Combination Therapy of Intrabony Periodontal Defects Using an Er:YAG Laser and Enamel Matrix Protein Derivative: A Case Series

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Purpose: The aim of the present series of case reports was to evaluate the combination therapy of intrabony periodontal defects using an Er:YAG laser (ERL) and enamel matrix protein derivative (EMD).

Materials and Methods: Fifteen patients with chronic periodontitis, each of whom displayed one intrabony defect, were randomly treated with access flap surgery and root surface/defect debridement using ERL (16.5 J/cm^2) followed by the application of EMD. The following clinical parameters were recorded at baseline and at 6 months: plaque index (PI), bleeding on probing (BOP), probing pocket depth (PD), gingival recession (GR), and clinical attachment level (CAL).

Results: Healing was uneventful in all patients. At 6 months after therapy, the sites treated with ERL and EMD showed a mean PD reduction from 8.5 ± 1.1 mm to 4.7 ± 0.8 mm and a mean CAL change from 10.5 ± 1.4 mm to 7.6 ± 1.4 mm ($p < 0.001$).

Conclusion: Within the limits of this case series, it may be concluded that the combination of ERL and EMD may improve clinical healing of intrabony periodontal defects over a period of 6 months.

Keywords: case series, enamel proteins, intrabony defects, lasers, periodontal regeneration, therapeutic use.

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According to the cause-related concept of periodontal therapy, the main objective of treatment is to control infection and thereby arrest disease progression.[1] Ideally, periodontal therapy does not only include arresting the disease but also regeneration of the periodontal attachment, including cementum, a functionally oriented periodontal ligament, and alveolar bone.[2] Several treatment modalities, such as the use of different types of bone grafts, root surface demineralization, guided tissue regeneration (GTR), or the application of growth factors have been employed with varying degrees of success in order to predictably accomplish this goal.[3-7] Enamel matrix derivative (EMD) has also been introduced as an alternative modality in regenerative periodontal treatment.[8] The rationale for the clinical use of EMD is the observation that enamel matrix proteins (EMPs) are deposited along the surface of developing tooth roots prior to cementum formation.[8] It has been suggested that EMPs are involved in the formation of acellular extrinsic fiber cementum and may trigger differentiation of progenitor cells into cementoblasts.[8] Recent data have also indicated that EMD may contain additional mitogenic factors, such as TGF, and BMP-like growth factors, that stimulate fibroblastic proliferation and contribute to the induction of biomineralization during periodontal regeneration. [9-12] Indeed, histological findings from both animals[13] and humans[14-16] have shown that the application of EMD onto a debrided root surface may promote periodontal regeneration. Furthermore, clinical results have shown that treatment of intrabony defects with EMD may result in clinical outcomes comparable to those following GTR therapy.[17,18] Moreover, clinical trials have reported improved gains in clinical attachment levels compared to access flap surgery alone.[18-20]

Usually, periodontally diseased root surfaces are debrided using hand instruments. However, the formation of a smear layer after mechanical scaling and root planing (SRP) has been reported to be detrimental to periodontal tissue healing, as it may inhibit reattachment of cells to the root surface.[21,22] In order to improve the biocompatibility, root surface conditioning with various substances, such as ethylene diamine tetraacetic acid gel (EDTA) at neutral pH, citric and ortho-phosphoric acids, has been proposed.[21-24] The biological rationale of using EDTA for root surface conditioning is based on findings from experimental studies, indicating that EDTA operating at neutral pH appeared to effectively remove the smear layer produced by SRP and to selectively remove mineral from the dentin or cementum surface, exposing a collagenous matrix.[21,22,25,26] In contrast, etching with citric and phosphoric acids appeared to remove not only the mineral component but also the collagenous matrix.[21,22,25,26] The exposure of a collagenous matrix may also be favorable for retention of biologically active substances, such as EMD.[27]

In recent years, the use of laser radiation has also been expected to serve as an alternative or adjunctive treatment to conventional periodontal therapy. Various hypothetically advantageous characteristics, such as hemostatic effects, improved calculus removal, and bactericidal effects against periodontopathic pathogens, might lead to improved treatment outcomes.[28] Close attention has been paid to the clinical applicability of the Er:YAG laser (ERL) with a wavelength of 2.940 nm in the near infrared spectrum.[29] This laser system effectively removes calculus from periodontally diseased root surfaces without producing thermal side effects in adjacent tissues.[30-32] The absence of thermal damage was most likely due to the optical characteristics of its wavelength of 2.940 nm, since the ERL laser theoretically has a 10 and 15,000 to 20,000 times higher absorption coefficient of water than the CO_2 and the Nd:YAG lasers, respectively.[33,34] Additionally, recently published studies have reported a lack of cementum removal when laser instrumentation was performed under in vivo conditions.[31,32,35] Furthermore, several studies have reported antimicrobial effects against periodontopathic bacteria and the removal of lipopolysaccharides by ERL radiation.[36-40] These findings suggest that root surface debridement and detoxification using ERL may also faciliate a precipitation of EMD. Indeed, preliminary results from a pilot study have shown that the combination therapy ERL+EMD resulted in statistically significant clinical attachment level gains.[41] However, data on clinical outcomes following treatment of intrabony defects with ERL+EMD are still limited.

Therefore, the aim of the present series of case reports was to evaluate the combination therapy of intrabony periodontal defects using ERL and EMD.

MATERIALS AND METHODS

Study Population

Fifteen patients suffering from chronic periodontitis were included in the present series of case reports. The patient population comprised 7 men and 8 women (mean age: 49 ± 12 years). The inclusion criteria were: (1) no systemic diseases that could influence the outcome of the therapy; (2) a good level of oral hygiene (plaque index <1[42]); (3) compliance with the maintenance program; and (4) presence of one intrabony defect with a probing depth ≥ = 6 mm and an intrabony component of ≥ = 3 mm as detected on radiographs.

Patients who reported smoking only occasionally were not considered as smokers.[43] According to the given definition, there were 6 smokers included in the present study.

Three months prior to surgery, each patient was given thorough oral hygiene instructions, and full mouth supra- and subgingival scaling using ERL (KEY2/KEY3, KaVo, Biberach, Germany).

Clinical Measurements

The following clinical parameters were assessed 1 week prior to and 6 months after the surgical procedure using a periodontal probe (PCP 12, Hu-Friedy; Leimen, Germany): plaque index (PI),[42] bleeding on probing (BOP), probing depth (PD), gingival recession (GR), and clinical attachment level (CAL). The measurements were made at 6 sites per tooth: mesiovestibular (mv), midvestibular (v), distovestibular (dv), mesiolingual (ml), midlingual (l), and distolingual (dl) by one blinded and calibrated investigator. Five patients, each showing two pairs of contralateral teeth (single- and multi-rooted) with probing depths 6 mm at at least one site of each tooth, were used to calibrate the examiner. The examiner evaluated the patients on 2 separate occasions, 48 h apart. Calibration was accepted if measurements at baseline and at 48 h were within a millimeter 90% or more of the time. The cemento-enamel junction (CEJ) was used as the reference point. In cases where the CEJ was not visible, a restoration margin was used for these measurements. The study reports only measurements at the same deepest point of the selected defect. Prior to surgery and 6 months postoperatively, periapical radiographs were taken using the long-cone paralleling technique.

Surgical Procedure

All surgical procedures were performed under local anesthesia. Following intracrevicular incisions, full-thickness mucoperiosteal flaps were raised vestibularly and orally. Both the removal of the granulation tissue from the intrabony defects and debridement of root surfaces was performed using the ERL (KEY2/KEY3 without feedback system; KaVo) device emitting a pulsed infrared radiation at a wavelength of 2.94 µm. The laser beam was guided under water irrigation with a specially designed periodontal handpiece and a chisel-shaped glass fiber tip (size 0.5 x 1.65 mm) (2061, KaVo). Laser parameters were set at 160 mJ/pulse and 10 pulses/s, and pulse energy at the tip was approximately 136 mJ/pulse (16.5 J/cm^2).[44-46] Debridement of the root surfaces was performed from coronal to apical in parallel paths with an inclination of the fiber tip of 15 to 20 degrees[47] to the root surface. EMD (Emdogain, Straumann; Waldenburg, Switzerland) was applied to the root surfaces and into the defects according to the instructions given by the manufacturer. Finally, the flaps were repositioned coronally and closed with vertical or horizontal mattress sutures (Figs 1a to 1h). During surgery, the following measurements were made: distance from the CEJ to the bottom of the defect (CEJ-BD) and distance from the CEJ to the most coronal extension of the alveolar bone crest (CEJ-BC). The intrabony component (INTRA) of the defects was defined as (CEJ-BD) minus (CEJ-BC).

Postoperative Care

Postoperative care consisted of 0.2% chlorhexidine rinses twice a day for 4 weeks. The sutures were removed 10 days after surgery. Recall appointments were scheduled every second week during the first 2 postoperative months, and once a month for the rest of the observation period. Neither probing nor subgingival instrumentation was performed during the first six months after surgery.

Statistical Analysis

A software package (SPSS 13.0, SPSS; Chicago, IL, USA) was used for the statistical analysis. The primary outcome variable was CAL. In the calculations, only the deepest site per tooth was included. The data rows were examined with the Kolmogorow-Smirnow test for normal distribution. Accordingly, the paired t-test was used to statistically evaluate the changes from baseline to 6 months.

RESULTS

The depth and configurations of the intrabony components as assessed during surgery is presented in Table 1. Before surgery, one tooth received additional root canal treatment due to a combined periodontal and endodontic lesion.

Postoperative healing was considered as generally uneventful. Minor complications were related to the usual postoperative swelling and occurred within the first days after surgery. Neither allergic reactions nor suppuration or abscesses were observed in any of the patients.

Mean clinical parameters at baseline and after 6 months are summarized in Table 2. Mean PI remained low throughout the study period. In general, ERL+EMD resulted in statistically significant improvements of all clinical parameters investigated. In particular, BOP improved statistically significantly from 100% at baseline to 33% after 6 months ($p < 0.001$). Mean PD changed

Fig 1a Clinical situation at baseline.

Fig 1b Acute periodontal inflammation on the lingual aspect of tooth 32.

Fig 1c Bilateral intrasulcular incisions and mobilization of mucoperiosteal flaps.

Fig 1d Two-wall intrabony defect on the lingual aspect of tooth 32.

Fig 1e Removal of granulation tissue and root surface debridement using ERL.

Fig 1f Removal of a thin blood film from the root surface.

Fig 1g Application of EMD.

Fig 1h Wound closure.

Table 1 Baseline defect characteristics in mm (mean ± SD) (n = 15 patients)

Treatment	CEJ-BD	CEJ-BC	INTRA	1-2-wall	2-wall	3-wall
ERL+EMD	12.5 ± 1.4	8.7 ± 1.6	3.8 ± 0.9	4	9	2

Table 2 Clinical parameters (mean ± SD) at baseline and after 6 months (n = 15 patients)

	Baseline	6 months	Differencep	P value
PI	0.9 ± 0.7	0.6 ± 0.6	0.3 ± 0.2	> 0.05
BOP (%)	100%	33%	67%	< 0.001
PD (mm)	8.5 ± 1.1 mm	4.7 ± 0.8 mm	3.9 ± 0.9 mm	< 0.001
GR (mm)	2.0 ± 0.9 mm	2.9 ± 1.0 mm	0.9 ± 0.6 mm	< 0.001
CAL (mm)	10.5 ± 1.4 mm	7.6 ± 1.4 mm	3.0 ± 1.2 mm	< 0.001

from 8.5 ± 1.1 mm at baseline to 4.7 ± 0.8 mm after 6 months ($p < 0.001$), and mean CAL changed from 10.5 ± 1.4 mm at baseline to 7.6 ± 1.4 mm after 6 months ($p < 0.001$) (Figs 1i and 1j). Radiological observation revealed a decreased translucency within the intrabony component of each defect investigated (Figs 1k, 1l, and 2).

The sample size was too small to draw any conclusions regarding the effects of smoking on clinical outcomes.

DISCUSSION

The findings of the present case series have shown that treatment of intrabony periodontal defects with ERL+EMD may lead to clinically important and statistically significant PD reductions and CAL gains. The fact that all defects treated in this study healed uneventfully suggests that this treatment modality was well tolerated. The present results corroborate recent findings from a pilot study evaluating the clinical use of ERL+EMD and SRP+EDTA+EMD for the treatment of intrabony periodontal defects.[41] In particular, the sites treated with ERL+EMD showed a reduction in mean

Fig 1i Clinical situation after 6 months.

Fig 1j Healthy periodontal conditions also at the lingual aspect.

Fig 1k Preoperative radiograph.

Fig 1l Postoperative radiograph at 6 months.

PD from 8.6 ± 1.2 mm at baseline to 4.6 ± 0.8 mm after 6 months, and a change in mean CAL from 10.7 ± 1.3 mm at baseline to 7.5 ± 1.4 mm after 6 months. In the group treated with SRP+EDTA+EMD, the mean PD was reduced from 8.1 ± 0.8 mm at baseline to 4.0 ± 0.5 mm after 6 months, and the mean CAL changed from 10.4 ± 1.1 mm at baseline to 7.1 ± 1.2 mm after 6 months. In both groups, the differences between baseline and 6 months were statistically significant. However, the differences between the groups were not statistically significant.

The finding that treatment of intrabony periodontal defects with EMD may result in the short term (up to 1 year) in statistically significantly improvements in PD and CAL compared to baseline is in agreement with already reported results.[17,19,20,48] In particular, Hejil et al[19] reported a mean CAL gain of 2.1 mm after 8 months (baseline CAL: 9.4 mm: INTRA: 4.8 mm). There was a statistically significant difference between EMD- and placebo-treated sites. Similarly, Pontoriero et al[20] reported a mean CAL gain of 2.9 mm for EMD-treated sites after 1 year with a statistically significant

Fig 2a Preoperative radiograph.

Fig 2b Postoperative radiograph at 6 months. Before surgery, tooth 33 received an additional root canal treatment due to a combined periodontal and endodontic lesion.

difference between EMD- and placebo-treated sites (baseline CAL: 9.1 mm; INTRA 4.2 mm). Froum et al[48] reported a 4.26 mm CAL gain (baseline CAL: not reported; INTRA: 5.63 mm), and Sculean et al[17] reported a 3.4 mm CAL gain (baseline CAL: 10.6; INTRA 3.8 mm). In these clinical trials, the most frequently employed root conditioning agent was 24% EDTA.[17,20,48]

Furthermore, the present results obtained with ERL+EMD are in accordance with those from a case report evaluating EMD with neither etching nor chemical preparation of the root surfaces for the treatment of intrabony periodontal defects. At 12 months, the results demonstrated a mean PD reduction of 4.4 ± 1.3 mm and a mean CAL gain of 3.6 ± 1.2 mm.[49] These findings might also be supported by the results of a recent study, which failed to show statistically significant differences in terms of PD reduction and CAL gain following regenerative surgery with either EDTA+EMD or EMD alone.[50] All these data, taken together with the results of the present study seem to indicate that the clinical results may rather be attributed to the effect of EMD than to the root surface conditioning itself. In this context, however, it is important to realize that the presented clinical results need to be supported by histologic evidence, since it is still unclear to what extent the CAL gains obtained following ERL+EMD represent real periodontal regeneration rather than defect fill without new connective tissue attachment. Furthermore, the stability of the obtained CAL gains over time must be evaluated in further clinical studies.

Within the limits of a case report study, it may be concluded that the combination of ERL and EMD may improve clinical healing of intrabony periodontal defects over a period of 6 months.

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