

Review Article

Oxidative Adjuncts in Endodontic Disinfection: A Narrative Review of Ozone and Photodynamic Therapy Against *Enterococcus faecalis* Biofilms

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Abstract

Background and Aim: Persistent endodontic infections remain a major clinical challenge due to the resilience of biofilm-forming microorganisms, particularly *Enterococcus faecalis*, which can survive conventional chemomechanical disinfection and persist within the complex anatomy of the root canal system. This narrative review provides a mechanism-based comparison of two oxidative adjunctive strategies—ozone therapy and photodynamic therapy (PDT)—focusing on their antimicrobial effects against *E. faecalis* biofilms. Both modalities act through oxidative pathways that induce damage to microbial cells independent of bacterial metabolic activity, making them theoretically suitable for targeting biofilm-associated infections. Evidence indicates that PDT demonstrates more consistent antimicrobial efficacy, attributed to its controlled generation of reactive oxygen species and localized activation within biofilm structures. In contrast, ozone therapy exhibits variable antimicrobial performance, primarily limited by its instability, rapid decomposition, and restricted penetration into mature biofilms. Despite their oxidative potential, neither modality reliably achieves complete eradication of *E. faecalis* when used as standalone treatments, and both remain less effective than sodium hypochlorite irrigation in conventional protocols. Current findings support their role as adjunctive rather than primary disinfection strategies. This review highlights the need for standardized clinical protocols and well-designed in vivo studies to better define their translational value and optimize their integration into multimodal endodontic disinfection approaches.

Keywords: *Enterococcus faecalis*, Biofilm, Endodontics, Ozone therapy, Photodynamic therapy, Oxidative disinfection, Narrative review

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Introduction

Persistent apical periodontitis continues to challenge contemporary endodontic practice, not due to a lack of antimicrobial agents, but because of the remarkable adaptability of microbial biofilms within the root canal system¹⁻³. These structured microbial communities are not merely clusters of bacteria but highly organized, dynamic ecosystems capable of resisting chemical, mechanical, and environmental stressors. As a result, complete eradication of infection remains elusive, particularly in anatomically complex regions such as dentinal tubules, lateral canals, and apical ramifications.

Among the diverse endodontic pathogens, *Enterococcus faecalis* has emerged as a key contributor to post-treatment disease and retreatment failure^{4,5}. Its clinical relevance lies not only in its prevalence but in its biological resilience: the ability to survive prolonged nutrient deprivation, tolerate high alkaline environments, invade dentinal tubules, and persist within mature biofilms. Importantly, the biofilm mode of growth fundamentally alters bacterial behavior, conferring a level of antimicrobial tolerance that cannot be predicted from planktonic models alone. This discrepancy highlights a critical gap between laboratory efficacy and clinical reality.

Conventional chemomechanical preparation, centered on instrumentation and irrigants such as sodium hypochlorite, remains the foundation of root canal disinfection. While highly effective under ideal conditions, its antimicrobial performance is inherently limited by restricted penetration into biofilm-protected niches and the intricate architecture of the root canal system. Increasing concentrations or exposure times may enhance antimicrobial activity but at the cost of cytotoxicity and potential damage to periapical tissues. Consequently, the focus of modern endodontic research has shifted from simply increasing antimicrobial strength to enhancing biofilm disruption and improving targeted microbial inactivation.

Within this evolving paradigm, adjunctive disinfection strategies such as ozone therapy and photodynamic therapy (PDT) have gained attention as non-conventional approaches that rely on oxidative mechanisms rather than direct chemical

toxicity⁶⁻¹². Ozone therapy exerts its antimicrobial effect through the rapid oxidation of cellular components, while PDT achieves microbial killing via the localized generation of reactive oxygen species following light activation of a photosensitizer. Although both modalities share a common oxidative basis, their interaction with biofilm architecture, depth of penetration, and dependence on application parameters differ substantially.

Despite a growing body of literature, current evidence remains fragmented and, at times, contradictory. Most studies evaluate antimicrobial outcomes in isolation, often under simplified experimental conditions that do not adequately reflect the structural and physiological complexity of endodontic biofilms. Moreover, direct comparisons between ozone therapy and PDT are limited, and their relative performance is rarely interpreted within a unified mechanistic framework. As a result, their true clinical value remains difficult to define.

This narrative review aims to move beyond a descriptive comparison and instead provide a mechanism-driven and biofilm-oriented synthesis of the antimicrobial effects of ozone therapy and PDT against *E. faecalis*. By integrating microbiological behavior, physicochemical properties of the treatment modalities, and clinically relevant considerations, this review seeks to clarify not only whether these approaches are effective but also how, when, and to what extent they can meaningfully enhance root canal disinfection.

Biofilm Biology of *Enterococcus faecalis*: Why Conventional Disinfection Fails: Persistent endodontic infection is not solely a consequence of microbial presence, but rather of microbial organization. Within the root canal system, bacteria exist predominantly as biofilms—structured, surface-adherent communities embedded in a self-produced extracellular polymeric matrix. This mode of growth fundamentally alters microbial behavior, transforming otherwise susceptible organisms into highly resistant populations. In this context, *E. faecalis* exemplifies a pathogen whose clinical persistence is closely linked to its biofilm-forming capacity (Table 1)¹³⁻¹⁵. A defining feature of *E. faecalis* biofilms is their structural and functional heterogeneity¹⁶. Cells within the biofilm are not uniform; instead, they exhibit

Table 1. Key biological factors contributing to *E. faecalis* biofilm resistance in root canal systems.

Biofilm Feature	Description	Mechanism of Resistance	Impact on Root Canal Disinfection
Extracellular Polymeric Substance (EPS) Matrix	Complex matrix of polysaccharides, proteins, lipoteichoic acids, and extracellular DNA	Acts as a physical and chemical barrier limiting diffusion of antimicrobial agents	Reduces penetration and effectiveness of irrigants such as sodium hypochlorite
Metabolic Heterogeneity	Gradients of nutrient and oxygen availability within biofilm layers	Presence of slow-growing or dormant cells with reduced metabolic activity	Decreases susceptibility to antimicrobials targeting active cellular processes
Persister Cells	Subpopulation of dormant, highly tolerant bacterial cells	Survive antimicrobial exposure without genetic resistance	Serve as a reservoir for recolonization after treatment
Deep Dentinal Tubule Penetration	Ability of <i>E. faecalis</i> to invade dentinal tubules (up to hundreds of micrometers)	Physical protection from mechanical instrumentation and irrigants	Limits access of disinfecting agents to infected areas
Biofilm Structural Organization	Multilayered, spatially structured microbial community	Creates microenvironments with variable pH, oxygen, and antimicrobial exposure	Promotes survival of bacteria in protected niches
Alkaline pH Tolerance	Ability to survive high pH environments (e.g., calcium hydroxide)	Maintains cellular homeostasis under stress conditions	Reduces effectiveness of intracanal medicaments
Virulence Factors	Expression of aggregation substance, enterococcal surface protein (Esp), gelatinase	Enhances adhesion, biofilm formation, and resistance to host defenses	Facilitates persistence and reinfection
Quorum Sensing and Cell Communication	Cell-to-cell signaling within biofilm	Coordinates gene expression related to survival and resistance	Promotes adaptive responses to antimicrobial stress
Limited Fluid Dynamics in Root Canal System	Stagnant or low-flow conditions in complex anatomy	Reduces distribution of irrigants and antimicrobial agents	Leads to incomplete disinfection of lateral canals and apical regions
Interaction with Root Canal Anatomy	Colonization of isthmuses, lateral canals, and apical deltas	Physical shielding from instrumentation and irrigation	Allows persistence and reinfection after treatment

gradients of metabolic activity, oxygen availability, and nutrient access. Bacteria located in deeper layers enter a slow-growing or dormant state, rendering them significantly less susceptible to antimicrobial agents that primarily target active cellular processes. This metabolic stratification creates a reservoir of “persister” cells capable of surviving even aggressive chemomechanical disinfection.

In addition to physiological adaptation, the extracellular polymeric substance (EPS) matrix plays a critical protective role. Composed of polysaccharides, proteins, lipoteichoic acids, and extracellular DNA, the EPS matrix acts as a physical and chemical barrier that limits the diffusion of antimicrobial agents. Irritants such as sodium hypochlorite may be neutralized or diluted before reaching deeper biofilm layers, resulting in incomplete bacterial elimination. This diffusion limitation is particularly relevant in dentinal tubules, where biofilms can extend hundreds of micrometers

from the canal lumen.

E. faecalis further enhances its survival through specific virulence factors, including aggregation substance, enterococcal surface protein (Esp), and gelatinase, which facilitate adhesion, biofilm maturation, and resistance to host defenses¹⁷. Its ability to withstand alkaline environments—such as those created by calcium hydroxide dressings—distinguishes it from many other endodontic pathogens and contributes to its persistence in previously treated canals.

Another critical aspect is the interaction between biofilm and root canal microanatomy. The complexity of the canal system, including isthmuses, apical deltas, and lateral canals, creates protected niches that limit access for mechanical instrumentation. Within these regions, biofilms remain largely undisturbed, allowing recolonization of the main canal space after treatment. This spatial limitation underscores the inadequacy of approaches that rely solely on direct chemical contact.

Importantly, most antimicrobial evaluations are performed on planktonic bacteria or immature biofilms, which do not accurately replicate clinical conditions. Mature *E. faecalis* biofilms demonstrate significantly higher tolerance, leading to an overestimation of disinfection efficacy in laboratory studies. This discrepancy represents a major barrier to translating experimental findings into predictable clinical success.

Collectively, these factors highlight a central challenge in endodontic disinfection: failure is not due to insufficient antimicrobial potency alone, but to the inability of current strategies to effectively penetrate, disrupt, and inactivate complex biofilm systems. This realization has driven the exploration of adjunctive approaches, particularly those based on oxidative mechanisms, which may offer advantages in targeting biofilm structure and function beyond the scope of conventional chemical action.

PDT in Root Canal Disinfection: Mechanisms and Evidence: PDT has emerged as a promising adjunctive approach in endodontic disinfection, primarily due to its ability to generate localized oxidative stress in a controlled and targeted manner^{18,19}. Its antimicrobial action relies on the interaction among a photosensitizing agent, a specific wavelength of light, and molecular oxygen, which produces reactive oxygen species (ROS), including singlet oxygen and free radicals. These highly reactive molecules induce irreversible damage to microbial cellular components such as membrane lipids, proteins, and nucleic acids, ultimately leading to cell death²⁰⁻²².

From a microbiological perspective, PDT offers a distinct advantage in the context of biofilm-associated infections. Unlike conventional antimicrobial agents that rely on metabolic activity or specific biochemical targets, PDT exerts a non-specific oxidative effect, making it effective against both actively dividing bacteria and dormant persister cells within mature biofilms. This property is particularly relevant for *Enterococcus faecalis*, which is known for its ability to survive in nutrient-deprived conditions and maintain viability within deeply embedded biofilm layers.

Experimental studies have consistently demonstrated that PDT can significantly reduce *E. faecalis* counts

in infected root canal systems. The degree of bacterial reduction, however, is influenced by multiple variables, including the type and concentration of the photosensitizer (e.g., methylene blue or toluidine blue O), the wavelength and power of the light source, irradiation time, and the accessibility of the biofilm. In general, better antimicrobial outcomes are observed when PDT is used in combination with conventional irrigants, suggesting a synergistic interaction between chemical debridement and photoactivated oxidation.

Despite these promising findings, PDT is not without limitations. One of the primary challenges is limited light penetration into complex root canal anatomy and deeper dentinal tubules, which may restrict its effectiveness in clinically relevant infection sites. Additionally, the uneven distribution of photosensitizers within the biofilm matrix can lead to suboptimal ROS generation in certain regions, resulting in incomplete microbial inactivation. These factors contribute to variability in reported outcomes across different studies.

Nevertheless, compared to other adjunctive modalities, PDT has demonstrated relatively more consistent antimicrobial performance against *E. faecalis*, particularly in in vitro biofilm models²³⁻²⁵. Some studies have reported substantial reductions in bacterial viability, reaching levels significantly lower than those achieved with conventional irrigation alone, although complete eradication remains rare. Importantly, its safety profile is favorable, as it selectively targets microbial cells while minimizing damage to surrounding host tissues.

Another key advantage of PDT is its lack of microbial resistance development. Since the mechanism of action involves physical and chemical destruction rather than receptor-mediated targeting or metabolic inhibition, the likelihood of bacteria developing adaptive resistance is minimal. This feature positions PDT as a valuable adjunct in the long-term management of persistent endodontic infections.

In summary, PDT represents a biologically targeted, oxidative disinfection strategy that addresses several limitations of conventional endodontic irrigants. However, its clinical efficacy remains dependent on protocol optimization and anatomical accessibility, and it should therefore be considered as a complementary rather than standalone disinfection

approach.

Ozone Therapy in Root Canal Disinfection

Evidence and Clinical Constraints: Ozone therapy has been investigated as an adjunctive disinfection modality in endodontics due to its strong oxidative potential and broad-spectrum antimicrobial activity²⁶⁻²⁸. Ozone (O₃), a triatomic oxygen molecule, exerts its biological effects through the oxidation of cellular components, including phospholipids, lipoproteins, and intracellular enzymes. This oxidative disruption leads to increased membrane permeability, loss of cellular integrity, and eventual microbial cell death. Unlike conventional irrigants that rely on specific biochemical interactions, ozone acts via a non-selective oxidative mechanism, which theoretically enables activity against a broad range of microorganisms, including *Enterococcus faecalis*.

In the context of endodontic infections, ozone has been evaluated in different forms, including gaseous ozone, ozonated water, and ozonated oils. Among these, aqueous ozone is often considered more clinically applicable due to improved handling and reduced toxicity. Experimental studies have demonstrated that ozone can significantly reduce bacterial loads in infected root canals; however, its efficacy against mature *E. faecalis* biofilms remains variable and generally less predictable than that of conventional sodium hypochlorite irrigation.

A key limitation of ozone therapy lies in its high instability and rapid decomposition. Ozone has a very short half-life and decomposes quickly into oxygen, which significantly limits its sustained antimicrobial activity within the root canal environment. This transient nature reduces its ability to maintain effective concentrations over time, particularly in anatomically complex regions where diffusion is restricted. As a result, its penetration into deeper layers of mature biofilms and dentinal tubules is often insufficient to achieve complete microbial eradication.

Another important constraint is the limited interaction with biofilm architecture. While ozone can oxidize exposed bacterial cells, its ability to disrupt the EPS matrix is less pronounced compared to other oxidative or mechanical adjuncts. The EPS matrix acts as a diffusion barrier, further reducing

ozone accessibility to embedded bacterial communities. Consequently, bacterial survival within protected biofilm niches may persist even after ozone exposure.

Clinical and laboratory studies have reported heterogeneous outcomes, with some demonstrating moderate antimicrobial effects and others showing minimal additional benefit when ozone is used alongside conventional irrigation protocols. This variability is largely attributed to differences in ozone concentration, exposure time, delivery systems, and study design. Importantly, in most comparative studies, ozone therapy has not consistently demonstrated superiority over sodium hypochlorite, nor has it shown clear advantages over other adjunctive modalities in reducing *E. faecalis* biofilm viability.

Despite these limitations, ozone therapy offers certain advantages, particularly its biocompatibility and minimal residual toxicity. Unlike strong chemical irrigants, ozone decomposes into oxygen without leaving harmful residues, making it a relatively safe adjunct in endodontic procedures. This safety profile has contributed to continued interest in its clinical application, particularly in cases where reduced cytotoxicity is desired.

Overall, ozone therapy represents an oxidative antimicrobial strategy with theoretical advantages but practical limitations. Its effectiveness is constrained by instability, limited biofilm penetration, and inconsistent clinical outcomes. As a result, its role in modern endodontics is best understood as a supplementary measure rather than a primary disinfection strategy, particularly when compared to more consistently effective modalities such as PDT.

Direct Comparative Analysis

Ozone Therapy versus PDT in *E. faecalis* Biofilm

Disinfection: Although both ozone therapy and PDT are based on oxidative antimicrobial mechanisms, their interaction with *E. faecalis* biofilms differs substantially in terms of consistency, depth of action, and reproducibility of outcomes (Table 2)^{26,29}. A direct comparison of the available evidence reveals that these differences are not merely methodological but are rooted in fundamental physicochemical and biological properties of each modality.

Table 2. Mechanistic and clinical comparison of ozone therapy and photodynamic therapy in endodontic disinfection.

Parameter	Ozone Therapy	Photodynamic Therapy (PDT)
Primary mechanism of action	Direct oxidation via ozone (O ₃)	Photoactivated generation of reactive oxygen species (ROS)
Key reactive species	Ozone, secondary oxygen radicals	Singlet oxygen (¹ O ₂), free radicals
Dependence on bacterial metabolism	No	No
Target of action	Cell membrane lipids, proteins, enzymes	Cell membranes, proteins, nucleic acids
Effect on biofilm structure (EPS)	Limited disruption	Partial disruption; may enhance permeability
Activity against persister cells	Moderate	Moderate to high
Penetration into biofilm/dentinal tubules	Limited (due to rapid decomposition)	Moderate (depends on photosensitizer diffusion + light penetration)
Spatial control of antimicrobial action	Low (diffuse effect)	High (localized activation by light)
Stability and duration of action	Very low (short half-life)	Controlled (dependent on irradiation time)
Forms of application	Gas, ozonated water, ozonated oils	Photosensitizer + laser/LED light
Protocol sensitivity	Moderate	High (photosensitizer, wavelength, time)
Consistency of antimicrobial outcomes	Variable	More consistent
Effectiveness vs <i>E. faecalis</i> biofilm	Moderate	Moderate to high
Effectiveness as standalone therapy	Limited	Limited
Adjunctive potential with NaOCl	Moderate	High
Comparison with sodium hypochlorite	Inferior	Inferior (alone), enhanced when combined
Cytotoxicity	Low	Low
Residual effects	None (decomposes to O ₂)	Minimal
Risk of microbial resistance	Very low	Very low
Main limitations	Instability, poor penetration, inconsistent outcomes	Light penetration limits, protocol variability
Clinical applicability	Limited adjunct	Promising adjunct (especially in retreatment cases)

From an antimicrobial efficacy perspective, both approaches demonstrate measurable reductions in *E. faecalis* viability; however, PDT generally exhibits more consistent and reproducible biofilm reduction across studies. This consistency is largely attributed to its controlled activation mechanism, in which reactive oxygen species are generated in situ following light activation of a photosensitizer. This allows for localized and sustained oxidative stress within the biofilm microenvironment, improving the likelihood of targeting both superficial and deeper bacterial layers when adequate photosensitizer penetration is achieved.

In contrast, ozone therapy demonstrates a more variable antimicrobial profile, with outcomes strongly dependent on delivery method, concentration, and exposure time. Its rapid decomposition limits sustained oxidative activity,

resulting in a narrower therapeutic window. Consequently, while ozone may effectively reduce planktonic bacterial populations or superficial biofilm layers, its ability to achieve uniform disruption of mature, structured biofilms is less predictable.

When considering biofilm penetration and structural disruption, PDT again demonstrates a relative advantage. The generation of reactive oxygen species occurs in close proximity to microbial cells that have absorbed the photosensitizer, allowing for targeted oxidative damage within defined regions of the biofilm. Furthermore, there is evidence that PDT may contribute to partial disruption of the EPS matrix, thereby enhancing access to deeper bacterial populations. Ozone, although a strong oxidant, lacks this level of spatial targeting and may be neutralized before fully interacting with deeper biofilm layers due to diffusion barriers and organic load within the canal system.

Another important distinction lies in clinical reproducibility. PDT protocols, although varied, are generally more standardized in terms of light wavelength, exposure duration, and photosensitizer usage. This relative standardization contributes to more consistent inter-study outcomes. Conversely, ozone therapy protocols exhibit greater heterogeneity, particularly regarding concentration, delivery systems (gas, aqueous, oil), and exposure conditions, which contributes to variability in reported antimicrobial efficacy.

Despite these differences, it is important to emphasize that neither modality achieves predictable or complete eradication of *E. faecalis* biofilms when used as a standalone treatment. Both ozone therapy and PDT demonstrate significantly improved outcomes when used as adjuncts to conventional chemomechanical preparation, particularly sodium hypochlorite irrigation. This suggests that their primary clinical value lies in enhancing disinfection rather than replacing standard protocols.

Importantly, the apparent superiority of PDT over ozone therapy should not be interpreted as a definitive clinical hierarchy but rather as a reflection of current experimental evidence under specific laboratory conditions. Variations in biofilm models, dentinal substrate, and methodological parameters continue to limit direct comparability across studies. Therefore, conclusions regarding relative efficacy must be interpreted cautiously.

In summary, while both ozone therapy and PDT offer oxidative strategies for targeting *E. faecalis* biofilms, PDT demonstrates greater consistency, improved spatial control of antimicrobial activity, and more reproducible experimental outcomes. Ozone therapy, although biocompatible and conceptually appealing, is constrained by instability and limited biofilm penetration. Collectively, the evidence supports the role of both modalities as adjunctive tools within a multimodal disinfection strategy, with PDT currently showing more favorable characteristics for clinical translation.

Clinical Implications: The management of persistent endodontic infections requires a shift from a purely antimicrobial paradigm toward a biofilm-centered clinical strategy. The available evidence suggests that neither ozone therapy nor PDT should

be considered as standalone disinfecting agents capable of reliably eliminating *E. faecalis* biofilms. Instead, their clinical value lies in their ability to function as adjunctive modalities that enhance the effectiveness of conventional chemomechanical preparation.

In clinical practice, sodium hypochlorite irrigation remains the cornerstone of root canal disinfection due to its well-established tissue dissolution and broad-spectrum antimicrobial properties³⁰⁻³². However, its limitations in penetrating complex anatomical spaces and disrupting mature biofilms justify the integration of supplementary approaches. Within this context, PDT appears to offer a more predictable adjunctive benefit, particularly in cases of persistent or retreatment infections where residual biofilm may be suspected. Its controlled activation and localized oxidative effect allow it to target residual microbial niches that may persist after standard irrigation.

Ozone therapy, while biologically safe and easy to apply, demonstrates less consistent clinical benefit due to its instability and limited penetration capacity. Its role may therefore be more appropriate in specific clinical scenarios where mild adjunctive disinfection is desired, rather than in cases involving established or refractory biofilm-associated infections. Importantly, the variability in protocols across studies highlights the need for caution when extrapolating laboratory findings directly into clinical decision-making.

Overall, the clinical implication of current evidence is not the replacement of existing disinfection protocols, but rather their strategic enhancement through adjunctive oxidative therapies, with PDT showing comparatively stronger translational potential than ozone therapy.

Future Perspectives: Despite increasing interest in oxidative adjunctive therapies, several critical gaps remain before ozone therapy and PDT can be fully integrated into evidence-based endodontic protocols. One of the primary challenges is the lack of standardized treatment parameters. Variations in photosensitizer type and concentration, light wavelength, irradiation time, and ozone delivery systems significantly limit the comparability of existing studies and contribute to inconsistent outcomes.

Future research should prioritize the development of

standardized, reproducible protocols, particularly for PDT, to enable more accurate assessment of its true clinical efficacy. In addition, most of the available evidence comes from in vitro biofilm models that do not fully replicate the complex physicochemical environment of the root canal system. There is a clear need for well-designed in vivo and randomized clinical trials that evaluate not only microbial reduction but also long-term clinical outcomes such as healing of apical periodontitis and retreatment success rates.

Another promising direction is the exploration of combination strategies, in which ozone therapy and PDT are integrated with conventional irrigants or adjunctive activation systems such as ultrasonic or sonic irrigation. Such multimodal approaches may overcome the limitations of individual modalities by enhancing penetration, disrupting the extracellular polymeric matrix, and improving antimicrobial distribution within the canal system.

Furthermore, advances in nanotechnology and targeted drug delivery may offer opportunities to improve photosensitizer penetration and stability, potentially enhancing the effectiveness of PDT in deep biofilm layers. Similarly, innovations in ozone delivery systems may help address current limitations related to instability and rapid decomposition.

Ultimately, future endodontic disinfection strategies are likely to move toward a biofilm-targeted, multimodal therapeutic model, in which oxidative therapies serve as adjuncts within a broader, mechanism-based approach to microbial control. Within this evolving framework, PDT currently appears to have greater translational potential, although both modalities require further rigorous investigation before definitive clinical recommendations can be established.

Conclusion

Both ozone therapy and PDT are promising oxidative adjuncts for disinfecting root canal systems, particularly against resilient *E. faecalis* biofilms frequently implicated in persistent endodontic infections. Current evidence indicates that while both modalities exhibit antimicrobial activity, their clinical performance is inconsistent when used in isolation and is strongly influenced by

methodological and biological factors.

Among the two approaches, PDT demonstrates comparatively greater antimicrobial reliability, likely due to its controlled activation mechanism and more predictable generation of reactive oxygen species within the biofilm environment. Ozone therapy, although biocompatible and conceptually attractive, is limited by its instability, rapid decomposition, and reduced penetration into mature biofilm structures. Importantly, neither modality achieves complete or predictable eradication of *E. faecalis* biofilms as a standalone treatment, and both remain inferior to sodium hypochlorite in conventional chemomechanical disinfection protocols.

Overall, the available evidence supports the use of ozone therapy and PDT as adjunctive, biofilm-targeted strategies rather than primary disinfection methods. Their greatest clinical value likely lies in their integration into multimodal endodontic protocols aimed at enhancing disruption of residual biofilm following conventional irrigation. Future research should focus on protocol standardization and well-designed clinical trials to better define their true translational potential and long-term therapeutic impact.

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Conflict of interest

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

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References

1. Wen YH, Lin YX, Zhou L, Lin C, Zhang L. The immune landscape in apical periodontitis: From mechanism to therapy. *Int Endod J.* 2024;57(11):1526-45.
2. Nair PN. On the causes of persistent apical periodontitis: a review. *Int Endod J.* 2006;39(4):249-81.
3. Nair PN. Pathogenesis of apical periodontitis and the causes of endodontic failures. *Crit Rev Oral Biol Med.* 2004;15(6):348-81.
4. Stuart CH, Schwartz SA, Beeson TJ, Owatz CB. *Enterococcus faecalis*: its role in root canal treatment failure and current concepts

in retreatment. *J Endod.* 2006;32(2):93-8.

5. Prada I, Micó-Muñoz P, Giner-Lluesma T, Micó-Martínez P, Collado-Castellano N, Manzano-Saiz A. Influence of microbiology on endodontic failure. Literature review. *Med Oral Patol Oral Cir Bucal.* 2019;24(3):364-72.

6. Ozkan C, Derelioglu SS, Hanci H, et al. Evaluation of the Antimicrobial Efficacy of Sodium Hypochlorite, Ozone, Diode Laser, and Photodynamic Therapy Against *Enterococcus faecalis* and *Streptococcus mutans*: An In-Vitro Study. *Oral Health Prev Dent.* 2026;24:12-7.

7. Luza A, Salvatierra MES, Montero SCL, et al. Antibiofilm Effect of Curcuma longa L. Extract in Antimicrobial Photodynamic Therapy against *Enterococcus faecalis*. *Photodiagnosis Photodyn Ther.* 2026:105515.

8. Perin MLC, Duarte ML, Silva KS, et al. Efficacy of antimicrobial photodynamic therapy for bacterial reduction in endodontics of primary teeth: an in vitro study. *Lasers Med Sci.* 2025;40(1):322.

9. Gunasekaran G, Madhubala MM, Nayanthara GS, Mahalaxmi S. Photodynamic antibacterial evaluation of polydopamine nanoparticle optimised Curcumin Longa against endodontic biofilm-An in-vitro study. *Aust Endod J.* Apr 2025;51(1):90-102.

10. Namba AM, Santos ELS, Garcia MT, et al. Farnesol as a potentiator of antimicrobial photodynamic inactivation on *Enterococcus faecalis*. *Photodiagnosis Photodyn Ther.* 2022;39:102928.

11. Moreira SA, Nunes JB, Colombo FA, Fonseca N, Viola NV. Radiographic and antimicrobial evaluation of enterococcus *Faecalis* and *Actinomyces Israelii* micro-organisms after photodynamic therapy (aPDT). *Photodiagnosis Photodyn Ther.* 2021;35:102433.

12. Tuncay Ö, Dinçer AN, Kuştarıcı A, Er Ö, Dinç G, Demirbuga S. Effects of ozone and photo-activated disinfection against *Enterococcus faecalis* biofilms in vitro. *Niger J Clin Pract.* Nov-Dec 2015;18(6):814-8.

13. Willett JLE, Dunny GM. Insights into ecology, pathogenesis, and biofilm formation of *Enterococcus faecalis* from functional genomics. *Microbiol Mol Biol Rev.* Mar 27 2025;89(1):e0008123.

14. Suryaetha K, Chandrika SK, Thomas S. Comprehensive genomics depict accessory genes encoding pathogenicity and biofilm determinants in *Enterococcus faecalis*. *Future Microbiol.* 2021;16(3):175-84.

15. Ballering KS, Kristich CJ, Grindle SM, Oromendia A, Beattie DT, Dunny GM. Functional genomics of *Enterococcus faecalis*: multiple novel genetic determinants for biofilm formation in the core genome. *J Bacteriol.* Apr 2009;191(8):2806-14.

16. Dasgupta M, Paul R, Chowdhury P, et al. Management of

Enterococcus faecalis biofilms: a combinatorial approach with phytochemical. *Braz J Microbiol.* 2025;56(2):1131-43.

17. Francisco PA, Fagundes P, Lemes-Junior JC, Lima AR, Passini MRZ, Gomes B. Pathogenic potential of *Enterococcus faecalis* strains isolated from root canals after unsuccessful endodontic treatment. *Clin Oral Investig.* 2021;25(9):5171-9.

18. Aebischer D, Szpara J, Bartusik-Aebischer D. Advances in Medicine: Photodynamic Therapy. *Int J Mol Sci.* 2024;25(15).

19. Nunes LP, Nunes GP, Ferrisse TM, et al. Antimicrobial photodynamic therapy in endodontic reintervention: A systematic review and meta-analysis. *Photodiagnosis Photodyn Ther.* 2022;39:103014.

20. Kwiatkowski S, Knap B, Przystupski D, et al. Photodynamic therapy - mechanisms, photosensitizers and combinations. *Biomed Pharmacother.* 2018;106:1098-107.

21. Rkein AM, Ozog DM. Photodynamic therapy. *Dermatol Clin.* 2014;32(3):415-425, x.

22. Plotino G, Grande NM, Mercade M. Photodynamic therapy in endodontics. *Int Endod J.* 2019;52(6):760-774.

23. Gholami L, Shahabi S, Jazaeri M, Hadilou M, Fekrazad R. Clinical applications of antimicrobial photodynamic therapy in dentistry. *Front Microbiol.* 2022;13:1020995.

24. Das U, Giri J, Ayer A. Effectiveness of antimicrobial photodynamic therapy in endodontic infection: a systematic review and meta-analysis. *BMC Oral Health.* 2025;26(1):116.

25. de Oliveira BP, Aguiar CM, Câmara AC. Photodynamic therapy in combating the causative microorganisms from endodontic infections. *Eur J Dent.* 2014;8(3):424-30.

26. Meire MA, Bronzato JD, Bomfim RA, Gomes B. Effectiveness of adjunct therapy for the treatment of apical periodontitis: A systematic review and meta-analysis. *Int Endod J.* Oct 2023;56 Suppl 3:455-74.

27. Sen S, Sen S. Ozone therapy a new vista in dentistry: integrated review. *Med Gas Res.* 2020;10(4):189-92.

28. Azarpazhooh A, Limeback H. The application of ozone in dentistry: a systematic review of literature. *J Dent.* 2008;36(2):104-16.

29. Nunes IA, Novais TM, Garcia PP, et al. Antimicrobial action of ozonated water and photodynamic therapy with sonic activation in root canals infected with *Enterococcus faecalis*. *J Clin Exp Dent.* Jun 2022;14(6):e486-e491.

30. Nasiri K, Wrbas KT. Management of sodium hypochlorite accident in root canal treatment. *J Dent Sci.* Apr 2023;18(2):945-6.

31. Singh PK. Root canal complications: 'the hypochlorite accident'. *Sadj.* 2010;65(9):416-9.

32. Cai C, Chen X, Li Y, Jiang Q. Advances in the Role of Sodium Hypochlorite Irrigant in Chemical Preparation of Root Canal Treatment. *Biomed Res Int.* 2023;2023:8858283.