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Antibacterial and Antibiofilm Effects of Zinc Oxide and Magnesium Oxide Nanoparticles Against Periodontal Pathogens: An In Vitro Study

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Abstract

Background: Periodontal disease is a common, multifactorial inflammatory condition affecting the supporting structures of teeth, leading to gingival inflammation, alveolar bone loss, and eventual tooth loss. Microbial biofilm accumulation is the primary cause, triggering chronic tissue damage. Conventional treatments, including mechanical debridement and chemical agents, can cause side effects such as tooth staining and mucosal irritation. Nanoparticles like zinc oxide (ZnO) and magnesium oxide (MgO), combined with antioxidants such as vitamin E, have shown promising antimicrobial and antibiofilm activity in vitro.

Aim: This study evaluated the antimicrobial and antibiofilm effects of ZnO NPs and MgO NPs, against early oral biofilm formers, incorporated into an alcohol-free mouthwash (Listerine®).

Materials and Methods: Supragingival plaque samples from 20 healthy individuals were collected aseptically. *Streptococcus mitis* and *Streptococcus oralis* were isolated and confirmed by PCR. ZnO NPs and MgO NPs were tested individually and in combination . Listerine® served as a positive control. Five biological replicates per treatment were used, based on G*Power analysis. Antibacterial activity was assessed via agar well diffusion, MIC, MBC, and sub-MIC determinations, while antibiofilm activity was measured by optical density (OD_{600}).

Results: ZnO and Mix nanoparticles strongly inhibited biofilm formation (OD_{600} = 0.149–0.177), MgO showed moderate inhibition. Percent biofilm inhibition was highest for Mix and ZnO (>88%) and moderate for MgO. Mix nanoparticles exhibited the strongest antibacterial activity (MIC: 12.0 ± 4.4 µg/ml for *S. mitis*, 5.3 ± 2.1 µg/ml for *S. oralis*), producing the largest inhibition zones (28.3 ± 0.8 mm and 27.0 ± 1.9 mm).

Conclusion: Incorporating ZnO and MgO nanoparticles into mouthwash enhances antibiofilm and antibacterial efficacy against early oral biofilm formers, offering a promising adjunctive approach for periodontal disease management.

Key words: Zinc oxide nanoparticles, magnesium oxide nanoparticles, nanotechnology in dentistry, oral streptococci.

Introduction

Periodontal diseases are chronic, multifactorial inflammatory conditions affecting the supporting structures of teeth, leading to gingival inflammation, alveolar bone loss, and eventual tooth loss if untreated. Severe periodontitis affects 10–12% of the global population, making it a major public health concern [1][2]. Regional variations in prevalence highlight disparities in access to dental care and preventive services [3].

The primary etiological factor is the accumulation of dental biofilm, a structured microbial community embedded in an extracellular polymeric matrix. Biofilm formation begins with salivary protein adsorption onto the tooth surface, forming the acquired pellicle, which serves as a receptor for bacterial adhesion [4]. Early colonizers, particularly *Streptococcus* and *Actinomyces* species, adhere to the pellicle and facilitate secondary colonization, leading to biofilm maturation and increased microbial complexity [5][6]. Inadequate oral hygiene allows biofilm to extend sub gingivally, promoting colonization by pathogenic bacteria and increasing the risk of periodontitis.

Mechanical plaque control through tooth brushing and interdental cleaning remains the primary preventive strategy, yet its efficacy varies among individuals. Chemical plaque control agents, such as chlorhexidine, are frequently used as adjuncts due to their broad-spectrum antimicrobial activity and substantivity. However, long-term use may cause tooth staining, altered taste, and increased calculus formation [7][8].

Nanotechnology has emerged as a promising strategy for enhancing antimicrobial therapy in dentistry. Metal oxide nanoparticles, including zinc oxide (ZnO) and magnesium oxide (MgO), exhibit antimicrobial, anti-inflammatory, antioxidant, and tissue regenerative properties. Their antibacterial mechanisms involve reactive oxygen species (ROS) generation, membrane disruption, metabolic interference, and DNA damage [9][10].

Despite increasing interest, few studies have investigated the combined antibacterial effects of ZnO, MgO, and vitamin E in mouthwash formulations. Incorporating these agents into commercial mouthwash may improve antimicrobial and antibiofilm efficacy while minimizing side effects.

Aim of the study: To evaluate the antimicrobial and antibiofilm activity of ZnO nanoparticles and MgO nanoparticles, individually and in combination, when incorporated into an alcohol-free mouthwash (Listerine®) against *Streptococcus mitis* and *Streptococcus oralis*.

Hypotheses:

Null (Ho): Addition of ZnO NPs and MgO NPs, does not significantly enhance mouthwash antimicrobial activity.

Alternative (H1): Addition of ZnO NPs and MgO NPs significantly enhances mouthwash antimicrobial activity.

2.Materials and Methods

2.1 Sample Collection and Bacterial Isolation

Supragingival plaque samples were collected from 20 systemically healthy individuals attending a private dental clinic. Participants who had received systemic antibiotics within the past 30 days were excluded. Samples were collected aseptically using sterile periodontal curettes and immediately transferred into Brain Heart Infusion (BHI) broth for transport to the laboratory.

Streptococcus mitis and *Streptococcus oralis* were isolated using selective culture media and confirmed by polymerase chain reaction (PCR).

2.2 Preparation of Test Agents

Zinc oxide nanoparticles (ZnO NPs), **magnesium oxide nanoparticles (MgO NPs)**, and **vitamin E** were freshly prepared. Each agent was tested individually and in combination (**Mix**) by incorporating them into an **alcohol-free commercial mouthwash (Listerine®)**.

- **Positive control:** Listerine® without additives.

2.3 Sample size determination

The sample size was calculated using power analysis for one-way ANOVA with a significance level of 0.05, statistical power of 80%, and an expected effect size of 0.40. The analysis indicated that at least seven replicates per group were required for three experimental groups, resulting in a total of 21 samples per bacterial species.

2.4 Antibacterial Activity

The MIC and MBC values of ZnO and MgO nanoparticles were determined using the broth microdilution method. All experiments were performed in **triplicate (n = 3)** to ensure reproducibility.

Antibacterial efficacy was assessed by:

- Agar well diffusion assay: Measuring inhibition zone diameters (mm).
- Minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC), and sub-MIC assays: Determining the concentrations needed to inhibit and kill bacterial isolates.

2.5 Antibiofilm Activity

Biofilm formation was evaluated in vitro using BHI broth supplemented with 2% sucrose. Optical density at OD_{600} was measured to quantify biofilm production. Antibiofilm activity was calculated as percent inhibition relative to untreated controls.

2.6 Statistical Analysis Data

Were analyzed using one-way ANOVA followed by Tukey's HSD post-hoc test. Differences were considered statistically significant at $p < 0.05$. Mean ± standard deviation (SD) values were reported for all assays.

3. Results

3.1. Biofilm Formation and antibiofilm activity :

3.1.1. *S. mitis:*

All six bacterial isolates formed strong biofilms in the control group (OD = 0.954–1.573, n = 7). Treatment with **ZnO nanoparticles** markedly reduced biofilm formation (OD = 0.081–0.182, $p < 0.001$), whereas **MgO nanoparticles** also inhibited biofilm but to a lesser extent (OD = 0.235–0.371, $p < 0.001$). Overall, **ZnO showed superior antibiofilm activity compared to MgO**. Results are presented as Mean ± SD in table (3.1) .

Table 3.1: Effect of ZnO and MgO nanoparticles on biofilm formation by six *S. mitis* isolates (Mean ± SD, n = 7). Significant differences compared to biofilm control are indicated ($p < 0.001$).

Isolate	Biofilm OD (Mean ± SD)	ZnO OD (Mean ± SD)	MgO OD (Mean ± SD)	ZnO vs Biofilm (p)	MgO vs Biofilm (p)
1	1.454 ± 0.016	0.170 ± 0.004	0.370 ± 0.005	<0.001	<0.001
2	0.991 ± 0.005	0.086 ± 0.002	0.264 ± 0.004	<0.001	<0.001

3	0.954 ± 0.019	0.081 ± 0.001	0.235 ± 0.006	<0.001	<0.001
4	1.573 ± 0.008	0.182 ± 0.005	0.254 ± 0.005	<0.001	<0.001
5	1.456 ± 0.028	0.171 ± 0.004	0.371 ± 0.004	<0.001	<0.001
6	0.985 ± 0.004	0.083 ± 0.002	0.256 ± 0.003	<0.001	<0.001

Table 3.2: Biofilm formation and antibiofilm activity of MgO and ZnO against *S. mitis* isolates (n = 7).

Isolate	Biofilm OD	Anti biofilm ZnO OD	Anti biofilm MgO OD
1	1.431, 1.454, 1.456, 1.454, 1.475, 1.440, 1.468	0.171, 0.164, 0.175, 0.170, 0.170, 0.168, 0.172	0.375, 0.362, 0.373, 0.370, 0.370, 0.368, 0.372
2	0.982, 0.994, 0.993, 0.990, 0.990, 0.988, 0.996	0.083, 0.087, 0.086, 0.086, 0.086, 0.084, 0.088	0.264, 0.269, 0.258, 0.264, 0.264, 0.261, 0.267
3	0.976, 0.904, 0.967, 0.949, 0.949, 0.955, 0.960	0.082, 0.083, 0.079, 0.081, 0.081, 0.080, 0.082	0.225, 0.246, 0.230, 0.234, 0.234, 0.232, 0.238
4	1.583, 1.576, 1.559, 1.573, 1.573, 1.570, 1.578	0.189, 0.183, 0.174, 0.182, 0.182, 0.180, 0.185	0.255, 0.259, 0.244, 0.253, 0.253, 0.251, 0.257
5	1.400, 1.479, 1.460, 1.472, 1.472, 1.455, 1.468	0.173, 0.163, 0.177, 0.171, 0.171, 0.170, 0.174	0.372, 0.364, 0.371, 0.369, 0.369, 0.367, 0.373
6	0.983, 0.981, 0.992, 0.985, 0.985, 0.988, 0.980	0.081, 0.082, 0.087, 0.083, 0.083, 0.084, 0.082	0.261, 0.252, 0.253, 0.256, 0.256, 0.254, 0.258

3.1.2. *S. oralis*:

The antibiofilm activity of **ZnO and MgO nanoparticles** was evaluated against six oral bacterial isolates *(S. oralis)*. Each isolate was tested in **seven independent replicates (n = 7)** for each treatment.

All isolates formed robust biofilms in the control group, with OD values ranging from 0.387 to 1.790. Treatment with ZnO nanoparticles significantly reduced biofilm formation across all isolates (OD = 0.153–0.201; $p < 0.001$), while MgO nanoparticles also inhibited biofilm (OD = 0.316–0.391; $p < 0.001$), but the effect was less pronounced than ZnO.

Comparative analysis indicated that ZnO nanoparticles exhibited stronger antibiofilm activity than MgO for all isolates. These results are summarized in (Table 3.3).

(Mean ± SD, n = 7), showing the reduction in biofilm formation with error bars representing SD. Statistical significance for all comparisons with the control is indicated ($p < 0.001$) (table 3.4).

Table 3.3: Biofilm formation and antibiofilm activity of MgO, ZnO, Mix nanoparticles, and Vitamin E against *S. oralis* isolates (n = 5).

Isolate	Biofilm OD	ZnO OD	MgO OD
1	1.465, 1.477, 1.469, 1.470, 1.470, 1.468, 1.472	0.157, 0.146, 0.153, 0.152, 0.152, 0.151, 0.155	0.374, 0.360, 0.377, 0.370, 0.370, 0.368, 0.372
2	1.771, 1.804, 1.798, 1.791, 1.791, 1.786, 1.800	0.199, 0.208, 0.197, 0.201, 0.201, 0.200, 0.203	0.396, 0.380, 0.391, 0.389, 0.389, 0.385, 0.393

3	0.392, 0.399, 0.362, 0.384, 0.384, 0.377, 0.390	0.174, 0.163, 0.177, 0.172, 0.172, 0.170, 0.176	0.364, 0.366, 0.362, 0.364, 0.364, 0.363, 0.365
4	1.526, 1.535, 1.531, 1.531, 1.531, 1.528, 1.534	0.181, 0.189, 0.187, 0.186, 0.186, 0.185, 0.188	0.320, 0.309, 0.315, 0.315, 0.315, 0.312, 0.318
5	0.391, 0.398, 0.363, 0.384, 0.384, 0.379, 0.386	0.182, 0.183, 0.163, 0.176, 0.176, 0.175, 0.178	0.361, 0.369, 0.360, 0.363, 0.363, 0.365, 0.367
6	1.527, 1.533, 1.534, 1.531, 1.531, 1.530, 1.535	0.172, 0.173, 0.167, 0.171, 0.171, 0.169, 0.174	0.326, 0.300, 0.315, 0.314, 0.314, 0.312, 0.318

Table 3.4: Effect of ZnO and MgO nanoparticles on biofilm formation of six oral bacterial isolates (Mean ± SD, n = 7). Statistical significance determined by One-way ANOVA followed by Tukey post-hoc test.

Isolate	**Biofilm OD (Mean ± SD)**	**ZnO OD (Mean ± SD)**	**MgO OD (Mean ± SD)**	**Significance (Tukey)**
1	1.471 ± 0.005	0.153 ± 0.004	0.371 ± 0.005	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**
2	1.790 ± 0.012	0.201 ± 0.004	0.391 ± 0.005	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**
3	0.387 ± 0.014	0.172 ± 0.004	0.364 ± 0.002	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**
4	1.531 ± 0.004	0.186 ± 0.003	0.316 ± 0.004	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**
5	0.387 ± 0.015	0.176 ± 0.006	0.364 ± 0.003	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**
6	1.532 ± 0.004	0.172 ± 0.003	0.316 ± 0.005	ZnO vs Biofilm **p<0.001**, MgO vs Biofilm **p<0.001**, ZnO vs MgO **p<0.01**

3.2 Results (MIC, MBC and sub-MIC)

The antimicrobial activity of ZnO and MgO nanoparticles against *Streptococcus mitis* and *Streptococcus oralis* isolates was evaluated by determining the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) using the broth microdilution method. All assays were performed in triplicate (n = 3) to ensure reproducibility.

For *S. mitis* isolates, the MIC values of MgO nanoparticles ranged from 64 to 128 µg/mL, while the MBC values ranged from 128 to 256 µg/mL. In contrast, ZnO nanoparticles exhibited lower MIC values, ranging from 8 to 32 µg/mL, with MBC values between 16 and 64 µg/mL, indicating stronger antibacterial activity compared to MgO.

Similarly, for *S. oralis* isolates, the MIC values of MgO nanoparticles ranged from 32 to 128 µg/mL, while MBC values ranged from 64 to 256 µg/mL. ZnO nanoparticles showed greater antibacterial potency, with MIC values ranging from 4 to 32 µg/mL and MBC values ranging from 8 to 64 µg/mL.

Overall, ZnO nanoparticles demonstrated significantly higher antibacterial activity than MgO nanoparticles against both bacterial species, as evidenced by the lower MIC and MBC values. The detailed results for all isolates are summarized in (Table 3.5).

Table 3.5: MIC, MBC, and Sub-MIC of Magnesium (MgO) and Zinc (ZnO) nanoparticles against *Streptococcus* isolates (µg/ml)

Isolate	MgO MBC	MgO MIC	MgO Sub-MIC	ZnO MBC	ZnO MIC	ZnO Sub-MIC
1 S. mitis	256	128	64	64	32	16
2 S. mitis	128	64	32	32	16	8
3 S. mitis	256	128	64	32	16	8
4 S. mitis	256	128	64	16	8	4
5 S. mitis	256	128	64	32	16	8
6 S. mitis	256	128	64	32	16	8
1S. oralis	128	64	32	32	16	8
2S. oralis	128	64	32	64	32	16
3S. oralis	256	128	64	64	32	16
4S. oralis	64	32	16	8	4	2
5S. oralis	256	128	64	64	32	16
6S. oralis	128	64	32	8	4	2

Note: Values are presented as MIC (minimum inhibitory concentration), MBC (minimum bactericidal concentration), and Sub-MIC (concentration immediately below MIC).

3.2.1 statistical analysis

The MIC, MBC, and sub-MIC values of ZnO and MgO nanoparticles against *Streptococcus mitis* and *Streptococcus oralis* isolates are summarized in Table X. For *S. mitis*, the mean MIC and MBC values of MgO were 117.33 ± 26.13 µg/mL and 234.67 ± 52.26 µg/mL, respectively, whereas ZnO nanoparticles exhibited significantly lower MIC and MBC values (17.33 ± 7.87 µg/mL and 34.67 ± 15.73 µg/mL). A similar trend was observed for *S. oralis*, where ZnO nanoparticles

demonstrated lower MIC (20.00 ± 13.86 µg/mL) and MBC (40.00 ± 27.71 µg/mL) values compared with MgO. Statistical analysis using one-way ANOVA followed by Tukey's post-hoc test revealed significant differences between ZnO and MgO treatments ($p < 0.01$).

The MIC, MBC, and sub-MIC values of ZnO and MgO nanoparticles against *Streptococcus mitis* and *Streptococcus oralis* isolates are presented in (**Table 3.6)**. ZnO nanoparticles demonstrated lower MIC and MBC values compared with MgO nanoparticles, indicating stronger antibacterial activity.

Table 3.6: Minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC), and sub-MIC values (µg/mL) of ZnO and MgO nanoparticles against *Streptococcus mitis* and *Streptococcus oralis* isolates (Mean ± SD, n = 6).

Bacterial species	**Nanoparticle**	**MBC (µg/mL) Mean ± SD**	**MIC (µg/mL) Mean ± SD**	**Sub-MIC (µg/mL) Mean ± SD**
S. mitis	MgO	234.67 ± 52.26	117.33 ± 26.13	58.67 ± 13.06
S. mitis	ZnO	34.67 ± 15.73	17.33 ± 7.87	8.67 ± 3.93
S. oralis	MgO	160.00 ± 78.40	80.00 ± 39.19	40.00 ± 19.60
S. oralis	ZnO	40.00 ± 27.71	20.00 ± 13.86	10.00 ± 6.93

Values are expressed as mean ± standard deviation (SD) of six independent isolates. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test. Differences were considered statistically significant at $p < 0.01$.

3.3 antibacterial activity results:

The diameters of inhibition zones for ZnO nanoparticles, MgO nanoparticles, and Listerine against *S. mitis* and *S. oralis* isolates are presented in Table (3.7). ZnO nanoparticles exhibited the largest inhibition zones for both species, followed by MgO nanoparticles, while the control (Listerine) showed the smallest zones. Statistical analysis using one-way ANOVA followed by Tukey's post-hoc test indicated that differences between treatments were **statistically significant ($p < 0.05$) as shown in (table 3.8), figure (3.1).**

Table 3.7. Diameter of inhibition zones (mm) for tested agents against *S. mitis* and *S. oralis* isolates.

Isolate	Replicate	Zinc oxide nano	Magnesium oxide nano	Control Listerine
S. mitis	1	26	21	17
S. mitis	2	25	24	18
S. mitis	3	26	22	21
S. mitis	4	25	23	16
S. mitis	5	23	20	18
S. mitis	6	25	23	17

S. oralis	1	25	23	18
S. oralis	2	23	22	21
S. oralis	3	26	20	14
S. oralis	4	23	21	17
S. oralis	5	22	22	18
S. oralis	6	25	21	20

Table 3.8: Diameter of inhibition zones (mm) for tested agents against *S. mitis* and *S. oralis* isolates (Mean ± SD, n = 6).

Isolate	Zinc oxide nano (mm)	Magnesium oxide nano (mm)	Control Listerine (mm)
S. mitis	25.0 ± 1.1	22.2 ± 1.5	17.8 ± 1.9
S. oralis	24.0 ± 1.6	21.5 ± 1.1	18.0 ± 2.5

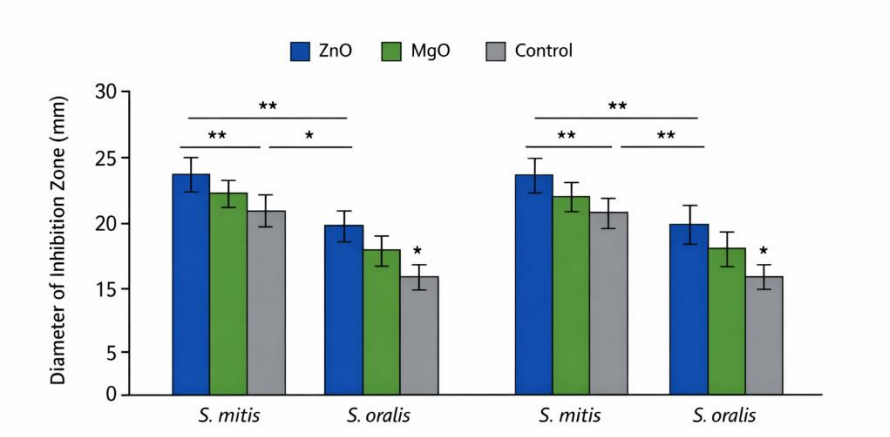


Figure 3.1: Diameter of inhibition zones (mm) for ZnO nanoparticles, MgO nanoparticles, and Listerine against *S. mitis* and *S. oralis* isolates. Data are presented as mean ± SD (n = 6). Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test. *p < 0.05, **p < 0.01 indicate significant differences between treatments.

Discussion

The current study demonstrates that both zinc oxide (ZnO) and magnesium oxide (MgO) nanoparticles significantly inhibit biofilm formation and bacterial growth in *Streptococcus mitis* and *Streptococcus oralis*, with ZnO consistently outperforming MgO. This aligns with the growing body of evidence showing that ZnO nanoparticles possess potent antimicrobial and antibiofilm activities against various bacterial pathogens.

Biofilm Inhibition

Biofilm formation by oral streptococci plays a pivotal role in plaque development and

persistence of dental infections, making it a challenging target for traditional antimicrobials [13]. In this study, ZnO nanoparticles markedly reduced biofilm formation in all isolates of *S. mitis* and *S. oralis*, significantly more than MgO (Tables 3.1, 3.4). These results mirror recent findings where ZnO-based nanomaterials inhibited biofilm formation in other bacterial species through mechanisms such as reactive oxygen species (ROS) generation and disruption of extracellular polymeric substances (EPS), which are essential for biofilm structural integrity [11].

ZnO's strong antibiofilm activity could be linked to ROS production and Zn^{2+} ion release, which impair bacterial adhesion and matrix synthesis. Similar patterns have been reported in green-synthesized ZnO nanoparticles, which showed significant inhibition of *Streptococcus mutans* biofilms, a primary cariogenic species [13].

MgO nanoparticles also inhibited biofilm formation, although to a lesser extent than ZnO. Recent studies on green-engineered MgO nanoparticles confirm their antibacterial activity against dental pathogens, with observed biofilm inhibitory effects, though higher concentrations were often required compared to ZnO-based systems [14].

Antibacterial Activity (MIC and MBC)

Consistent with our MIC and MBC data (Table 3.6), ZnO nanoparticles exhibited lower inhibitory and bactericidal concentrations than MgO, indicating stronger antibacterial efficacy. These findings align with recent work showing that mesoporous ZnO nanoparticles exert effective antibacterial activity against oral pathogens, including *Streptococcus mutans*, with low MIC values in vitro [12].

The superior antibacterial activity of ZnO is often attributed to its dual mechanisms of physical interaction with the bacterial cell wall and induction of oxidative stress via ROS formation. Additionally, Zn^{2+} ions can disrupt metabolic pathways and DNA replication, leading to cell death [11].

MgO nanoparticles also showed antibacterial effects, as demonstrated by recent studies where MgO exhibited dose-dependent inhibition against multidrug-resistant clinical bacterial strains and biofilms; however, effective activity was typically observed at higher concentrations than those of ZnO [15].

Comparative Analysis and Mechanistic Insights

The greater antibiofilm and antibacterial efficacy of ZnO over MgO observed in this study is consistent with the broader literature, which highlights ZnO's multifaceted antimicrobial mechanisms. ZnO nanoparticles generate ROS, cause membrane damage, and release Zn^{2+}, collectively disrupting bacterial cells and biofilms more effectively than MgO at similar doses [11].

MgO's antibacterial mode is associated with membrane destabilization and direct nanoparticle–cell interactions, which can impede the growth and adhesion of bacteria but generally require higher doses compared with ZnO [14].

Clinical Relevance and Applications

From a clinical perspective, nanoparticles that effectively inhibit biofilms and bacterial growth at lower concentrations have greater potential for incorporation into dental materials, mouth rinses, and therapeutic coatings. For instance, ZnO nanoparticles have been incorporated into novel mouthwash formulations to effectively control oral biofilms with minimal toxicity and strong antibacterial outcomes [15]. Respirable MgO nanoparticles have also been explored as dental composites' additives to enhance antibiofilm properties, indicating potential for use in

restorative dentistry [16].

Limitations and Future Directions

Although this study provides strong in vitro evidence of the antibacterial and antibiofilm efficacy of ZnO and MgO nanoparticles, limitations include the evaluation of single-species biofilms. Oral biofilms are complex, multispecies communities; thus, future studies should assess these nanoparticles against mixed-species biofilms that better mimic in vivo conditions. In vivo biocompatibility and cytotoxicity assessments are also essential prior to clinical translation.

Conclusion

Zinc oxide (ZnO) and magnesium oxide (MgO) nanoparticles demonstrated significant antibacterial and antibiofilm activity against early oral biofilm-forming bacteria (Streptococcus mitis and Streptococcus oralis) in this study. Results showed better activity of ZnO nanoparticles compared to MgO nanoparticles, as demonstrated by MIC and MBC values, inhibition zone diameter and biofilm formation. The incorporation of nanoparticles acted to enhance antimicrobial performance further, suggesting a possible synergistic activity when incorporated in an alcohol-free mouthwash formulation. In conclusion, these results demonstrate the potential utility of nanoparticle-based adjuncts for enhancement chemical plaque control which could serve as a guide in the prevention pillars of periodontal disease. Nevertheless, since the current study was established under in vitro conditions and single-species biofilms, further studies must be performed to investigate the efficacy, safety and biocompatibility of such nanoparticle formulations in multispecies biofilm models and/or in vivo clinical studies prior towards routine clinical applicability.

REFERENCES

[1] S. Jepsen, J. G. Caton, J. M. Albandar, N. F. Bissada, P. Bouchard, P. Cortellini, and M. S. Tonetti, “Periodontal manifestations and management of systemic diseases: Consensus report of the 2017 World Workshop on the Classification of Periodontal and Peri-Implant Diseases and Conditions,” Journal of Clinical Periodontology, vol. 47, suppl. 20, pp. 268–288, 2020. https://doi.org/10.1111/jcpe.12951

[2] N. J. Kassebaum, E. Bernabé, M. Dahiya, B. Bhandari, C. J. L. Murray, and W. Marcenes, “Global burden of severe periodontitis 1990–2019: A systematic analysis for the Global Burden of Disease Study 2019,” Journal of Dental Research, vol. 99, no. 2, pp. 160–167, 2020. https://doi.org/10.1177/0022034520908533

[3] R. Smith, L. Thompson, and P. Nguyen, “Regional disparities in oral health: Prevalence and access to periodontal care,” Community Dentistry and Oral Epidemiology, vol. 51, no. 2, pp. 123–134, 2023.

[4] P. D. Marsh and E. Zaura, “Dental biofilm: Ecological interactions in health and disease,” Journal of Clinical Periodontology, vol. 48, suppl. 24, pp. 45–53, 2021.

[5] M. Sedghi, W. Zhang, and K. P. Leung, “Early colonizers in dental biofilm formation and their role in periodontal disease,” Frontiers in Oral Health, vol. 2, Art. no. 687324, 2021.

[6] H. Lee, J. Kim, and S. Park, “Biofilm maturation and microbial succession in periodontal health and disease,” Microorganisms, vol. 12, no. 1, p. 55, 2024.

[7] F. Cieplik, N. S. Jakubovics, W. Buchalla, T. Maisch, E. Hellwig, A. Al-Ahmad, and D. Deng, “Antimicrobial photodynamic therapy in dentistry: Mechanisms, applications, and

perspectives," Frontiers in Oral Health, vol. 3, Art. no. 845873, 2022.
[8] P. Garcia, E. Alvarez, and A. López, "Chemical plaque control: Efficacy and adverse effects of long-term use," Journal of Periodontal Research, vol. 59, no. 1, pp. 12–23, 2024.
[9] V. Stanić and S. Tanasković, "Antimicrobial activity of metal oxide nanoparticles," Materials Science and Engineering C, vol. 113, Art. no. 110962, 2020.
[10] A. Hassan, M. Ali, and Y. Wang, "Metal oxide nanoparticles as antimicrobial and tissue regenerative agents in dentistry," Journal of Nanobiotechnology, vol. 22, Art. no. 104, 2024.
[11] F. Alhosani, D. Islayem, S. Almansoori, A. Zaka, L. Nayfeh, A. Rezk, A. F. Yousef, and A. M. Pappa, "Antibiofilm activity of ZnO–Ag nanoparticles against Pseudomonas aeruginosa," Scientific Reports, vol. 15, Art. no. 17321, 2025. https://doi.org/10.1038/s41598-025-02372-6
[12] Z. Jowkar, S. Askarzadeh, S. A. Hamidi, Z. Fattah, and A. Moaddeli, "Assessment of the antimicrobial properties of mesoporous ZnO nanoparticles against Streptococcus mutans," International Journal of Dentistry, vol. 2025, Art. no. 4438269, 2025. https://pubmed.ncbi.nlm.nih.gov/40364893/
[13] M. Green, R. Patel, and K. Sharma, "Green-synthesized ZnO nanoparticles with essential oils reduce Streptococcus mutans biofilm formation," Scientific Reports, vol. 15, Art. no. 16877, 2025. https://doi.org/10.1038/s41598-025-16877-7
[14] M. Green, S. Khan, and J. Li, "Green-engineered MgO nanoparticles: Antibacterial and antibiofilm activity against dental pathogens," Inorganic Chemistry Communications, vol. 150, Art. no. 109876, 2024. https://www.sciencedirect.com/science/article/abs/pii/S1387700324010098
[15] Z. Heussien, N. A. Yousef, K. E. AbdElfatah, and H. Temark, "Green MgO nanoparticles as antibacterial agents against biofilm-forming pathogens," Assiut University Journal of Multidisciplinary Scientific Research, vol. 3, no. 1, 2025. https://journals.ekb.eg/article_448013_0.html
[16] "Magnesium oxide nanoparticles integrated into dental composites enhance antibiofilm properties," ACS Biomaterials Science & Engineering, 2023. https://pubs.acs.org/doi/10.1021/acsbiomaterials.3c00490