experienced periodontist (M. F.) conducted all clinical measurements. He and the patients had no information about the group assignments (double-blinding). IBM SPSS Statistics 26 (IBM) was used for statistical evaluation. Each patient contributed to the evaluation with one infrabony periodontal defect. Therefore, the patient was considered as a statistical unit. The defect was evaluated using the software program. The defect depth was calculated using a specialized software program.

The patients were recruited from February 2014 to 2020. The test group included 20 patients (11 female, 9 male) with a mean age of 54.34 ± 13.40 years (range: 25.27 – 85.63 years) and the control group 26 patients (14 female, 6 male) with a mean age of 52.37 ± 15.24 years (range: 22.94 – 73.83 years). There were no significant differences between both groups regarding age, gender or smoking habits (test group: one smoker; control group: two smokers). There were five dropouts due to endodontic involvement (n = 1; test group), development of endo-periodontal lesion (n = 1; test group), non-attendance to the follow-up visits (n = 2; test group), and need for retreatment after abscess formation (n = 1; control group). Besides this, incomplete data sets were generated in two patients due to new pregnancy (n = 1; control group) and nonattendance to the follow-up visit after 12 months (n = 1; control group). Thus, 35 patients were included in the statistical evaluation at t1, and 33 patients at t2.

4.2 | Baseline defect characteristics

There was a comparable distribution of defects between maxilla (test group: n = 5; control group: n = 6) and mandible (test group: n = 15; control group: n = 14) in both groups (Table 51). Also, one-wall (test group: n = 4; control group: n = 4), two-wall (test group: n = 13; control group: n = 14), and three-wall defects (test group: n = 3; control group: n = 2) were comparably distributed. The remaining clinical and radiographic variables also showed no significant group-dependent differences (Table 1).

5 | INTRAOPERATIVE MEASUREMENTS

No significant differences were found between the two groups for the intraoperative measurements. INFRA was 6.74 ± 1.89 mm in the test group and 7.30 ± 2.36 mm in the control group. The surgical intervention lasted on average 60.84 ± 16.84 min in the test group and 60.71 ± 10.99 min in the control group.

5.1 | Clinical and radiographic outcome at 6-months and 12-months follow-up visits

When considering the entire study population, no significant differences were found between the test and control group for the primary
outcome variable ΔCALt0−t2 and secondary outcome variable RBGt0−t2. Intragroup comparison using the GLM revealed that there was a significant PPD reduction (both groups: \( p < .001 \)), RED increase (test group: \( p = .012 \); control group: \( p < .001 \)), CAL gain (both groups: \( p < .001 \)) and RBG (both groups: \( p < .001 \)) in the two groups. The intergroup comparison showed that the temporal changes in RED differed significantly between the groups (GLM using “group” as a between-subjects factor: \( p = .031 \)). Regarding all patients, RED increase between t0 and t1 (ΔREDt0−t1) was significantly greater in the control group (\( p = .020 \); t test for independent samples). Further variables examined did not differ significantly in the intergroup comparison (Tables 2 and S2).

The COM showed that six defects (37.5%) of the test group and five defects (29.4%) of the control group had been successful treated (relevant CAL gain, no residual pocket). Relevant CAL gain in combination with a residual pocket was observed in nine defects (56.3%) of the test group and five defects (29.4%) of the control group. Treatment failure was determined in one defect (6.3%) of the test group and five defects (29.4%) of the control group (Table 3).

We hypothesized that especially noncontaining defects with a large defect angle would benefit from the GTPT. Considering patients with baseline radiographic defect angle >22°, there was a significantly greater CAL gain in the test group than in the control group. Thus, ΔCALt0−t1 was 3.08 ± 1.38 mm in the test group and 1.77 ± 1.54 mm in the control group (\( p = .032 \); t test for independent samples), and ΔCALt0−t2 was 3.62 ± 0.96 mm in the test group and 2.18 ± 1.83 mm in the control group (\( p = .034 \); t test for independent samples). When evaluating patients with a baseline radiographic defect angle ≤22°, no significant differences between the two groups were found for any of the examined parameters (Table S4).

In the next step, multivariate linear regression with backward elimination was performed (Table 4). The previous univariate linear regression showed that the variables PPDt0, defect angle, and INFRA had a substantial influence on ΔCALt0−t2 and were included in the multivariate linear regression. The variables PPDt0 (\( p = .001 \)) and group (\( p = .064 \)) remained in the last model and, thus, were identified as variables with the greatest influence on ΔCALt0−t2. Furthermore, univariate linear regression revealed that the variables PPDt0, defect angle, and INFRA had the greatest influence on RBGt0−t2. Following backward elimination, INFRA (\( p = .003 \)) and PPDt0 (\( p = .033 \)) were identified as variables with the greatest influence on RBGt0−t2.

### 6 | DISCUSSION

The hypothesis of the present study was that the GTPT would result in significantly more CAL gain than the double flap approach with resection of the defect-filling granulation tissue 12 months after regenerative periodontal surgery. However, the study failed to find a

## TABLE 2 Changes of PPD, RED, CAL, and RBG between baseline (t0) and the follow-up visits 6 months (t1) and 12 months (t2) after surgery

| Parameter         | \( \Delta t0−t1 \)                        | \( \Delta t0−t2 \)                        | t0−t2                           | t1−t2                           |
|-------------------|----------------------------------------|----------------------------------------|---------------------------------|---------------------------------|
|                   | Test group (mean ± SD)                  | Control group (mean ± SD)              | p value (95% CI)                | p value (95% CI)                |
| All patients      |                                        |                                        |                                 |                                 |
| PPD (mm)          | 3.81 ± 1.52                            | 3.63 ± 2.29                            | .789 (−1.18; 1.55)              | .438 ± 1.36                     |
| RED (mm)          | −0.50 ± 0.73                           | −1.26 ± 1.05                           | .020 (.13; 1.40)                | −0.63 ± 0.72                    |
| CAL (mm)          | 3.31 ± 1.58                            | 2.37 ± 1.92                            | .126 (−.28; 2.17)               | 3.75 ± 1.24                     |
| RBG (mm)          | 2.40 ± 1.65                            | 2.53 ± 2.08                            | .834 (−1.45; 1.17)              | 3.06 ± 1.74                     |
| Patients with defect angle >22° |                                        |                                        |                                 |                                 |
| PPD (mm)          | 3.62 ± 1.39                            | 2.77 ± 1.24                            | .113 (−.22; 1.91)               | 4.31 ± 1.25                     |
| RED (mm)          | −0.54 ± 0.78                           | −1.00 ± 0.71                           | .126 (−.14; 1.06)               | −0.69 ± 0.75                    |
| CAL (mm)          | 3.08 ± 1.38                            | 1.77 ± 1.54                            | .032 (1.2; 2.49)                | 3.62 ± 0.96                     |
| RBG (mm)          | 2.21 ± 1.10                            | 1.79 ± 0.95                            | .307 (−.41; 1.25)               | 2.98 ± 1.45                     |

Note: All patients: Δt0−t1: test group: \( n = 16 \), control group: \( n = 19 \); Δt0−t2: test group: \( n = 16 \), control group: \( n = 17 \). Patients with defect angle >22°: Δt0−t1: test group: \( n = 13 \), control group: \( n = 13 \); Δt0−t2: test group: \( n = 13 \), control group: \( n = 11 \).

**Abbreviations:** CAL, clinical attachment level; PPD, probing pocket depth; RBG, radiographic bone gain; RED, recession depth.

## TABLE 3 Composite outcome measure differentiated by group

| Residual PPDt2 | ≤4 mm | >4 mm |
|----------------|-------|-------|
| CAL gain t0−t2| ≥3 mm | Test group: 6 (37.5%) control group: 5 (29.4%) | Test group: 9 (56.3%) control group: 5 (29.4%) |
|                | <3 mm | Test group: 0 (0%) control group: 2 (11.8%)    | Test group: 1 (6.3%) control group: 5 (29.4%) |

**Abbreviations:** CAL, clinical attachment level; PPD, probing pocket depth.
significant difference between the test and control group for the primary outcome variable (ΔCALt0–t2) (Table 2).

The intragroup comparison using the GLM revealed that significant PPD reduction (test group: 4.38 ± 1.36 mm; control group: 4.06 ± 2.38 mm), CAL gain (test group: 3.75 ± 1.24 mm; control group: 3.06 ± 0.74 mm), and RBG (test group: 3.06 ± 1.74 mm; control group: 3.27 ± 2.19 mm) were achieved in both groups 12 months after surgery. Recently, a meta-analysis on the clinical performance of minimally invasive periodontal surgery (MIPS) for the treatment of infrabony defects was published (Clementini et al., 2019). The 18 studies included showed a PPD reduction of 4.24 mm, a RED increase of 0.44 mm, and a CAL gain of 3.89 mm, which is comparable to the results of our study. The lower CAL gain and higher RED increase observed in our study may be explained by the fact that the patient-related factors HI and FMBS were not as good as in other studies, in which MIPS was applied (Cortellini et al., 2017; Ribeiro et al., 2011). Assessment of the treatment outcome using the COM revealed that relevant CAL gain ≥ 3 mm was achieved in 15 of 16 (93.8%) defects in the test group but only in 10 of 17 (58.8%) defects in the control group. In addition, there were five cases (29.4%) of treatment failure in the control group but only one case (6.3%) in the test group. This distribution shows how reliably GTPT generates relevant CAL gain and how rarely treatment failures occur.

Regarding baseline defect characteristics, advanced periodontal defects with a mean PPD >9 mm and a mean INFRA >6 mm were present in both groups. A systematic review has shown that deeper periodontal defects (>4 mm) are associated with more RBG than shallower defects (≤4 mm) (Nibali et al., 2021). Consistent with these findings, we observed by multivariate linear regression that PPDt0 and particularly INFRAx-ray had the greatest influence on RBG 12 months after surgery. This agrees with the results of other studies that also found a positive correlation between RBG and INFRAx-ray (Ilgenli et al., 2007; Liñares et al., 2006; Meyle et al., 2011). There is evidence that the defect morphology plays a crucial role in the outcome of regenerative periodontal therapy (Cortellini et al., 2008; Losada et al., 2017; Meyle et al., 2011). Comprehensive characterization of infrabony defects requires further factors such as the number of residual bone walls and the radiographic defect angle. In the present study, two residual bone walls were preserved in most of the defects (test group: n = 13; control group: n = 14). However, it was sometimes difficult to determine the number of residual bone walls beyond doubt during the intraoperative assessment. Thus, every one- or two-wall defect has a more or less pronounced three-wall component in the deeper area of the defect. To capture the heterogeneity of the defects in a more differentiated way, we decided to perform a separate analysis with subgroups determined by

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**Table 4** Multivariate linear regression analysis with backward elimination using ΔCALt0–t2 and RBGt0–t2 as dependent variables

| Model | Included variables | Regression coefficient β | Significance | Adjusted R² |
|-------|-------------------|--------------------------|--------------|-------------|
| 1     | Group             | -1.045                   | 0.063        | .277        |
|       | Defect angle      | -0.016                   | 0.653        |             |
|       | INFRA             | .018                     | 0.928        |             |
|       | PPDt0             | .524                     | 0.039        |             |
| 2     | Group             | -1.042                   | 0.058        | .301        |
|       | Defect angle      | -0.017                   | 0.600        |             |
|       | PPDt0             | .538                     | 0.008        |             |
| 3     | Group             | -.974                    | 0.064        | .318        |
|       | PPDt0             | .590                     | 0.001        |             |

Abbreviations: ΔCALt0–t2, clinical attachment level gain between baseline and 12 months after surgery; RBGt0–t2, radiographic bone gain between baseline and 12 months after surgery; INFRA, clinical infrabony component; INFRAx-ray, radiographic infrabony component; PPDt0, probing pocket depth at baseline.
radiographic defect angle. A large defect angle is known to have a negative impact on the clinical and radiographic outcome (Eickholz et al., 2004; Losada et al., 2017). Accordingly, Tsitoura et al. reported that the probability of achieving CAL gain ≥ 4 mm is 2.5 times higher in defects with a defect angle ≤ 22° than in those with a defect angle ≥ 36° (Tsitoura et al., 2004). Our evaluation of patients with a defect angle > 22° revealed that a significantly greater CAL gain was achieved in the test group. This difference was detectable after 6 and 12 months and was mainly a result of PPD reduction. The fact that the use of EMD + graft material does not lead to more PPD reduction and greater CAL gain in the treatment of noncontainment defects using either EMD or EMD + graft material and differentiated the preservation and precise repositioning of the coronal part provided additional stability to the papillary soft tissue and facilitated the subsequent wound healing process.

Bone substitutes are frequently applied in regenerative therapy of advanced infrabony defects to prevent soft tissue collapse into the defect and, thus, preserve space for regeneration (Kao et al., 2015). There is conflicting data on whether EMD + graft material results in more CAL gain and greater PPD reduction than EMD alone. A recently published meta-analysis looked at the treatment of infrabony defects using either EMD or EMD + graft material and differentiated the treatment outcome by flap design (Trombelli et al., 2021). They reported that EMD + graft material resulted in more CAL gain in minimally invasive variants (EMD: 3.69 mm; EMD + graft: 4.10 mm) and papilla preservation variants (EMD: 3.08 mm; EMD + graft: 3.65 mm) than EMD alone. However, other studies provide evidence that the use of EMD + graft material does not lead to more PPD reduction and greater CAL gain in the treatment of noncontaining defects (Hoffmann et al., 2016; Losada et al., 2017; Pietruska et al., 2012). These observations raise the question of whether it would have been better to use EMD + graft material in the control group instead of EMD alone.

Another significant limitation of the present study was that patients with containing defects (three-wall defects, defect angle ≤ 22°) were also included. This was probably the main reason why no significant differences (apart from ΔRED10–12) were found between the test and control group when all study participants were considered. Another possible explanation could be that EMDs were used in both groups. Furthermore, the present study does not provide any information on whether periodontal regeneration or repair actually occurred during the wound healing process. Interestingly, there was a smaller RBG in the GTPT group compared to the control group, when all patients were considered. This was the case despite greater PPD reduction, greater CAL gain, and lower RED increase in the GTPT group. In contrast, patients with defect angle > 22° tended to have a larger RBG in the test group. These contradictory data lead to the question of what happens to the granulation tissue during wound healing and what influence EMDs play in its maturation process. Animal studies could provide important information to clarify these questions.

7 CONCLUSIONS

We conclude that the GTPT may show its advantages over the conventional technique especially in advanced cases characterized by unfavorable defect morphology, namely few residual bone walls and large defect angle. Conversely, if sufficient residual bone walls and/or small defect angles are still present, removal of the granulation tissue does not represent a disadvantage for the regenerative healing process.

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CONFLICT OF INTERESTS

The authors declare that there are no conflict of interests.

ETHICS STATEMENT

The present study has been approved by the Institutional Review Board (Ethics Committee of Hannover Medical School, reference number: 5556) and all participants signed an informed consent according to the Declaration of Helsinki.

AUTHOR CONTRIBUTIONS

Knut Adam conceived the study, performed the surgical procedures, analyzed and interpreted the data, and drafted the manuscript. Hüsamettin Günay participated in the study design, supervised the surgical interventions, and critically reviewed the manuscript. Bernhard Vaske conducted the sample size calculation and assisted in performing the statistical analysis. Marco Flohr performed the statistical analysis. Marco Flohr performed the statistical analysis.
measurements of the clinical parameters and critically reviewed the manuscript. Ingmar Staufenbiel conceived the study, performed the surgical procedures, and critically reviewed the manuscript. All authors read and approved the final version of the manuscript.

DATA AVAILABILITY STATEMENT
Data generated during the study are not publicly available but can be obtained from the corresponding author on reasonable request.

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REFERENCES
Adam, K., Gousoopoulou, E., Bakopoulou, A., Leyhausen, G., Volk, J., Staufenbiel, I., Günday, H., Schertl, P. P. J., & Geurtsen, W. (2019). Characterization of cells derived from inflamed intra-bony periodontal defects. Deutsche Zahnärztliche Zeitschrift International, 1, 182–194. https://doi.org/10.3238/dzz-int.2019.0182-0194
Adam, K., Volk, J., Bakopoulou, A., Gousoopoulou, E., Staufenbiel, I., Günday, H., & Geurtsen, W. (2020). Cells from granulation tissue of intra-bony periodontal defects reveal neurogenic and angiogenic differentiation potential and express the embryonic transcription factors NANG, OCT4 and SOX2. Deutsche Zahnärztliche Zeitschrift International, 2, 82–94. https://doi.org/10.3238/dzz-int.2020.0082-0094
Ainamo, J., & Bay, I. (1975). Problems and proposals for recording gingivitis and plaque. International Dental Journal, 25(4), 229–235.
Clementini, M., Ambrosi, A., Cicciarelli, V., De Risi, V., & de Sanctis, M. (2019). Clinical performance of minimally invasive periodontal surgery in the treatment of infrabony defects: Systematic review and meta-analysis. Journal of Clinical Periodontology, 46(12), 1236–1253. https://doi.org/10.1111/jcpe.13201
Cortellini, P., Buti, J., Pini Prato, G., & Tonetti, M. S. (2017). Periodontal regeneration compared with access flap surgery in human intra-bony defects 20-year follow-up of a randomized clinical trial: Tooth retention, periodontitis recurrence and costs. Journal of Clinical Periodontology, 44(1), 58–66. https://doi.org/10.1111/jcpe.12638
Cortellini, P., Nieri, M., Prato, G. P., & Tonetti, M. S. (2008). Single minimally invasive surgical technique with an enamel matrix derivative to treat multiple adjacent intrabony defects: Clinical outcomes and patient morbidity. Journal of Clinical Periodontology, 33(7), 605–613. https://doi.org/10.1111/j.1600-051X.2008.01242.x
Cortellini, P., Pini Prato, G., & Tonetti, M. S. (1993). Periodontal regeneration of human infrabony defects. I. Clinical measures. Journal of Periodontology, 64(4), 254–260. https://doi.org/10.1902/jop.1993.64.4.254
Cortellini, P., & Tonetti, M. S. (2007). A minimally invasive surgical technique with an enamel matrix derivative in the regenerative treatment of intra-bony defects: A novel approach to limit morbidity. Journal of Clinical Periodontology, 34(1), 87–93. https://doi.org/10.1111/j.1600-051X.2006.01020.x
Cortellini, P., & Tonetti, M. S. (2009). Improved wound stability with a modified minimally invasive surgical technique in the regenerative treatment of isolated interdental infrabony defects. Journal of Clinical Periodontology, 36(2), 157–163. https://doi.org/10.1111/j.1600-051X.2008.01352.x
Cortellini, P., Tonetti, M. S., Lang, N. P., Suvan, J. E., Zucchelli, G., Vangsted, T., Silvestri, M., Rossi, R., McClain, P., Fonzar, A., Dubravec, D., & Adriaens, P. (2001). The simplified papilla preservation flap in the regenerative treatment of deep intrabony defects: Clinical outcomes and postoperative morbidity. Journal of Periodontology, 72(12), 1702–1712. https://doi.org/10.19072/jop.2001.72.12.1702
Eickholz, P., Hörr, T., Klein, F., Hassfeld, S., & Kim, T. S. (2004). Radiographic parameters for prognosis of periodontal healing of infrabony defects: Two different definitions of defect depth. Journal of Periodontology, 75(3), 399–407. https://doi.org/10.19072/jop.2004.75.3.399
Gousoopoulou, E., Bakopoulou, A., Apatzidou, D. A., Leyhausen, G., Volk, J., Staufenbiel, I., Geurtsen, W., & Adam, K. (2021). Evaluation of stemness properties of cells derived from granulation tissue of peri-implantitis lesions. Clinical and Experimental Dental Research, 7, 739–753. https://doi.org/10.1002/cre.2406
Günay, H., Staufenbiel, I., Geurtsen, W., & Adam, K. (2019). The granulation tissue preservation technique in regenerative therapy of peri-implantitis—a treatment concept with case reports. Deutsche Zahnärztliche Zeitschrift International, 1, 4–15. https://doi.org/10.3238/dzz-int.2019.004-0015
Günay, H., Weinspach, K., Geurtsen, W., & Staufenbiel, I. (2013). Relevance of the intralesional granulation tissue in regenerative periodontal surgery—case reports. Deutsche zahnärztliche Zeitschrift, 68(9), 526–537. https://doi.org/10.3238/dzz.2013.0526-0537
Harrel, S. K. (1998). A minimally invasive surgical approach for periodontal bone grafting. International Journal of Periodontics and Restorative Dentistry, 18(2), 161–169.
Harrel, S. K., & Rees, T. D. (1995). Granulation tissue removal in routine and minimally invasive procedures. Compendium of Continuing Education in Dentistry, 16(9), 960, 962, 964 passim.
Heitz-Mayfield, L. J., Trombelli, L., Heitz, F., Needleman, I., & Moles, D. (2002). A systematic review of the effect of surgical debridement vs non-surgical debridement for the treatment of chronic periodontitis. Journal of Clinical Periodontology, 29(Suppl 3), 92–102. discussion 160–2. https://doi.org/10.1034/j.1600-051x.29.s3.5.x
Hoffmann, T., Al-Machot, E., Meyle, J., Jervae-Storm, P. M., & Jepsen, S. (2016). Three-year results following regenerative periodontal surgery of advanced intrabony defects with enamel matrix derivative alone or combined with a synthetic bone graft. Clinical Oral Investigations, 20(2), 357–364. https://doi.org/10.1007/s00784-015-1522-4
Ilgenli, T., Dündar, N., & Kal, B. I. (2007). Demineralized freeze-dried bone allograft and platelet-rich plasma vs platelet-rich plasma alone in infrabony defects: A clinical and radiographic evaluation. Clinical Oral Investigations, 11(1), 51–59. https://doi.org/10.1007/s00784-006-0083-y
Kao, R. T., Naes, S., & Reynolds, M. A. (2015). Periodontal regeneration—infrabony defects: A systematic review from the AAP Regeneration Workshop. Journal of Periodontology, 86(2 Suppl), S77–S104. https://doi.org/10.1902/jop.2015.130685
Liñares, A., Cortellini, P., Lang, N. P., Suvan, J., & Tonetti, M. S. European Research Group on Periodontology (ErgoPerio). (2006). Guided tissue regeneration/deproteinized bovine bone mineral or papilla preservation flaps alone for treatment of intrabony defects. II: Radiographic predictors and outcomes. Journal of Clinical Periodontology, 33(5), 351–358. https://doi.org/10.1111/j.1600-051X.2006.00911.x
Lindhe, J., & Nyman, S. (1985). Scaling and granulation tissue removal in periodontal therapy. Journal of Clinical Periodontology, 12(5), 374–388. https://doi.org/10.1111/j.1600-051X.1985.tb00928.x
Losada, M., González, R., García, Á. P., Santos, A., & Nart, J. (2017). Treatment of non-contained infrabony defects with enamel matrix derivative alone or in combination with biphasic calcium phosphate bone graft: A 12-month randomized controlled clinical trial. Journal of Periodontology, 88(5), 426–435. https://doi.org/10.19072/jop.2016.160459
Meyle, J., Hoffmann, T., Topoll, H., Heinz, B., Al-Machot, E., Jervae-Storm, P. M., Meiss, C., Eickholz, P., & Jepsen, S. (2011). A multi-centre
randomized controlled clinical trial on the treatment of intra-bony defects with enamel matrix derivatives/synthetic bone graft or enamel matrix derivatives alone: Results after 12 months. Journal of Clinical Periodontology, 38(7), 652–660. https://doi.org/10.1111/j.1600-051x.2011.01726.x

Nibali, L., Sultan, D., Arena, C., Pelekos, G., Lin, G. H., & Tonetti, M. (2021). Periodontal intra-bony defects: Systematic review of healing by defect morphology following regenerative surgery. Journal of Clinical Periodontology, 48(1), 100–113. https://doi.org/10.1111/jcp.13381

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

Park, J. C., Kim, J. M., Jung, I. H., Kim, J. C., Choi, S. H., Cho, K. S., & Kim, C. S. (2011). Isolation and characterization of human periodontal ligament (PDL) stem cells (PDLSCs) from the inflamed PDL tissue: In vitro and in vivo evaluations. Journal of Clinical Periodontology, 38(8), 721–731. https://doi.org/10.1111/j.1600-051x.2011.01716.x

Petruska, M., Pietruski, J., Nagy, K., Breccx, M., Arweiler, N. B., & Sculean, A. (2012). Four-year results following treatment of intra-bony periodontal defects with an enamel matrix derivative alone or combined with a biphasic calcium phosphate. Clinical Oral Investigations, 16(4), 1191–1197. https://doi.org/10.1007/s00784-011-0611-2

Reynolds, M. A., Aichelmann-Reidy, M. E., Branch-Mays, G. L., & Gunsolley, J. C. (2003). The efficacy of bone replacement grafts in the treatment of periodontal osseous defects. A systematic review. Annals of Periodontology, 8(1), 227–265. https://doi.org/10.1902/annals.2003.8.1.227

Ribeiro, F. V., Casarin, R. C., Palma, M. A., Júnior, F. H., Sallum, E. A., & Casati, M. Z. (2011). Clinical and patient-centered outcomes after minimally invasive non-surgical or surgical approaches for the treatment of intra-bony defects: A randomized clinical trial. Journal of Periodontology, 82(9), 1256–1266. https://doi.org/10.1902/jop.2011.100680

Ribeiro, F. V., Casarin, R. C., Palma, M. A., Júnior, F. H., Sallum, E. A., & Casati, M. Z. (2013). Clinical and microbiological changes after minimally invasive therapeutic approaches in intra-bony defects: A 12-month follow-up. Clinical Oral Investigations, 17(7), 1635–1644. https://doi.org/10.1007/s00784-012-0855-5

Sanz, M., Herrera, D., Kebschull, M., Chapelle, I., Jepsen, S., Beglundh, T., Sculean, A., & Tonetti, M. S. (2020). EFP workshop participants and methodological consultants. Treatment of stage I-III periodontitis—the EFP S3 level clinical practice guideline. Journal of Clinical Periodontology, 47(Suppl 22), 4–60. https://doi.org/10.1111/jcpe.13290

Stavropoulos, A., Kostopoulos, L., Nyengaard, J. R., & Karring, T. (2003). Deproteinized bovine bone (Bio-Oss) and bioactive glass (Biogran) arrest bone formation when used as an adjunct to guided tissue regeneration (GTR): An experimental study in the rat. Journal of Clinical Periodontology, 30(7), 636–643. https://doi.org/10.1034/j.1600-051x.2003.00093.x

Trombelli, L., Farina, R., Franceschetti, G., & Calura, G. (2009). Single-flap approach with buccal access in periodontal reconstructive procedures. Journal of Periodontology, 80(2), 353–360. https://doi.org/10.1902/jop.2009.080420

Trombelli, L., Farina, R., Vecchietti, R., Maietti, E., & Simonelli, A. (2020). A simplified composite outcome measure to assess the effect of periodontal regenerative treatment in intraosseous defects. Journal of Periodontology, 91(6), 723–731. https://doi.org/10.1002/jper.19-0127

Trombelli, L., Simonelli, A., Quaranta, A., Tu, Y. K., Li, H., Agusto, M., Jiao, X. J., & Farina, R. (2021). Effect of flap design for enamel matrix derivative application in intraosseous defects. JDR Clinical and Translational Research, 6(2), 184–194. https://doi.org/10.1177/2380084420934731

Tsitoura, E., Tucker, R., Suvan, J., Laurell, L., Cortellini, P., & Tonetti, M. (2004). Baseline radiographic defect angle of the intra-bony defect as a prognostic indicator in regenerative periodontal surgery with enamel matrix derivative. Journal of Clinical Periodontology, 31(8), 643–647. https://doi.org/10.1111/j.1600-051x.2004.00555.x

Wachtel, H., Schenk, G., Böhm, S., Wenig, D., Zuhr, O., & Hürzeler, M. B. (2003). Microsurgical access flap and enamel matrix derivative for the treatment of periodontal intra-bony defects: A controlled clinical study. Journal of Clinical Periodontology, 30(6), 496–504. https://doi.org/10.1034/j.1600-051x.2003.00013.x

SUPPORTING INFORMATION

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