RESEARCH ARTICLE

Received / Geliş Tarihi: 17.11.2025
Accepted / Kabul Tarihi: 04.02.2026
Published / Yayın Tarihi: 30.04.2026

Aydın Dental Journal - Volume 12 Issue 1 - April 2026 (1-11)

Aydın Dental Journal

Journal homepage: http://dergipark.ulakbim.gov.tr/adj

DOI: 10.17932/IAU.DENTAL.2015.009/dental_v012i1001

Effectiveness of Ascorbic Acid in Eliminating Orange-Brown Precipitate from Root Canals

Kök Kanallarından Turuncu-Kahverengi Çökeltiyi Uzaklaştırmada Askorbik Asitin Etkinliği

Beyza Tunç[1], İrem Özçelik[1], Ceren Özcan Diker[2], Kürşat Er[3*]

ABSTRACT

Objectives: To determine the effect of ascorbic acid (AA) on the removal of orange-brown precipitate (para-chloroaniline) (PCA) formed by the interaction of sodium hypochlorite (NaOCl) and chlorhexidine (CHX) during root canal treatment.

Materials and Method: PCA was obtained by mixing 1.25 mL of 2.5% NaOCl and 1.25 mL of 2% CHX in glass tubes. Different concentrations of AA ranging from 5-30% were prepared and the effect of these solutions on PCA solubility was determined by gravimetric method. In addition, a deep eutectic solvent system (DES) was obtained by mixing certain mole ratios of AA, glycerin and deionized water at 70 °C for 1 h and cooling to room temperature at the end of the time. The performance of DES on the solubility of PCA was evaluated using the same method.

Results: In control group prepared with a certain volume of physiological saline applied to PCA at constant volume at 25 °C, it was determined that PCA dissolved approximately 27%. When a certain volume of AA was applied to the PCA the same way, it was determined that this rate increased to approximately 55% with the increase in the concentration of AA. In addition, a solubility of over 80% was achieved in PCA to which a certain volume of DES was applied.

Conclusion: High concentrations of AA can be used as an irrigating solvent for the removal of PCA and it was also found that DES system has the potential to be a more effective solvent for PCA solubilization compared to AA.

Keywords: *Ascorbic acid, Precipitate, Chlorhexidine, Root canal irrigation, Para-chloroaniline, Sodium hypochlorite*

ÖZET

Amaç: Bu çalışmanın amacı, kök kanal tedavisi sırasında sodyum hipoklorit ve klorheksidin solüsyonlarının etkileşimi sonucu oluşan turuncu-kahverengi para kloroanilin çökeltisinin (PKA) uzaklaştırılmasında askorbik asitin (para-kloroailin) (AA) etkisini belirlemektir.

Gereç ve Yöntem: PKA, laboratuvar koşullarında cam tüplerde 1.25 mL %2.5'lik sodyum hipoklorit ve 1.25 mL %2'lik klorheksidin çözeltileri karıştırılarak elde edildi. %5-30 arasında değişen farklı konsantrasyonlarda AA çözeltileri hazırlandı ve bu çözeltilerin kök kanalındaki PKA çözünürlüğü üzerindeki etkisi gravimetrik yöntemle belirlendi. Ayrıca, belirli mol oranlarında AA, gliserin ve deiyonize suyun (AA: Gli: H_2O) 70 °C'de 1 saat karıştırılması ve sürenin sonunda oda sıcaklığına soğutulmasıyla derin ötektik bir çözücü sistemi (DÖÇ) elde edildi. DES'in PCA çözünürlüğü üzerindeki performansı aynı yöntem kullanılarak değerlendirildi.

Bulgular: 25 °C'de sabit hacimde sodyum hipoklorit ve klorheksidin ile hazırlanan PKA'ya belirli hacimde serum fizyolojik uygulanarak hazırlanan kontrol grubunda, PKA'nın yaklaşık %27 oranında çözündüğü belirlendi. Aynı şekilde hazırlanan PKA'ya belirli hacimde AA uygulandığında, AA konsantrasyonundaki artışla bu oranın yaklaşık %55'e çıktığı belirlendi. Ayrıca, belirli hacimde DÖÇ uygulanan PKA'da %80'in üzerinde bir çözünürlük elde edildi.

Sonuç: Kanal içerisinde oluşan PKA'nın uzaklaştırılmasında irrigasyon çözücüsü olarak yüksek konsantrasyonlu AA solüsyonunu kullanılabilir. Ayrıca, DÖÇ sisteminin AA'ya kıyasla PKA'nın çözündürülmesinde daha etkili bir çözücü olma potansiyeline sahip olduğu bulunmuştur.

Anahtar Kelimeler: *Askorbik asit, Çözelti, Klorheksidin, Kök kanal irrigasyonu, Para-kloroanilin, Sodyum hipoklorit*

[1] Research Assistant, Department of Endodontics, Faculty of Dentistry, Akdeniz University, Antalya, Türkiye.
[2] Research Assistant, Department of Chemistry, Faculty of Science , Akdeniz University, Antalya, Türkiye.
[3] Professor, Department of Endodontics, Faculty of Dentistry, Akdeniz University, Antalya, Türkiye.
***Corresponding Author:** Kürşat Er **e-mail:** qursater@hotmail.com **ORCID:** (0000-0002-0667-4909) Department of Endodontics, Faculty of Dentistry, Akdeniz University, İstanbul, Türkiye.

Introduction

The goal of root canal treatment (RCT) is to eliminate bacteria from infected root canals and prevent reinfection. Biomechanical cleaning and shaping of the root canal significantly reduces bacterial counts; however, due to the anatomical complexity of the root canal system, organic and inorganic debris and bacteria cannot be completely removed, and bacteria often persist in root canal systems. Therefore, various irrigants are used to help remove residual debris, necrotic tissue, microorganisms, and the smear layer. Sodium hypochlorite (NaOCl), chlorhexidine (CHX), citric acid, and phosphoric acid are some of these irrigants.[1-3]

NaOCl is the most frequently used irrigant in endodontics. Its concentration can range from 0.5% to 8.5%, with 2.5% being the most common.[4] It has both tissue-dissolving and antibacterial capabilities. While higher concentrations of NaOCl enhance its antimicrobial efficacy, they may also lead to irritation of periapical tissues.[5] CHX is a positively charged, antimicrobial irrigant. In higher concentrations, it demonstrates bactericidal effects, while at lower doses, it exhibits bacteriostatic effects.[6] Despite having similar antimicrobial properties to NaOCl with lower toxicity, CHX lacks tissue-dissolving properties.[7,8]

The combination of NaOCl and CHX has been proposed as an irrigation protocol to enhance antimicrobial properties during RCT. However, a problem with this combination is that CHX irrigation in the presence of NaOCl in the canal produces a dense orange-brown precipitate.[3,9] Clinically, this precipitate is difficult to remove from the canal walls and can cause esthetic tooth discoloration. Another concern is that this precipitate can form a chemical smear layer, obstruct dentinal tubules, and compromise the seal of root canal fillings, particularly those using resin-based sealer.[9] Furthermore, its cytotoxic and carcinogenic potential on human tissues is a major concern. To prevent or remove this precipitate, researchers have tested a range of intermediate irrigants and final rinses (including chelators (EDTA, citric acid), organic acids and phytic acid, alcohol (ethanol or isopropyl alcohol), saline/water rinses, and newer agents) with variable efficacy: chelators and alcohols generally perform better at dissolving or displacing the deposit than water alone.[9,10] Several in vitro studies[11,12] specifically report that brief intermediate irrigation with EDTA or citric acid, or a short flush with high-concentration alcohol, significantly reduces residual precipitate compared with no intermediate rinse. In addition, activation techniques such as passive ultrasonic irrigation or sonic/EDDY activation enhance the mechanical disruption and removal of the precipitate versus syringe irrigation alone, improving penetration of the irrigant into irregularities and increasing the cleanliness of canal walls.[13,14] Taken together, these studies linking these strategies to improved long-term outcomes remain limited. Therefore, the search for new solutions and techniques continues. The aim of this study was to determine the effect of ascorbic acid (AA) (C Vitamin) and AA-containing deep eutectic systems (DES) in removing orange-brown precipitate (para-chloroaniline) (PCA) formed by the interaction of NaOCl and CHX during RCT.

Materials and Methods

Chemical agents

AA, used to prepare the irrigant, was supplied from BDH (British Drug Houses, London, UK), and the glycerol required for the preparation of the deep eutectic solvent system was supplied from Riedel-de Haen (Seelze, Germany). The 2.5% NaOCl required to produce parachloroaniline (PCA) was supplied from Cerkamed Medical (Chloraxid, Stalowa Wola, Poland). The 2% CHX was supplied from Jefix Dental (Istanbul, Türkiye). The physiological saline solution used in the control group was supplied by Turkfleks (Ankara, Türkiye). Deionized water was used throughout the study.

Preparation of solutions

In this study, specific amounts of AA solid were weighed separately. These amounts were dissolved in deionized water to prepare 25 mL AA solutions of 5%, 10%, 15%, 20%, 25%, and 30% by weight/volume. To investigate the effects of these solutions on the solubility of PCA, 2.5 mL of NaOCl and 2.5 mL of CHX were added to 6 separate test tubes, and PCA formation was achieved. Then, 5 mL of AA solution at a specific concentration was added to each test tube. The tubes containing AA were vortexed for 10 sec to ensure thorough dispersion. Photographs of the test tubes were then taken. The same process was repeated with 5 mL of AA solutions at specific concentrations added to each test tube to determine whether the AA volume affected PCA solubility. The control group of this study was prepared by adding physiological serum instead of AA solution to PCA prepared with 2.5 mL of NaOCl and 2.5 mL of CHX in a separate test tube.

Test process

The study also investigated the effects of AA-based deep eutectic solvents (DES) on the solubility of PCA. For this purpose, AA, glycerol (Gly), and H_2O were added to separate capped glass vials at molar ratios of 1:4:10 (DES-1) and 1:10:10 (DES-2). The DES-1 system was stirred at 70 °C on a magnetic stirrer for 1 h, and the DES-2 system was stirred at 60 °C on a magnetic stirrer for 2 h. After allowing the mixture to cool to room temperature, both DESs were determined to have a homogeneous, transparent, and fluid structure.[15-17] To determine whether the prepared DESs could be used to dissolve PCA, 2.5 mL of DES-1 and DES-2 were added to PCA prepared with 1.25 mL of NaOCl and 1.25 mL of CHX in separate test tubes, and photographs were taken of the test tubes.

The amounts of PCA precipitated by mixing NaOCl and CHX and dissolved by adding various solutions were determined gravimetrically. For this purpose, eight 50 mL capped centrifuge tubes were weighed empty. 1.25 mL of NaOCl and 1.25 mL of CHX were added to each tube. Centrifugation was performed for 10 min at a rotation speed of 7500 rpm to precipitate the resulting PCA. After centrifugation, the liquid remaining on the precipitated PCA was removed by decantation, and the centrifuge tubes containing PCA were reweighed. The total amount of precipitated PCA was calculated from the difference between these weighing results and the weights of the empty tubes. Then, 5%, 10%, 15%, 20%, 25%, and 30% AA solutions were added to 6 tubes separately, DES-1 was added to one tube, and 2.5 mL of physiological serum was added to the control tube. The tubes were vortexed and ultrasonicated sequentially to completely disperse the precipitated PCA in the liquid portion. They were then centrifuged again at a rotation speed of 7500 rpm for 10 min. To determine the amount of PCA precipitated by centrifugation, the liquid in the tubes was removed by decantation and reweighed. The amount of PCA remaining undissolved in the centrifuge tube was determined from the difference between the final weighing values and the weights of the empty tubes. Thus, precipitated PCA, insoluble PCA, and % solubility values were calculated.

Results

Photographs showing the effect of AA concentration and applied AA volume on the solubility of PCA and including the control group are given in Figure 1, and photographic images of test tubes to which a total of 10 mL of AA solution at certain concentrations and physiological serum were added are given in Figure 2. An examination of Figures 1 and 2 reveals that PCA dissolves better with increasing the concentration of the AA solution added to the PCA formed with NaOCl and CHX. It appears that increasing the volume of AA applied does not significantly change the solubility of PCA, but diluting the medium facilitates visual monitoring of the remaining undissolved PCA. Figure 2 compared to the control group using physiological saline, clearly demonstrates that AA can be used as a highly effective irrigating solution for PCA, regardless of the applied concentration.

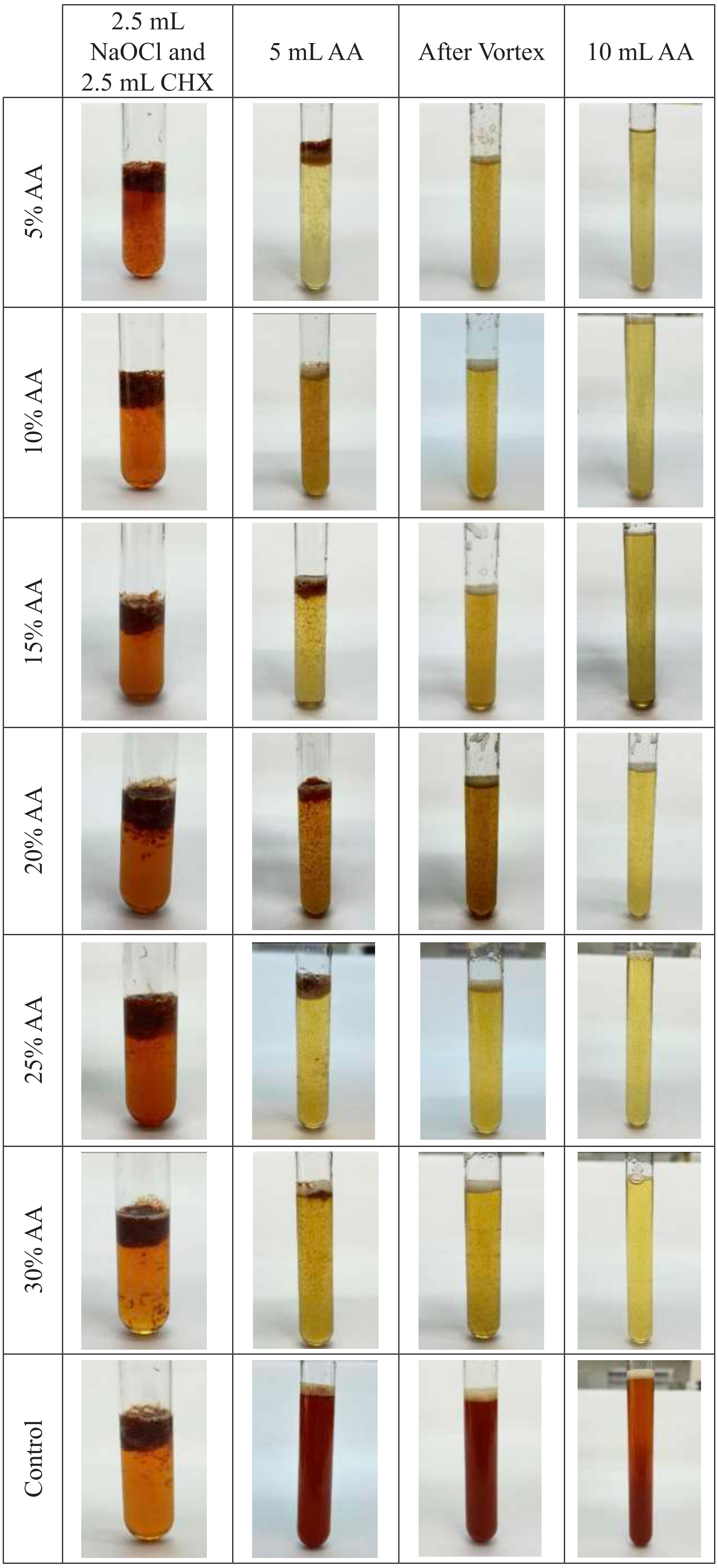

Figure 1. Photographs showing the effect of AA concentration and applied AA volume on the resolution of PCA and including the control group.

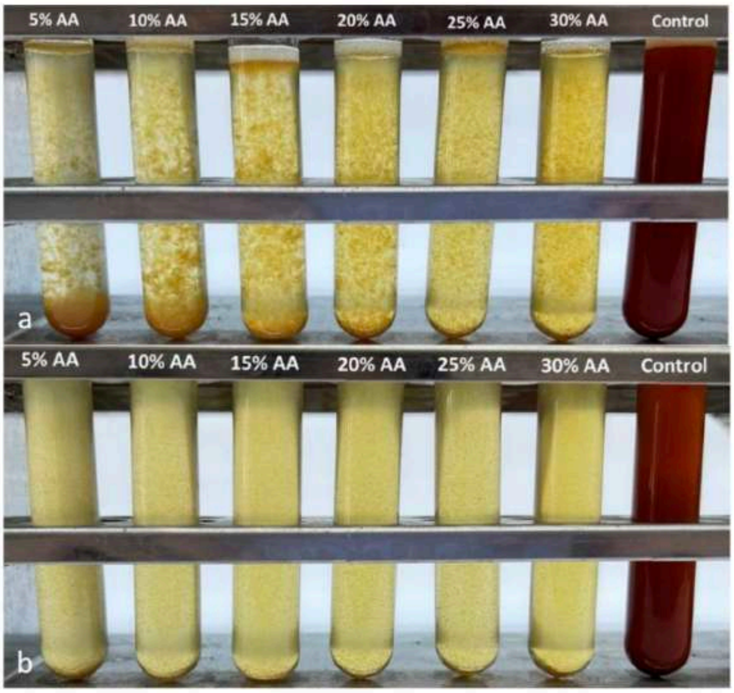

Figure 2. Photographs of test tubes to which 5 mL of AA (a) and 10 mL of AA (b) were added at specific concentrations.

The effects of deep eutectic solvents prepared using different ratios of AA, Gly, and H_2O on the dissolution of PCA were investigated. Photographs showing the effects of DES-1 and DES-2 on the solubility of PCA are given in Figure 3. The photographs in Figure 3 show that both DES-1 and DES-2 significantly dissolve PCA. It is particularly clear that DES-1 is the best dissolving PCA system among the DES systems, both compared to the AA solutions at different concentrations shown in Figure 2, and that it can be used as a highly effective irrigation solution for removing PCA formed in the canal.

Table 1. Precipitated PCA, insoluble PCA and % solubility values.

Solution	Precipitate PCA (g)	Insoluble PCA (g)	Solubility (%)
%5 AA	0.2247	0.1485	33.912
%10 AA	0.1919	0.1252	34.758
%15 AA	0.2746	0.1748	36.344
%20 AA	0.2081	0.1226	41.086
%25 AA	0.2150	0.1225	43.023
%30 AA	0.1939	0.0836	56.885
DES-1	0.1816	0.0339	81.333
Control	0.1828	0.1323	27.626

The amounts of dissolved PCA in the solvent systems applied to the precipitated PCA were determined gravimetrically. The calculated values for precipitated PCA, insoluble PCA, and % solubility are given in Table 1. When the values in Table 1 are examined, it is seen that the 27.626% solubility of PCA achieved by applying physiological saline solution increases from 33.912% to 56.885% with increasing concentrations of AA solution applied instead of physiological saline from 5% to 30%, and these numerical data confirm the results obtained in Figures 1 and 2. From the data presented in Table 1 it is clearly seen that a high solubility of 81.333% was achieved with the deep eutectic solvent system prepared with AA:Gly:H_2O at a 1:4:10 molar ratio, coded as DES-1. However, during the studies, it was determined that the homogeneous DES-1 lost its stability after remaining at room conditions for more than 6 h. This suggests that if the reaction conditions for DES-1 prepared with AA:Gly:H_2O are optimized, it has the potential to be used as an irrigation solution providing high dissolution for PCA.

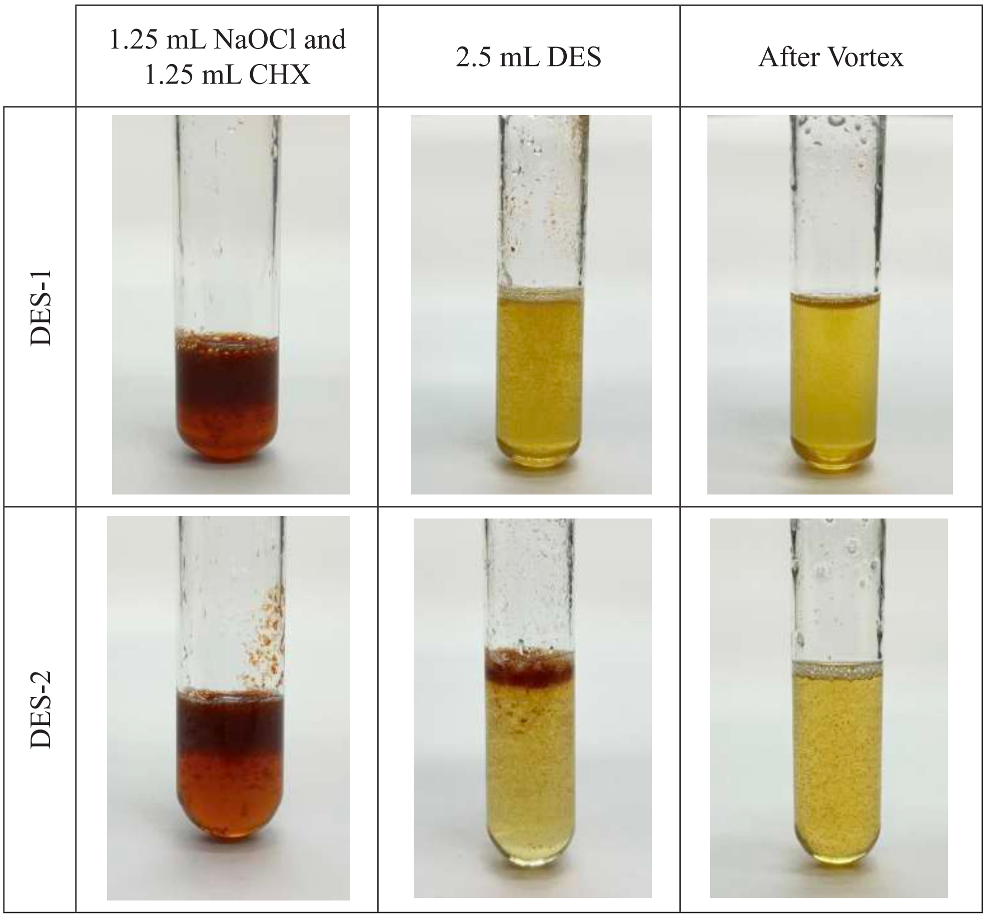

Figure 3. Photos showing the effect of DES-1 and DES-2 on the resolution of PCA.

Discussion

The interaction between NaOCl and CHX in the root canals is a well-documented clinical challenge in endodontics.[9] While both are highly effective irrigants, their combination results in the formation of an orange-brown PCA, which can obstruct dentinal tubules, compromise the seal of root canal fillings, and potentially pose cytotoxic risks.[9,18-23] The search for an effective and safe solvent to remove this precipitate is ongoing, as existing intermediate rinses like EDTA, QMix, citric acid, and alcohol show variable efficacy and additionally several activation techniques (e.g. EndoActivator, PIPS).[24-31] This study investigated the potential of AA and a DES as novel agents for PCA solubilization.

AA is a water-soluble antioxidant and an essential cofactor for collagen biosynthesis, catecholamine production and iron absorption; humans cannot synthesize it endogenously and must obtain it from the diet.[32] In the oral and dental context, AA's roles are twofold: systemically it supports connective-tissue formation and modulates inflammatory responses that are central to periodontal disease and wound healing, and locally it has been investigated as an adjunct to promote healing after tooth extraction and implant surgery.[32-35] Clinical and experimental studies suggest that perioperative or short-term oral supplementation with AA can improve extraction-socket healing and may accelerate soft-tissue repair in implant sites, likely through enhanced collagen deposition and antioxidant effects.[33,35] At the same time, formulations of AA (especially acidic tablets or powders) may pose an erosion risk to enamel if used improperly, so its dental application requires attention to dosage form and timing relative to oral hygiene.[36] Recent narrative and systematic reviews propose that AA is a promising adjunct in periodontal therapy and oral wound management, but they emphasize that high-quality, larger randomized clinical trials are still needed to define optimal dosing, delivery methods, and long-term outcomes in dental practice.[34] Recent researchs have highlighted the multifaceted role of AA in endodontic and restorative dentistry. Diederich et al.[37] demonstrated that AA functions as a growth and differentiation factor for dental stem cells, thereby supporting its potential application in regenerative endodontic therapies. Complementing these biological findings, Grazioli et al.[38] reported that antioxidant solutions, including AA, can counteract the adverse effects of NaOCl on dentin, significantly improving the bond strength of adhesive systems. Similarly, Albashaireh et al.[39] showed that conditioning solutions such as AA and QMix enhance the adhesion of glass fiber-reinforced composite posts to root dentin, underscoring the clinical relevance of antioxidant-mediated dentin conditioning.

A DES is a mixture of two or more substances (often a hydrogen-bond donor and a hydrogen-bond acceptor) that, when mixed in a precise molar ratio, produces a liquid with a substantially lower melting point than the separate components. They function similarly to ionic liquids, but at a lower cost and with less environmental impact. They are commonly utilized in medicines, chemical synthesis, extraction, nanoparticle production, and biomaterials. AA-containing DES is a DES system in which AA is a constituent of the eutectic mixture. It can behave as a hydrogen-bond donor, acceptor, or functional additive with antioxidant activity. These are beneficial because they have significant antioxidant capabilities, are biodegradable, are considered green solvents, and may stabilize reactive molecules (for example, metal ions and nanoparticles).[40] In this study, we investigated the effects of AA-based DES on the solubility of PCA. For this purpose, AA, glycerol (Gly), and H_2O were added to separate capped glass vials at molar ratios of 1:4:10 (DES-1) and 1:10:10 (DES-2).

The results demonstrate a clear concentration-dependent effect of AA on PCA solubility. While the control group, using physiological saline, achieved a solubility of approximately 27.6%, the application of AA solutions significantly increased this rate. Specifically, the solubility increased from 33.9% to 5% AA to a maximum of 56.9% at 30% AA concentration. This finding is visually supported by photographic evidence, which shows a marked reduction in the PCA as the AA concentration increases (Fig. 1 and 2). The observation that increasing the volume of AA did not significantly alter the solubility suggests that the chemical concentration, rather than the total volume, is the primary factor driving the dissolution process. This is critical finding for clinical application, as it suggests that a higher concentration of AA, even in a standard irrigation volume, would be more effective.

The use of DES represents a novel approach in this context. DES are often considered green solvents with tunable properties.[15-17] The AA-based DES system, specifically DES-1 (AA:Gly:H_2O at 1:4:10 molar ratio), demonstrated superior performance compared to all tested AA solutions. The gravimetric analysis showed that DES-1 achieved a high solubility of 81.3%, which is a substantial improvement over the 30% AA (56.9%). This high efficacy is also visually evident, with DES-1 showing the best dissolution among all tested systems (Fig. 3). This suggests that the unique solvent properties of the DES, which may include enhanced hydrogen bonding or altered polarity compared to a simple aqueous AA, are highly effective at breaking down the PCA. However, a significant limitation was identified regarding the stability of the DES-1 system, which lost its homogeneity after approximately 6 h at room temperature. This instability necessitates further research into optimizing the preparation and storage conditions of the DES-1 system to ensure its clinical viability. Future studies should focus on modifying the molar ratios or incorporating stabilizing agents to extend the shelf life of this highly effective solvent.

In summary, this study confirms that AA is an effective solvent for PCA, with its efficacy increasing with concentration. More importantly, the DES system based on AA shows a significantly higher potential for PCA removal, although its stability requires further optimization.

Conclusion

Based on the gravimetric and visual analysis, the following conclusions can be drawn:

1. AA is an effective solvent for the orange-brown PCA formed by the interaction of NaOCl and CHX. The efficacy of AA in dissolving PCA is directly proportional to its concentration.
2. The DES system prepared with AA, glycerol, and water (DES-1) is a significantly more effective solvent for PCA.
3. The DES-1 system has the potential to be a superior irrigating solvent for PCA removal compared to aqueous AA, but its stability at room temperature must be improved before clinical application.
4. High concentrations of AA (e.g., 30%) can be considered a viable and biocompatible irrigating solvent for the removal of PCA formed in the root canal.

Acknowledgements
None.

Ethical Approval
Ethical approval was not required for this study as it did not involve any interventions on human participants, the use of personal data, or animal experiments. The study was conducted in accordance with relevant regulations and ethical principles.

Sources of Funding
This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors

Conflict of Interest
The authors declare no conflicts of interest regarding the subject matter or materials discussed in this article.

Authorship Contributions
Idea/Concept: K.E Design: K.E, B.T, İ.Ö Control/Supervision: K.E Literature Review: B.T, İ.Ö Materials B.T, İ.Ö, C.Ö.D Data Collection and/or Processing: B.T, İ.Ö, C.Ö.D Analysis and/or Interpretation: K.E Writing the Article: B.T, İ.Ö, C.Ö.D Critical Review: K.E

References

1. Erkal D, Çakmak YE, Pistol AM, Er K. Pulp tissue dissolution capacities of two hypochlorite-based irrigants at different temperatures: an in vitro study. *BMC Oral Health.* 2025;25(1):1104.
2. Tekinarslan D, Er K, Eğin M, Dinçer T, Kiliç AO. The effect of various irrigants on mixed biofilms in dentinal tubules: a confocal laser scanning microscopy study. *Microsc Res Tech.* 2025;88(9):2586-93.
3. Er K, Koç S, Erkal D, Tekinarslan D, Kesmez Ö, Demir F, Borbely E. Effect of precipitate formation in the root canal on the accuracy of electronic apex locators. *J Oral Sci.* 2025;67(4):161-6.
4. Cullen JK, Wealleans JA, Kirkpatrick TC, Yaccino JM. The effect of 8.25% sodium hypochlorite on dental pulp dissolution and dentin flexural strength and modulus. *J Endod.* 2015;41:920-4.
5. Gernhardt CR, Eppendorf K, Kozlowski A, Brandt M. Toxicity of concentrated sodium hypochlorite used as an endodontic irrigant. *Int Endod J.* 2004;37:272-80.
6. Agrawal VS, Rajesh M, Sonali K, Mukesh P. A contemporary overview of endodontic irrigants: a review. *J Dent App.* 2014;1:105-15.
7. Rôças IN, Siqueira JF Jr. Comparison of the in vivo antimicrobial effectiveness of sodium hypochlorite and chlorhexidine used as root canal irrigants: a molecular microbiology study. *J Endod.* 2011;37:143-50.
8. Klein U, Kleier DJ. Sodium hypochlorite accident in a pediatric patient. *Pediatr Dent.* 2013;35:534-8.
9. Drews D-J, Nguyen AD, Diederich A, Gernhardt CR. The interaction of two widely used endodontic irrigants, chlorhexidine and sodium hypochlorite, and its impact on the disinfection protocol during root canal treatment. *Antibiotics (Basel).* 2023;12(3):589.
10. Kim J-W. Precipitate from a combination of sodium hypochlorite and chlorhexidine. *Restor Dent Endod.* 2012;37(3):185-6.
11. Keles A, Ors SA, Yilmaz Z. Effect of various solutions on the removal of orange-brown precipitate formed by interaction of sodium hypochlorite and chlorhexidine with or without ultrasonic activation. *Niger J Clin Pract.* 2020;23:381-5.
12. Krishnamurthy S, Sudhakaran S. Evaluation and prevention of the precipitate formed on interaction between sodium hypochlorite and chlorhexidine. *J Endod.* 2010;36(7):1154-7.
13. Arslan H, Gok T, Saygili G, Altintop H, Akçay M, Çapar ID. Evaluation of effectiveness of various irrigating solutions on removal of calcium hydroxide mixed with 2% chlorhexidine gel and detection of orange-brown precipitate after removal. *J Endod.* 2014;40(11):1820-3.
14. İnce Yusufoğlu S, Keskin NB. Efficacy of EDDY, passive ultrasonic irrigation and manual irrigation on the removal of orange-brown precipitate. *Int Dent Res.* 2021;11(1):1-5.
15. Koigerova A, Gosteva A, Samarov A, Tsvetov N. Deep eutectic solvents based on carboxylic acids and glycerol or propylene glycol as green media for extraction of bioactive substances from Chamaenerion angustifolium (L.) Scop. *Molecules.* 2023;28:6978.
16. Rasool MH, Ahmad M, Ayoub M, Abbas MA. A novel ascorbic acid based natural deep eutectic solvent as a drilling mud additive for shale stabilization. *Processes.* 2023;11:1135.
17. Kutlu N, Yılmaz MS, İşci A, Şakıyan Ö. Utilization of deep eutectic solvents in the extraction of bioactive compounds from food materials. *Niğde Ömer Halisdemir Univ. J Engineer Sci.* 2021;10(2):591-7.
18. Jeong JW, Sarmast ND, Terlier T, van der Hoeven R, Holland JN, Parikh N. Assessment of the cytotoxic effects and chemical composition of the insoluble precipitate formed from sodium hypochlorite and chlorhexidine gluconate. *Int Endod J.* 2021;54(10):1892-1901.
19. Basrani BR, Manek S, Sodhi RN, Fillery E, Manzur A. Interaction between sodium hypochlorite and chlorhexidine gluconate. *J Endod.* 2007;33(8):966-9.
20. Akisue E, Tomita VS, Gavini G, Poli de Figueiredo JA. Effect of the combination of sodium hypochlorite and chlorhexidine on dentinal permeability and scanning electron microscopy precipitate observation. *J Endod.* 2010;36(5):847-50.
21. Abusteit OE. Evaluation of resin sealer penetration of dentin following different final rinses for endodontic irrigation using confocal laser scanning microscopy. *Aust Endod J.* 2021;47(2):195-201.
22. Patil P, Aminoshariae A, Harding J, Montagnese TA, Mickel A. Determination of mutagenicity of

the precipitate formed by sodium hypochlorite and chlorhexidine using the ames test. *Aust Endod J.* 2016;42(1):16-21.

23. Homayouni H, Majd NM, Zohrehei H, Mosavari B, Adel M, Dajmar R, Homayouni A. The effect of root canal irrigation with combination of sodium hypochlorite and chlorhexidine gluconate on the sealing ability of obturation materials. *Open Dent J.* 2014;8:184-7.

24. Guneser MB, Dincer AN, Arslan D. Comparison of conventional syringe, canalbrush, EndoActivator, photon-induced photoacoustic streaming, and manual instrumentation in removing orange-brown precipitate: an in vitro study. *Photomed Laser Surg.* 2017;35(6):311-6.

25. Bueso V, Parikh N, Terlier T, Holland JN, Sarmast ND, Jeong JW. Comparative evaluation of intermediate solutions in prevention of brown precipitate formed from sodium hypochlorite and chlorhexidine gluconate. *Clin Exp Dent Res* 2022; 8(6): 1591-7.

26. Alberto APL, Oliveira DDS, Oliveira HE, Maciel ACC, Belladonna FG, Silva EJNLD. Does sodium thiosulphate avoid the formation of the brown-coloured precipitate as an intermediate irrigant between NaOCl and chlorhexidine? *Aust Endod J.* 2022;48(1):72-76.

27. Metri M, Hegde S, Dinesh K, Indiresha HN, Nagaraj S, Bhandi SH. Comparative evaluation of two final irrigation techniques for the removal of precipitate formed by the interaction between sodium hypochlorite and chlorhexidine. *J Contemp Dent Pract.* 2015;16(11):850-3.

28. Boppré LM, Menezes Savaris J, Catherine Maiola E, Peressoni Vieira-Schuldt D, da Fonseca Roberti Garcia L, da Silveira Teixeira C, Antunes Bortoluzzi E. Can heated distilled water effectively prevent precipitate formation between NaOCl and CHX? *Int J Dent.* 2024; 2024:6612675.

29. Maiola EC, Boppré LM, Savaris JM, Dias Junior LCL, Garcia LDFR, Teixeira CDS, Bortoluzzi EA. Did in-between rinsing and agitating with distilled water prevents precipitate formation by the interaction between sodium hypochlorite and chlorhexidine canal irrigants? *Microsc Res Tech.* 2024;87(2):315-25.

30. Arslan H, Uygun AD, Keskin A, Karatas E, Seçkin F, Yıldırım A. Evaluation of orange-brown precipitate formed in root canals after irrigation with chlorhexidine and QMix and spectroscopic analysis of precipitates produced by a mixture of chlorhexidine/NaOCl and QMix/NaOCl. *Int Endod J.* 2015;48(12):1199-203.

31. Magro MG, Kuga MC, Aranda-Garcia AJ, Victorino KR, Chávez-Andrade GM, Faria G, Keine KC, Só MV. Effectiveness of several solutions to prevent the formation of precipitate due to the interaction between sodium hypochlorite and chlorhexidine and its effect on bond strength of an epoxy-based sealer. *Int Endod J.* 2015;48(5):478-83.

32. Murererehe J, Uwitonze AM, Nikuze P, Patel J, Razzaque MS. Beneficial effects of Vitamin C in maintaining optimal oral health. *Front Nutr.* 2022;8:805809.

33. Li X, Tang L, Lin YF, Xie GF. Role of vitamin C in wound healing after dental implant surgery in patients treated with bone grafts and patients with chronic periodontitis. *Clin Implant Dent Relat Res.* 2018;20(5):793-8.

34. Ruzijevaite G, Acaite E, Jagelaviciene E. Therapeutic impact of ascorbic acid on oral and periodontal tissues: a systematic literature review. *Medicina.* 2024;60(12):2041.

35. Pisalsitsakul N, Pinnoi C, Sutanthavibul N, Kamolratanakul P. Taking 200 mg vitamin C three times per day improved extraction socket wound healing parameters: a randomized clinical trial. *Int J Dent.* 2022;2022:6437200.

36. Bhasin S, Singh S, Thomas MS, Mahabala KY, Shenoy R. Evaluation of the impact of acidic medications and fluoride-containing mouthwash on the enamel surface using quantitative light-induced fluorescence, microhardness, and scanning electron microscopy: an in vitro study. *BMC Oral Health.* 2025;25(1):141.

37. Diederich A, Fründ HJ, Trojanowicz B, Santos AN, Nguyen AD, Hoang-Vu C, Gernhardt CR. Influence of ascorbic acid as a growth and differentiation factor on dental stem cells used in regenerative endodontic therapies. *J Clin Med.* 2023;12(3):1196.

38. Grazioli G, de León Cáceres E, Tessore R, Lund RG, Monjarás-Ávila AJ, Lukomska-Szymanska M, Hardan L, Bourgi R, Cuevas-Suárez CE. In vitro bond strength of dentin treated with sodium hypochlorite: effects of antioxidant solutions. *Antioxidants (Basel).* 2024;13(9):1116.

39. Albashaireh ZS, Taha NA, Albashaireh KZ. The effect of ascorbic acid, QMix and other conditioning solutions on the bond strength of adhesively luted glass fiber-reinforced composite posts to root dentin, a laboratory study. *J Dent.* 2024;149:105286.

40. Song J-E, Jun S-H, Ryoo J-Y, Kang N-G. Formulation of ascorbic acid and betaine-based therapeutic deep eutectic system for enhanced transdermal delivery of ascorbic acid. *Pharmaceutics*. 2024;16(5):687.