J Clin Periodontol 2010; 37: 59–79 doi: 10.1111/j.1600-051X.2009.01499.x

Journal of Clinical Periodontology

Review Article

Do bone grafts or barrier membranes provide additional treatment effects for infrabony lesions treated with enamel matrix derivatives? A network meta-analysis of randomized-controlled trials

Yu-Kang Tu[1,2], Andrew Woolston[2] and Clovis Mariano Faggion, Jr.[3]

[1]Department of Periodontology, Leeds Dental Institute, Leeds, UK; [2]Centre for Epidemiology & Biostatistics, Division of Biostatistics, University of Leeds, Leeds, UK; [3]Department of Prosthodontics, School of Dentistry, University of Heidelberg, Heidelberg, Germany

Tu Y-K, Woolston A, Faggion CM Jr. Do bone grafts or barrier membranes provide additional treatment effects for infrabony lesions treated with enamel matrix derivatives? A network meta-analysis of randomized-controlled trials. J Clin Periodontol 2010; 37: 59–79. doi: 10.1111/j.1600-051X.2009.01499.x.

Abstract

Background/Aim: Network meta-analyses of randomized-controlled trials were undertaken to investigate whether enamel matrix derivatives (EMD) in conjunction with other regenerative materials yield better treatment outcomes than EMD alone in the treatment of infrabony defects ≥ 3 mm.

Material and Methods: A literature search was conducted using the Medline, EMBASE, LILACS and CENTRAL databases up to and including December 2008. Treatment outcomes were changes in probing pocket depth (PPD), clinical attachment level (CAL) and infrabony defect depth. Different types of bone grafts (or barrier membranes) were first treated as a group and then separately.

Results: Twenty-eight studies were included in the review. EMD plus bone grafts and EMD plus membranes attained 0.24 mm [95% high probability density (HPD) intervals: -0.38, 0.65] and 0.07 mm (95% HPD intervals: -1.26, 1.04) more PPD reduction than EMD alone, respectively. For CAL gain, EMD plus bone grafts and EMD plus membranes attained 0.46 mm (95% HPD intervals: -0.17, 0.83) and 0.15 mm (95% HPD intervals: -1.37, 0.30), respectively. When different types of bone grafts and barrier membranes were treated separately, EMD with bovine bone grafts showed greater treatment effects.

Conclusion: There was little evidence to support the additional benefits of EMD in conjunction with other regenerative materials.

Key words: enamel matrix protein derivatives; network meta-analysis; randomized-controlled trials

Accepted for publication 4 October 2009

Conflict of interest and source of funding statement
The authors declare that they have no conflict of interests.

The first author was funded by the United Kingdom government's Higher Education Funding Council for England (HEFCE) and currently holds a UK Research Council Fellowship. The second author is supported by a UK Medical Research Council studentship.

In the last decade, the introduction of enamel matrix derivatives (EMD) in the treatment of periodontal lesions has led periodontal regeneration into a new era of tissue engineering. While its clinical efficacy is well established, little is known about the molecular mechanisms of its activities (Esposito et al. 2005, Bosshardt 2008, Palmer et al. 2008, Trombelli & Farina 2008). EMD has been received with great enthusiasm, and this has been shown in the numerous publications in the literature (Kalpitis & Ruben 2002, Venezia et al. 2004, Esposito et al. 2005). EMD has been used for treatments of periodontal lesions, such as furcation involvement (Hoffmann et al. 2006) and infrabony defects (Esposito et al. 2005, Tu et al. 2008).

In several recent studies, EMD has been used in conjunction with other regenerative materials such as barrier membranes and bone grafts in the treatment of infrabony lesions (Zucchelli et al. 2003, Gurinsky et al. 2004, Sculean et al. 2005, Bokan et al. 2006, Kuru et al. 2006), presumably assuming that combination therapy is likely to yield better treatment outcomes than the use of EMD alone. Nevertheless, as recognized by the Consensus Report in a recent European Workshop on Periodontology (Palmer et al. 2008), the high number of possible combinations being tested makes it difficult to disentangle the role of different components of combination therapy. Moreover, it is possible that, under some circumstances, a combination might antagonize the bioactive agent (Palmer et al. 2008).

The use of combination therapy also raised the issue of cost-effectiveness. Most regenerative materials are expensive, and combination therapy increases the treatment cost and surgical time. To justify the uses of these expensive treatments, combination therapy is expected to provide substantial, additional treatment effects than the use of EMD alone.

Several recent systematic reviews compared EMD with periodontal flap operation in the treatment of infrabony lesions (Kalpitis & Ruben 2002, Venezia et al. 2004, Esposito et al. 2005, Tu et al. 2008), but only one broad qualitative systematic review has considered whether or not EMD in conjunction with the uses of barrier membranes and/or bone grafts provides better treatment outcomes than EMD alone (Trombelli & Farina 2008). As there are many combination therapies being tested in the literature, the aim of this study was to conduct a network meta-analysis, an emerging new methodology for evidence synthesis, to investigate whether the combination therapy yields better clinical outcomes in the treatment of infrabony defects compared with the use of EMD alone.

Material and Methods

Literature search

We adopted modified selection criteria for the inclusion of randomized-controlled trials (RCTs) in this meta-analysis, as reported by a recently updated review on the efficacy of EMD in the treatment of periodontal infrabony lesions (Esposito et al. 2005). For studies to be included, the patients/defects needed to be randomly allocated to either the test or the control groups, and treatment outcomes were measured at baseline and at least 6 months after the treatments. Where a trial reported results at 6-month of follow-up and results longer than that, the results at 6 months or later were all used in the meta-analysis if the numbers of studies are considered sufficient (≥ 4) for the meta-analysis. Studies that included treatment of shallow infrabony defects <3 mm were excluded. Three groups of studies were included in this meta-analysis: the first group is studies comparing EMD or possible combinations to flap operation; the second included studies comparing combination therapy with EMD alone; and the third included studies comparing different combination therapies. While studies in the second group provided direct evidence to the comparisons between combination therapies and EMD alone, studies in the other two groups as a whole provided evidence for indirect comparisons in the network meta-analysis. For instance, while the aim is to compare two active treatments A and B, results from studies comparing A with placebo with B to placebo nevertheless provide evidence for indirect comparisons between A and B. In our network meta-analysis, flap operation is considered as the ''placebo'' group. Note that for studies of the third group to be included, EMD has to be one component of the combination therapy. For example, studies that compared EMD with guided tissue regeneration (GTR) were not included. An electronic search of the database MEDLINE, EMBASE, LILACS and CENTRAL from January 1996 to December 2008 was undertaken to identify relevant studies using the following key words: ''emdogain'', ''enamel matrix proteins'', ''infrabony, intrabony, or intraosseous''. The reference lists of previously published systematic reviews on EMD (Kalpitis & Ruben 2002, Venezia et al. 2004, Esposito et al. 2005, Tu et al. 2006, Trombelli & Farina 2008) were cross-checked. Electronic searching of the journal websites of three major periodontal journals, Journal of Clinical Periodontology, Journal of Periodontology and Journal of Periodontal Research, was also undertaken. Electronic literature search and data extractions were undertaken in duplicate, and quality assessment of included studies, such as randomization, allocation concealment, blinding, intention to treat and sample size calculation, was carried out independently by two authors (Y. K. T. and C. M. F.). Disagreements on study inclusions and quality assessment were resolved by discussions between two authors.

We also searched for non-published and published RCTs in ClinicalTrials.gov and used the internet search engine ''Google'' in English, French, Spanish, German and Italian. We used the key word Emdogain for articles published in the language of the original search engine (e.g. articles in Italian in Google Italy, articles in German in Google Germany, etc.).

Network meta-analyses

Network meta-analysis is a methodology for the statistical synthesis of direct and indirect comparisons of different treatments (Lumley 2002, Psaty et al. 2003, Lu & Ades 2004, Caldwell et al. 2005, Glenny et al. 2005, Chou et al. 2006, Coleman et al.

2008, Thijs et al. 2008, Cipriani et al. 2009). The treatment outcomes used in our meta-analysis were changes in probing pocket depth (PPD), clinical attachment level (CAL) and infrabony defect depth recorded at least 6 months after the lesions were treated by periodontal regeneration. Changes in the bone level might be measured clinically or radiographically. When the standard deviations (SD) for the outcomes were not available, they were derived from the SD of baseline and 12-month measurements by assuming that their correlations were 0.5 based on data in a previous review (Tu et al. 2005). When multiple lesions were treated within one patient, the standard errors of means (SEM) for treatment effects were derived from the reported SD by using the number of patients as the unit of analysis. Because the commercial product of EMD has evolved over the last decade and there are many different combination therapies, the network meta-analysis was undertaken in two stages. In the first stage, the following assumptions were made in constructing the network: firstly, it was assumed that there was no difference in treatment effects between treatment groups that did or did not carry out root conditioning procedures before the application of EMD, and that the vehicle for EMD, propylene glycol alginate (PGA), was not effective in periodontal regeneration. Secondly, there was no difference in the additional treatment effects for EMD in conjunction with different types of bone grafts. Thirdly, there was no difference in the additional treatment effects for EMD in conjunction with different types of barrier membranes. In the second stage, the second and third assumptions were relaxed by estimating possible different additional treatment effects among studies that used different types of bone grafts or barrier membranes.

The network meta-analyses were undertaken using the random-effects modelling as described by Whitehead (2002) and Glenny et al. (2005). The basic random-effects model is given as

$$y_{ij} = \hat{y} + u + v + uv + \varepsilon_{ij} \quad (1)$$

where y_{ij} is the observed treatment effect of treatment group i in study j, $\hat{y}$ is the mean treatment effect of the reference group (i.e. defects treated with EMD alone), u is the fixed effects of different treatment groups (i.e. different combination therapies or flap operation), v is the study-level random effects, uv is the random effects for the interaction between treatment groups and studies and ε_{ij} is the residuals. Details of the rationales and estimations of this model have been described elsewhere (Whitehead 2002, Glenny et al. 2005, Brown & Prescott 2006). When studies reported treatment outcomes at different follow-ups after surgery, the length of follow-up was modelled as both random and fixed effects. Study design (parallel groups *versus* split mouth) was also modelled as both random and fixed effects to account for the heterogeneity in treatment effects. To explore the potential bias caused by regression to the mean (Tu & Gilthorpe 2007), the baseline measurements of treatment outcomes were included as covariates in a further analysis. Standard pair-wise meta-analyses of direct comparisons among different combination therapies, EMD alone and flap operation were also carried out and compared with the results from the network meta-analysis. The inverse of SEM for treatment effects was used as the weights in the meta-analysis. Corresponding authors of original studies were contacted by email for clarifications of dubious data and requests for missing data.

The random-effects network meta-analysis was performed using the function lmer in the package lme4 (Bates & Maechler 2009) for the statistical software R (version 2.9.1, R Foundation for Statistical Computing, Vienna, Austria) with the estimation procedure of restricted maximum likelihood. Markov Chain Monte Carlo methods with 50,000 simulations were then used to obtain 95% high probability density (HPD) intervals (interpreted as confidence intervals) (Baayen 2008). The standard pair-wise random-effects meta-analysis was performed using statistical software STATA (version 10.1, StataCorp LP, College Station, TX, USA).

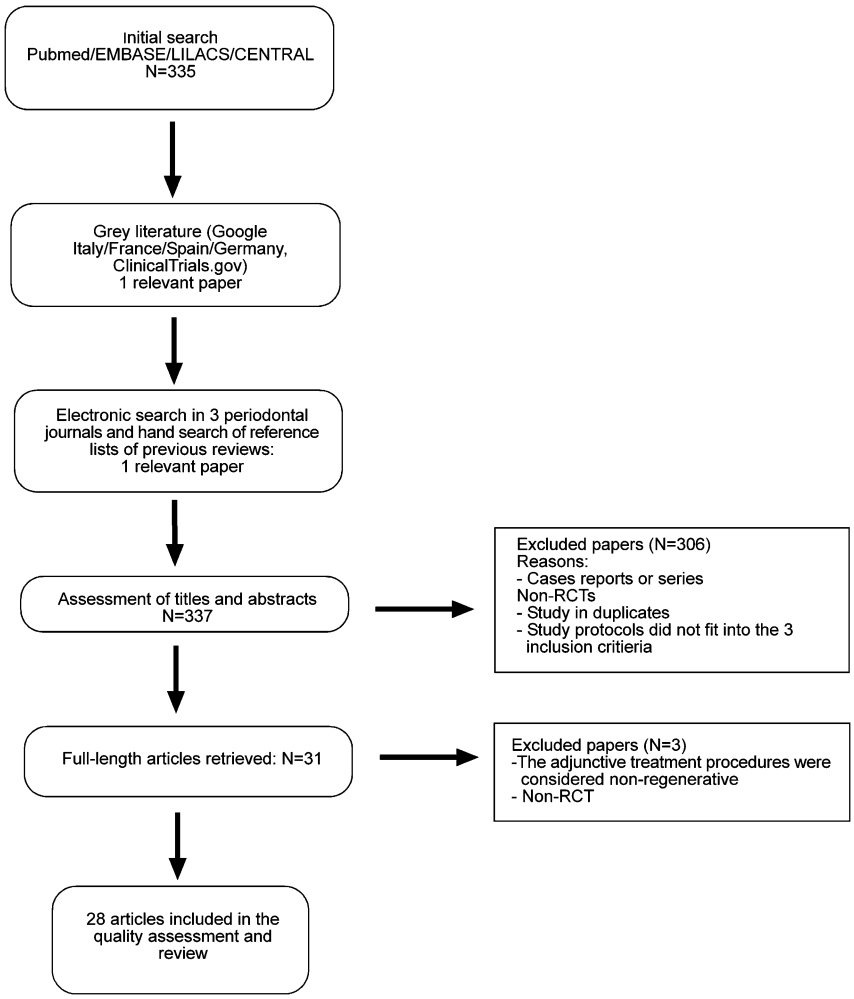

Fig. 1. Flowchart for literature search and identifications of articles for review.

Table 1. Quality assessment of randomized-controlled trials (RCTs) included for network meta-analysis

	Described as randomized	Randomization methods	Allocation concealment method	Patient blinding*	Caregiver blinding†	Examiner blinding	All patient accounted for at end of study	Analysis accounts for patient losses	Sample size/ statistical power calculation
Bokan et al. (2006)	Yes/abstract	Unclear	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Camargo et al. (2001)	Yes/text	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Chambrone et al. (2007)	Yes/title	Unclear	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Yes. Statistical power = 0.8
Döri et al. (2005)	Yes/abstract	Yes/adequate	Unclear	Unclear	Not applicable	Unclear	Yes	Not applicable	Seems post hoc analysis. Statistical power = 0.7
Döri et al. (2008)	Yes/abstract	Yes/block approach	Unclear	NA	Not applicable	Yes/adequate	Yes	Not applicable	Statistical power = 0.8
Francetti et al. (2004)	Yes/abstract	Yes/adequate	Unclear	Yes	Not applicable	Yes/adequate	Yes: 12 months No: 24 months	Not applicable for 12 months Unclear for 24 months	Not reported
Francetti et al. (2005)	Yes/abstract	Yes/adequate	Yes/adequate	Unclear	Not applicable	Unclear	Yes	Unclear	Not reported
Froum et al. (2001)	Talk on flip coin in text	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Unclear	Unclear	Not reported
Guida et al. (2007)	Yes/abstract	Unclear	Unclear	Unclear	Not applicable	Radiographical: yes/adequate Clinical: no	Yes	Not applicable	Statistical power = 0.85
Gurinsky et al. (2004)	Yes/abstract	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Heijl et al. (1997)	Yes/abstract	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Yes: 8 months No: 16 months	Not applicable: 8 months Unclear: 16 months	A statistical power = 0.8 was specified based on site-level analysis.
Jepsen et al. (2008)	Yes/title	Yes/adequate	Yes/adequate	Unclear	Not applicable	Yes/adequate	Yes	Yes	Yes. Statistical power = 0.8
Kuru et al. (2006)	Yes/text	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not clear. Seems post hoc
Minabe et al. (2002)	Yes/text	Unclear	Unclear	NA	Not applicable	Yes/adequate (except for defect morphology)	Yes	Not applicable	Not reported
Lekovic et al. (2000)	Yes/text	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Lekovic et al. (2001)	Yes/text	Yes/adequate	Unclear	NA	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Okuda et al. (2000)	Yes/abstract	Yes/adequate	Unclear	Yes	Not applicable	Unclear	Yes	Not applicable	Not reported
Pontoriero et al. (1999)	Yes/abstract	Unclear	Unclear	NA	Not applicable	Unclear	Yes	Not applicable	Not reported
Rösing et al. (2005)	Talk on flip coin in text	Yes/adequate	Unclear	Unclear	Not applicable		Yes	Unclear	Not clear. Seems post hoc

						Clinical: unclear Radiographical: yes/adequate			
Sculean et al. (2001b)	Yes/abstract	Toss of a coin after surgical debridement	Unclear	NA	Not applicable	Yes/adequate	Maybe yes (Not explicit)	Not clear	Seems post hoc. Statistical power = 0.72
Sculean et al. (2005)	Yes/abstract	Yes/adequate	Unclear	Unclear	Not applicable	Maybe but not explicit	Maybe yes (not explicit)	Not applicable	Not clear. Seems post hoc
Silvestri et al. (2000)	Yes/text	Unclear (there is just a citation)	Unclear	NA‡	Not applicable	Unclear	Yes	Not applicable	Not reported
Sipos et al. (2005)	Yes/abstract	Yes/adequate	Unclear	NA	Not applicable	Yes/adequate	Yes	Unclear	Not clear. Seems post hoc
Tonetti et al. (2002)	Yes/title	Yes/adequate	Yes/adequate	Unclear	Not applicable	Unclear	Yes	Unclear	Yes. Statistical power = 0.9
Velasquez-Plata et al. (2002)	Yes/text	Unclear	Unclear	Unclear	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Wachtel et al. (2003)	Yes/abstract	Yes/adequate	Unclear	Unclear	Not applicable	Yes/adequate	Unclear	Unclear	Not clear. Seems post hoc
Zucchelli et al. (2002)	Yes/text	Yes/adequate	Unclear	NA‡	Not applicable	Yes/adequate	Yes	Not applicable	Not reported
Zucchelli et al. (2003)	Yes/text	Yes/adequate	Unclear	Unclear	Not applicable	Unclear	Yes	Not applicable	Treatment difference threshold not described. Statistical power = 0.85

*Patient blinding is considered not possible, when EMD is compared with EMD in conjunction with GTR or platelet-rich plasma.

†Although it can be argued that caregiver blinding might be possible (but might not feasible) for the comparisons between EMD and flap operation by applying PGA gel to sites treated with flap operation (e.g. Heijl et al. 1997), this was only doable after the preparation of the commercial product of EMD no longer required mixing EMD powders with PGA gel before the application. Therefore, caregiver blinding is considered not possible for all comparisons, as different materials and procedures are used for different groups.

‡There was another group treated with GTR.

EMD, enamel matrix derivatives; PGA, propylene glycol alginate; GTR, guided tissue regeneration; NA, not applicable.

Table 2. Summary of studies included in the network meta-analysis for probing pocket depth (PPD) reduction

Study	Treatment groups	Treatment effects (mm)	SD	F/u	Study design	Baseline PPD
Bokan et al. (2006)	EMD+24% EDTA	3.90	1.30	12	Parallel group	8.60
Bokan et al. (2006)	EMD+24% EDTA+Cerasorb (TCP)	4.10	1.20	12	Parallel group	8.60
Bokan et al. (2006)	Flap operation	3.80	1.80	12	Parallel group	9.80
Döri et al. (2005)	EMD+24% EDTA+Bio-Oss	4.80	0.90	12	Parallel group	7.90
Döri et al. (2005)	EMD+24% EDTA+Cerasorb (TCP)	4.60	0.80	12	Parallel group	7.80
Döri et al. (2008)	EMD+24% EDTA+Bio-Oss	5.90	1.30	12	Parallel group	8.80
Döri et al. (2008)	EMD+24% EDTA+Bio-Oss+PRP	5.80	1.80	12	Parallel group	8.80
Francetti et al. (2004)	EMD+24% EDTA	4.71	1.60	12	Parallel group	7.86
Francetti et al. (2004)	Flap operation	2.57	1.27	12	Parallel group	6.71
Francetti et al. (2005)	EMD+24% EDTA	4.00	2.01	12	Parallel group	8.06
Francetti et al. (2005)	Flap operation	3.00	1.47	12	Parallel group	7.11
Froum et al. (2001)	EMD+citric acid	4.94	0.45	12	Split mouth	7.99
Froum et al. (2001)	Flap operation	2.24	0.91	12	Split mouth	7.32
Guida et al. (2007)	EMD+24% EDTA+autogenous bone grafts	5.10	1.70	12	Parallel group	9.10
Guida et al. (2007)	EMD+24% EDTA	5.60	1.70	12	Parallel group	9.60
Heijl et al. (1997)	EMD	3.30	1.40	16	Split mouth	7.80
Heijl et al. (1997)	Flap operation+PGA	2.60	1.20	16	Split mouth	7.80
Minabe et al. (2002)	EMD+37% orthophosphoric acid gel+GTR (Tissue Guide)	4.30	1.60	12	Parallel group	7.90
Minabe et al. (2002)	EMD+37% orthophosphoric acid gel	3.80	0.90	12	Parallel group	7.80
Okuda et al. (2000)	EMD+37% orthophosphoric acid gel	3.00	0.97	12	Split mouth	6.33
Okuda et al. (2000)	Flap op+PGA+37% orthophosphoric acid gel	2.22	0.81	12	Split mouth	6.22
Pontoriero et al. (1999)	EMD+24% EDTA	4.40	1.05	12	Split mouth	8.00
Pontoriero et al. (1999)	Flap operation+PGA	3.50	1.25	12	Split mouth	7.90
Rösing et al. (2005)	EMD+24% EDTA	4.17	1.59	12	Split mouth	7.57
Rösing et al. (2005)	Flap op+PGA+24% EDTA	4.39	1.50	12	Split mouth	7.38
Sculean et al. (2001b)	EMD+24% EDTA+GTR (Resolut)	4.30	1.40	12	Parallel group	8.60
Sculean et al. (2001b)	EMD+24% EDTA	4.10	1.70	12	Parallel group	8.40
Sculean et al. (2001b)	Flap operation	3.70	1.40	12	Parallel group	8.60
Sculean et al. (2005)	EMD+24% EDTA+Perioglass	4.20	1.40	12	Parallel group	8.50
Sculean et al. (2005)	EMD+24% EDTA	4.50	2.00	12	Parallel group	8.50
Silvestri et al. (2000)	EMD+17% EDTA	4.90	1.79	12	Parallel group	7.70
Silvestri et al. (2000)	Flap operation	1.40	1.26	12	Parallel group	7.70
Sipos et al. (2005)	EMD+24% EDTA+GTR (ePTFE)	3.02	1.55	12	Split mouth	7.32
Sipos et al. (2005)	EMD+24% EDTA	2.86	0.75	12	Split mouth	6.95
Tonetti et al. (2002)	EMD+24% EDTA	3.90	1.70	12	Parallel group	8.00
Tonetti et al. (2002)	Flap operation+24% EDTA	3.30	1.70	12	Parallel group	7.70
Wachtel et al. (2003)	EMD+24% EDTA	3.90	1.40	12	Split mouth	7.00
Wachtel et al. (2003)	Flap op	2.10	1.10	12	Split mouth	6.50
Zucchelli et al. (2002)	EMD+24% EDTA	5.10	0.70	12	Parallel group	9.20
Zucchelli et al. (2002)	Flap operation+24% EDTA	4.50	1.00	12	Parallel group	8.90
Zucchelli et al. (2003)	EMD+24% EDTA+Bio-Oss	6.20	0.40	12	Parallel group	9.40
Zucchelli et al. (2003)	EMD+24% EDTA	5.80	0.80	12	Parallel group	9.20
Camargo et al. (2001)	EMD+24% EDTA+Bio-Oss	3.91	1.39	6	Split mouth	7.30
Camargo et al. (2001)	Flap op	1.59	1.36	6	Split mouth	7.09
Chambrone et al. (2007)	EMD+24% EDTA	3.75	1.12	6	Split mouth	6.42
Chambrone et al. (2007)	Flap op	4.08	0.98	6	Split mouth	6.08
Gurinsky et al. (2004)	EMD+24% EDTA	4.00	0.30	6	Parallel group	7.50
Gurinsky et al. (2004)	EMD+24% EDTA+DFDBA	3.60	0.20	6	Parallel group	7.50
Jepsen et al. (2008)	EMD+BoneCeramic (60%HA+40%TCP)	1.93	1.80	6	Parallel group	6.90
Jepsen et al. (2008)	EMD	2.55	1.80	6	Parallel group	7.10
Lekovic et al. (2000)	EMD+24% EDTA+Bio-Oss	3.40	1.34	6	Split mouth	7.46
Lekovic et al. (2000)	EMD+24% EDTA	1.88	1.40	6	Split mouth	7.24
Lekovic et al. (2001)	EMD+24% EDTA+BIO-Oss+GTR (Bio-Gide)	4.85	1.50	6	Split mouth	8.39
Lekovic et al. (2001)	Flap op	2.87	0.87	6	Split mouth	8.39
Velasquez-Plata et al. (2002)	EMD+24% EDTA+Bio-Oss	4.00	0.80	6	Split mouth	6.90
Velasquez-Plata et al. (2002)	EMD+24% EDTA	3.80	1.20	6	Split mouth	6.60
Guida et al. (2007)*	EMD+24% EDTA+autogenous bone grafts	4.60	1.30	6	Parallel group	9.10
Guida et al. (2007)*	EMD+24% EDTA	5.10	1.90	6	Parallel group	9.60
Minabe et al. (2002)*	EMD+37% orthophosphoric acid gel+GTR (Tissue Guide)	4.20	1.73	6	Parallel group	7.90
Minabe et al. (2002)*	EMD+37% orthophosphoric acid gel	3.60	1.13	6	Parallel group	7.80
Rösing et al. (2005)*	EMD+24% EDTA	3.72	1.40	6	Split mouth	7.57
Rösing et al. (2005)*	Flap op+24% EDTA	4.02	1.36	6	Split mouth	7.38
Sipos et al. (2005)*	EMD+24% EDTA+GTR (ePTFE)	2.95	1.29	6	Split mouth	7.32
Sipos et al. (2005)*	EMD+24% EDTA	2.26	1.03	6	Split mouth	6.95

Table 2. (Contd.)

Study	Treatment groups	Treatment effects (mm)	SD	F/u	Study design	Baseline PPD
Wachtel et al. (2003)*	EMD+24% EDTA	3.30	1.20	6	Split mouth	7.00
Wachtel et al. (2003)*	Flap op	2.20	0.80	6	Split mouth	6.50
Kuru et al. (2006)	EMD+24% EDTA	5.03	0.89	8	Parallel group	9.47
Kuru et al. (2006)	EMD+24% EDTA+Perioglass	5.73	0.80	8	Parallel group	9.77
Heijl et al. (1997)	EMD	3.30	1.30	8	Split mouth	7.80
Heijl et al. (1997)	Flap op+PGA	2.60	1.30	8	Split mouth	7.80

*These studies reported treatment outcomes at 6 (or 8) and 12 (or 16) months from baseline.
F/u, length of follow-up in months.; PGA, propylene glycol alginate; EDTA, ethylenediaminetetraacetic acid; EMD, enamel matrix derivatives; TCP, tricalcium phosphates; PRP, platelet-rich plasma; DFDBA, demineralized freeze-fried bone allografts; SD, standard deviation.

Table 3. Summary of studies included in the network meta-analysis for CAL gain

Study	Treatment groups	Treatment effects (mm)	SD	F/u	Study design	Baseline CAL
Bokan et al. (2006)	EMD+24% EDTA	3.70	1.00	12	Parallel group	10.30
Bokan et al. (2006)	EMD+24% EDTA+Cerasorb (TCP)	4.00	1.00	12	Parallel group	9.80
Bokan et al. (2006)	Flap operation	2.10	1.40	12	Parallel group	10.20
Döri et al. (2005)	EMD+24% EDTA+Bio-Oss	4.30	0.80	12	Parallel group	8.80
Döri et al. (2005)	EMD+24% EDTA+Cerasorb (TCP)	4.10	0.80	12	Parallel group	8.80
Döri et al. (2008)	EMD+24% EDTA+Bio-Oss	5.00	0.90	12	Parallel group	10.50
Döri et al. (2008)	EMD+24% EDTA+Bio-Oss+PRP	4.80	1.30	12	Parallel group	10.80
Francetti et al. (2004)	EMD+24% EDTA	4.14	1.35	12	Parallel group	9.43
Francetti et al. (2004)	Flap operation	2.29	0.95	12	Parallel group	8.29
Francetti et al. (2005)	EMD+24% EDTA	3.41	2.15	12	Parallel group	8.91
Francetti et al. (2005)	Flap operation	1.96	2.17	12	Parallel group	7.98
Froum et al. (2001)	EMD+citric acid	4.26	0.56	12	Split mouth	*
Froum et al. (2001)	Flap operation	2.75	0.93	12	Split mouth	*
Guida et al. (2007)	EMD+24% EDTA+autogenous bone grafts	4.90	1.80	12	Parallel group	10.30
Guida et al. (2007)	EMD+24% EDTA	4.60	1.30	12	Parallel group	10.60
Heijl et al. (1997)	EMD	2.30	1.60	16	Split mouth	9.40
Heijl et al. (1997)	Flap operation+PGA	1.70	1.20	16	Split mouth	9.30
Minabe et al. (2002)	EMD+37% orthophosphoric acid gel+GTR (Tissue Guide)	3.00	1.30	12	Parallel group	8.50
Minabe et al. (2002)	EMD+37% orthophosphoric acid gel	2.60	1.00	12	Parallel group	8.60
Okuda et al. (2000)	EMD+37% orthophosphoric acid gel	1.72	1.07	12	Split mouth	6.72
Okuda et al. (2000)	Flap op+PGA+37% orthophosphoric acid gel	0.83	0.86	12	Split mouth	6.83
Pontoriero et al. (1999)	EMD+24% EDTA	3.00	1.00	12	Split mouth	9.10
Pontoriero et al. (1999)	Flap operation+PGA	1.80	1.21	12	Split mouth	8.60
Rösing et al. (2005)	EMD+24% EDTA	2.01	1.96	12	Split mouth	12.93
Rösing et al. (2005)	Flap op+PGA+24% EDTA	2.16	2.57	12	Split mouth	13.47
Sculean et al. (2001b)	EMD+24% EDTA+GTR (Resolut)	3.40	1.10	12	Parallel group	10.00
Sculean et al. (2001b)	EMD+24% EDTA	3.40	1.50	12	Parallel group	10.60
Sculean et al. (2001b)	Flap operation	1.70	1.50	12	Parallel group	10.10
Sculean et al. (2005)	EMD+24% EDTA+Perioglass	3.20	1.70	12	Parallel group	10.40
Sculean et al. (2005)	EMD+24% EDTA	3.90	1.80	12	Parallel group	10.20
Silvestri et al. (2000)	EMD+17% EDTA	4.50	1.58	12	Parallel group	9.10
Silvestri et al. (2000)	Flap operation	1.20	1.03	12	Parallel group	8.70
Sipos et al. (2005)	EMD+24% EDTA+GTR (ePTFE)	1.65	1.29	12	Split mouth	11.10
Sipos et al. (2005)	EMD+24% EDTA	1.28	2.04	12	Split mouth	10.94
Tonetti et al. (2002)	EMD+24% EDTA	3.10	1.50	12	Parallel group	9.40
Tonetti et al. (2002)	Flap operation+24% EDTA	2.50	1.50	12	Parallel group	9.10
Wachtel et al. (2003)	EMD+24% EDTA	3.60	1.60	12	Split mouth	7.70
Wachtel et al. (2003)	Flap op	1.70	1.40	12	Split mouth	7.20
Zucchelli et al. (2002)	EMD+24% EDTA	4.20	0.90	12	Parallel group	9.90
Zucchelli et al. (2002)	Flap operation+24% EDTA	2.60	0.80	12	Parallel group	10.00

Table 3. (Contd.)

Study	Treatment groups	Treatment effects (mm)	SD	F/u	Study design	Baseline CAL
Zucchelli et al. (2003)	EMD+24% EDTA+Bio-Oss	5.80	1.10	12	Parallel group	10.30
Zucchelli et al. (2003)	EMD+24% EDTA	4.90	1.00	12	Parallel group	10.10
Camargo et al. (2001)	EMD+24% EDTA+Bio-Oss	3.45	1.35	6	Split mouth	*
Camargo et al. (2001)	Flap op	1.43	1.31	6	Split mouth	*
Chambrone et al. (2007)	EMD+24% EDTA	2.67	2.10	6	Split mouth	13.42
Chambrone et al. (2007)	Flap op	1.84	2.12	6	Split mouth	12.42
Gurinsky et al. (2004)	EMD+24% EDTA	3.20	0.30	6	Parallel group	8.10
Gurinsky et al. (2004)	EMD+24% EDTA+DFDBA	3.00	0.30	6	Parallel group	8.20
Jepsen et al. (2008)	EMD+BoneCeramic(60% HA+40% TCP)	1.31	1.80	6	Parallel group	9.30
Jepsen et al. (2008)	EMD	1.83	1.80	6	Parallel group	10.10
Lekovic et al. (2000)	EMD+24% EDTA+Bio-Oss	3.12	1.40	6	Split mouth	0.00
Lekovic et al. (2000)	EMD+24% EDTA	1.74	1.35	6	Split mouth	0.00
Lekovic et al. (2001)	EMD+24% EDTA+BIO-Oss+GTR (Bio-Gide)	3.84	1.15	6	Split mouth	*
Lekovic et al. (2001)	Flap op	1.50	0.81	6	Split mouth	*
Velasquez-Plata et al. (2002)	EMD+24% EDTA+Bio-Oss	3.40	0.90	6	Split mouth	*
Velasquez-Plata et al. (2002)	EMD+24% EDTA	2.90	0.90	6	Split mouth	*
Guida et al. (2007)†	EMD+24% EDTA+autogenous bone grafts	4.40	1.50	6	Parallel group	10.30
Guida et al. (2007)†	EMD+24% EDTA	4.40	1.30	6	Parallel group	10.60
Minabe et al. (2002)†	EMD+37% orthophosphoric acid gel+GTR (Tissue Guide)	2.90	1.80	6	Parallel group	8.50
Minabe et al. (2002)†	EMD+37% orthophosphoric acid gel	2.70	1.44	6	Parallel group	8.60
Rösing et al. (2005)†	EMD+24% EDTA	1.58	1.83	6	Split mouth	12.93
Rösing et al. (2005)†	Flap op+24% EDTA	2.42	2.54	6	Split mouth	13.47
Sipos et al. (2005)†	EMD+24% EDTA+GTR (ePTFE)	1.45	1.60	6	Split mouth	7.32
Sipos et al. (2005)†	EMD+24% EDTA	0.94	1.51	6	Split mouth	6.95
Wachtel et al. (2003)†	EMD+24% EDTA	2.80	1.30	6	Split mouth	7.70
Wachtel et al. (2003)†	Flap op	2.00	1.30	6	Split mouth	7.20
Kuru et al. (2006)	EMD+24% EDTA	4.06	1.06	8	Parallel group	*
Kuru et al. (2006)	EMD+24% EDTA+Perioglass	5.17	0.85	8	Parallel group	*
Heijl et al. (1997)	EMD	2.10	1.50	8	Split mouth	9.40
Heijl et al. (1997)	Flap op+PGA	1.50	1.10	8	Split mouth	9.30

*These studies did not report baseline CAL, and so they were excluded from the adjusted network meta-analysis.

†These studies reported treatment outcomes at 6 (or 8) and 12 (or 16) months from baseline.

PRP, platelet-rich plasma; DFDBA, demineralized freeze-fried bone allografts; F/u, length of follow-up in months; PGA, propylene glycol alginate; TCP, tri-calcium phosphates; EDTA, ethylenediaminetetraacetic acid; EMD, enamel matrix derivatives; CAL, clinical attachment level; SD, standard deviation.

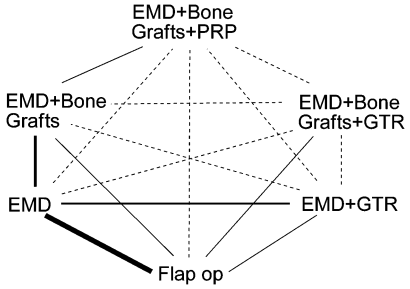

Fig. 2. Network for the comparisons among different combination therapies, enamel matrix derivatives (EMD) alone and flap operation. Dotted lines refer to those comparisons that have not been tested directly in randomized-controlled trials. The width of the solid lines is in proportion to the amount of evidence available in the literature.

Results

The flow of literature search is shown in Fig. 1. Eventually, 31 studies were identified from the literature search. After retrieving full articles, one study was excluded because its design was to test the effects of different post-surgical antibiotics regimes (Sculean et al. 2001a); one was excluded because it aimed to test whether or not root conditioning with 24% EDTA before the application of EMD made any difference in the treatment outcomes (Sculean et al. 2006), and another was excluded because it is unclear whether it is an RCT and none of the SDs of treatment effects were reported (Forabosco et al. 2003). A study comparing EMD with two different bone grafts (Döri et al. 2005) was only included in the second-stage network meta-analysis. Table 1 shows the results of trial quality assessment for the studies included. Of the 28 studies, only 14 reported changes in infrabony defect depths measured clinically or radiographically. Tables 2–4 present summaries of the studies included. Most studies reported no serious complications, except mild swel-

Table 4. Summary of studies included in the network meta-analysis for clinical or radiographical infrabony defect fill

Study	Treatment groups	Tx effects (mm)	SD	F/u	Study design	Baseline depth
Francetti et al. (2004)	EMD+24% EDTA	2.96	1.13	12	Parallel group	5.93
Francetti et al. (2004)	Flap op	1.44	0.74	12	Parallel group	4.81
Francetti et al. (2005)	EMD+24% EDTA	3.09	2.13	12	Parallel group	5.76
Francetti et al. (2005)	Flap op	1.86	1.60	12	Parallel group	5.26
Froum et al. (2001)	EMD+citric acid	3.83	0.61	12	Split mouth	5.63
Froum et al. (2001)	Flap op	1.47	0.71	12	Split mouth	4.11
Heijl et al. (1997)	EMD	2.20	1.60	16	Split mouth	7.10
Heijl et al. (1997)	Flap op+PGA	−0.20	0.60	16	Split mouth	6.50
Rösing et al. (2005)	EMD+24% EDTA	1.55	2.22	12	Split mouth	6.95
Rösing et al. (2005)	Flap op+24% EDTA	1.39	1.33	12	Split mouth	6.56
Zucchelli et al. (2003)	EMD+24% EDTA+Bio-Oss	5.20	1.10	12	Parallel group	6.10
Zucchelli et al. (2003)	EMD+24% EDTA	4.30	1.50	12	Parallel group	6.10
Guida et al. (2007)	EMD+24% EDTA+autogenous bone grafts	4.30	1.30	12	Parallel group	6.50
Guida et al. (2007)	EMD+24% EDTA	4.30	2.40	12	Parallel group	6.50
Sipos et al. (2005)	EMD+24% EDTA+GTR (ePTFE)	1.58	1.92	12	Split mouth	12.90
Sipos et al. (2005)	EMD+24% EDTA	1.63	1.21	12	Split mouth	12.70
Camargo et al. (2001)	EMD+24% EDTA+Bio-Oss	3.82	1.52	6	Split mouth	7.30
Camargo et al. (2001)	Flap op	1.06	1.02	6	Split mouth	7.09
Jepsen et al. (2008)	EMD+BoneCeramic(60% HA+40% TCP)	2.07	2.10	6	Parallel group	6.70
Jepsen et al. (2008)	EMD	2.07	1.20	6	Parallel group	6.90
Lekovic et al. (2000)	EMD+24% EDTA+Bio-Oss	3.78	1.41	6	Split mouth	*
Lekovic et al. (2000)	EMD+24% EDTA	1.37	1.18	6	Split mouth	*
Lekovic et al. (2001)	EMD+24% EDTA+Bio-Oss+ GTR (Bio-Gide)	4.79	1.37	6	Split mouth	*
Lekovic et al. (2001)	Flap op	1.73	0.91	6	Split mouth	*
Velasquez-Plata et al. (2002)	EMD+24% EDTA+Bio-Oss	4.00	0.80	6	Split mouth	5.30
Velasquez-Plata et al. (2002)	EMD+24% EDTA	3.10	1.00	6	Split mouth	4.90
Gurinsky et al. (2004)	EMD+24% EDTA+DFDBA	3.70	0.20	6	Parallel group	4.90
Gurinsky et al. (2004)	EMD+24% EDTA	2.60	0.40	6	Parallel group	5.20
Sipos et al. (2005)†	EMD+24% EDTA+GTR (ePTFE)	1.25	1.99	6	Split mouth	12.90
Sipos et al. (2005)†	EMD+24% EDTA	1.41	2.20	6	Split mouth	12.70
Heijl et al. (1997)†	EMD	0.90	0.60	8	Split mouth	7.10
Heijl et al. (1997)†	Flap op+PGA	−0.10	0.40	8	Split mouth	6.50
Rösing et al. (2005)†	EMD+24% EDTA	0.33	2.35	6	Split mouth	6.95
Rösing et al. (2005)†	Flap op+24% EDTA	1.20	1.03	6	Split mouth	6.56

*These studies did not report baseline CAL, and so they were excluded from the adjusted network meta-analysis.

†These studies reported treatment outcomes at 6 (or 8) and 12 (or 16) months from baseline.

TCP, tri-calcium phosphates; PRP, platelet-rich plasma; DFDBA, demineralized freeze-fried bone allografts; F/u, length of follow-up in months; PGA, propylene glycol alginate; EDTA, ethylenediaminetetraacetic acid; EMD, enamel matrix derivatives; CAL, clinical attachment level; SD, standard deviation.

ling and pain after surgery, and the most serious post-operative complication was membrane exposure in lesions where a GTR procedure was undertaken along with the application of EMD (Sculean et al. 2001b, Sipos et al. 2005).

Stage 1 network meta-analysis

Twenty-seven studies were grouped into six nodes in the network meta-analysis, yielding 15 possible pairs of comparisons (Fig. 2). Evidence of direct comparisons was only available in seven of the 15 pairs (Figs 2 and 3). Fourteen studies reported treatment outcomes at 6–8 months after surgery and 21 studies reported results at 12–16 months (Tables 2–4). To simplify the presentation and analysis, results at 6–8 months were pooled together as the 6-month group, and results at 12–16 months were pooled together as the 12-month group. Two studies reported results at 24 months and one at 36 months after surgery. As the numbers of studies reporting results at 24 months or longer were too few to yield robust and meaningful results, the meta-analyses were only conducted on 6- or 12-month groups. The length of follow-up in months (6- *versus* 12-month group) was included as both fixed and random effects in the network meta-analysis, and results from the tradition pair-wise meta-analysis were based on the stratifications of follow-up lengths.

For PPD reduction and CAL gain, four studies compared EMD with flap operation after 6–8 months of follow-up, and 13 studies did that after 12–16 months of follow-up. Six studies compared EMD with EMD in conjunction with bone grafts after 6–8 months, and four studies compared these treatments after 12–16 months of follow-up. No more than three studies were found for the other five comparisons. For infrabony defect fill, four studies compared EMD with EMD in conjunction with bone grafts at 6 months, and five studies compared FMD with flap operation at 12 months. No more than two studies were found for other comparisons.

The results from network meta-analysis showed that the mean PPD reduction for EMD alone at 12 months was 4.48 mm, which was 0.93 mm (95% HPD intervals: 0.56, 1.54) greater than that for flap operation (Table 5). EMD in

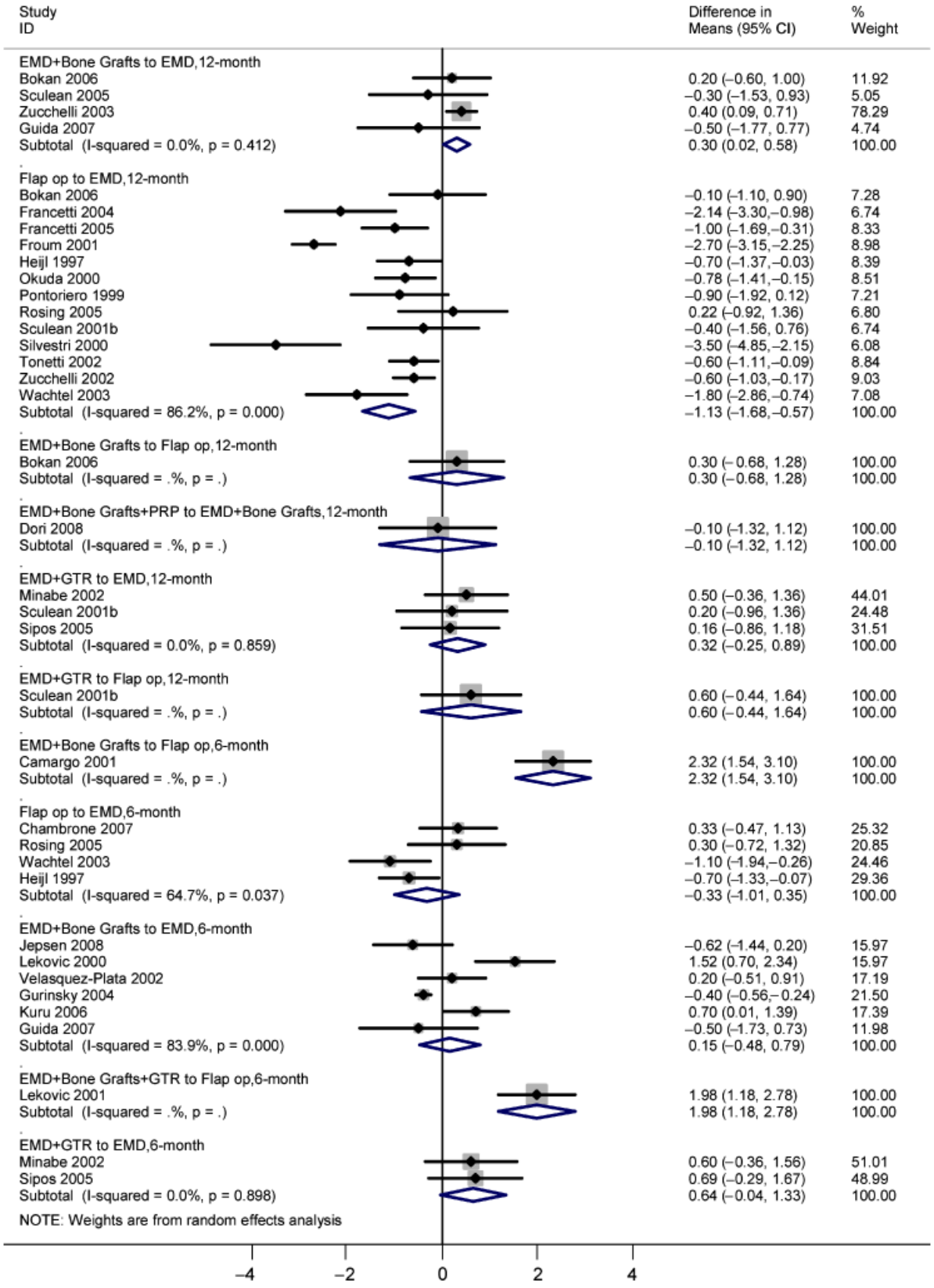

Fig. 3. Forest plot of the standard pair-wise meta-analysis for probing pocket depth reduction when different types of bone grafts were grouped together and different types of barrier membranes were grouped together.

Table 5. Results of network and standard meta-analysis for PPD reduction

	Unadjusted network meta-analysis		Adjusted network meta-analysis		Standard pair-wise meta-analysis: 12 months		Standard pair-wise meta-analysis: 6 months	
	estimates	95% HPD intervals	estimates	95% HPD intervals	estimates	95% CI	estimates	95% CI
Stage 1								
Intercept	4.48*	(− 3.14, 11.52)	− 1.67	(− 8.62, 4.44)				
EMD+bone grafts	0.24	(− 0.38, 0.65)	0.24	(− 0.35, 0.46)	0.30	(0.02, 0.58)	0.15	(− 0.48, 0.79)
EMD+bone grafts+GTR	1.17	(− 0.97, 3.42)	0.96	(− 0.95, 2.47)				
EMD+bone grafts+PRP	0.62	(− 2.10, 3.83)	0.57	(− 1.63, 2.99)				
EMD+GTR	0.07	(− 1.26, 1.04)	0.03	(− 0.93, 0.82)	0.32	(− 0.25, 0.89)	0.64	(− 0.04, 1.33)
Flap op	− 0.93	(− 1.54, − 0.56)	− 0.76	(− 1.19, − 0.43)	− 1.13	(− 1.68, − 0.57)	− 0.33	(− 1.01, 0.35)
F/u = 6 months	− 0.23	(− 10.59, 9.96)	− 0.22	(− 8.99, 8.12)				
Split-mouth design	− 0.77	(− 1.32, − 0.06)	0.06	(− 0.45, 0.72)				
Stage 2								
Intercept	4.49*	(− 2.57, 11.57)	− 0.86	(− 7.23, 4.87)				
EMD+autogenous bones	− 0.05	(− 1.50, 1.94)	− 0.11	(− 1.48, 1.31)	− 0.50	(− 1.77, 0.77)	− 0.50	(− 1.73, 0.73)
EMD+Bio-Oss	0.78	(0.15, 1.37)	0.70	(0.09, 1.07)	0.40	(0.09, 0.71)	0.84	(− 0.45, 2.14)
EMD+Bio-Oss+Bio-Gide	1.28	(− 0.71, 3.21)	1.01	(− 0.75, 2.46)				
EMD+Bio-Oss+PRP	1.01	(− 1.58, 3.73)	0.85	(− 1.29, 2.97)				
EMD+TCP	0.04	(− 1.21, 1.03)	0.26	(− 0.81, 1.02)	0.20	(− 0.60, 1.00)		
EMD+DFDBA	− 0.53	(− 1.20, 0.21)	− 0.26	(− 0.88, 0.30)			− 0.40	(− 0.56, − 0.24)
EMD+HA	− 1.49	(− 3.59, 0.02)	− 0.93	(− 2.62, 0.34)			− 0.62	(− 1.44, 0.20)
EMD+Perioglass	0.43	(− 0.45, 1.96)	0.16	(− 0.79, 1.20)	− 0.30	(− 1.53, 0.93)	0.70	(0.01, 1.39)
EMD+ePTFE	− 0.04	(− 1.88, 1.58)	− 0.17	(− 1.66, 1.11)	0.16	(− 0.86, 1.18)	0.69	(− 0.29, 1.67)
EMD+Resolut	− 0.10	(− 2.34, 1.77)	− 0.13	(− 2.02, 1.31)	0.20	(− 0.96, 1.36)		
EMD+Tissue Guide	0.22	(− 1.34, 1.68)	0.35	(− 0.92, 1.51)	0.50	(− 0.36, 1.36)	0.60	(− 0.36, 1.56)
Flap op	− 0.87	(− 1.44, − 0.56)	− 0.74	(− 1.17, − 0.44)	− 1.13	(− 1.68, − 0.57)	− 0.33	(− 1.01, 0.35)
F/u = 6 months	− 0.22	(− 10.24, 9.73)	− 0.21	(− 7.88, 8.20)				
Split-mouth design	− 0.88	(− 1.35, − 0.17)	− 0.14	(− 0.65, 0.49)				

*The mean PPD reduction for the reference treatment group, i.e. defects treated with EMD alone.

Baseline PPD is the covariate in the adjusted network meta-analysis. Note that HPD intervals are not necessarily symmetrical around the estimates of fixed effects.

CI, confidence intervals; F/u, length of follow-up in months; EMD, enamel matrix derivatives; HPD, high probability density; PRP, platelet-rich plasma; DFDBA, demineralized freeze-fried bone allografts; PPD, probing pocket depth

conjunction with bone grafts achieved 0.24 mm (95% HPD intervals: − 0.38, 0.65) greater PPD reduction, while EMD in conjunction with GTR showed no additional treatment effects. EMD in conjunction with bone grafts and GTR seemed to achieve the greatest treatment effects (1.17 mm more than EMD alone, 95% HPD intervals: − 0.97, 3.42), but the confidence intervals were large. The adjustment of baseline PPD in general reduced the differences in the treatment effects between combination therapies and EMD alone (Table 5). The estimated PPD reduction for EMD at 6 months was 0.23 mm less than that at 12 months, but the confidence intervals are very large (95% HPD intervals: − 9.96, 10.59). Studies using the split-mouth design seemed to report less favourable results than those using the parallel-group design. Results from standard pair-wise meta-analysis showed that EMD alone achieved significantly greater PPD reduction than flap operation at 12 months, while EMD in conjunction with bone grafts showed greater PPD reduction than EMD alone at 12 months (Table 5, Fig. 3).

For CAL gain, network meta-analysis showed that EMD alone achieved 3.57 mm at 12 months, which was 1.27 mm (95% HPD intervals: 0.89, 1.71) greater than that for flap operation (Table 6). EMD in conjunction with bone graft achieved 0.46 mm (95% HPD intervals: − 0.13, 0.83) greater CAL gain, while EMD in conjunction with GTR showed no additional the treatment effects. The adjustment of baseline CAL in general reduced the differences in treatment effects between combination therapies and EMD alone (Table 6). Studies using the split-mouth design seemed to report less favourable results than those using the parallel-group design. Results from standard meta-analysis showed that EMD alone achieved significantly greater CAL gain than flap operation at 12 months. EMD in conjunction with bone grafts showed 0.36 mm (95% CI: − 0.22, 0.95) greater CAL gain than EMD alone at 12 months (Table 6, Fig. 4).

For infrabony defect fill, network meta-analysis showed that EMD alone achieved 3.10 mm at 12 months, which was 1.53 mm (95% HPD intervals: 0.91, 2.13) greater than that for flap operation (Table 7). EMD in conjunction with bone graft achieved 1.10 mm (95% HPD intervals: 0.56, 1.82) greater defect fill, while EMD in conjunction with GTR showed no additional treatment effects. The adjustment of baseline CAL has only a small impact on the estimated treatment effects between combination therapies and EMD alone (Table 4). Studies using the split-mouth design seemed to report less favourable results than those using the parallel-group design. Results from standard meta-analysis showed that EMD alone achieved significantly greater defect fill than flap operation at 12 months, while EMD in conjunction with bone grafts achieved a small additional defect fill than EMD alone (Table 7, Fig. 5).

Table 6. Results of network and standard meta-analysis for CAL gain

	Unadjusted network meta-analysis		Adjusted network meta-analysis		Standard pair-wise meta-analysis: 12 months		Standard pair-wise meta-analysis: 6 months	
	estimates	95% HPD intervals	estimates	95% HPD intervals	estimates	95% CI	estimates	95% CI
Stage 1								
Intercept	3.57*	(− 2.29, 9.88)	2.14	(− 3.70, 7.33)				
EMD+bone grafts	0.46	(− 0.13, 0.83)	0.20	(− 0.29, 0.63)	0.36	(− 0.22, 0.95)	0.36	(− 0.23, 0.95)
EMD+bone grafts+GTR	1.10	(− 0.34, 2.61)	†					
EMD+bone grafts+PRP	0.44	(− 1.17, 2.59)	0.15	(− 1.24, 2.04)				
EMD+GTR	0.15	(− 1.37, 0.56)	0.18	(− 1.04, 0.64)	0.27	(− 0.28, 0.82)	0.32	(− 0.49, 1.14)
Flap op	− 1.27	(− 1.71, − 0.89)	− 1.19	(− 1.55, − 0.76)	− 1.36	(− 1.72, − 1.00)	− 0.53	(− 1.04, − 0.03)
F/u = 6 months	− 0.20	(− 9.59, 7.72)	− 0.16	(− 7.54, 7.08)				
Split-mouth design	− 0.75	(− 1.25, − 0.13)	− 1.01	(− 1.61, − 0.35)				
Stage 2								
Intercept	3.62*	(− 2.03, 9.37)	2.22	(− 2.55, 6.04)				
EMD+autogenous bones	0.33	(− 0.67, 2.32)	0.38	(− 0.50, 1.94)	0.30	(− 0.88, 1.48)	0.00	(− 1.06, 1.06)
EMD+Bio-Oss	0.93	(0.48, 1.54)	1.07	(0.55, 1.83)	0.90	(0.37, 1.43)	0.90	(0.04, 1.76)
EMD+Bio-Oss+Bio-Gide	1.18	(− 0.04, 2.51)	†					
EMD+Bio-Oss+PRP	0.82	(− 0.65, 2.57)	0.89	(− 0.40, 2.28)				
EMD+TCP	0.56	(− 0.39, 1.27)	0.67	(− 0.06, 1.34)	0.30	(− 0.33, 0.93)		
EMD+DFDBA	− 0.23	(− 0.85, 0.34)	− 0.21	(− 0.71, 0.36)			− 0.20	(− 0.38, − 0.02)
EMD+HA	− 0.91	(− 2.98, − 0.08)	− 0.86	(− 2.73, − 0.12)			− 0.52	(− 1.34, 0.30)
EMD+Perioglass	0.54	(− 0.03, 2.13)	− 0.62	(− 2.14, 0.97)	− 0.70	(− 1.95, 0.55)	1.11	(0.33, 1.89)
EMD+ePTFE	0.03	(− 2.37, 0.70)	− 0.08	(− 1.88, 0.68)	0.37	(− 1.06, 1.80)	0.51	(− 0.78, 1.80)
EMD+Resolut	0.17	(− 1.53, 1.26)	0.23	(− 1.18, 1.13)	0.00	(− 0.98, 0.98)		
EMD+Tissue Guide	0.16	(− 1.39, 0.97)	0.19	(− 1.03, 0.92)	0.40	(− 0.34, 1.14)	0.20	(− 0.86, 1.26)
Flap op	− 1.19	(− 1.54, − 0.82)	− 1.12	(− 1.41, − 0.74)	− 1.36	(− 1.72, − 1.00)	− 0.53	(− 1.04, − 0.03)
F/u = 6 months	− 0.20	(− 8.53, 7.50)	− 0.17	(− 6.00, 5.67)				
Split-mouth design	− 0.90	(− 1.27, − 0.27)	− 1.03	(− 1.49, − 0.44)				

*The mean CAL gain for the reference treatment group, i.e. defects treated with EMD alone.

†Only one study is included in this group, and it does not report baseline CAL.

Baseline CAL is the covariate in the adjusted network meta-analysis. Note that HPD intervals are not necessarily symmetrical around the estimates of fixed effects.

CAL, clinical attachment level; EMD, enamel matrix derivatives; CI, confidence intervals; F/u, length of follow-up in months; DFDBA, demineralized freeze-fried bone allografts; HPD, high probability density; PRP, platelet-rich plasma; GTR, guided tissue regeneration.

Stage 2 network meta-analysis

Twenty-eight studies were grouped into 12 nodes in the network meta-analysis, yielding 66 possible pairs of comparisons. Evidence of direct comparisons was only available in 14 of the 66 pairs (Figs 6 and 7). Thirteen studies compared EMD with flap operation at 12 months, and four compared the two at 6 months. No more than two studies were found for the other comparisons. Because only one or two studies provided direct evidence for these comparisons, the confidence intervals were generally large. EMD in conjunction with bovine bone grafts showed 0.78 mm (95% HPD intervals: 0.15, 1.37) greater PPD reduction, while other bone grafting materials did not show significantly additional treatment effects. EMD in conjunction with bovine bone grafts and a resorbable membrane (Bio-Gide®, Osteohealth, Shirley, NY, USA) seemed to achieve the greatest treatment effects (1.28 mm greater than EMD alone, 95% HPD intervals: − 0.71, 3.21), but the confidence intervals were large. The adjustment of baseline PPD reduced the differences in the treatment effects between combination therapies and EMD alone (Table 5). Standard pair-wise meta-analysis found a small additional effect for EMD in conjunction with bovine bone grafts (Table 5, Fig. 6).

EMD in conjunction with bovine bone grafts showed 0.93 mm (95% HPD intervals: 0.48, 1.54) greater CAL gain than EMD alone, and EMD in conjunction with Perioglass® showed 0.54 mm (95% HPD intervals: − 0.03, 2.13) greater gain than EMD alone. However, in the adjusted analysis, Perioglass did not show an additional benefit. Standard meta-analysis based on one study found significantly better results for EMD in conjunction with bovine bone grafts (Table 6, Fig. 7).

EMD in conjunction with bovine bone grafts showed 1.46 mm (95% HPD intervals: 0.69, 2.62) greater defect fill than EMD alone, and EMD in conjunction with bovine bone grafts and BioGuide® membrane showed 1.91 mm (95% HPD intervals: 0.05, 4.40) greater gain than EMD alone. The adjustment of baseline defect depth did not yield substantial changes in the estimates. Standard meta-analysis found significantly better results for EMD in conjunction with bovine bone grafts (Table 7, Fig. 8).

Discussion

In this study, the network meta-analysis did not find substantially additional benefits for EMD in conjunction with bone grafts or barrier membranes in the treatments of infrabony defects, while both network and standard meta-analyses showed that EMD achieved better treatment outcomes than flap operation. The latter is in agreement with previous meta-analyses on the treatment effects of EMD (Kalpitis & Ruben 2002, Espo-

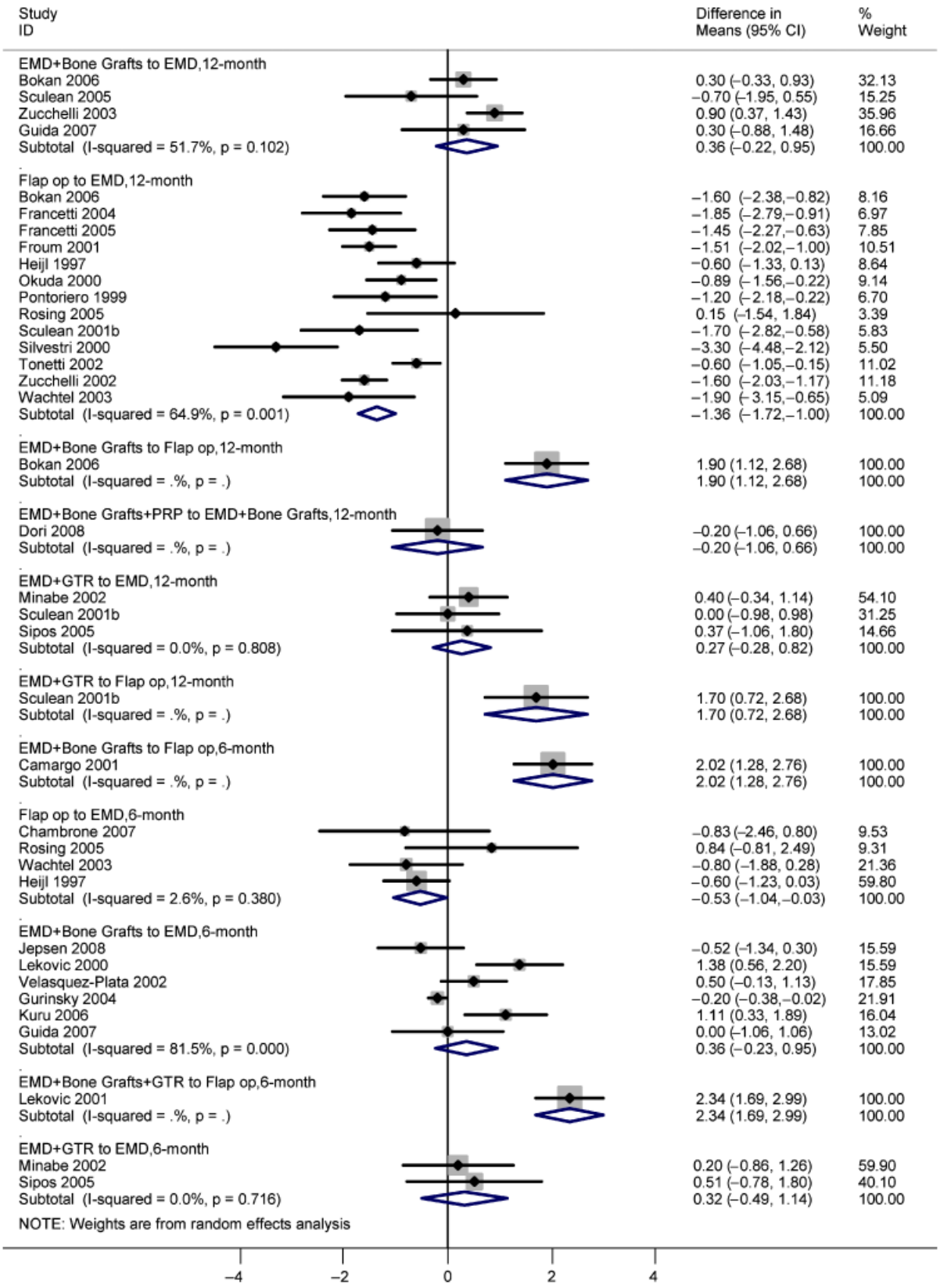

Fig. 4. Forest plot of the standard pair-wise meta-analysis for clinical attachment level gain when different types of bone grafts were grouped together and different types of barrier membranes were grouped together.

Table 7. Results of network and standard meta-analysis for clinical or radiographical infrabony defect fill

	Unadjusted network meta-analysis		Adjusted network meta-analysis		Standard pair-wise meta-analysis: 12 months		Standard pair-wise meta-analysis: 6 months	
	estimates	95% HPD intervals	estimates	95% HPD intervals	estimates	95% CI	estimates	95% CI
Stage 1								
Intercept	3.10*	(− 3.31, 9.49)	4.22	(− 2.26, 10.34)				
EMD+bone grafts	1.10	(0.56, 1.82)	0.91	(0.40, 1.67)	0.70	(− 0.04, 1.43)	1.10	(0.40, 1.79)
EMD+bone grafts+GTR	1.77	(− 0.02, 4.27)	†					
EMD+GTR	− 0.54	(− 3.30, 1.79)	− 0.11	(− 2.57, 2.74)	− 0.05	(− 1.38, 1.28)	− 0.16	(− 1.90, 1.58)
Flap op	− 1.53	(− 2.13, − 0.91)	− 1.78	(− 2.47, − 1.09)	− 1.70	(− 2.34, − 1.05)	− 0.19	(− 2.00, 1.63)
F/u = 6 months	− 0.25	(− 10.13, 8.01)	− 0.25	(− 8.81, 8.18)				
Split-mouth design	− 0.43	(− 1.45, 0.60)	− 0.27	(− 1.45, 0.85)				
Stage 2								
Intercept	3.15*	(− 3.40, 9.60)	4.24	(− 2.14, 10.47)				
EMD+autogenous bones	0.87	(− 1.55, 3.37)	0.89	(− 1.41, 3.34)	0.00	(− 1.47, 1.47)	0.00	(− 1.06, 1.06)
EMD+Bio-Oss	1.46	(0.69, 2.62)	1.22	(0.34, 2.46)	0.90	(0.23, 1.57)	1.64	(0.16, 3.12)
EMD+Bio-Oss+Bio-Gide	1.91	(0.05, 4.40)	†					
EMD+DFDBA	1.03	(0.09, 2.03)	0.93	(− 0.13, 1.88)			1.10	(0.90, 1.30)
EMD+HA	− 0.29	(− 2.48, 1.77)	− 0.29	(− 2.27, 1.76)			− 0.00	(− 0.78, 0.78)
EMD+ePTFE	− 0.55	(− 3.16, 1.90)	− 0.09	(− 2.62, 2.82)	− 0.05	(− 1.38, 1.28)	− 0.16	(− 1.90, 1.58)
Flap op	− 1.46	(− 2.09, − 0.86)	− 1.74	(− 2.39, − 1.03)	− 1.70	(− 2.34, − 1.05)	− 0.19	(− 2.00, 1.63)
F/u = 6 months	− 0.23	(− 9.73, 8.78)	− 0.24	(− 8.95, 8.06)				
Split-mouth design	− 0.59	(− 1.57, 0.48)	− 0.37	(− 1.59, 0.79)				

*The mean defect fill for the reference treatment group, i.e. defects treated with EMD alone.

†Only one study is included in this group, and it does not report baseline defect depth.

Baseline defect depth is the covariate in the adjusted network meta-analysis. Note that HPD intervals are not necessarily symmetrical around the estimates of fixed effects.

CI, confidence intervals; F/u, length of follow-up in months; HPD, high probability density; EMD, enamel matrix derivatives; CI, confidence intervals; GTR, guided tissue regeneration; DFDBA, demineralized freeze-fried bone allografts.

sito et al. 2005, Tu et al. 2008). The HPD intervals for the differences between combination therapies and EMD alone were generally large, and this is probably due partly due to the limited number of studies included in this meta-analysis and partly to the heterogeneity in the observed differences (Figs 3–8). When different types of bone grafts and barrier membranes were examined separately, the network meta-analysis suggested that the use of bovine bone grafts may provide an additional 1 mm PPD reduction, CAL gain and defect fill that was similar to that shown by direct comparisons. The network meta-analysis also suggested that EMD in conjunction with bovine bone grafts and resorbable Bio-Gide® membrane might provide additional treatment effects, while no direct comparison has been performed yet.

In this study, the network meta-analysis was undertaken in two stages by treating different types of bone grafts (or barrier membranes) as a group first and then separately. The considerable variations in the estimates seemed to suggest that it might not be appropriate to assume that the clinical effects of different bone grafts (or barrier membranes) were identical. Moreover, the networking meta-analysis also suggested that it is possible that, under some circumstances, a combination might reduce the treatment effects of EMD (Palmer et al. 2008). Our results suggest that EMD in conjunction with GTR does not show additional benefits compared with EMD alone. Moreover, the study that used a non-resorbable membrane in conjunction with EMD showed a high incidence of early membrane exposure after surgery that might compromise the treatment outcomes (Sipos et al. 2005). Early exposure of membranes was also observed by another study that used a resorbable membrane in conjunction with EMD (Sculean et al. 2001b). Considering the additional cost and surgical time, the cost-effectiveness of this combination therapy seems to be questionable. In contrast, one study that used a combination of EMD, bovine bone grafts and a resorbable membrane showed better treatment outcomes than flap operation, and based on indirect evidence, the network meta-analysis suggested that this combination may achieve better outcomes than EMD alone. However, as both membranes and bovine bone grafts are used in this combination therapy, further studies are required to validate this finding and to evaluate its cost-effectiveness.

The most consistent finding in our meta-analysis is the additional benefits shown by EMD in conjunction with bovine bone grafts. While it is plausible that this combination may enhance periodontal regeneration (Simunek et al. 2008, Trombelli & Farina 2008), bone grafts may act as fillers for infrabony defects and therefore yield better short-term clinical and radiographical measurements (Wang & Cooke 2005). From a cost-effectiveness perspective, it is not clear how much difference in treatment effects is required to justify the use of combination therapies. Our analysis and previous systematic review showed that EMD alone achieved about 1 mm more PPD reduction and CAL gain than flap operation (publications in the literature (Kalpitis & Ruben 2002, Venezia et al. 2004, Esposito et al. 2005). To justify the additional cost and surgical time (and potential complications after surgery), we may expect the combination therapies to provide a similar number of additional benefits than EMD alone.

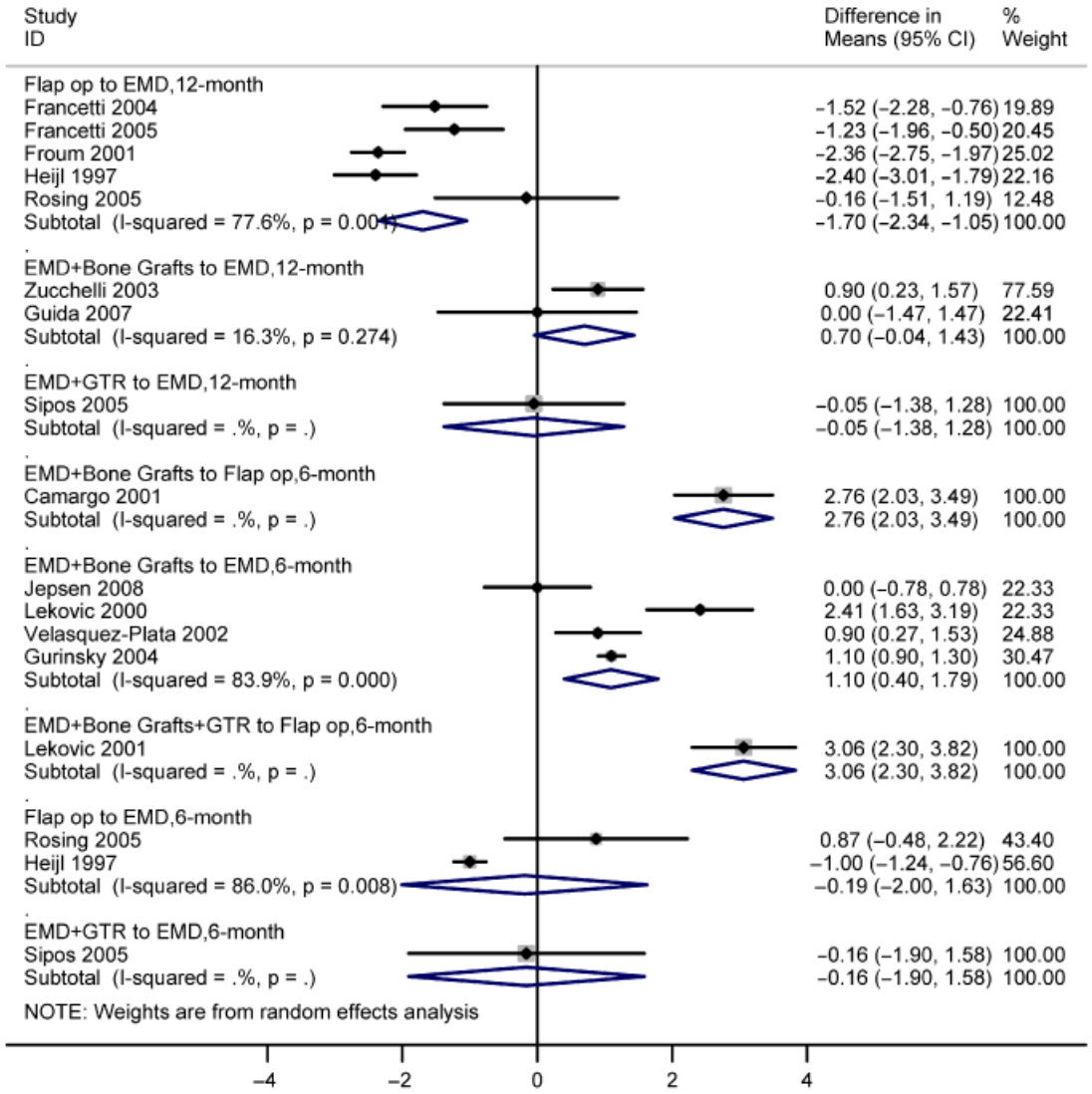

Fig. 5. Forest plot of the standard pair-wise meta-analysis for infrabony defect fill when different types of bone grafts were treated as separate groups and different types of barrier membranes were treated as separate groups.

In contrast to traditional pair-wise meta-analysis, network analysis took into account the indirect comparisons in its estimations. For instance, when estimating the differences between active treatments A and B, standard pair-wise meta-analysis only uses the evidence from clinical trials that randomly allocated patients to either A or B (i.e. direct comparison). However, in addition to the evidence from the direct comparison, network meta-analysis also takes into account the results such as those from the studies that compare A with placebo or B with placebo. The differences in the results from A-placebo and B-placebo clinical trials then provide indirect evidence of A–B comparison. If results from trials comparing A with placebo are better than those comparing B with placebo, it may be inferred that A is better than B. Network meta-analysis therefore uses a broader evidence base than standard pair-wise meta-analysis in the synthesis of research findings with a view to providing more robust estimates. Some studies suggested that adjusted network meta-analysis might be less biased than direct comparisons (Song et al. 2008).

It is important to note that the randomization in the included studies is not broken in the network meta-analysis (Lumley 2002, Caldwell et al. 2005, Glenny et al. 2005, Sutton et al. 2008), and this is different from those meta-analyses that summarize results for each arm of treatments from different studies and then test the differences between treatment arms (Venezia et al. 2004, Cheng et al. 2007). An assumption for network meta-analysis is that there is a consistency across evidence, i.e. if treatment A had been included in the B-to-placebo trials, the estimate of the difference between A and placebo would be comparable with those produced by A-to-placebo trials and vice versa (Sutton et al. 2008). This assumption is the same as that made by the standard pair-wise meta-analysis, where all the estimates of treatment effects across trials are exchangeable and the observed heterogeneity is attributed to random variations (Sutton et al. 2008).

One motivation for undertaking network (also known as mixed treatments) meta-analysis is that indirect comparisons can provide estimates for treatment comparisons that have not been tested in head-to-head trials without breaking randomization (Sutton et al. 2008). For instance, in the network shown in Fig. 2,

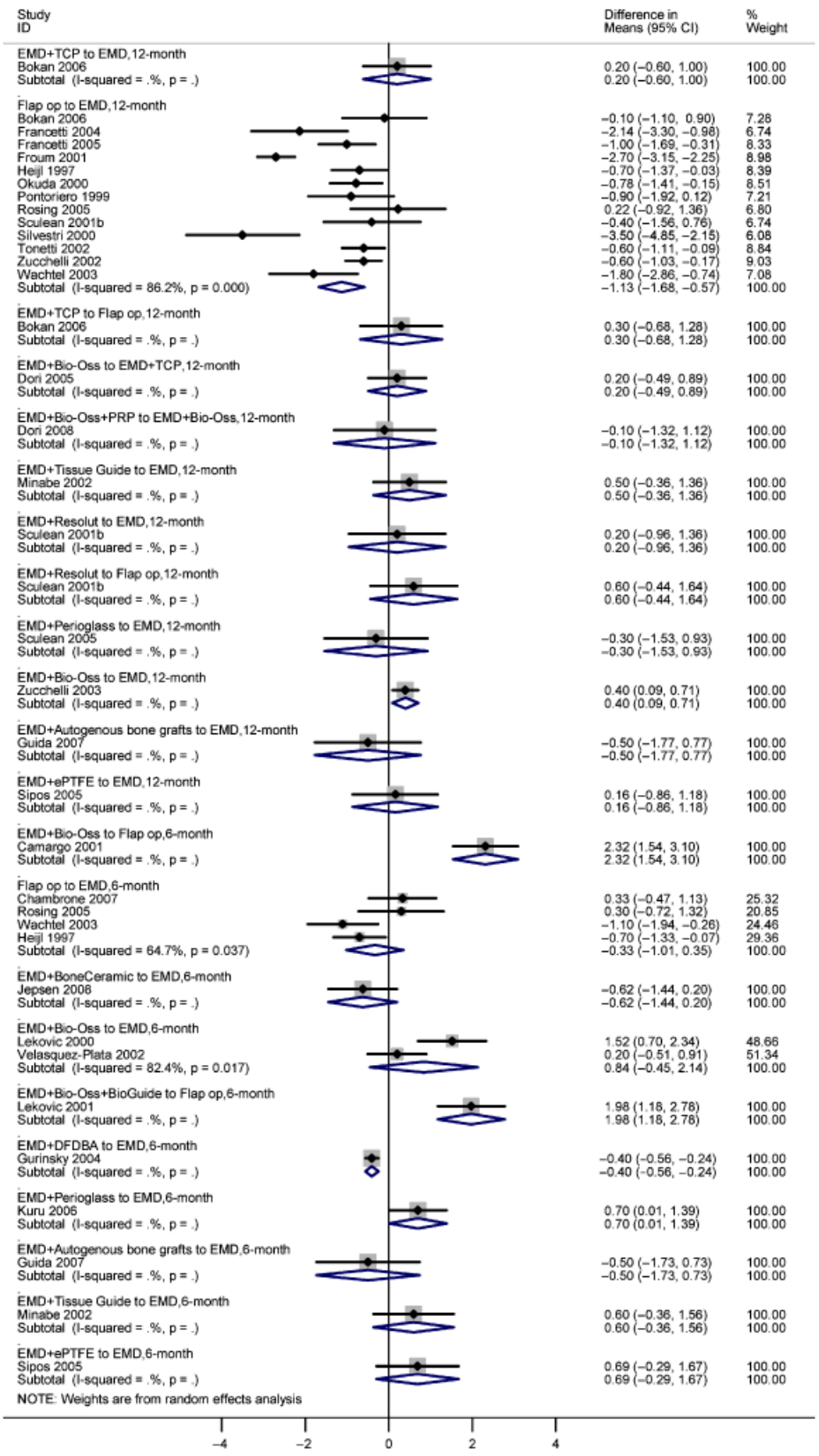

Fig. 6. Forest plot of the standard pair-wise meta-analysis for probing pocket depth reduction when different types of bone grafts were treated as separate groups and different types of barrier membranes were treated as separate groups.

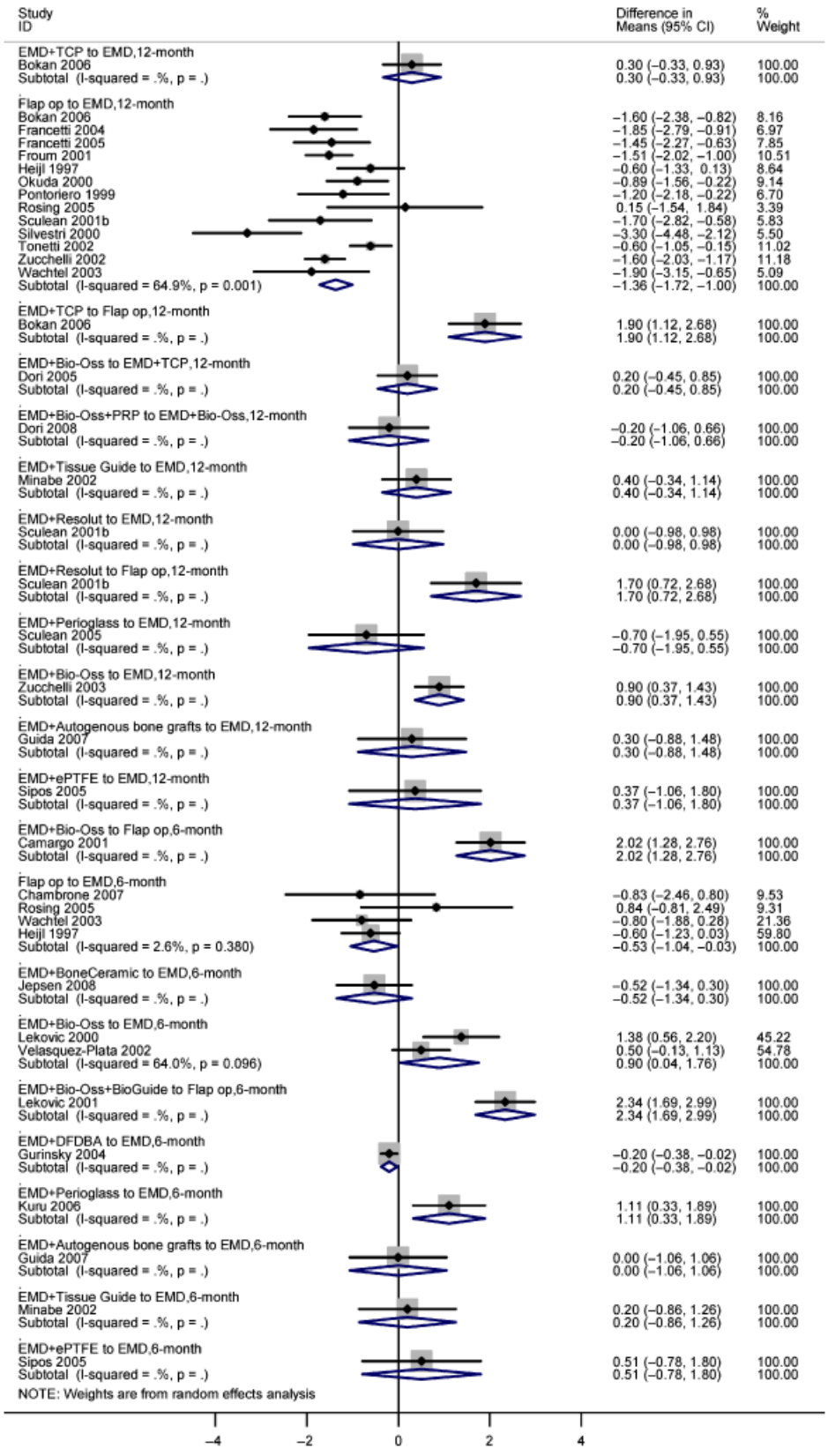

Fig. 7. Forest plot of the standard pair-wise meta-analysis for clinical attachment level gain when different types of bone grafts were treated as separate groups and different types of barrier membranes were treated as separate groups.

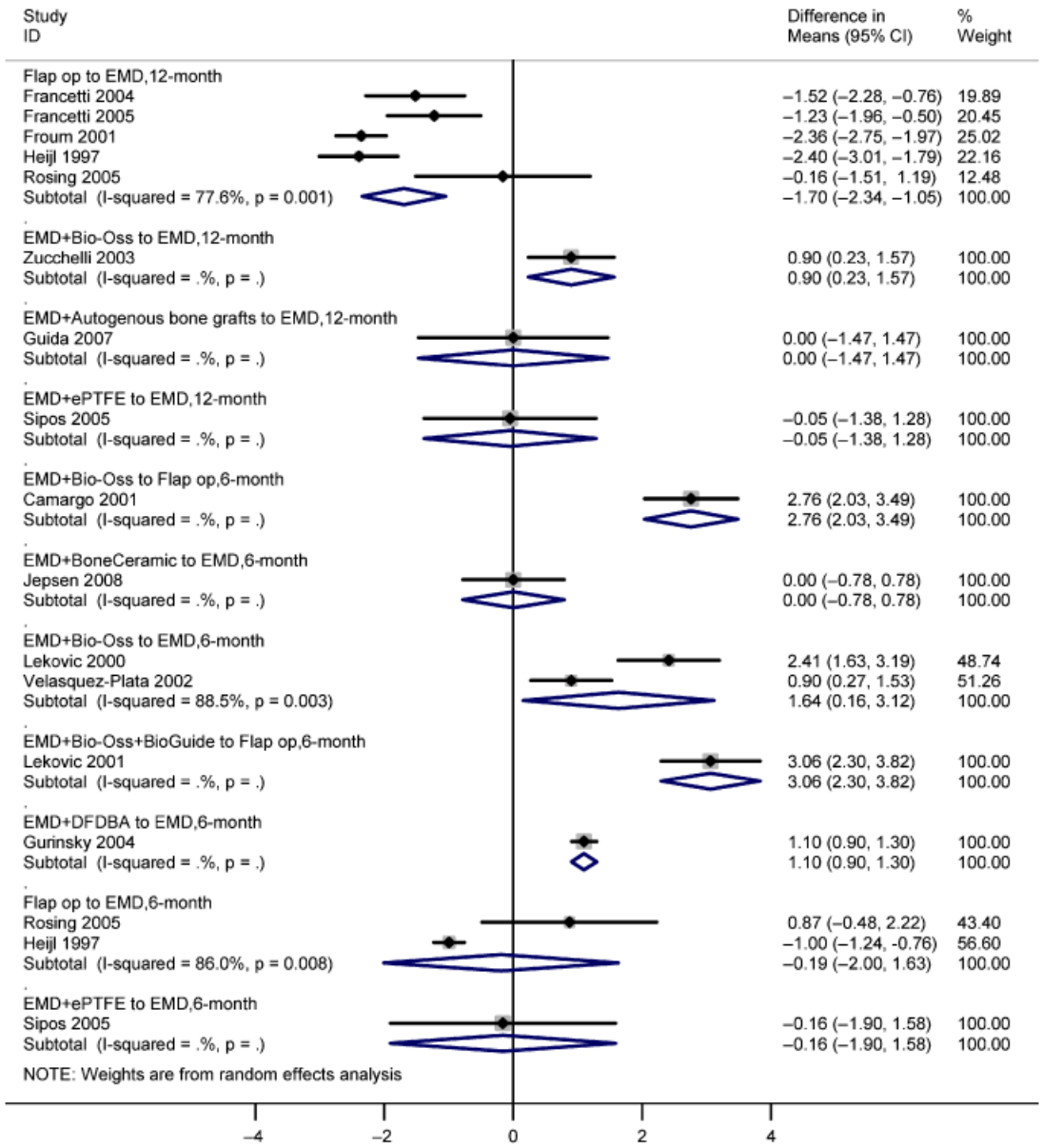

Fig. 8. Forest plot of the standard pair-wise meta-analysis for infrabony defect fill when different types of bone grafts were treated as separate groups and different types of barrier membranes were treated as separate groups.

evidence of direct comparisons is only available in seven out of the 15 possible comparisons. Results from the random effects meta-analysis nevertheless provide clues regarding what might have been observed if these comparisons had been made in clinical trials. Another motivation is that network and mixed treatments meta-analysis provide a framework for more than two treatments to be compared simultaneously, using the *same* evidence base. For instance, in this study, the network meta-analysis allows us to estimate the differences in the treatment effects between all possible combination therapies and EMD alone. If we wish to use direct evidence for these comparisons, we would only include trials with multiple arms testing all combination therapies. At the moment, no such study has been performed yet.

Network and mixed treatments analysis is an emerging new methodology for evidence synthesis and is still under development. It is only in recent years that the use of this methodology has become widespread (Bucher et al. 1997, Lumley 2002, van Houwelingen et al. 2002, Psaty et al. 2003, Caldwell et al. 2005, Glenny et al. 2005, Chou et al. 2006, Coleman et al. 2008, Thijs et al. 2008, Cipriani et al. 2009). In this study, random-effects modelling with the procedure of restricted maximum likelihood was used to obtain the point estimates, and then Markov Chain Monte Carlo methods were used to obtain Bayesian HPD intervals (Baayen et al. 2008). This is because the confidence intervals for estimates in the random-effects models based on classical likelihood methods may be too narrow for small samples, and the distributions of model estimates may not follow a symmetrical distribution. Different random-effects models for network meta-analysis have been formulated (Lumley 2002, van Houwelingen et al. 2002, Whitehead 2002, Glenny et al. 2005), and a more complex

Bayesian network meta-analysis approach has been proposed (Lu & Ades 2004, Ades et al. 2006, Salanti et al. 2008). While network meta-analysis should be viewed as an exploratory method for research hypothesis generation and its results have to be interpreted with caution, it is nevertheless a valuable tool for evidence synthesis when more than two treatment options are available, which is a very common scenario in periodontal and dental care. As discussed previously, certain assumptions are made by network meta-analysis, and if these assumptions do not hold, the validity of its results may be questionable. However, these assumptions are not different from those made by standard pair-wise meta-analysis, and both require sound evidence base, i.e. high-quality RCT.

A common problem with systematic review is that some important information such as SD or SEM of treatment effects is not available for meta-analysis. As SD or SEM is usually used as weights in meta-analysis; studies not reporting them have to be excluded. In this study, we made a few assumptions in order to derive SD and SEM from available information, and this may be viewed as a way of imputing missing data. However, our sensitivity analysis with the adjustment of baseline values was based on RCTs with complete data.

In summary, this network meta-analysis only found a small additional treatment effect for combination therapies compared with EMD alone in the treatment of infrabony defects when different bone grafts were treated as a group or different barrier membranes were treated as a group. Among different bone grafts, the network meta-analysis suggested that the use of bovine bone grafts might provide some additional benefits, and this will need to be confirmed in a large clinical trial with sufficient statistical power before any firm conclusion and recommendation can be made. Further research is also required to undertake an economic analysis of cost-effectiveness for these expensive combination therapies.

References

Ades, A. E., Sculpher, M., Sutton, A., Abrams, K., Cooper, N., Welton, N. & Lu, G. (2006) Bayesian methods for evidence synthesis in cost-effectiveness analysis. *Pharmacoeconomics* **24**, 1–19.

Baayen, R. H. (2008) *Language R: Data Sets and Functions with ''Analyzing Linguistic Data: A Practical Introduction to Statistics''. R Package Version 0.953*. Vienna, Austria: Foundation for Statistical Computing.

Baayen, R. H., Davison, D. J. & Bates, D. M. (2008) Mixed-effects modeling with crossed random effects for subjects and items. *Journal of Memory and Language* **59**, 390–412.

Bates, D. & Maechler, M. (2009) lme4: Linear mixed-effects models using S4 classes. R package version 0.999375-31. Available at http://lme4.r-forge.r-project.org (accessed 17 June 2009)

Bokan, I., Bill, J. S. & Schlagenhauf, U. (2006) Primary flap closure combined with Emdogain alone or Emdogain and Cerasorb in the treatment of intra-bony defects. *Journal of Clinical Periodontology.* **33**, 885–893.

Bosshardt, D. D. (2008) Biological mediators and periodontal regeneration: a review of enamel matrix proteins at the cellular and molecular levels. *Journal of Clinical Periodontology* **35** (Suppl. 8), 87–105.

Brown, H. & Prescott, R. (2006) *Applied Mixed Models in Medicine*, 2nd edition, pp. 203–213. Chichester: Wiley.

Bucher, H. C., Guyatt, G. H., Griffith, L. E. & Walter, S. D. (1997) The results of direct and indirect treatment comparisons in meta-analysis of randomized controlled trials. *Journal of Clinical Epidemiology* **50**, 683–691.

Caldwell, D. M., Ades, A. E. & Higgins, J. P. (2005) Simultaneous comparison of multiple treatments: combining direct and indirect evidence. *British Medical Journal* **331**, 897–900.

Camargo, P. M., Lekovic, V., Weinlaender, M., Vasilic, N., Kenney, E. B. & Madzarevic, M. (2001) The effectiveness of enamel matrix proteins used in combination with bovine porous bone mineral in the treatment of intrabony defects in humans. *Journal of Clinical Periodontology* **28**, 1016–1022.

Chambrone, D., Pasin, I. M., Conde, M. C., Panutti, C., Carneiro, S. & Lima, L. A. (2007) Effect of enamel matrix proteins on the treatment of intrabony defects: a split-mouth randomized controlled trial study. *Brazilian Oral Research* **21**, 241–246.

Cheng, Y.-F., Chen, J.-W., Lin, S.-J. & Lu, H.-K. (2007) Is coronally positioned flap procedure adjunct with enamel matrix derivative or root conditioning a relevant predictor for achieving root coverage? A systemic review. *Journal of Periodontal Research* **42**, 474–485.

Chou, R., Fu, R., Huffman, L. H. & Korthuis, P. T. (2006) Initial highly-active antiretroviral therapy with a protease inhibitor versus a non-nucleoside reverse transcriptase inhibitor: discrepancies between direct and indirect meta-analyses. *Lancet* **368**, 1503–1515.

Cipriani, A., Furukawa, T. A., Salanti, G., Geddes, J. R., Higgins, J. P., Churchill, R., Watanabe, N., Nakagawa, A., Omori, I. M., McGuire, H., Tansella, M. & Barbui, C. (2009) Comparative efficacy and acceptability of 12 new-generation antidepressants: a multiple-treatments meta-analysis. *Lancet* **373**, 746–758.

Coleman, C. I., Baker, W. L., Kluger, J. & White, C. M. (2008) Antihypertensive medication and their impact on cancer incidence: a mixed treatment comparison meta-analysis of randomized controlled trials. *Journal of Hypertension* **26**, 622–629.

Döri, F., Arweiler, N., Gera, I. & Sculean, A. (2005) Clinical evaluation of an enamel matrix protein derivative combined with either a natural bone mineral or β-tricalcium phosphate. *Journal of Periodontology* **76**, 2236–2243.

Döri, F., Nikolidakis, D., Húszár, T., Arweiler, N. B., Gera, I. & Sculean, A. (2008) Effect of platelet-rich plasma on the healing of intrabony defects treated with an enamel matrix protein derivative and a natural bone mineral. *Journal of Clinical Periodontology* **35**, 44–50.

Esposito, M., Grusovin, M. G., Coulthard, P. & Worthington, H. V. (2005) Enamel matrix derivative (Emdogain®) for periodontal tissue regeneration in intrabony defects (Cochrane Review). *The Cochrane Database of Systematic Reviews* 2005, Issue 4, Art. No.: CD003875.pub2, doi: 10.1002/14651858.CD003875.pub2.

Forabosco, A., Spinato, S., Diacci, S. & Grippo, A. (2003) Clinical comparison between guided tissue regeneration and induced tissue regeneration. *Minerva Stomatologica* **52**, 105–110.

Francetti, L., Del Fabbro, M., Basso, M., Testori, R. & Weinstein, R. (2004) Enamel matrix proteins in the treatment of intrabony defects. A prospective 24-month clinical trial. *Journal of Clinical Periodontology* **31**, 52–59.

Francetti, L., Trombelli, L., Lombardo, G., Guida, L., Cafiero, C., Roccuzzo, M., Carusi, G. & Del Fabbro, M. (2005) Evaluation of efficacy of enamel matrix derivative in the treatment of intrabony defects: a 24-month multicenter study. *International Journal of Periodontics and Restorative Dentistry* **25**, 461–473.

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. *Journal of Periodontology* **72**, 25–34.

Glenny, A. M., Altman, D. G., Song, F., Sakarovitch, C., Deeks, J. J., D'Amico, R., Bradburn, M. & Eastwood, A. J. (2005) Indirect comparisons of competing interventions. *Health Technology Assessment* **9** (26), 1–134.

Guida, L., Annunziata, M., Belardo, S., Farina, R., Scabbia, A. & Trombelli, L. (2007) Effect of autogenous cortical bone particulate in conjunction with enamel matrix derivative in the treatment of periodontal intraosseous defects. *Journal of Periodontology* **78**, 231–238.

Gurinsky, B. S., Mills, M. P. & Mellonig, J. T. (2004) Clinical evaluation of demineralized freeze-dried bone allograft and enamel matrix derivative versus enamel matrix derivative alone for the treatment of periodontal osseous

defects in humans. *Journal of Periodontology* **75**, 1309–1318.

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

Hoffmann, T., Richter, S., Meyle, J., Gonzales, J. R., Heinz, B., Arjomand, M., Sculean, A., Reich, E., Jepsen, K., Jepsen, S. & Boedeker, R. H. (2006) A randomized clinical multicentre trial comparing enamel matrix derivative and membrane treatment of buccal class II furcation involvement in mandibular molars. Part III: patient factors and treatment outcome. *Journal of Clinical Periodontology* **33**, 575–583.

Jepsen, S., Topoll, H., Rengers, H., Heinz, B., Teich, M., Hoffmann, T., Machot, E. A., Meyle, J. & Jervøe-Storm, P. M. (2008) Clinical outcomes after treatment of intrabony defects with EMD/synthetic bone graft or EMD alone. A multicenter randomized controlled clinical trial. *Journal of Clinical Periodontology* **35**, 420–428.

Kalpitis, C. D. R. & Ruben, M. P. (2002) Treatment of intrabony periodontal defects with enamel matrix derivative: a literature review. *Journal of Periodontology* **73**, 1360–1376.

Kuru, B., Yilmaz, S., Argin, K. & Noyan, U. (2006) Enamel matrix derivative alone or in combination with a bioactive glass in wide intrabony defects. *Clinical Oral Investigations* **10**, 227–234.

Lekovic, V., Camargo, P. M., Weinlaender, M., Kenney, E. B. & Vasilic, N. (2001) Combination use of bovine porous bone mineral, enamel matrix proteins, and a bioabsorbable membrane in intrabony periodontal defects in humans. *Journal of Periodontology* **72**, 583–589.

Lekovic, V., Camargo, P. M., Weinlaender, M., Nedic, M., Aleksic, Z. & Kenney, E. B. (2000) A comparison between enamel matrix proteins used alone or in combination with bovine porous bone mineral in the treatment of intrabony periodontal defects in humans. *Journal of Periodontology* **71**, 1110–1116.

Lu, G. & Ades, A. E. (2004) Combination of direct and indirect evidence in mixed treatment comparisons. *Statistics in Medicine* **23**, 3105–3124.

Lumley, T. (2002) Network meta-analysis for indirect treatment comparisons. *Statistics in Medicine* **21**, 2313–2324.

Minabe, M., Kodama, T., Kogou, T., Takeuchi, K., Fushimi, H., Sugiyama, T. & Mitarai, E. (2002) A comparative study of combined treatment with a collagen membrane and enamel matrix proteins for the regeneration of intraosseous defects. *International Journal of Periodontics and Restorative Dentistry* **22**, 595–605.

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

Palmer, R. M. & Cortellini, P.Group B of European Workshop on Periodontology. (2008) Periodontal tissue engineering and regeneration: Consensus Report of the Sixth European Workshop on Periodontology. *Journal of Clinical Periodontology* **35** (Suppl. 8), 83–86.

Pontoriero, R., Wennstrom, 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.

Psaty, B. M., Lumley, T., Furberg, C. D., Schellenbaum, G., Pahor, M., Alderman, M. H. & Weiss, N. S. (2003) Health outcomes associated with various antihypertensive therapies used as first-line agents: a network meta-analysis. *Journal of the American Medical Association* **289**, 2534–2544.

Rösing, C. K., Aass, A. M., Mavropoulos, A. & Gjermo, P. (2005) Clinical and radiographic effects of enamel matrix derivative in the treatment of intrabony periodontal defects: a 12-month longitudinal placebo-controlled clinical trial in adult periodontitis patients. *Journal of Periodontology* **76**, 129–133.

Salanti, G., Higgins, J. P., Ades, A. E. & Ioannidis, J. P. (2008) Evaluation of networks of randomized trials. *Statistical Methods in Medical Research* **17**, 279–301.

Sculean, A., Blaes, A., Arweiler, N., Reich, E., Donos, N. & Brecx, M. (2001a) The effect of postsurgical antibiotics on the healing of intrabony defects following treatment with enamel matrix proteins. *Journal of Periodontology* **72**, 190–195.

Sculean, A., Berakdar, M., Willershausen, B., Arweiler, N. B., Becker, J. & Schwarz, F. (2006) Effect of EDTA root conditioning on the healing of intrabony defects treated with an enamel matrix protein derivative. *Journal of Periodontology* **77**, 1167–1172.

Sculean, A., Pietruska, M., Schwarz, F., Willershausen, B., Arweiler, N. B. & Auschill, T. M. (2005) Healing of human intrabony defects following regenerative periodontal therapy with an enamel matrix protein derivative alone or combined with a bioactive glass. A controlled clinical study. *Journal of Clinical Periodontology* **32**, 111–117.

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

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

Simunek, A., Kopecka, D., Somanathan, R. V., Pilathadka, S. & Brazda, T. (2008) Deproteinized bovine bone versus beta-tricalcium phosphate in sinus augmentation surgery: a comparative histologic and histomorphometric study. *International Journal of Oral and Maxillofacial Implants* **23**, 935–942.

Sipos, P. M., Loos, B. G., Abbas, F., Timmerman, M. F. & van der Velden, U. (2005) The combined use of enamel matrix proteins and a tetracycline-coated expanded polytetrafluoroethylene barrier membrane in the treatment of intra-osseous defects. *Journal of Clinical Periodontology* **32**, 765–772.

Song, F., Harvey, I. & Lilford, R. (2008) Adjusted indirect comparison may be less biased than direct comparison for evaluating new pharmaceutical interventions. *Journal of Clinical Epidemiology* **61**, 455–463.

Sutton, A., Ades, A. E., Cooper, N. & Abrams, K. (2008) Use of indirect and mixed treatment comparisons for technology assessment. *Pharmacoeconomics* **26**, 753–767.

Thijs, V., Lemmens, R. & Fieuws, S. (2008) Network meta-analysis: simultaneous meta-analysis of common antiplatelet regimens after transient ischaemic attack or stroke. *European Heart Journal* **29**, 1086–1092.

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. *Journal of Clinical Periodontology* **29**, 317–325.

Trombelli, L. & Farina, R. (2008) Clinical outcomes with bioactive agents alone or in combination with grafting or guided tissue regeneration. *Journal of Clinical Periodontology* **35** (Suppl. 8), 117–135.

Tu, Y. K., Baelum, V. & Gilthorpe, M. S. (2005) The problem of analysing the relationship between change and initial value in oral health research. *European Journal of Oral Sciences* **113**, 271–278.

Tu, Y. K. & Gilthorpe, M. S. (2007) Revisiting the relation between change and initial value: a review and evaluation. *Statistics in Medicine* **26**, 443–457.

Tu, Y. K., Maddick, I., Kellett, M., Clerehugh, V. & Gilthorpe, M. S. (2006) Evaluating the quality of active-control trials in periodontal research. *Journal of Clinical Periodontology* **33**, 151–156.

Tu, Y. K., Tugnait, A. & Clerehugh, V. (2008) Is there a temporal trend in the reported treatment efficacy of periodontal regeneration? A meta-analysis of randomized-controlled trials. *Journal of Clinical Periodontology* **35**, 139–146.

Velasquez-Plata, D., Scheyer, E. T. & Mellonig, J. T. (2002) Clinical comparison of an enamel matrix derivative used alone or in combination with a bovine-derived xenograft for the treatment of periodontal osseous defects in humans. *Journal of Periodontology* **73**, 433–440.

van Houwelingen, H. C., Arends, L. R. & Stijnen, T. (2002) Advanced methods in meta-analysis: multivariate approach and meta-regression. *Statistics in Medicine* **21**, 589–624.

Venezia, E., Goldstein, M., Boyan, B. D. & Schwartz, Z. (2004) The use of enamel matrix derivative in the treatment of periodontal defects: a literature review and meta-analysis. *Critical Reviews in Oral Biology and Medicine* **15**, 382–402.

Wachtel, H., Schenk, G., Böhm, S., Weng, D., Zuhr, O. & Hürzeler, M. B. (2003) Microsurgical access flap and enamel matrix derivative for the treatment of periodontal intrabony defects: a controlled clinical study. *Journal of Clinical Periodontology* **30**, 496–504.

Wang, H. L. & Cooke, J. (2005) Periodontal regeneration techniques for treatment of periodontal diseases. *Dental Clinics of North American* **49**, 637–659.

Whitehead, A. (2002) *Meta-Analysis of Controlled Clinical Trials*, pp. 241–257. Chichester: Wiley.

Zucchelli, G., Amore, C., Montebugnoli, L. & De Sanctis, M. (2003) Enamel matrix proteins and bovine porous bone mineral in the treatment of intrabony defects: a comparative controlled clinical trial. *Journal of Periodontology* **74**, 1725–1735.

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

Address:
Yu-Kang Tu
Centre for Epidemiology & Biostatistics Division of Biostatistics
University of Leeds
Room 8.01, Level 8
Worsley Building, Clarendon Way
Leeds LS2 9JT
UK
E-mail: y.k.tu@leeds.ac.uk

Clinical Relevance

Scientific rationale for the study: Although EMD in conjunction with other regenerative materials has been used in a few clinical trials, it is not clear whether the use of these combination therapies provides substantial, additional treatment effects than the use of EMD alone. The rationale for this study was to conduct a network meta-analysis to estimate the additional treatment effects provided by combination therapies.

Principal findings: The network meta-analysis of randomized clinical trials showed that combination therapies did not give rise to substantial, additional benefits in the treatment of infrabony defects. However, EMD in conjunction with bovine bone grafts might provide greater treatment effects.

Practical implications: Combinations of different regenerative materials in the treatment of infrabony defects do not necessarily yield better treatment outcomes than single therapy.