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A minimally invasive surgical technique with an enamel matrix derivative in the regenerative treatment of intra-bony defects: a novel approach to limit morbidity

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Abstract

Aims: This study was undertaken to describe a new surgical approach (minimally invasive surgical technique, MIST) and to evaluate preliminarily its clinical performance and patient perception associated with the application of enamel matrix derivative (EMD) in the treatment of isolated deep intra-bony defects.

Methods: Thirteen deep isolated intra-bony defects in 13 patients were surgically accessed with the MIST. This technique was designed to limit the mesio-distal flap extension and the corono-apical reflection in order to reduce the surgical trauma and increase flap stability. The incision of the defect-associated papilla was performed according to the principles of the papilla preservation techniques. EMD was applied on the debrided root surfaces. Stable primary closure of the flaps was obtained with internal modified mattress sutures. Surgery was performed with the aid of an operating microscope and microsurgical instruments. Clinical outcomes were collected at baseline and at 1 year. Intra-operative and post-operative patient perception was also recorded.

Results: Early wound healing was uneventful: primary wound closure was obtained and maintained in all sites with the exception of one site with a small wound dehiscence at week 1. No oedema or haematoma were noted. Patients did not report any pain. Three patients experienced slight discomfort for 2-days post-operatively. The 1-year clinical attachment level (CAL) gain was 4.8 ± 1.9 mm. The 1-year percent resolution of the defect was $88.7 \pm 20.7\%$, and reached 100% of the baseline intra-bony component in seven sites. Residual probing depths (PD) were 2.9 ± 0.8 mm. Differences between baseline and 1-year CAL and PD were both clinically and statistically highly significant ($p < 0.0001$). A minimal increase of 0.1 ± 0.9 mm in gingival recession between baseline and 1 year was recorded ($p = 0.39$).

Conclusions: This case cohort indicates that MIST associated with EMD resulted in excellent clinical improvements while limiting patient morbidity. These preliminary findings need to be confirmed in a larger study.

Key words: clinical trial; microsurgery; osseous defects; periodontal diseases; periodontal regeneration

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Periodontal regeneration of intra-bony defects has been achieved with different principles: these include barrier membranes (Nyman et al. 1982, Gottlow et al. 1986), demineralized freeze-dried bone allograft (DFDBA, Bowers et al. 1989), combination of barrier membranes and grafts (Camelo et al. 1998, Mellonig 2000), and enamel matrix derivative (EMD, Mellonig 1999, Yukna & Mellonig 2000). Data from controlled clinical trials and meta-analyses from systematic reviews demonstrate that the cited approaches provide added benefits in terms of clinical attachment level

(CAL) gain and probing pocket depth (PPD) reduction as compared with access flap alone (Needleman et al. 2002, Murphy & Gunsolley 2003, Trombelli et al. 2002, Tonetti et al. 2004a).

In the last decade, a special emphasis has been focused on the design and performance of surgical procedures for periodontal regeneration. Specific surgical approaches have been proposed to preserve the soft tissues and to reach a stable primary closure of the wound in order to seal the area of regeneration from the oral environment (Cortellini et al. 1995, 1999). In fact, flap dehiscence at regenerative sites is a frequent occurrence with barrier membranes (Cortellini et al. 1993a, 2001, Tonetti et al. 1998), bone grafts (Sanders et al. 1983), combination of barriers and grafts (Tonetti et al. 2004a), and, to a lesser extent, with EMD (Tonetti et al. 2002, Sanz et al. 2004). Exposure and thus contamination of the regenerative material is a critical issue because it has been associated with reduced clinical outcomes (Nowzari et al. 1995, De Sanctis et al. 1996). In order to further increase surgical effectiveness, the use of operating microscopes and microsurgical instruments has been suggested (Cortellini & Tonetti 2001, 2005). The use of a microsurgical approach in combination with different regenerative materials resulted in maintenance of primary wound closure in more than 92% of the treated sites for the whole healing period. Data from the cited studies are very promising, even though controlled trials are desirable to estimate the magnitude of the real benefits of a microsurgical approach. Along with other advantages, the use of magnification and optimal illumination of the surgical field greatly improves the visual acuity and the control of the surgical instruments, making it possible to perform surgery with reduced flap reflection. This could confer several potential advantages for the surgery, the healing process, and the patient's perception of the procedure. The surgery could be less invasive, shorter in time and less demanding, the healing process could be favoured by the improved wound stability of minimally mobilized flaps, and the patients could benefit from a procedure with potentially reduced intra-operative and post-operative morbidity.

A minimally invasive surgery (MIS) has been proposed in 1995 (Harrel & Ress) with the aim to produce minimal wounds, minimal flap reflection, and gentle handling of the soft and hard tissues in periodontal surgery. Data from case reports show clinical improvements in terms of pocket depth reduction, attachment level gain, and minimal increase of recession after application of the MIS in different types of defects (Harrel 1998, Harrel & Nunn 2001). A recent case report from the same group (Harrel et al. 2005) showed CAL gains of 4.05 mm following application of MIS and EMD in 16 patients presenting multiple sites with deep pockets associated with different defect morphologies, including furcation involvements.

Recently, a modified surgical approach, the ''MIS technique (MIST)'', has been specifically designed to treat isolated intra-bony defects with periodontal regeneration. Background foundations for this technique are the concepts of the MIS (Harrel & Ress 1995), the application of largely tested papilla preservation techniques [modified papilla preservation technique (MPPT) Cortellini et al. 1995, simplified papilla preservation flap (SPPF)], and the application of passive internal mattress sutures to seal the regenerating wound from the oral environment. The main objectives of the MIST are the following: (1) reduce surgical trauma, (2) increase flap/wound stability, (3) allow stable primary closure of the wound, (4) reduce surgical chair time, and (5) minimize patient discomfort and side effects.

The aim of the present study was to describe the surgical approach and preliminarily evaluate the clinical performances and the patient perception of the ''MIST'' associated with the application of EMD in the treatment of isolated deep intra-bony defects.

Materials and Methods

Study population and experimental design

Patients with advanced periodontal disease, in general good health, presenting with at least one deep intra-bony defect were considered eligible for this study. Patients were included after completion of cause-related therapy consisting of scaling and root planing, motivation, and oral-hygiene instructions. Flap surgery for pocket elimination was performed, when indicated, in the remaining portions of the dentition of each patient before the regenerative treatment. All subjects gave informed written consent.

Inclusion/exclusion criteria were as follows:

1. *Absence of relevant medical conditions*. Patients with uncontrolled or poorly controlled diabetes, unstable or life-threatening conditions, or requiring antibiotic prophylaxis were excluded.
2. *Smoking status:* only non-smokers were included.
3. *Defect anatomy*: presence of at least one tooth with PPD and CAL loss of at least 5 mm associated with an intra-bony defect of at least 2 mm.
4. *Good oral hygiene*: full-mouth plaque score (FMPS) $\leqslant 20\%$.
5. *Low levels of residual infection*: full-mouth bleeding score (FMBS) $\leqslant 20\%$.
6. *Compliance*: only patients with optimal compliance, as assessed during the cause-related phase of therapy, were selected.
7. *Endodontic status*: teeth had to be vital or properly treated with root canal therapy.

Three months after completion of periodontal therapy, baseline clinical measurements were recorded. The experimental sites were accessed with the MIST and carefully debrided. Measurements were taken during surgery to characterize the defect anatomy. Ethylenediamine tetraacetic acid (EDTA) and EMD (Emdogain, Institute Straumann AG, Basel, Switzerland) were applied on the instrumented and dried root surfaces and flaps were sutured with modified internal mattress sutures. Patients were enrolled in a stringent post-operative supportive care programme with weekly recalls for 6 weeks, and then included in a 3-month periodontal supportive care programme for 1 year.

Clinical measurements at baseline and at 1-year follow-up visit

The following clinical parameters were evaluated at baseline before regenerative therapy and at the 1-year follow-up visit by an independent clinician: FMPS were recorded as the percentage of total surfaces (four aspects per tooth) that revealed the presence of plaque (O'Leary et al. 1972). Bleeding on probing (BOP) was assessed dichotomously

and FMBS were then calculated (Cortellini et al. 1993a).

PPD and recession of the gingival margin (REC) were recorded to the nearest millimetre at the deepest location of the selected inter-proximal site. All measurements and BOP were taken with a pressure-sensitive manual periodontal probe at 0.3 N (Brodontic probe equipped with a PCP-UNC 15 tip, Hu-Friedy, Chicago, IL, USA). CAL were calculated as the sum of PD and REC. The radiographic defect angle of each defect was measured on a periapical radiograph, as previously described (Tonetti et al. 1993). Chair-time of each surgical procedure was recorded. Primary closure of the flaps was evaluated at completion of surgery and at weekly recalls for a period of 6 weeks, along with the potential presence/absence of oedema and/or haematoma. Patients were questioned about the subjective perception of intra-operative pain and/or discomfort at completion of surgery, and of post-operative pain and/or discomfort 1 week after surgery (Tonetti et al. 2004b).

Clinical characterization of the intra-bony defects

Defect morphology was characterized intra-surgically in terms of distance between the cemento-enamel junction and the bottom of the defect (CEJ-BD) and total depth of the intra-bony component of the defect (INFRA), essentially as previously described (Cortellini et al. 1993b). The depths of the three-, two-, and one-wall sub-components were also recorded.

Surgical approach (MIST)

Flap elevation

The defect-associated inter-dental papilla was accessed either with the SPPF (Cortellini et al. 1999) or the MPPT (Cortellini et al. 1995). The SPPF was performed whenever the width of the inter-dental space was 2 mm or narrower, while the MPPT was applied at inter-dental sites wider than 2 mm. The inter-dental incision (SPPF or MPPT) was extended to the buccal and lingual aspects of the two teeth adjacent to the defect. These incisions were strictly intra-sulcular to preserve all the height and width of the gingiva, and their mesio-distal extension was kept at a minimum to allow the corono-apical elevation of a very small full-thickness flap with the objective to expose just 1–2 mm of the defect-associated residual bone crest. When possible, only the defect-associated papilla was accessed and vertical-releasing incisions were avoided. With these general rules in mind, different clinical pictures were encountered in different treated defects (Fig. 1).

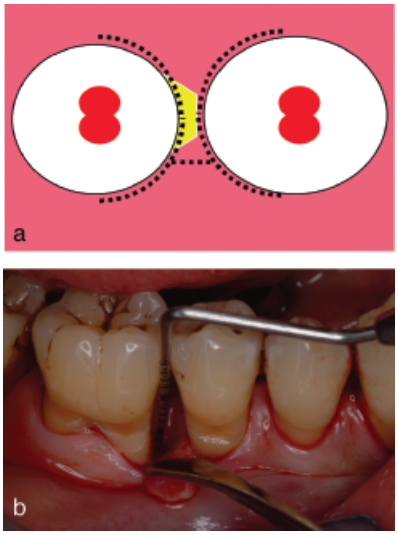

Fig. 1. Drawing (a) depicts the ideal flap design suggested to access a pure inter-proximal three-wall intra-bony defect. It includes the incision through the inter-dental papilla (MPPT) and the intra-sulcular incision running from the inter-proximal space to the mid-buccal and mid-lingual sides of the defect-associated teeth. A clinical example of a flap designed according to the above principles is shown in (b).

The shortest mesio-distal extension of the incision and the minimal flap reflection occurred when the intra-bony defect was a pure three-wall, or had shallow two- and/or one-wall sub-components allocated entirely in the inter-proximal area. In these instances, the mesio-distal incision involved only the defect-associated papilla and part of the buccal and lingual aspects of the two teeth neighbouring the defect. The full-thickness flap was elevated minimally, just to expose the buccal and lingual bone crest delimiting the defect in the inter-dental area (Fig. 2).

A larger corono-apical elevation of the full-thickness flap was necessary when the coronal portion of the intra-bony defect had a deep two-wall component. The corono-apical extension of the flap was kept to a minimum at the aspect where the bony wall was preserved (either buccal or lingual), and extended more apically at the site where the bony wall was missing (lingual or buccal), the objective being to reach and expose 1–2 mm of the residual bone crest.

When a deep one-wall defect was approached, the full-thickness flap was elevated to the same extent on both the buccal and the lingual aspects.

When the position of the residual buccal/lingual bony wall(s) was very deep and difficult or impossible to reach with the above-described minimal incision of the defect-associated inter-dental space, the flap(s) was (were) further extended mesially or distally involving one extra inter-dental space to obtain a larger flap reflection. The same approach was used when the bony defect also extended to the buccal or the palatal side of the involved tooth, or when it involved the two inter-proximal spaces of the same tooth. In the latter instance, a second inter-proximal papilla was accessed, either with an SPPF or an MPPT, according to indications. Vertical-releasing incisions were performed when flap reflection caused tension at the extremities of the flap(s). The vertical-releasing incisions were always kept very short and within the attached gingiva (never involving the muco-gingival junction). The overall aim of this approach was to avoid using vertical incisions whenever possible or to reduce at minimum their number and extent when there was a clear indication for them. Periosteal incisions were never performed.

Defect debridement and EMD application

The defects were debrided with a combined use of mini curettes (Gracey, Hu-Friedy) and power-driven instruments (Soniflex Lux, Kavo, Germany), and the roots were carefully planed. During the instrumentation, the flaps were slightly reflected, carefully protected with periosteal elevators and frequent saline irrigations. At the end of instrumentation, EDTA was applied on the instrumented root surface for 2 min. After that, the defect area was carefully rinsed with saline and finally EMD was applied on the dried root surface. Then, the flaps were repositioned.

Flap suturing technique

The suturing approach in most of the instances consisted of a single modified

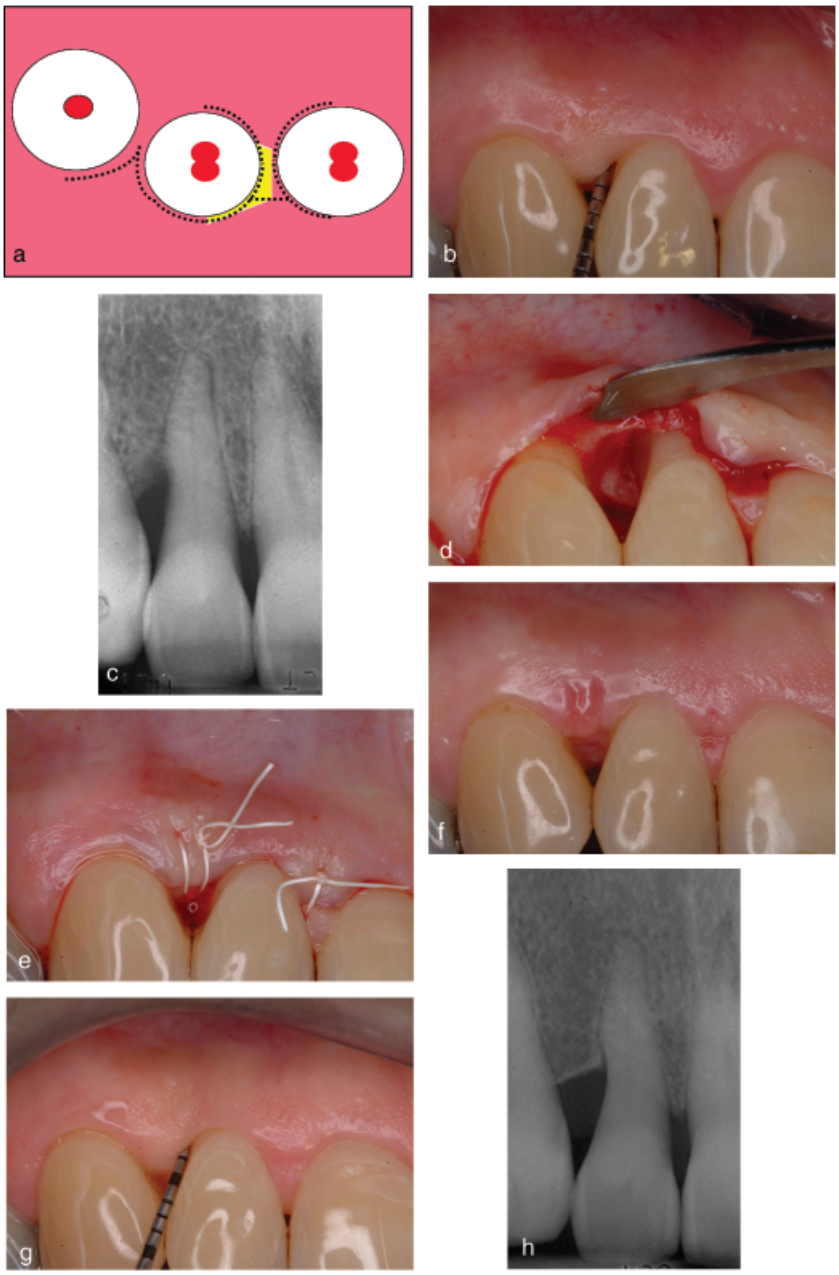

Fig. 2. Drawing (a) illustrates a different flap design indicated by the finding of a buccal extension of the intra-bony defect. The access to the buccal portion of the defect requires an extension of the flap to the neighbouring inter-dental papilla. Clinical picture (b) showing a pocket of 6 mm distal to the right lateral incisor and the associated radiographic view of the defect (c). The buccal flap involves the defect-associated inter-dental papilla and the papilla mesial to the lateral incisor. On the palatal side, the flap is restricted to the defect-associated papilla. The minimal flap elevation is sufficient for a careful debridement of the combined two- and three-wall defect (d). Primary closure of the wound is obtained with a modified internal mattress suture (e). Primary closure is maintained at the 1-week suture removal (f). The 1-year clinical photograph shows a 3 mm probing depth and no increase in gingival recession as compared with baseline (g). Baseline (c) and 1-year radiographs show the defect on the distal of the lateral incisor and the 1-year resolution of the intra-bony component of the defect (h).

internal mattress suture at the defect-associated inter-dental area to reach primary closure of the papilla in the absence of any tension (Cortellini & Tonetti 2001, 2005). When a second inter-dental space had been accessed, the same suturing technique was used to obtain primary closure in this area. Vertical-releasing incisions were sutured with simple passing sutures. The buccal and lingual flaps were re-positioned at their original level, without any coronal displacement to avoid any additional tension in the healing area.

All the surgical procedures were performed with the aid of an operating microscope (Global Protege, St. Louis, MO, USA) at a magnification of × 4– × 16 (Cortellini & Tonetti 2001, 2005). Microsurgical instruments were utilized, whenever needed, as a complement to the normal periodontal set of instruments. Incisions were carried out using delaminating microsurgical blades (M6900, Advanced Surgical Technologies, Sacramento, CA, USA). 6-0 e-PTFE (Goretex, WL Gore & Associates, Flagstaff, AZ, USA) sutures were preferred to obtain primary closure of the inter-dental tissues.

Post-operative period

A protocol for the control of bacterial contamination consisting of doxicycline (100 mg bid for 1 week), 0.12% chlorhexidine mouth rinsing three times per day, and weekly prophylaxis was prescribed (Tonetti et al. 2002). Patients were requested to avoid brushing, flossing, and chewing in the treated area for periods of 3–4 weeks. Then, patients resumed full oral hygiene. At the end of the ''early healing phase'', patients were placed on a 3-month recall system for 1 year.

Data analysis

Data were expressed as means ± standard deviation of 13 defects in 13 patients. No data points were missing. Comparisons between baseline and 1 year data were made using the paired *t*-test ($\alpha = 0.05$). CAL gain), residual pocket depth, and position of the gingival margin were the primary outcome variables. Percentage fill of the baseline intra-bony component of the defect was calculated as: CAL% = (CAL gains)/INFRA × 100.

Results

Patient and defect characteristics at baseline

Thirteen intra-bony defects in 13 subjects (mean age 43.1 ± 9.8, range 34–63 years, nine females, and non-smoker), who met the admission criteria, were included in this case cohort.

FMPS and FMBS at baseline were 11 ± 3.4% and 6.8 ± 2.9%, respectively

(Table 1). CAL of 8.7 ± 2.7 mm and probing depths (PD) of 7.7 ± 1.8 mm on average were recorded (Table 1). The radiographic defect angle was $29 \pm 7^{\circ}$. Distance from the CEJ-BD was 9.6 ± 2.8 mm, and the INFRA was 5.5 ± 1.5 mm (Table 1).

Table 1. Baseline patient and defect characteristics

Variables	Mean ± SD	Minimum	Maximum
FMPS (%)	11 ± 3.4	6	19
FMBS (%)	6.8 ± 2.9	3	12
PPD (mm)	7.7 ± 1.8	5	10
REC (mm)	1 ± 1.5	0	5
CAL (mm)	8.7 ± 2.7	5	13
CEJ-BD (mm)	9.6 ± 2.8	6	15
INFRA (mm)	5.5 ± 1.5	3	8
Three-wall (mm)	3.1 ± 1.6	0	6
Two-wall (mm)	1.7 ± 1.3	0	3
One-wall (mm)	0.7 ± 1	0	3
X-Ray angle (°)	29 ± 7	19	42

FMPS, full-mouth plaque scores; FMBS, full-mouth bleeding scores; PPD, probing pocket depth; REC, recession of the gingival margin ; CAL, clinical attachment level; CEJ-BD, cemento-enamel junction and the bottom of the defect ; INFRA, intra-bony component of the defect.

Table 2. Clinical outcomes at baseline and 1 year after treatment

Variables	Baseline	1 year	Difference	Significance*
PPD (mm)	7.7 ± 1.8	2.9 ± 0.8	4.8 ± 1.8	$p<0.0001$
REC (mm)	1 ± 1.5	0.9 ± 2.1	0.1 ± 0.9	$p=0.39$
CAL (mm)	8.7 ± 2.7	3.8 ± 2.2	4.8 ± 1.9	$p<0.0001$

*Paired *t*-test.

PPD, probing pocket depth; REC, recession of the gingival margin; CAL, clinical attachment level.

Design of the surgical flap and surgical chair-time

The SPPF was used in six sites, while the MPPT was applied in seven cases. An incision restricted to the defect-associated papilla was performed in four cases associated with pure three-wall defects. The flap was further extended buccally and/or lingually in nine cases presenting with one and/or two-wall sub-components, due to a deeper location of the bony wall. A vertical-releasing incision was performed in two cases to help flap reflection. The average surgical chair-time was 55.4 ± 6.5 min. (range 45–66 min.).

Primary closure of the flap and post-operative period

In all treated sites, primary closure was obtained at completion of the surgical procedure. At the 1-week suture removal follow-up, one site accessed with SPPF presented with a small inter-dental gap between the two edges of the papilla. At week 2, the papilla found open at week 1 was closed. All the other sites remained closed during the 6 weeks of the early healing period. No oedema or haematoma was noted in any of the treated sites. Patients were questioned at the end of surgery and at week 1 about the intra-operative and post-operative period and reported no pain. Three patients reported very limited discomfort in the first 2 days of the first post-operative week. Ten out of 13 described the first postoperative week as uneventful, reporting that they had no feeling of having been surgically treated after the second post-operative day.

One-year clinical outcomes

The 13 patients presented at the 1-year follow-up visit with FMPS and FMBS of $6.9 \pm 2.5\%$ (range 2.3–11) and $4.3 \pm 2.4\%$ (range 2.3–7), respectively. The differences in FMPS and FMBS between baseline and 1 year were statistically significant ($p = 0.002$ and 0.009, respectively).

The 1-year CAL was 3.8 ± 2.2 mm with a CAL gain of 4.8 ± 1.9 mm (range 3–8 mm). Differences in CAL between baseline and 1 year were clinically and statistically highly significant ($p<0.0001$). The 1-year CAL% was $88.7 \pm 20.7\%$, with a range of 50–114.3%. CAL% reached 100% of the baseline intra-bony component of the defect in seven sites.

Residual PD were 2.9 ± 0.8 mm, with an average pocket depth reduction of 4.8 ± 1.8 mm. Differences between baseline and 1-year PD were clinically and statistically highly significant ($p<0.0001$). Only three sites showed a residual PD of 4 mm; all the other sites resulted with a 1-year PPD of 3 mm or less.

A minimal average change of 0.1 ± 0.9 mm in the position of the gingival margin between baseline and 1 year was observed. This difference was not statistically significant ($p = 0.39$).

Discussion

This clinical cohort study illustrates the procedure, the clinical outcomes, and the patient perception of a novel surgical approach (MIST) designed to treat isolated deep intra-bony defects in combination with EMD.

The results preliminarily illustrate the potential benefits of this approach. In fact, a consistent amount of CAL gain was observed at 1 year (4.8 ± 1.9 mm in defects with an intra-bony component of 5.5 ± 1.5 mm) associated with substantial stability of the gingival margin and very shallow residual PD (Table 2). Very interestingly, the percent fill of the baseline intra-bony component of the defects in terms of CAL gain reached 100% in seven sites out of 13, indicating a complete resolution of the lesion in 54% of the cases. The average 1-year CAL% was $88.7 \pm 20.7\%$. Historical comparisons with other regenerative approaches clearly indicate that the results of this study in terms of CAL gains and defect resolution are in the top percentiles of the published evidence (Cortellini & Tonettti 2000, 2005, Rosen et al. 2000, Murphy & Gunsolley 2003).

These outcomes were obtained applying the MIST. This approach was designed to minimize surgical trauma and could entail for several advantages. Minimal flap elevation and reflection without involvement of the muco-gingival junction and without periosteal incisions potentially reduces some of the surgery-associated side-effects, like the amount of local bleeding during surgery, the ample exposure of both bone and connective tissue surfaces, the tension at the delicate inter-dental edges of the wound at the time of

suturing, and the amount of post-operative swelling and/or haematoma. These are relevant issues both for the healing process of the regenerative procedure and for the patient intra-operative and post-operative comfort. In this study, the early healing period was uneventful in all the cases (no oedema and or haematoma was detected in any of the treated sites) and only in one patient the defect-associated inter-dental papilla was found slightly open at 1 week. This very favourable healing process agrees well with the very promising clinical outcomes observed in this population. This hypothesis is supported by evidence of reduced clinical outcomes associated with a high prevalence of early complications (Murphy 1995, Nowzari et al. 1995, De Sanctis et al. 1996, Sanz et al. 2004).

A possible positive role in the healing process could also have been played by the improved post-operative wound stability due to the minimal flap elevation and the stable suturing technique. A study on dogs (Hiatt et al. 1968) demonstrated that the tensile strength of the tooth-gingival flap inter-face is minimal at the beginning of healing and increases up to a maximum within 2 weeks. These data support the hypothesis that wound integrity in the early healing phase is primarily dependent on the stability of the flaps and on the suturing technique. In an experimental model using heparin to pharmacologically impair the formation of the fibrin clot in dogs, Wikesjo & Nilveus (1990) and Haney et al. (1993) demonstrated that the fibrin clot and early wound stability play fundamental roles in periodontal wound healing. These studies suggest that providing adequate wound stability is a relevant factor for the establishment of a connective tissue attachment rather than a long junctional epithelium. In the present study, an improved stability of the healing area was attempted with a modified surgical approach designed to minimize flap mobility and tension following the application of sutures and to improve the gingival seal on top of the healing area.

The application of the MIST also resulted in a surgical chair-time of 55.4 ± 6.5 min. This time favourably compares with the average surgical chair-time of 80 ± 34 min. reported in a clinical trial on regeneration with EMD application in deep intra-bony defects (Tonetti et al. 2004b). In the latter study, the application of EMD was allowed by the elevation of more conventional and ample flaps associated with papilla preservation techniques (Cortellini et al. 1995, 1999), involving multiple inter-dental spaces and frequently the mucogingival junction; vertical-releasing incisions were often applied to allow a full reflection and an ample exposure of the defect area.

The patient perception of the procedure was very promising. No one reported any negative feeling during surgery that was described as being very easy and fast. Post-operative pain was not experienced by any of the patients. Only three subjects reported some ''discomfort'' during the first 2 post-operative days. The majority reported that the post-operative week was uneventful to the point that they could forget having been surgically treated in their mouth. This very positive patient perception is probably due to the very limited surgical trauma, to the rather short operative time, and to the lack of complications.

The clinical performance of this novel surgical approach requires the use of a magnification aid like an operating microscope associated with optimal illumination of the surgical field. In fact, the very limited flap reflection allows for a very limited angle of vision of the defect that can be accessed and instrumented mainly from the coronal side. This is especially true in deep defects: the deeper the defect, the more difficult the vision of the surgical area without a proper magnification and illumination. In addition, the very limited surgical access requires the adoption of very small surgical instruments to minimize tissue damage and to reach easily both the bony walls and the root surface. Surgical skill and experience are also probably very important requirements for the proper application of MIST.

In conclusion, this case cohort demonstrates the potential efficacy of MIST associated with EMD in the treatment of isolated deep intra-bony defects. A larger study is necessary to confirm and extend the reported positive preliminary outcomes.

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Clinical Relevance

Scientific rationale for study: There is a need to develop surgical approaches able to favour wound stability and primary closure of the flaps in order to improve the healing potential of regenerative therapy, and to reduce surgical trauma and patient side-effects. This could enhance clinical outcomes and patients' acceptance of the procedure.

Principal findings: Use of MIST and EMD resulted in remarkable clinical improvements, with the complete resolution of 54% of the treated intra-bony defects. Patients reported no post-operative pain and minimal discomfort.

Practical implications: Application of MIST in combination with EMD in the treatment of isolated deep intra-bony defects could help clinicians in increasing the rate of clinical success and the patients' acceptance of the procedure.