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ANTIMICROBIAL EFFICACY OF CHLORHEXIDINE AND SODIUM HYPOCHLORITE IN ROOT CANAL DISINFECTION

Abstract

We aimed to compare the antimicrobial efficacy of chlorhexidine (CHX) and sodium hypochlorite (NaOCl), 2 irrigants routinely used in root canal therapy of permanent teeth. Bacteria have a fundamental role in the pathogenesis of pulp periapical diseases. However, elimination of bacteria from an infected canal is a difficult process that requires the use of various instrumentation techniques, irrigants, and intracanal medications. Remnant pulp can serve as a nutrient source for remaining bacteria
Keywords: *chlorhexidine, endodontic treatment, meta-analysis, root canal irrigant, root canal disinfection, sodium hypochlorite*

Qələndər Xanlar oğlu Əliyev

Xlorheksidin və natrium hipoxloridin antimikrobiyal effektivliyi və kök kanallarının dezinfeksiyası

Xülasə

Daimi dişlərin kanal müalicəsində müntəzəm olaraq istifadə edilən 2 irriqant olan xlorheksidin (CHX) və natrium hipoxloritin (NaOCl) antimikrobiyal effektivliyini müqayisə etmək məqsədi daşıdıq. Bakteriyalar pulpa periapikal xəstəliklərin patogenezində əsas rol oynayır. Bununla belə, infeksiyalaşmış kanaldan bakteriyaların aradan qaldırılması müxtəlif cihaz üsullarının istifadəsini tələb edən çətin bir prosesdir. Qalıq pulpa bakteriyalar üçün qida mənbəyi kimi xidmət edə bilər. Kanalda qalan toxumalar da ola bilər irriqantların antibakterial təsirini məhdudlaşdırmaq irriqantlar və intrakanal dərmanlar.
Açar sözlər: *xlorheksidin, endodontik müalicə, meta-analiz, kök kanalının irriqantı, kök kanalının dezinfeksiyası, natrium hipoxlorit*

Introduction

Electronic databases, including PubMed, EMBASE, Web of Science, and Cochrane Library, were searched for randomized controlled trials published until March 2020. The meta-analysis of relative risk (RR) and standardized mean difference (SMD) was performed using a random effects model with a 95% confidence interval (CI). Subgroup analysis was performed for culture and molecular methods of bacterial detection.

Our findings suggest that both CHX and NaOCl can reduce bacterial infections after irrigation without any significant difference in antimicrobial efficacy between them. Although CHX and NaOCl showed similar efficacy, their molecular mechanisms were different. Therefore, they can be used as the main antibacterial root canal irrigants. However, our results were limited by inconsistencies among retrieved articles and a lack of clinically relevant outcomes. Further well-designed clinical studies are warranted to supplement our results.

Bacteria and their by-products are the main etiologic factors for pulpal and periapical diseases (Ahmed, Versiani, De-Deus, Dummer, 2017: 761-770). The goals of endodontic treatment are to achieve complete disinfection and prevent reinfection in the root canal system and periapical tissues. Sterilization of root canals is limited by the presently available techniques, instruments, and irrigants (Alves, Rochas, Almeida, 2012: 871-877). Thus, the focus should be on reducing intracanal bacterial populations to levels that are compatible with periapical tissue healing. Chemomechanical preparation, including both mechanical instrumentation and chemical irrigation, is crucial for decreasing bacterial population. Mechanical instrumentation alone is insufficient to yield effective disinfection (Alves, Rochas, Almeida, 2012: 871-877) because the complexity of root canal anatomy (Bui, Baumgartner, Mitchell, 2008: 181-185; Byström, Happonen, Sjögren, 1987: 58-63) prevents the accessibility of instrumentation and provides a shelter for microorganisms (Byström, Happonen, Sjögren, 1987: 58-63). The bacteria remaining in the root canal at the time of root filling cause persistent infection and treatment failure. Therefore, to achieve adequate disinfection, mechanical instrumentation should be supplemented with chemical irrigation methods.

Sodium hypochlorite (NaOCl) is the most widely used irrigant during endodontic treatment because of its effective antimicrobial (Dahlén, 2017: 51-89; Delany, Patterson, Miller, Newton, 1982; 518-523) and tissue-dissolving properties (Dutner, Mines, Anderson, 2012: 37-40). Several concentrations of NaOCl ranging from 0.5%–5.25% were found in the endodontic literature, and the most widely used concentration is 2.5%. Although higher concentrations of NaOCl may exert stronger antimicrobial activity and tissue-dissolving properties (Dutner, Mines, Anderson, 2012: 37-40), they can lead to increased cytotoxicity[16] and periapical tissue irritation (Gatot, Arbelle, Leiberman, 1991: 573-574).

Chlorhexidine (CHX) is an alternative irrigant to NaOCl because of its broad-spectrum antimicrobial activity (Gomes, Martinho, Vianna, 2009: 1350-1353) and considerably lower toxicity than NaOCl[20]. The most widely used concentration of CHX for root canal therapy is 2%. In contrast to NaOCl, high concentrations of CHX exert a bactericidal effect, whereas low concentrations provide only a bacteriostatic effect (Goncalves, Rodrigues, Andrade Junior, 2016: 527-553). CHX can be used either as a gel or solution with the same effectiveness. It exhibits the unique property of substantivity; the positive charges of the CHX molecule bind to the negative charges on dental surfaces resulting in prolonged adherence, which in turn leads to long-lasting antimicrobial activity (Goncalves, Rodrigues, Andrade Junior, 2016: 527-553). However, as an endodontic irrigant, the lack of tissue-dissolving capacity of CHX is a considerable drawback (Higgins, Altman, Gotzsche, 2011: 928).

Recent literature shows no agreement on the antimicrobial efficacy of CHX versus NaOCl because various studies presented contradictory results. A previous systematic review on the comparison of antimicrobial efficacy of CHX and NaOCl concluded that the number of clinical studies was scarce and inconsistent (Kakehashi, Stanley, Fitzgerald, 1965: 340-349) and proposed that additional well-designed randomized controlled trials (RCTs) should be conducted. A meta-analysis is required to provide robust evidence and improve clinical outcomes. Therefore, we performed a systematic review followed by a meta-analysis of available RCTs investigating the antimicrobial efficacy of CHX and NaOCl in root canal disinfection to improve the outcome of endodontic treatment.

This study complies with the Preferred Reporting Items for Systematic Reviews and Meta-analysis Statement (Kuruvilla, Kamath, 1998: 427-476). İt is defined by the following characteristics: Population, participants with pulpal and or periapical disease who received endodontic treatment in permanent teeth; Intervention, CHX irrigant.

The inclusion criteria were RCTs that used irrigants in root canal therapy of permanent teeth with pulpal and/or periapical disease. These studies compared the antimicrobial effects between CHX and NaOCl irrigants and reported the outcome as bacterial reduction using bacterial cultivation and/or molecular microbiological methods. Studies that did not compare the individual effects of NaOCl and CHX and those performed in primary teeth or open apex teeth were excluded from this meta-analysis.

Relative risk (RR) was calculated for studies that reported the detection of samples showing positive and negative bacterial growth after irrigation. For studies reporting the number of bacteria before and after irrigation, the standardized mean difference (SMD) was calculated. The 95% confidence intervals (CIs) were calculated for RR and SMD to compare the antimicrobial efficacy of CHX and NaOCl. Subgroup analysis was conducted if sufficient data were obtained. The significance of any variation and degree of heterogeneity was determined by I^2 and chi-square statistics, respectively (Nair, Henry, Cano, Vera, 2005: 231-252). Pooled estimates were calculated with a random effects model using the DerSimonian-Laird method. Because of the low number of studies included, publication bias tests were not conducted. We attempted to perform trial sequential analysis to estimate the information size related to the imprecision of outcomes; however, it could not be performed because of low information size. Comprehensive Meta-Analysis Software Version 3 was used to compute the RR and SMD.

The strength of evidence was evaluated according to the Grading of Recommendations, Assessment, Development, and Evaluation approach using a summary of findings table constructed with grade pro Guideline Development Tool software (Evidence Prime, Inc, Seattle, WA). Each gradecriterion was assessed individually and then computed for the certainty of the evidence. To achieve transparency and implicity, the grade approach classifies the certainty of evidence into 1 of the following 4 grades: high, moderate, low, or very low.

One additional record (33) was identified in the reference list of a review article (Neelakantan, Herrera, Pecorari, Gomes, 2019: 19-22). After removing duplicates, the remaining 2110 records were screened for titles and abstracts. A total of 2099 irrelevant records were found and removed. Finally, 11 articles were assessed for eligibility by full-text reviewing. At this stage, 3 articles were excluded because of the following reasons: 1 study reported only endotoxin level as an outcome, another study was a non-RCT, and the last one investigated only the effect of gaseous ozone in combination with NaOCl and CHX(37).

Although the 8 studies contained statistical data, 6 studies provided the number of samples with positive and negative bacterial growth (Sigueira, Rôças, Favieri, 2000: 331-334) that could be included for RR analysis. One study reported only the number of visits that yielded negative culture, whereas another reported only secondary outcome parameters; therefore, these 2 studies were not suitable for the quantitative analysis. Also, 1 of the 6 studies only reported selective bacterial strains, whereas another provided only the categorized data of bacteria. Therefore, these 2 studies were not suitable for SMD analysis. Finally, 4 studies that revealed the number of bacteria before and after irrigation were eligible for SMD analysis (Ringel, Patterson, Newton, 1982: 19-27).

The characteristics of the RCTs are presented in Table 1. All the studies are single-center RCTs. They reported the concentrations and forms of irrigants. However, only 2 studies indicated the total amount of irrigants (Rôça, Provenzano, Neves, Sigueira, 2016: 943-947), whereas 4 studies reported the volume with instrumentation. In addition, 1 study did not specify the volume of irrigants and vaguely mentioned “a copious amount of irrigant”.

All the studies used sterile paper points, either dry or soaked in transporting media, for sample collection. Only data taken from samples within the first visit were extracted. Of the 8 studies included in our meta-analysis, only 1 used both culture and molecular methods. Four studies used molecular methods (Rôça, Provenzano, Neves, Sigueira, 2016: 943-947), whereas another 3 studies used culture methods. However, none of the studies investigated fungi.

However, 1 pair of studies was assessed as a single study because 1 of the studies was a continuation. Four studies were scored as an overall “low” risk of bias, although 2 of them showed a “high” risk of bias of allocation concealment domain. Another 4 studies were considered as an overall “unclear” risk of bias because none of them mentioned their randomization methods, allocation concealment, and irrigation in sufficient detail. In addition, 1 of them was assigned to “high” risk on the “other bias” domain. Supplemental Appendix S3 (available online at www.jendodon.com) explains the risk of bias assessment for individual studies.

The grade approach was used to rate the confidence of evidence obtained from our meta-analysis comparing the efficacy of the investigated irrigants on antibacterial parameters (Supplemental Table S1 is available online at www.jendodon.com). The total bacterial number reduction was graded as very low evidence based on the serious inconsistency, indirectness, and imprecision domains. The incidence of samples with positive bacterial growth after irrigation was graded as low evidence based on serious indirectness and imprecision domains.

In this study, we aimed to compare the antimicrobial efficacy of CHX and NaOCl irrigants in root canal therapy of permanent teeth. We found no significant differences in their antimicrobial efficacy.

Effective chemomechanical preparation using chemical substances can improve the clinical outcome and long-term success of endodontic treatment. Our meta-analysis included only RCTs, which are considered as the highest level in the hierarchy of evidence. Although numerous records were preliminarily retrieved from the 4 databases, only 8 RCTs remained eligible for a systematic review supplemented by the meta-analysis. However, inconsistent data (eg, infection types) were observed among the 8 RCTs. A single treated root canal with persistent infection can harbor a similar number of bacteria to that of untreated root canals with primary infection; however, the microbial diversity decreases after treatment in persistent infection (43). Other factors such as tooth type, mechanical preparation, final canal enlargement and taper, irrigation protocols, and bacterial identification methods were also heterogeneous.

Various irrigation parameters of CHX and NaOCl were used in the included RCTs, especially concentrations and volume of irrigants. Their antimicrobial effect depends on the frequency and contact time during irrigation (Rôças, Sigueira, 2011: 143-150). Larger volume or longer contact time and frequent exchange of irrigants could compensate for the effects of smaller concentrations. However, these parameters were not precisely described in the trials. Furthermore, other factors, such as formulations, activation techniques and devices (Sigueira, Rôças, 2007: 267-280) and multivariable ratios of all parameters, are strongly related to the resulting antibacterial efficacy. Because of the limited data, we were not able to describe how these confounding factors affect our meta-analysis and perform further subgroup analysis. Because all these factors play a major role in the effectiveness of irrigants, future RCTs should ensure to publish all the details mentioned earlier.

Regarding sample collection, sterile paper points obtain bacteria only from the main root canal, and there is a lack of information on bacteria colonizing the hidden areas of the complex root canal (Sjögren, Hägglund, Sundgvist, 1990: 498-504). Therefore, bacteria collected by this technique might not be perfect representatives of all bacterial populations in the entire root canal system. Nevertheless, at present, no better sampling method is available. Two bacterial detection methods after sample collection were described in the included studies. The first one is the culture method, which can estimate bacterial load and detect virulence factors or antibiotic susceptibility (Tatnall, Leigh, Gibson, 1990: 157-163). However, this method cannot characterize several microorganisms in parallel or identify uncultivable bacteria. The molecular method was introduced to overcome the limitations of semiqualitative culture techniques. Studies in this meta-analysis used either the culture or molecular method, except for 1 study in which both the methods for bacterial quantification were applied. In molecular methods, Taqman and SYBRGreen assays were also compared. However, the Taqman assay provides precise results when low target samples are used. Consequently, we selected culture method results for RR analysis and molecular method results from the Taqman assay for SMD analysis. According to the varied sensitivity of culture and molecular methods, subgroup analysis for SMD was performed separately (Supplemental Figure S1 is available online at www.jendodon.com). The results indicated no significant difference in changes in the bacterial count after chemomechanical preparation between these 2 methods. This finding is in line with that of a recent *in vitro* study (Xavier, Martinho, Chung, 2013: 959-964). Thus, both methods are appropriate for endodontic bacterial detection.

The controversies among the results of the included RCTs might also be influenced by sample size because improper sample sizes will not give sufficient power to detect any differences between interventions (Zahed, 2008: 329-341). Among the included RCTs, only 2 reported sample size calculation.

In the present meta-analysis using RR or SMD parameters, no significant difference was found in antibacterial efficacy between CHX and NaOCl treatments. Our analysis extended the basis of similar RR results in a previous meta-analysis published 8 years ago (34), which was based only on 2 articles. Our findings of bacterial reduction also closely corresponded with those of another meta-analysis (55), which showed that intracanal endotoxin levels decreased compared with the initial levels after applying CHX and NaOCl. However, they found that NaOCl was more effective in the reduction of gram-negative bacterial endotoxin than CHX, but none of the gram-positive bacterial parameters were investigated.

Notably, in the included studies, more than half of the samples exhibited negative bacterial growth after irrigation with CHX or NaOCl, suggesting that neither of them could completely eliminate the bacterial population from the root canal. Although CHX and NaOCl showed similar antibacterial effectiveness, their molecular mechanisms of action were different. Clinicians should take other properties such as the necrotic pulp-dissolving capacity of NaOCl (23) or the substantivity of CHX[22] into consideration. Based on these properties, the combination of CHX and NaOCl may be recommended for endodontic irrigation. However, during the simultaneous application, their mixture can cause precipitate formation, which might occlude the dentinal tubules (Zandi, Rodrigues, Kristoffersen, 2016: 1307-1313). Thus, the consecutive application of NaOCl and CHX with intermediate flushes between each irrigant is needed. The underlying discrepancy of the included studies might present certain limitations. Nonetheless, 4 studies retained an overall low risk of bias.

Although 2 of them did not perform the allocation concealment, the overall risk of bias resulted in low risk because of the prevalence of other low-risk key domains. The remaining 4 studies were considered unclear. One of these studies showed a high risk of other bias[24] because it did not clearly describe the method used to randomize patients with multiple teeth, whether individual teeth or patients were the units of randomization. It is possible that these examinations are not independent, and the outcome may be subjected to a clustering effect. The investigated teeth might also be prone to cross contamination.

None of the included studies reported patient-relevant outcomes such as clinical symptoms and their disappearance, which are related to the effectiveness of the root canal irrigants, that would directly provide a recommendation to clinicians. Only 1 study showed the success rate based on radiographic outcome during endodontic retreatment. Taking all of these reasons into account, it can be suggested that our meta-analysis is restricted by inconsistent and insufficient data from the included RCTs, resulting in the downgrading of certainty of our evidence. Therefore, further well-designed RCTs performed using different types of teeth and with proper sample size and all clinically relevant outcomes are required.

Conclusion

In conclusion, the obtained evidence suggested that both CHX and NaOCl significantly, but not completely, reduced endodontic infections during root canal therapy. They were found to be equally effective despite their different molecular mechanisms. Because the mixture of these 2 chemicals can cause precipitate formation, their consecutive application with intermediate flushes between each irrigant as well as the development of more potent antibacterial agents is proposed.

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