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Five-year results following treatment of intrabony defects with enamel matrix proteins and guided tissue regeneration

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Abstract

Background: Treatment with enamel matrix proteins (EMD) or guided tissue regeneration (GTR) has been shown to enhance periodontal regeneration. However, until now there are limited data on the long-term results following these treatment modalities.

Aim: The aim of the present clinical study was to present the 5-year results following treatment of intrabony defects with EMD, GTR, combination of EMD and GTR, and open flap debridement (OFD).

Material and Methods: Forty-two patients, each of whom displayed one intrabony defect of a probing depth of at least 6 mm, were randomly treated with one of the four treatment modalities. The following parameters were evaluated prior to surgery, at 1 year and at 5 years after: plaque index, gingival index, bleeding on probing, probing pocket depth (PPD), gingival recession, and clinical attachment level (CAL). No statistically significant differences in any of the parameters were observed at baseline between the four groups.

Results: The sites treated with EMD demonstrated a mean CAL gain of 3.4 ± 1.1 mm ($p < 0.001$) and of 2.9 ± 1.6 mm ($p < 0.001$) at 1 and 5 years, respectively. The sites treated with GTR showed a mean CAL gain of 3.2 ± 0.8 ($p < 0.001$) at 1 year and of 2.7 ± 0.9 mm ($p < 0.001$) at 5 years. The mean CAL gain at sites treated with EMD+GTR was 3.0 ± 1.0 mm ($p < 0.001$) and 2.6 ± 0.7 mm ($p < 0.001$) at 1 and 5 years, respectively. The sites treated with OFD demonstrated a mean CAL gain of 1.6 ± 1.0 mm ($p < 0.001$) at 1 year and 1.3 ± 1.2 mm ($p < 0.001$) at 5 years. At 1 year, the only statistically significant difference between the four different treatments was found in terms of PPD reduction and CAL gain between EMD and OFD ($p < 0.05$). However, at 5 years there were no statistically significant differences in any of the investigated parameters between the four different treatments.

Conclusion: Within the limits of the present study, it may be concluded that the short-term clinical results following treatment with EMD, GTR, EMD+GTR, and OFD can be maintained over a period of 5 years.

Key words: bioresorbable membranes; enamel matrix proteins; guided tissue regeneration; long-term results

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Regenerative periodontal treatment with an enamel matrix protein (EMD) derivative or guided tissue regeneration (GTR) have been shown to promote periodontal regeneration in animal and human intrabony defects (Nyman et al. 1982, Gottlow et al. 1986, Sculean et al. 1999a, b, 2000a, b, Yukna & Mellonig 2000). Clinical studies have demonstrated that both therapies may lead to additional gain of clinical attachment level (CAL) when compared with baseline or flap surgery alone (Cortellini et al. 1996a, Heijl et al. 1997, Tonetti et al. 1998, 2002, Heden et al. 1999, Pontoriero et al. 1999, Sculean et al. 1999a, c, 2001a, Okuda et al. 2000, Silvestri et al. 2000, Froum et al. 2001, Trombelli et

al. 2002, Zuchelli et al. 2002). When comparing the clinical results obtained in intrabony defects, no significant differences were found between EMD or GTR therapy (Pontoriero et al. 1999, Sculean et al. 1999b, d, 2001a, Silvestri et al. 2000, Zuchelli et al. 2002). However, the histological and clinical results were not additionally improved when EMD was combined with GTR (Sculean et al. 2000a, 2001a). Although on a short-term basis, these therapies have been shown to improve significantly probing depths and CALs; there are still very limited data on the long-term results following these regenerative techniques. Moreover, there are virtually no data from controlled clinical studies evaluating the treatment of intrabony defects with EMD or a combination of EMD+GTR over a period of up to 5 years.

Therefore, the aim of this paper was to present the 5-year results following treatment of human intrabony defects with EMD, GTR, combination of EMD+GTR, and open flap debridement (OFD).

Material and Methods

Study population

The study population has been described in detail previously (Sculean et al. 2001a). Briefly, a total of 56 patients were included in the study based on signed informed consent. The study was in accordance with the Helsinki Declaration of 1975, as revised in 1983. However, only 42 patients with a mean age of (47 ± 14.5 years) completed the 5-year evaluation. Reasons for dropouts were death (one patient in the EMD group), moving to another area (two patients in the GTR group and two in the EMD+GTR group), and non-compliance (the rest of nine patients). Therefore, in the following only the data of the 42 available patients are presented.

Each patient received verbal and written explanations about the possible risks of the study and the possibility to withdraw at any time. All patients signed an informed consent form.

The criteria for patient selection were (1) presence of one intrabony defect of a probing depth of at least 6 mm, (2) no systemic diseases that could interfere with periodontal healing, (3) no use of antibiotics the least 6 months prior to treatment, and (4) good level of oral hygiene (i.e. a plaque index score, PlI < 1). Three months prior to surgery, all patients received oral hygiene instructions and full-mouth supra- and subgingival scaling and root planing under local anesthesia.

The following clinical measurements were made one week prior to, at 1 year, and at 5 years after surgery by one blinded and previously calibrated examiner: PlI (Sillness & Löe 1964), gingival index (GI) (Löe 1967), bleeding on probing (BOP), probing pocket depth (PPD), gingival recession (GR), and CAL. The measurements were made at six sites per tooth: mesiovestibular (mv), midvestibular (v), distovestibular (dv), mesiolingual (ml), midlingual (ml), and distolingual (dl) with the same type of manual periodontal probe (PCP 12, Hu-Friedy, Chicago, IL, USA). The cemento-enamel junction (CEJ) was used as a fixed reference for the CAL measurements. In cases where the CEJ was not clearly visible, a restoration margin was used for these measurements.

Surgical procedure and postoperative care

All operative procedures were performed by the same surgeon (A.S.). The surgical technique and the postoperative protocol have been described in great detail previously (Sculean et al. 2001a). The defects were randomly assigned to one of the following treatments:

1. EMD: (Emdogain®, BIORA, AB, Malmö, Sweden) (11 patients).
2. GTR: (Resolut®, Gore-Tex, Flagstaff, AZ, USA) (11 patients).
3. Combination: EMD+GTR (10 patients).
4. OFD: 10 patients.

Supportive periodontal therapy

Recall appointments were scheduled every second week during the first 2 months following surgery and then once per month for the first year postoperatively. After the first year and during the rest of the observation period of 5 years, patients were recalled every 3 months. The recall appointments consisted mainly of reinforcement of oral hygiene measures and professional supragingival tooth cleaning.

Statistical analysis

The statistical analysis was performed using a software program (SPSS® for Windows 95, SPSS Inc., Chicago, IL, USA). The primary outcome variable was CAL. In the calculations only the deepest measure per tooth was included. The same site was measured at 1 and at 5 years. First ANOVA was conducted to determine whether differences existed among the groups. Then post hoc range test and pairwise multiple comparisons of all possible combinations were performed using Scheffé's F-test. The alpha error was set at 0.05. The paired t-test was used to compare the data from baseline to those at 1 and at 5 years for each treatment group. The power of the study, given 1 mm as a significant difference between groups, was calculated to be 0.70.

Results

The observations on the early postoperative healing were described in detail elsewhere (Sculean et al. 2001a). Briefly, there were no postoperative complications such as allergic reactions against EMD or the membrane material, nor suppuration or abscesses.

Frequency distributions for PlI, GI, and BOP for all four treatment groups at baseline and after 1 and 5 years are summarized in Tables 1–3. The mean PlI did not reveal a statistically significant difference between the groups at baseline and after 1 and 5 years. Although at 5 years the PlI increased slightly in both treatment groups, this difference was not found to be statistically significant compared with the baseline or with the 1-year results. A statistically significant difference was observed in both treatment groups, when comparing the 1- and 5-year GI and BOP to the baseline values ($p < 0.001$). However, no statistically significant differences were observed between the 1- and the 5-year results (Tables 1–3).

The distribution of the defects according to their configuration is presented in Table 4.

The baseline defect characteristics are presented in Table 5. No statistically significant difference in the initial depth of the intrabony component was found between the four groups.

At 1 year, the PPD decreased statistically significantly in all four groups ($p < 0.001$). Comparing the four groups, the only statistically significant difference was found between EMD and OFD ($p = 0.042$). At 5 years, the PPD was still statistically significantly improved

Table 1. Frequency distribution of PII at the treated sites, at baseline at 1 and at 5 years

PII	EMD			GTR			EMD+GTR			OFD		
	baseline	1 year	5 years	baseline	1 year	5 years	baseline	1 year	5 years	baseline	1 year	5 years
	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)
0	8 (73)	9 (82)	6 (55)	7 (64)	8 (73)	6 (55)	5 (50)	8 (80)	4 (40)	5 (50)	8 (80)	4 (40)
1	3 (27)	2 (18)	5 (45)	4 (36)	3 (27)	5 (45)	5 (50)	2 (20)	6 (60)	5 (50)	2 (20)	6 (60)
2												
3												
Total no.		11			11			10			10	

Table 2. Frequency distribution of GI at the treated sites, at baseline at 1 and at 5 years

GI	EMD			GTR			EMD+GTR			OFD		
	baseline	1 year	5 years	baseline	1 year	5 years	baseline	1 year	5 years	baseline	1 year	5 years
	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)
0	4 (36)	8 (73)	5 (45)	5 (45)	8 (73)	7 (71)	3 (30)	7 (70)	6 (60)	4 (40)	7 (70)	6 (60)
1	4 (36)	3 (28)	5 (45)	3 (28)	3 (28)	4 (29)	4 (40)	3 (30)	4 (40)	6 (60)	3 (30)	4 (40)
2	3 (28)		1 (10)	3 (28)			3 (30)					
3												
Total no.		11			11			10			10	

Table 3. Frequency distribution of BOP at the treated sites, at baseline at 1 and at 5 years

BOP	EMD			GTR			EMD+GTR			OFD		
	baseline	1 year	baseline	baseline	1 year	5 years	1 year	5 years	5 years	baseline	1 year	5 years
	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)	no. (%)
+	8 (64)	3 (36)	5 (45)	9 (82)	3 (36)	5 (45)	7 (70)	3 (30)	4 (40)	6 (60)	3 (30)	4 (40)
−	3 (36)	8 (64)	6 (55)	2 (18)	8 (64)	6 (55)	3 (30)	7 (70)	6 (60)	4 (40)	7 (70)	6 (60)
Total no.		11			11			10			10	

Table 4. Distribution and configuration of treated defects

	EMD	GTR	EMD+GTR	OFD
1–2 wall	3	3	2	3
2 wall	7	6	7	5
3 wall	1	2	1	2

Table 5. baseline defect characteristics expressed in mm (mean ± SD)

Treatment	PPD	GR	CAL	CEJ-BBD	CEJ-crest	Intrabony depth
EMD	8.2 ± 1.1	1.7 ± 1.3	9.9 ± 1.4	11.0 ± 1.9	7.1 ± 1.2	3.9 ± 1.5
GTR	8.3 ± 1.3	1.6 ± 1.4	9.9 ± 1.7	10.8 ± 1.8	7.0 ± 1.3	3.8 ± 1.7
EMD+GTR	8.4 ± 1.0	1.4 ± 0.8	9.8 ± 1.2	10.9 ± 1.8	7.2 ± 1.3	3.7 ± 1.5
OFD	8.2 ± 1.1	1.5 ± 0.7	9.7 ± 0.8	10.7 ± 1.9	6.9 ± 1.8	3.8 ± 1.2

compared with baseline ($p<0.001$) (Table 6). However, between the groups no statistically significant differences were found.

At 1 year, the GR increased statistically significantly ($p<0.001$) in all four groups compared with the baseline, but the difference between the groups was not significant (Table 6). No statistically significant differences between the four groups were found at 5 years.

At 1 year, the CAL improved statistically significantly in all four groups compared with the baseline ($p<0.001$) (Table 6). When comparing the groups the only statistically significant difference was found between EMD and OFD ($p=0.031$). At 5 years, the CAL was statistically significantly improved in all four groups compared with baseline ($p<0.001$). However, between the groups no statistically significant differences were found (Table 6). Due to the fact that the number of smokers was limited (one in the EMD group, one in the GTR group, two in the EMD+GTR group and one in the OFD group), no analysis was performed in order to evaluate the effect of smoking on the clinical outcome.

Table 6. Changes of clinical parameters at 1 and 5 years compared to baseline (mean ± SD)

Parameter	OFD	EMD	GTR	EMD+GTR
ΔPPD (mm) 1 year	3.3 ± 1.1	4.6 ± 1.2	4.4 ± 1.4	4.4 ± 0.8
ΔPPD (mm) 5 years	2.7 ± 1.2	4.3 ± 1.7	3.9 ± 1.6	4.0 ± 1.0
ΔGR (mm) 1 year	1.7 ± 0.5	1.3 ± 0.6	1.3 ± 1.0	1.5 ± 0.7
ΔGR (mm) 5 years	1.7 ± 0.5	1.3 ± 0.7	1.2 ± 1.0	1.5 ± 0.7
ΔCAL (mm) 1 year	1.6 ± 1.0	3.4 ± 1.1	3.2 ± 0.8	3.0 ± 1.0
ΔCAL (mm) 5 years	1.3 ± 1.2	2.9 ± 1.6	2.7 ± 0.9	2.6 ± 0.7

Discussion

The results of the present study have shown that treatment of intrabony defects with EMD, GTR, EMD+GTR, and OFD may result in statistically significant reductions in PPD and gains of CAL that can be maintained over a period of 5 years. At 1 year, the only statistically significant difference between the four different treatments was found in terms of PPD reduction and CAL gain between EMD and OFD ($p < 0.05$). At 5 years, however, there were no statistically significant differences in any of the investigated parameters between the four treatments.

The finding that treatment with EMD may result in statistically significant short-term improvements in terms of PPD reduction and CAL gain is consistent with previously published data (Heijl et al. 1997, Heden et al. 1999, Pontoriero et al. 1999, Sculean et al. 1999c, d, 2001a, Okuda et al. 2000, Silvestri et al. 2000, Froum et al. 2001, Tonetti et al. 2002, Trombelli et al. 2002, Zuchelli et al. 2002). It is important to realize that until now there are no published 5-year data on the clinical outcome following treatment with EMD or EMD+GTR and therefore, a comparison with other long-term studies is difficult. On the other hand, the present 5-year data on EMD treatment are in agreement with recent results from a case report study and a controlled split-mouth study evaluating this treatment modality over a period of 4 years (Sculean et al. 2001b, 2003). In a case report study evaluating a total of 46 consecutively treated intrabony defects in 33 patients, mean CAL gains of 3.0 and 3.2 mm were obtained at 1 and 4 years, respectively (Sculean et al. 2003). Comparable results were obtained in a 4-year follow-up split-mouth study comparing treatment with EMD to GTR. The results revealed that in the EMD group, the mean CAL gain was 3.4 mm at 1 year and 3.0 mm at 4 years (Sculean et al. 2001b).

The finding that the short-term outcome (after 1 year) following treatment with both GTR and access flap surgery did not show significant difference compared with the 5-year results corroborates previously reported data that have shown that the clinical improvements obtained with these treatments can be maintained on a long-term basis if an optimal patient and defect selection are accomplished and the patients are enrolled in an adequate maintenance program (Ramfjord et al. 1987, Cortellini et al. 1996b, Kahldahl et al. 1996, Tonetti et al. 1996, Sculean et al. 2001b, Eicholz & Hausmann 2002). This is in line with the fact that in none of the four groups differences in terms of PlI, GI, and BOP between the 1- and 5-year results were found. However, it has to be pointed out that in all four groups a slight, statistically not significant, loss of mean CAL was measured between the 1- and 5-year evaluation period that in turn was probably due to the increase (statistically not significant) of mean PPD. Although at 5 years, the increase in PlI, GI, and BOP did not reach statistical significance compared with the baseline, and with the 1-year values, it cannot be excluded that the plaque accumulation might have led to inflammation and loss of CAL.

When interpreting the present results, it also has to be noted that in the present follow-up study only 42 out of originally 56 patients were included. Therefore, it cannot be excluded that if available, the rest of the 14 patients might have yielded different clinical results at 5 years. The lack of these data might also serve as an explanation for the fact that in the present study, the only statistically significant difference between the four treatments was found at 1 year between EMD and OFD in terms of PPD reduction and CAL gain.

Although findings from histological studies have failed to show predictable regeneration of the attachment apparatus following OFD (Caton et al. 1980, Bowers et al. 1989, Sculean et al. 2000a), our results indicate that this treatment option may also lead to significant and stable clinical results on a long-term basis. Thus, treatment of intrabony defects with OFD may still be considered a valuable option for treating intrabony defects.

Another important factor that was demonstrated to influence strongly the outcome of regenerative periodontal treatment is smoking. However, due to the fact that only five out of the 42 patients were smokers, no conclusion can be drawn with respect to this issue.

In the present study, treatment with EMD+GTR has led to significant clinical improvements on both short- and long-term basis, but the improvements were not significantly higher than those obtained following treatment with EMD alone or GTR alone. Comparable results were obtained in recent experimental study in monkeys evaluating treatment of experimentally created intrabony defects with EMD, GTR, EMD+GTR, or OFD (Sculean et al. 2000a). The histological evaluation has indicated that treatment with EMD+GTR may enhance the formation of new attachment and new bone, but the amount of the newly formed tissues was not superior to treatment with EMD or GTR. All these data taken together may suggest that a combination of EMD+GTR does not seem to improve the results additionally.

Within the limits of the present study, it may be concluded that the short-term clinical results following treatment with EMD, GTR, EMD+GTR, and OFD can be maintained over a period of 5 years.

References

Bowers, G. M., Chadroff, B., Carnevale, R., Mellonig, J., Corio, R., Emerson, J., Stevens, M. & Romberg, E. (1989) Histologic evaluation of new attachment apparatus in humans. Part I. *Journal of Periodontology* **60**, 675–682.

Caton, J., Nyman, S. & Zander, H. (1980) Histometric evaluation of periodontal surgery. II. Connective tissue attachment after four regenerative procedures. *Journal of Clinical Periodontology* **7**, 224–231.

Cortellini, P., Pini Prato, G. P. & Tonetti, M. S. (1996a) Periodontal regeneration of human intrabony defects with bioresorbable membranes. A controlled clinical trial. *Journal of Periodontology* **67**, 217–223.

Cortellini, P., Pini Prato, G. P. & Tonetti, M. S. (1996b) Long-term stability of clinical attachment following guided tissue regenera-

tion and conventional therapy. *Journal of Clinical Periodontology* **23**, 106–111.

Eicholz, P. & Hausmann, E. (2002) Evidence for healing of periodontal defects 5 years after conventional and regenerative therapy: digital subtraction and bone level measurements. *Journal of Clinical Periodontology* **29**, 922–928.

Froum, S. J., Weinberg, M. A., Rosenberg, E. & Tarnow, D. (2001) A comparative study utilizing open flap debridement with and without enamel matrix derivative in the treatment of periodontal intrabony defects: a 12-month re-entry study. *Journal of Periodontology* **72**, 25–34.

Gottlow, J., Nyman, S., Lindhe, J., Karring, T. & Wennström, J. (1986) New attachment formation in the human periodontium by guided tissue regeneration. Case reports. *Journal of Clinical Periodontology* **13**, 604–616.

Heden, G., Wennström, J. & Lindhe, J. (1999) Periodontal tissue alterations following Emdogain® treatment of periodontal sites with angular bone defects. A series of case reports. *Journal of Clinical Periodontology* **26**, 855–860.

Heijl, L., Heden, G., Svardström, G. & Östgren, A. (1997) Enamel matrix derivative (Emdogain®) in the treatment of intrabony periodontal defects. *Journal of Clinical Periodontology* **24**, 705–714.

Kahldahl, W. B., Kalkwarf, K. L., Patil, K. D., Molvar, M. P. & Dyer, J. K. (1996) Long-term evaluation of periodontal therapy: I. Response to 4 therapeutic modalities. *Journal of Periodontology* **67**, 93–102.

Löe, H. (1967) The gingival index, the plaque index and the retention index systems. *Journal of Periodontology* **38**, 610–616.

Nyman, S., Lindhe, J., Karring, T. & Rylander, H. (1982) New attachment following surgical treatment of human periodontal disease. *Journal of Clinical Periodontology* **9**, 290–296.

Okuda, K., Momose, M., Miyazaki, A., Murata, M., Yokoyama, S., Yonezawa, Y., Wolff, L. & Yoshie, H. (2000) Enamel matrix derivative in the treatment of human intrabony osseous defects. *Journal of Periodontology* **71**, 1821–1828.

Pontoriero, R., Wennström, J. & Lindhe, J. (1999) The use of barrier membranes and enamel matrix proteins in the treatment of angular bone defects. A prospective controlled clinical study. *Journal of Clinical Periodontology* **26**, 833–840.

Ramfjord, S., Caffesse, R. G., Morrison, E. G., Hill, R., Kerry, G. J., Appleberry, E., Nissle, R. R. & Stults, J. (1987) Four modalities of periodontal treatment compared over 5 years. *Journal of Clinical Periodontology* **14**, 445–452.

Sculean, A., Chiantella, G. C., Miliauskaite, A., Brecx, M. & Arweiler, N. B. (2003) Four-year results following treatment of intrabony periodontal defects with an enamel matrix protein derivative. A report of 46 cases. *The International Journal of Periodontics & Restorative Dentistry* **23**, 345–351.

Sculean, A., Chiantella, G. C., Windisch, P. & Donos, N. (2000b) Clinical and histologic evaluation of treatment of intrabony defects with an enamel matrix protein derivative (Emdogain®). *The International Journal of Periodontics & Restorative Dentistry* **20**, 375–381.

Sculean, A., Donos, N., Blaes, A., Lauermann, M., Reich, E. & Brecx, M. (1999d) Comparison of enamel matrix proteins an bioabsorbable membranes in the treatment of intrabony defects. A split-mouth study. *Journal of Periodontology* **70**, 255–262.

Sculean, A., Donos, N., Brecx, M., Reich, E. & Karring, T. (2000a) Treatment of intrabony defects with enamel matrix proteins and guided tissue regeneration. An experimental study in monkeys. *Journal of Clinical Periodontology* **27**, 466–472.

Sculean, A., Donos, N., Chiantella, G. C., Windisch, P., Reich, E. & Brecx, M. (1999a) Treatment of intrabony defects with bioabsorbable membranes. A clinical and histologic study. *The International Journal of Periodontics & Restorative Dentistry* **19**, 501–509.

Sculean, A., Donos, N., Miliauskaite, A., Arweiler, N. & Brecx, M. (2001b) Treatment of Intrabony defects with enamel matrix proteins or bioabsorbable membranes. A 4-year follow-up split-mouth study. *Journal of Periodontology* **72**, 1695–1701.

Sculean, A., Donos, N., Windisch, P., Gera, I., Brecx, M., Reich, E. & Karring, T. (1999b) Healing of human intrabony defects following treatment with enamel matrix proteins or guided tissue regeneration. *Journal of Periodontal Research* **34**, 310–322.

Sculean, A., Reich, E., Chiantella, C. G. & Brecx, M. (1999c) Treatment of intrabony periodontal defects with an enamel matrix protein derivative (Emdogain®): A report of 32 cases. *The International Journal of Periodontics & Restorative Dentistry* **19**, 157–163.

Sculean, A., Windisch, P., Chiantella, G. C., Donos, N., Brecx, M. & Reich, E. (2001a) Treatment of intrabony defects with enamel matrix proteins and guided tissue regeneration. A prospective controlled clinical study. *Journal of Clinical Periodontology* **28**, 397–403.

Silness, J. & Löe, H. (1964) Periodontal disease in pregnancy (II). Correlation between oral hygiene and periodontal condition. *Acta Odontologica Scandinavica* **22**, 121–135.

Silvestri, M., Ricci, G., Rasperini, G., Sartori, S. & Cattaneo, V. (2000) Comparison of treatments of intrabony defects with enamel matrix derivative, guided tissue regeneration with a nonresorbable membrane and Widman modified flap. A pilot study. *Journal of Clinical Periodontology* **27**, 603–610.

Tonetti, M. S., Cortellini, P., Suvan, J. E., Adriaens, P., Baldi, C., Dubravec, D., Fonzar, A., Fourmousis, I., Magnani, C., Muller-Campanile, V., Patroni, S., Sanz, M., Vangsted, T., Zabalegui, I., Pini Prato, G. & Lang, N. P. (1998) Generalizability of the added benefits of guided tissue regeneration in the treatment of deep intrabony defects. Evaluation in a multi-center randomized controlled clinical trial. *Journal of Periodontology* **69**, 1183–1192.

Tonetti, M. S., Lang, N. P., Cortellini, P., Suvan, J. E., Adriaens, P., Dubravec, D., Fonzar, A., Fourmousis, I., Mayfield, L., Rossi, R., Silvestri, M., Tiedemann, C., Topoll, H., Vangsted, T. & Wallkamm, B. (2002) Enamel matrix proteins in the regenerative therapy of deep intrabony defects. A multicentre randomized controlled clinical trial. *Journal of Clinical Periodontology* **29**, 317–325.

Tonetti, M. S., Pini Prato, G. & Cortellini, P. (1996) Factors affecting the healing response of intrabony defects following guided tissue regeneration and access flap surgery. *Journal of Clinical Periodontology* **23**, 548–556.

Trombelli, L., Bottega, S. & Zuchelli, G. (2002) Supracrestal soft tissue preservation with enamel matrix proteins in treatment of deep intrabony defects. A report of 35 consecutively treated cases. *Journal of Clinical Periodontology* **29**, 433–439.

Yukna, R. A. & Mellonig, J. (2000) Histologic evaluation of periodontal healing in humans following regenerative therapy with enamel matrix derivative. A 10-case series. *Journal of Periodontology* **71**, 752–759.

Zuchelli, G., Bernardi, F., Montebugnoli, L. & DeSanctis, M. (2002) Enamel matrix proteins and guided tissue regeneration with titanium-reinforced expanded polytetrafluoroethylene membranes in the treatment of intrabony defects: a comparative controlled clinical trial. *Journal of Periodontology* **73**, 3–12.

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