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REVIEW

Hyaluronic acid as adjunctive to non-surgical and surgical periodontal therapy: a systematic review and meta-analysis

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Abstract

Objectives To evaluate the potential added benefit of the topical application of hyaluronic acid (HA) on the clinical outcomes following non-surgical or surgical periodontal therapy.

Materials and methods A systematic search was performed in Medline, Embase, Cochrane, Web of Science, Scopus and Grey literature databases. The literature search was preformed according to PRISMA guidelines. The Cochrane risk of bias tool was used in order to assess the methodology of the included trials. Weighted mean differences (WMDs) and 95% confidence intervals (CIs) between the treatment and controls were estimated using the random-effect model for amount of bleeding on probing (BOP), probing depth (PD) reduction and clinical attachment level (CAL) gain. In order to minimize the bias and to perform meta-analysis, only randomized clinical studies (RCTs) were selected.

Results Thirteen RCTs were included: 11 on non-surgical periodontal treatment and two on surgical periodontal treatment. Overall analysis of PD reduction, CAL gain and BOP reduction in non-surgical therapy with adjunctive HA presented WMD of − 0.36 mm (95% CI − 0.54 to − 0.19 mm; $p < 0.0001$), 0.73 mm (95% CI 0.28 to 1.17 mm; $p < 0.0001$) and − 15% (95% CI − 22 to − 8%; $p < 0.001$) respectively, favouring the application of HA. The overall analysis on PD and CAL gain in surgical therapy with adjunctive HA presented WMD of − 0.89 mm (95% CI − 1.42 to − 0.36 mm; $p < 0.0001$) for PD reduction and 0.85 mm (95% CI 0.08 to 1.62 mm; $p < 0.0001$) for CAL gain after 6–24 months favouring the treatment with HA. However, comparison presented considerable heterogeneity between the non-surgical studies and a high risk of bias in general.

Conclusions Within their limits, the present data indicate that the topical application of HA may lead to additional clinical benefits when used as an adjunctive to non-surgical and surgical periodontal therapy. However, due to the high risk of bias and heterogeneity, there is a need for further well-designed RCTs to evaluate this material in various clinical scenarios.

Clinical relevance The adjunctive use of HA may improve the clinical outcomes when used in conjunction with non-surgical and surgical periodontal therapy.

Keywords Hyaluronic acid · Hyaluronan · Periodontitis · Surgical periodontal therapy · Non-surgical periodontal therapy

Introduction

Hyaluronic acid (HA) is a major natural carbohydrate component of the extracellular matrix and can be found in the skin, the joints, the eyes and most other organs and tissues including the periodontium. Furthermore, it is present in body fluids like serum, saliva and gingival crevicular fluid and as a component of the soft and hard tissues [1]. In the periodontium, HA is synthesized by HA synthase enzymes present in various cells including fibroblasts and keratinocytes in the gingival and periodontal ligament, cementoblasts and osteoblasts [2, 3].

There is evidence that HA is bacteriostatic [4, 5], fungostatic [6], anti-inflammatory [7], anti-oedematous [8], osteoinductive [7, 9–11] and pro-angiogenetic [12]. These properties suggest

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HA to be an ideal material for wound healing [13]. In animal studies, HA showed promising results for connective tissue [14, 15] and bone repair [16, 17] and it facilitated re-epithelialization, formed a good elasticity of the connective tissue and increased microvascular density when used on full thickness surgical skin wounds. The use of HA on human skin wounds and of skin ulcers resulted in faster reduction of the wound size when compared with the controls [18, 19].

Since HA is a key molecule in inflammation, granulation tissue formation, epithelium formation and tissue remodelling, it was suggested to play also an important role in periodontal wound healing [16, 20].

The above-mentioned effects (anti-inflammatory, anti-oedematous and antibacterial) have also been shown in non-surgical periodontal therapy [21]. It is anticipated that the anti-inflammatory effect is due to the exogenous HA that acts as a scavenger by draining prostaglandins, metalloproteinases and other bioactive molecule [22]. HA has shown a positive effect on the reduction of plaque and sulcus bleeding index in patients with induced gingivitis [23, 24]. In patients with chronic periodontitis, the additional application of HA to non-surgical periodontal treatment (scaling and root planing) resulted in higher clinical improvements in terms of bleeding on probing (BOP) and probing depth (PD) reduction compared with SRP alone [25]. However, other studies have failed to show statistically significant differences in terms of bacterial profile when HA was applied subgingivally as adjunctive to SRP in chronic periodontitis patients [26].

Interestingly, some clinical reports and randomized clinical trials have shown additional benefits in terms of clinical attachment level (CAL) gain and PD reduction following the adjunctive use of HA during periodontal surgery [27–29].

Most recently, one systematic review [25], however without meta-analysis, concluded that the use of HA adjunctive to SRP and to periodontal surgery yielded positive effects on the clinical outcomes (i.e. PD, CAL, BOP and bone fill). According to the best of our knowledge, at present, no meta-analysis has been published on the effects of HA when applied in the frame on non-surgical and surgical periodontal therapy. Therefore, the aim of this systematic review including meta-analysis was to evaluate the potential clinical effects of HA when used in conjunction with non-surgical and surgical periodontal therapy.

Objectives

This systematic review had the following aims:

1. To evaluate the effect of HA application on clinical parameters in conjunction with non-surgical periodontal therapy.
2. To evaluate the effect of HA application on clinical parameters as adjunctive therapy to periodontal surgery.

The PICOS (Participants, Interventions, Comparisons, Outcomes, Study Designs) research question addressing the research objectives is presented in Table 1 [30].

Materials and methods

Search method and identification of studies

Studies reporting application of HA as adjunctive to periodontal non-surgical and surgical therapies were identified by electronically searching PubMed (NCBI), Embase, Cochrane, Web of Science, Scopus and Grey literature database (www.greynet.org, https://scholar.google.ch/, www.worldcat.org) from the earliest available date through April 2016. The search strategy used was a combination of MeSH terms and/or free text words, depending on the literature database. The key words used for electronic search were “periodontics” (MeSH) OR “periodontal disease” (MeSH) OR “periodontitis” (MeSH) AND “surgical procedures, operative” (MeSH) OR “periodontal therapy” (MeSH) AND “hyaluronic acid” (MeSH) OR “hyaluronan” OR “hyaluronate” (full search strategy: Appendix 1). Hand searching of eligible article references was also performed.

Two authors (J.C.I. and M.E.) selected and evaluated independently the articles during the entire selection process, and any disagreements between authors were resolved after discussion. If information within a study should be missing, the authors would be contacted per email.

Table 1 PICOS (Participants, Interventions, Comparisons, Outcomes, Study Designs)

Participants	Chronic periodontitis patients and healthy adults
Interventions	Application of hyaluronic acid in conjunction with periodontal therapy (either surgical or non-surgical)
Comparisons	Same periodontal procedure (either surgical or non-surgical) without hyaluronic acid
Outcomes	Clinical periodontal parameters (periodontal probing depth, BOP, clinical attachment gain)
Study designs	Randomized control trials in a parallel or split-mouth design

Inclusion criteria

The study inclusion criteria were as follows:

1. Study design—randomized controlled trials (parallel- or split-mouth design).
2. English language.
3. No year restriction.
4. Studies reporting application of HA as adjunctive to non-surgical and surgical periodontal therapy.
5. No combinations with biomaterials (e.g. bone substitute, membranes).
6. Minimum 3-month follow-up period for non-surgical treatment and minimum 6-month follow-up period for surgical treatment.
7. Studies reporting either on PD, CAL or BOP as outcomes.

Type of outcome measurements

The primary outcomes were changes in PD, CAL and BOP reported at different time points.

Data collection

The following data from each study were extracted and entered into an electronic spreadsheet:

Name of the authors, year of publication, total number of participants, total amount of treated sites, months of follow-up, BOP, PD, CAL and study design.

Risk of bias (quality) assessment

The Cochrane risk of bias tool [31] was used in order to assess the methodology of the included trials. Two authors (M.E., J.C.I.) independently assessed risk of bias on the following criteria:

1. Random sequence generation.
2. Allocation concealment.
3. Blinding of participants and the investigator.
4. Blinding of outcome assessments.
5. Incomplete outcome data.
6. Selective outcome reporting.
7. Other bias.

Each relevant domain per trail was judged either as low risk (if all criteria were met), unclear risk (only one criterion was missing) or high risk (two or more criteria were missing). As a proxy to publication bias, a funnel plot and the Egger tests were applied only for non-surgical studies outcome on PD, which was considered in ten trials. For the other outcomes and the surgical studies, publication bias could not be assessed because there were fewer than ten included studies.

Data analysis

The treatment outcomes used in the meta-analysis were changes in PD, CAL and BOP from baseline and at 3 months after periodontal pockets were treated by non-surgical therapy. For surgical therapy, only PD and CAL changes were assessed after a follow-up period of at least 6 months.

A correlation coefficient of 0.5 was used in order to calculate the standard deviations (SDs) of the mean difference of the before and after outcome changes. Weighted mean differences (WMDs) and 95% confidence intervals (CIs) between the treatment and controls were estimated using the random-effect model for the continuous outcome amount of BOP, PD reduction and CAL gain. For studies providing only the interquartile ranges (ICRs), the SD was estimated by dividing the ICR by 1.35 [31].

Results were presented as forest plots with weighted means and 95% CIs. Heterogeneity across studies was evaluated using I^2 statistic ($I^2 \geq 50\%$ denoting substantial heterogeneity). All statistical analyses were conducted using the "metan" family of commands in Stata 14.2 (Stata Corp, College Station, TX, USA). Statistical significance was set at $p < 0.05$.

The alternative research hypothesis of this study was that there are differences in the treatment outcomes between the intervention group (with HA) and the control group (without HA).

Results

Search results

A total of 438 studies were identified in six databases. After elimination of duplicates, 261 studies could be assessed. Two hundred forty-three studies had to be excluded in the process of title and abstract reading. Those studies were case series, written in a language other than English, had not an appropriate follow-up or used a combination with other biomaterials (e.g. membranes, bone substitutes). Eighteen full-text publications were further assessed for eligibility. After full eligibility assessment, five studies and the surgical part of one study were excluded (Table 2) and 13 studies were included in this review (search flow diagram: Fig. 1). Among the included studies, 11 clinical trials reported on the effect of HA in non-surgical therapy (scaling and root planing) in patients with chronic periodontitis [26, 32–41] (Table 3), and two studies reported on the effect of HA as adjunct to surgical periodontal therapy [27, 28] (Table 4). The surgical studies compared either flap surgery alone or flap surgery with HA delivery into intrabony defects.

Table 2 Excluded studies and reason for exclusion

Author	Reason for exclusion
Engström et al. (2001)	Only the surgical part was excluded—using a combination of a membrane and HA
Kaira et al. (2015)	Case report + combination of HA with amnion membrane
Mesa et al. (2002)	Study focused on effect of an HA gel on cell proliferation and inflammation
Pilloni et al. (2011)	Not patients with chronic periodontitis
Sandhu et al. (2015)	Case report + combination of HA with platelet-rich fibrin
Xi et al. (2014)	Language (Chinese)

Results of meta-analyses for non-surgical therapy

CAL gain

Nine studies [26, 32–34, 36, 37, 40, 41] reported data on CAL gain for sites treated with scaling and root planing either with or without the adjuvant use of HA after 3 months. Overall, the WMD was 0.73 mm (95% CI 0.28 to 1.17 mm; $p < 0.0001$), favouring the addition of HA. However, considerable heterogeneity was identified among studies (chi-squared test $p < 0.0001$) (Fig. 2).

PD reduction

Eight studies [26, 32–34, 36–38, 41] reported data on PD reduction for sites treated with the use of HA versus a control group without. The WMD of the eight studies was −0.36 mm (95% CI −0.54 to −0.19 mm; $p < 0.0001$), favouring the treatment with HA. Considerable heterogeneity was identified among studies (chi-squared test $p < 0.0001$) (Fig. 3).

BOP reduction

Five studies [26, 32, 34, 37, 41] reported data on BOP reduction in percentage of sites treated with HA versus a control group. Overall, the WMD was −15% (95% CI −22 to −8%; $p < 0.001$), favouring the treatment. Despite, considerable heterogeneity was identified among studies (chi-squared test $p < 0.0001$) (Fig. 4).

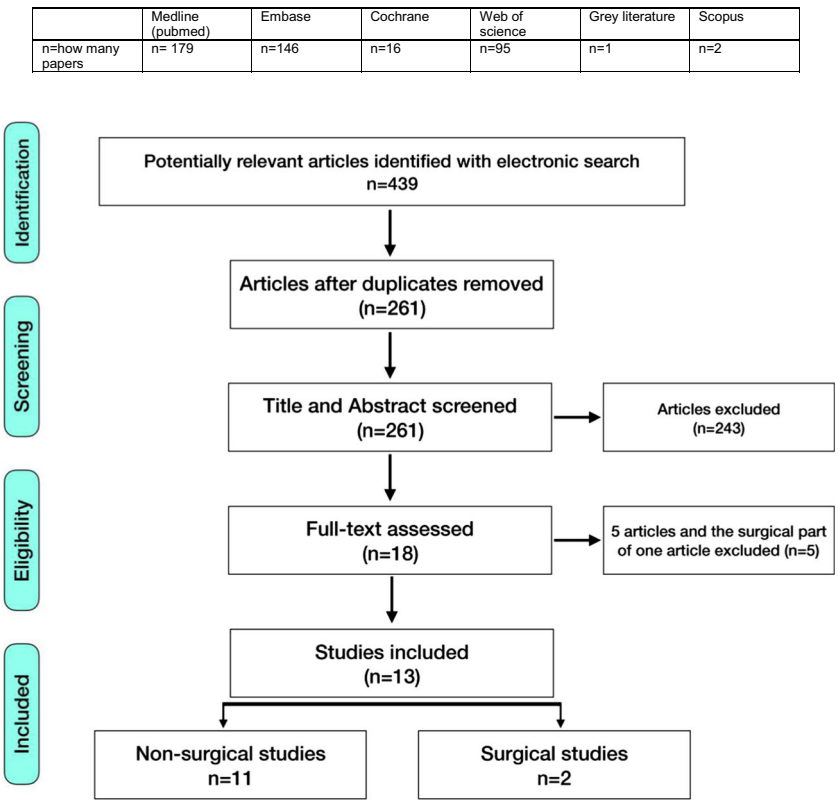

Fig. 1 Flow diagram describing the search and study inclusion process

Table 3 Characteristics of included studies—non-surgical therapy

Author (year)	Study design	Participants Control/test	Clinical parameters Control/test	Intervention	Follow-up	Outcomes	Site and funding
Bevilacqua (2012)	Split mouth	11 individuals	Average BOP (%) 72.7 72.7 Average PD (mm) 6.36 (5.86–6.87) 6.14 (5.7–6.58) Average CAL (mm) 5.91 (5–6.83) 5.91 (5–6.84)	1. SRP + placebo 2. SRP + HA	45 days 90 days	PD, CAL, BOP, PI	University, industry
Chauhan (2013)	Parallel groups	40 individuals 20/20	Average CAL (mm) 6.10 ± 0.38 6.13 ± 0.54 Average PD (mm) 5.93 ± 0.6 5.90 ± 0.27	1. SRP 2. SRP + HA	3 months	PD, CAL, GI, PI	University, industry
Eick (2013)	Parallel groups	34 individuals 17/17	Average PD (mm) 4.1 ± 0.4 mm 4.2 ± 0.4 mm Average BOP (%) 18.8 ± 11.1 mm 16.3 ± 8.7 mm Average CAL (mm) 5.7 ± 0.6 mm 5.5 ± 0.9 mm	1. SRP 2. SRP + HA + 2*d HA rinsing for 2 weeks	3 + 6 months	BOP, PI, CAL, PD	University, industry
Engström (2001)	Split mouth	9 individuals	Average PD (mm) 6.8 ± 1.5 6.4 ± 1.3	1. SRP 2. SRP + HA (3×)	2 weeks 1 month 3 months 6 months	BOP, PI, PD	University, industry
Gontiya and Galgali (2012)	Parallel groups	26 individuals 13/13	Average PD (mm) 6.42 ± 0.44 6.57 ± 0.45 Average CAL (mm) 8.56 ± 0.41 8.91 ± 0.41	1. SRP 2. SRP + HA (4×)	12 weeks	PD, CAL (RAL), PD	University
Johannsen (2009)	Split mouth	12 individuals	Average BOP (%) 58 (26) 74.5 (45.7) (IQR) Average CAL (mm) 4.5 (4.2–4.7) 4.4 (4.1–4.8) Average PD (mm) 4.2 (3.6–4.7) 4.2 (3.7–4.7)	1. SRP 2. SRP + HA (2×)	12 weeks	BOP, PI, CAL, PD	University, industry

Table 3 (continued)

Author (year)	Study design	Participants Control/test	Clinical parameters Control/test	Intervention	Follow-up	Outcomes	Site and funding
Koshal (2012)	Split mouth	52 individuals	Average PD (mm) 3.90 ± 0.93 3.82 ± 0.78	1. SRP + placebo 2. SRP + HA	3 months	GI, PD	University, industry
Polepalle (2015)	Split mouth	18 Individuals	Average PD (mm) 5.21 ± 0.54 4.99 ± 0.34 Average CAL (mm) 5.41 ± 0.65 5.40 ± 0.71	1. SRP 2. SRP + HA (2×)	12 weeks	GI, PI, PD, CAL	University
Rajan (2014)	Split mouth	33 individuals	Average PD (mm) 6.09 ± 1.26 6.33 ± 0.99 Average CAL (mm) 9.12 ± 1.67 10.18 ± 2.08	1. SRP 2. SRP + HA (2×)	4 weeks 12 weeks	GI, PI, PD, CAL	University
Wan (2004)	Parallel groups	56 individuals 28/28	Average BOP (%) 67.4% ± 21.2 71.3% ± 16.8 Average PD (mm) 2.5 ± 0.7 2.4 ± 0.5 Average PAL (mm) 12.7 ± 2.6 12.3 ± 2.2	1. SRP + placebo 2. SRP + HA (2×)	1 month 3 months	BOP, PI, PD, CAL (PAL)	University, industry
Xu (2004)	Split mouth	20 individuals	Average PD (mm) 5.2 ± 1.62 5.3 ± 1.61 Average CAL (mm) 5.4 ± 1.97 5.5 ± 1.79 Average BOP (mm) 72% 78%	1. SRP 2. SRP + HA (6×)	6 weeks 12 weeks	BOP, CAL, PD	Grant (German Academic Exchange Service), university

BOP bleeding on probing, *PD* probing depth, *CAL* clinical attachment level, *PI* plaque index, *HA* hyaluronic acid, *SRP* scaling and root planing, *GI* gingival index, *RAL* relative attachment level, *PAL* probing attachment level, *IQR* interquartile range

Table 4 Characteristics of included studies—surgical therapy

Author (year)	Study design	Participants Control/test	Clinical parameters Control/test	Intervention	Follow-up	Outcomes	Site and funding
Briguglio (2013)	Parallel groups	40 individuals 20/20	Average PD 8.0 ± 0.7 8.6 ± 1.5 Average CAL 8.3 ± 1.2 7.2 ± 1.5	1. IBD + EDTA 2. IBD + EDTA + HA	12 months 24 months	PD, CAL, BOP, PI	University
Fawzy El-Sayed (2012)	Split mouth	14 individuals 2 teeth per site	Average CAL 5.50 (5.00/8.00) 5.50 (2.00/7.00) (IQR) Average PD 5.00 (5.00/6.00) 5.00 (5.00/6.00) (IQR)	1. IBD 2. IBD + HA	3 months 6 months	CAL, GR, PD, GI, PI	Funded by the first author

BOP bleeding on probing, *PD* probing depth, *CAL* clinical attachment level, *PI* plaque index, *GI* gingival index, *HA* hyaluronic acid, *GR* gingival recessions, *IBD* intrabony defect, *EDTA* ethylenediaminetetraacetic acid, *IQR* interquartile range

Results of meta-analyses for surgical therapy

CAL gain

Two studies reported data on CAL gain for sites treated with HA versus a control group at 6 months and 24 months [27, 28]. The WMD was 0.85 mm (95% CI 0.08 to 1.62 mm; $p < 0.0001$), favouring the treatment. A low heterogeneity among studies was observed (chi-squared test $p = 0.822$) (Fig. 5).

PD reduction

Two studies [27, 28] reported data on PD reduction for sites treated with HA versus a control group at 6 months and 24 months. Overall, the WMD was − 0.89 mm (95% CI − 1.42 to − 0.36 mm; $p < 0.0001$), favouring the adjunctive use of HA. Furthermore, the comparison presented low heterogeneity among the two studies (chi-squared test $p = 0.714$) (Fig. 6).

BOP reduction

BOP was not measured in the included studies. Therefore, there are no results for BOP reduction in surgical therapy.

Results of risk of bias assessment

Results of the risk of bias assessment for the included RCTs are summarized in Table 5. Only three studies were

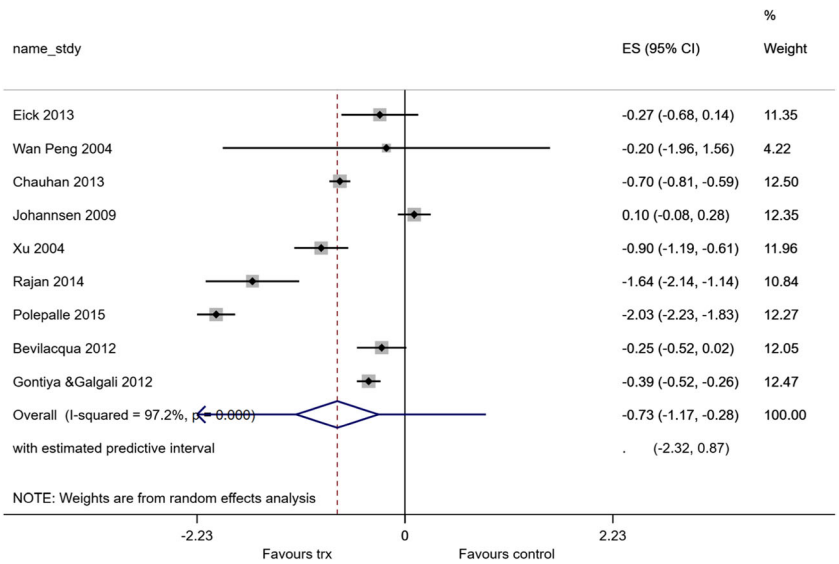

Fig. 2 Forest plots for CAL gain following non-surgical therapy after 3 months

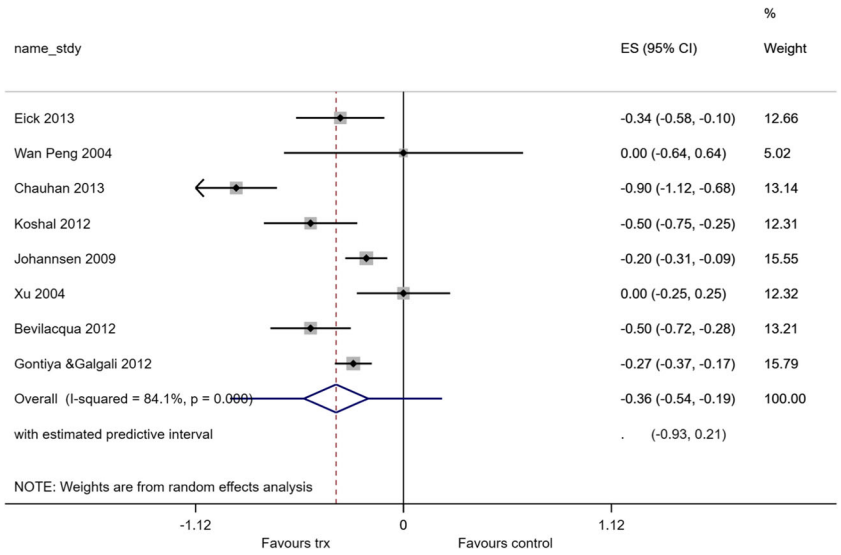

Fig. 3 Forest plot for PD reduction following non-surgical therapy after 3 months

assessed at low risk of bias while 11 studies were determined to be at high risk.

Publication bias

The Egger test was not significant suggesting that there was no evidence for small study effects. The funnel plot is asymmetric; however, it is difficult to assess whether this is due to publication bias as a number of reasons could be the reason for this asymmetry [42] (Fig. 1).

Discussion

The present systematic review including meta-analysis has evaluated the potential additional effects of local application of HA on the clinical outcomes of non-surgical and surgical periodontal therapy.

Thirteen RCTs fulfilled the inclusion criteria with adequate follow-up (3 months for non-surgical treatment and more than 6 months for surgical treatment).

Eleven RCTs have evaluated the effectiveness of HA adjunctively to non-surgical treatment on chronic periodontitis patients. Six out of the 11 studies were performed in a split-mouth design and five in a parallel group design. The application frequency of the different HA-containing products differed between the studies from one application during scaling and root planing to a repeated application during scaling and root planing and additional weekly applications up to 6 weeks.

The meta-analysis revealed that non-surgical treatment with adjunctive HA resulted in additional PD reduction (mean − 0.36 mm), CAL gain (mean 0.73 mm) and BOP reduction (mean − 15%) compared with conventional scaling and root

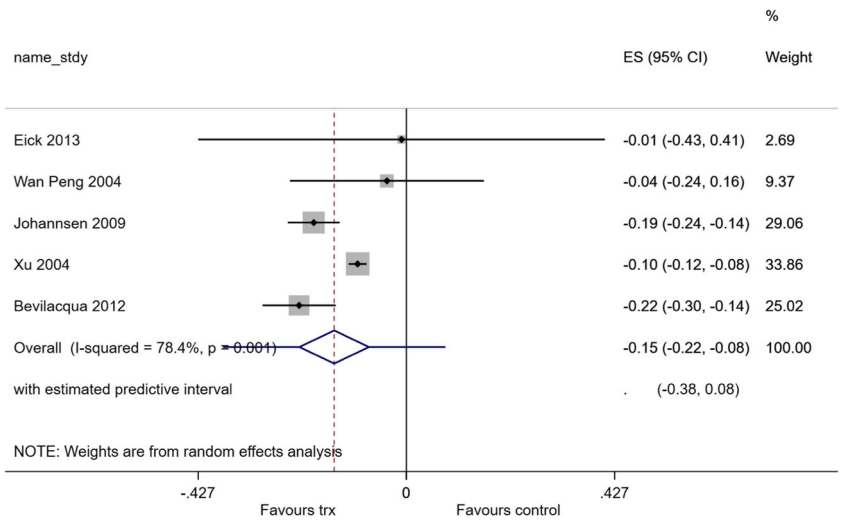

Fig. 4 Forest plot for BOP reduction following non-surgical therapy after 3 months

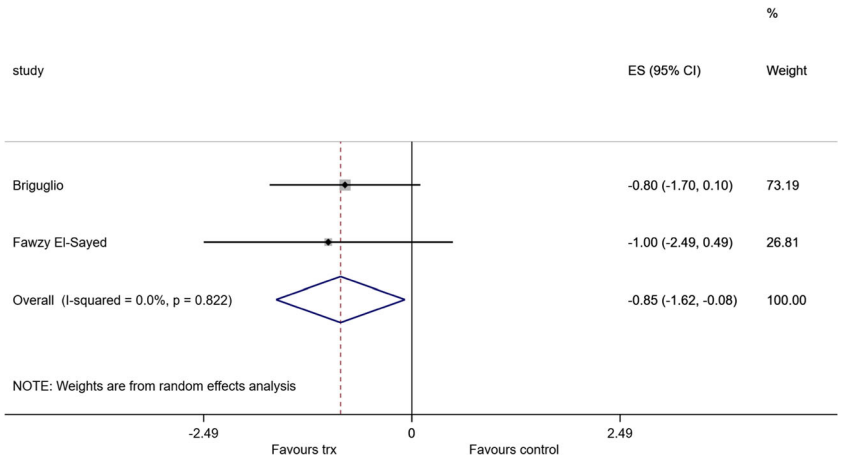

Fig. 5 Forest plot for CAL gain following surgical therapy after 6–24 months

planing after 3 months. If we are looking at results of a recently published systematic review [43] about additional CAL gain with different adjuncts compared with scaling and root planing alone (0.35 mm with systemic antimicrobials, PDT diode laser 0.53 mm, chlorhexidine chips 0.40 mm), HA could represent a suitable alternative to the most frequently used adjuvants. Nevertheless, there was an overall high risk of bias and a high heterogeneity among the studies.

The heterogeneity among the studies may be attributed to differences in the treatment protocol and the different types of products used. All the products contained high molecular weight HA with a concentration from 0.2 to 0.8%. It needs to be kept in mind that the most appropriate protocol, product and concentration for the clinical application of HA are still unknown. Moreover, in the included studies, there are different time points and different number of applications. Additionally, it is still unknown which formulation of HA (i.e. cross-linked or non-cross-linked) will give the best clinical result [44].

Two RCTs have evaluated the effectiveness of HA as an adjunctive to surgical treatment (open flap debridement (OFD)) in chronic periodontitis patients. One study was conducted as a split-mouth study and one with a parallel group design. In both studies, intrabony defects were treated with either OFD + HA (test) or OFD (control). The results have shown that after 6–24 months, the adjunctive application of HA yielded statistically significantly higher clinical improvements evidenced by PD reduction and CAL gain compared with OFD alone thus suggesting that HA has an added beneficial effect when used as an adjunct to periodontal surgery [27, 28]. It is generally accepted that angular bony defects, when left untreated, will worsen/progress over time, eventually leading to tooth loss [45]. The results of the present meta-analysis indicate that the use of HA in conjunction with OFD may provide an added clinical benefit evidenced by a further reduction in PD and gain of CAL gain in intrabony defects compared with OFD alone.

The added clinical improvements shown in the present meta-analysis are in line with the results from several preclinical and clinical studies. A case series of surgical periodontal therapy in conjunction with HA and autologous bone revealed good clinical outcome without the use of a membrane [46]. Furthermore, another case series showing promising results in intrabony defect treated with HA in conjunction with OFD

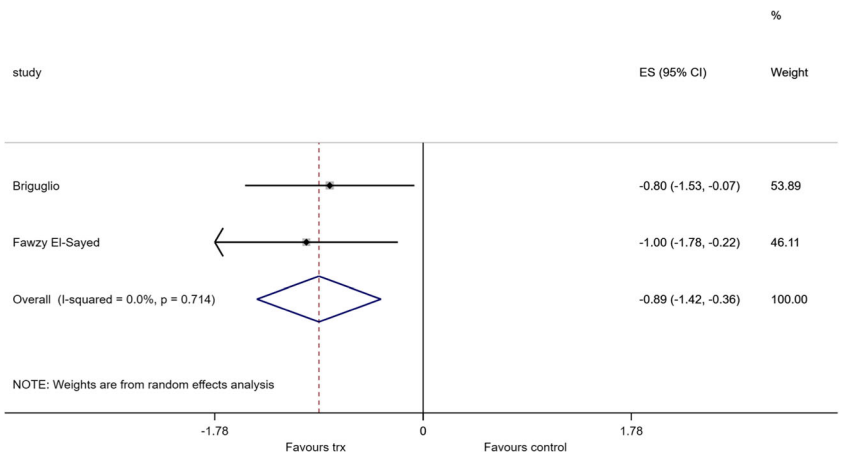

Fig. 6 Forest plot for PD reduction following surgical therapy after 6–24 months

Table 5 Results of quality assessment

	Random sequence generation (selection bias)	Allocation concealment (selection bias)	Blinding of participants and personnel (performance bias)	Blinding of outcome assessments (detection bias)	Incomplete outcome data (attrition bias)	Selective outcome reporting (reporting bias)	Other bias	General risk assessment
Bevilacqua (2012)	+	+	+	+	+	+	+	Low
Chauhan (2013)	+	+	−	?	+	+	−	High
Eick (2013)	+	+	−	+	−	+	?	High
Engström (2012)	+	+	−	+	−	+	?	High
Gontiya and Galgali (2012)	+	+	?	?	+	+	?	High
Johannsen (2009)	+	+	−	?	+	+	?	High
Polepalle (2015)	+	+	−	?	+	+	?	High
Rajan (2014)	+	?	+	+	?	?	?	High
Wan (2004)	+	+	+	+	+	+	+	Low
Xu (2004)	+	+	−	−	+	+	?	High
Kohal (2012)	+	−	+	−	+	+	?	High
Briguglio (2013)	+	+	+	+	+	+	+	Low
Fawzy El-Sayed (2012)	+	+	−	?	+	+	?	High

'+' = low risk; '?' = unclear risk; '-' = high risk

[29]. HA has shown to increase osteoblast activity by stimulating differentiation and migration of mesenchymal cells [6] and accelerate bone formation in a rabbit model [47]. Kim et al. reported that HA improved wound healing and bone formation in hemisectioned-performed extraction sockets with communication to periodontal lesions in a canine model [48].

Taken together, the positive outcomes reported in preclinical and clinical studies corroborate the results of the present meta-analysis and lend additional support to the capacity of HA to improve wound healing. Findings from medical field have shown that HA possesses a number of properties that are relevant in wound healing such as stabilizing the blot clot, lowering the inflammatory response, helping in neovascularization and angiogenesis and accelerating fibroblast migration and wound closure [49, 50].

The above-mentioned positive biologic effects of HA are also supported by the results of a recently published preclinical (i.e. in vitro) study which have demonstrated that HA enhanced expression of genes encoding type III collagen and transforming growth factor-β3, characteristic of scarless wound healing [44]. The application of HAs up-regulated the expression of genes encoding pro-proliferative, pro-migratory and pro-inflammatory factors in palatal and gingival fibroblasts while in palatal but not gingival fibroblasts, an indirect effect of HA on the expression of matrix metalloproteinases 2 and 3 was detected. Taken together, these preclinical data provide further support on the effects of HA to enhance the proliferative, migratory and wound-healing properties of cell types involved in periodontal wound healing/regeneration.

When discussing the role of HA on wound healing, it needs to be also pointed to the findings of a preclinical study in dogs, which have failed to show an advantage of using HA in periodontal surgery [51]. Following the application of HA in surgically created class III furcation defects, the histological analysis did not reveal any substantial formation of root cementum, periodontal ligament and bone. However, these negative findings are most likely due to the low regenerative potential of class III furcation defects [52].

It has also to be realized that the present systematic review and meta-analysis has a number of limitations, and therefore, the results need to be interpreted with caution. First of all, there is a significant heterogeneity

between the studies evaluating HA in non-surgical periodontal therapy due to study design, treatment time points, products and outcome assessments. Second, out of 13 RCTs evaluating the effects of HA in conjunction with surgical periodontal therapy, only two studies fulfilled the inclusion criteria (i.e. 11 had a high risk or unclear risk of bias), and thus, there is a need for well-designed, controlled clinical studies evaluating this material in conjunction with periodontal surgery.

Obviously, due to an overall high risk of bias and heterogeneity among the studies, there is a need for future well-designed RCTs to justify the benefits of using HA for non-surgical periodontal treatment. Last but not least, an appropriate protocol and the most adequate formulation of HA for clinical applications need to be tested and further evaluated.

Conclusion

Within their limits, the present data indicate that the topical application of HA may lead to additional clinical benefits when used as an adjunctive to non-surgical and surgical periodontal therapy. However, due to the high risk of bias and heterogeneity, there is a need for further well-designed RCTs to evaluate this material in various clinical scenarios.

Funding The work was supported by the authors' own institutions.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

Informed consent For this type of study (e.g. systematic review and meta-analysis), formal consent is not required.

Appendix 1 Full searching strategy

((“periodontics” [MeSH Terms] OR “periodontics” [All Fields] OR “periodontology” [All Fields]) OR (“periodontitis” [MeSH Terms] OR “periodontitis” [All Fields]) OR (“periodontal diseases” [MeSH Terms] OR “periodontal” [All Fields] AND “diseases” [All Fields]) OR “periodontal diseases” [All Fields] OR (“periodontal” [All Fields] AND “disease” [All Fields]) OR (“periodontal disease” [All Fields] OR “periodontal pocket” [MeSH Terms]) OR (“periodontal” [All Fields] AND “pocket” [All Fields]) OR (“periodontal pocket” [All Fields]) OR (furcation [All Fields] AND (“therapy” [Subheading] OR “therapy” [All Fields] OR “therapeutics” [MeSH Terms] OR “therapeutics” [All Fields])) OR (intrabony [All Fields] AND defect [All Fields]) OR (infrabony [All Fields] AND defect [All Fields]) OR (intraosseous [All Fields] AND defect [All Fields]) OR (periodontal [All Fields] AND (“surgery” [Subheading] OR “surgery” [All Fields] OR “surgical procedures, operative” [MeSH Terms] OR (“surgical” [All Fields] AND “procedures” [All Fields] AND “operative” [All Fields]) OR “operative surgical procedures” [All Fields] OR “surgery” [All Fields] OR “general surgery” [MeSH Terms] OR (“general” [All Fields] AND “surgery” [All Fields]) OR “general surgery” [All Fields])) OR (periodontal [All Fields] AND (“therapy” [Subheading] OR “therapy” [All Fields] OR “therapeutics” [MeSH Terms] OR “therapeutics” [All Fields])) OR (periodontal [All Fields] AND (“regeneration” [MeSH Terms] OR “regeneration” [All Fields]))) AND (hyaluron [All Fields] OR (“hyaluronic acid” [MeSH Terms] OR (“hyaluronic” [All Fields] AND “acid” [All Fields]) OR “hyaluronic acid” [All Fields]) OR (“hyaluronic acid” [MeSH Terms] OR (“hyaluronic” [All Fields] AND “acid” [All Fields]) OR “hyaluronic acid” [All Fields] OR “hyaluronan” [All Fields]) OR (“hyaluronic acid” [MeSH Terms] OR (“hyaluronic” [All Fields] AND “acid” [All Fields]) OR “hyaluronic acid” [All Fields] OR “hyaluronate” [All Fields]))

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