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Antibacterial efficacy of oxygen rich fluid against three early colonizing peri-implant bacteria

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The management of peri-implantitis is a relatively new area of research and clinical practice. As of yet, no specific treatment protocol has been proven to be absolutely successful or considered as the gold standard. Oxygenated agents have demonstrated potential benefits when they come to treating periodontal disease, however, studies regarding their use in the treatment of peri-implantitis are lacking. The purpose of this study is to evaluate the direct antimicrobial efficacy of Oxygen-rich fluid against three early peri-implant colonizing bacteria, namely, Escherichia coli (*E. coli*), Staphylococcus aureus (S. aureus), and Pseudomonas aeruginosa (P. aeruginosa). The study was designed to evaluate the minimum inhibitory concentration (MIC) of Oxygen-rich fluid BlueM against *E. coli*, S. aureus, and P. aeruginosa using micro-serial dilution method. BlueM was tested as the experimental group, while 0.2% Chlorhexidine (CHX) as the positive control, and sterile Mueller-Hinton broth (MHB) as the negative control. Each bacterial suspension was prepared at an optical density (OD600 of 0.1) and inoculated into separate rows of a 96-well plate. The plates were incubated at 35 °C for 48 h using the Bioscreen C assay reader, with optical density measurements recorded hourly. For the time-kill assay, the Oxygen-rich agent was tested at its MIC, 2xMIC, and 4xMIC levels. Added to the wells were 180 µL of MHB, 150 µL of the Oxygen-rich agent, and 10 µL of bacterial suspension. Growth was measured at 1, 2, 5, and 30 min. Serial dilutions were performed, and the plates were further incubated at 37 °C for 18–24 h, and colonies were counted. All tests were performed in triplicates. The data were evaluated by using ANOVA test followed by post hoc Tukey test. The within-group comparison at four-time intervals was compared using Repeated measure ANOVA test. The MIC of the Oxygen-rich fluid was assessed against *E. coli*, S. aureus, and P. aeruginosa. *E. coli* exhibited the highest susceptibility, with an MIC of 1.25 mg/L, followed by S. aureus at 2.5 mg/L, and P. aeruginosa at 5 mg/L. The time-kill assay showed a time-dependent reduction in bacterial counts, with significant decreases observed for all strains over the 30-minute testing period. However, despite testing multiple concentrations (MICx1, MICx2, MICx4), no significant differences in bacterial inhibition were observed across these concentrations, indicating that the fluid's antimicrobial effect is consistent across concentrations. The Oxygen-rich fluid demonstrated effective antimicrobial activity against *E. coli*, S. aureus, and P. aeruginosa, with no significant difference observed across concentrations. The fluid's time-dependent antibacterial effects suggest its potential for use in clinical settings and personal oral care, particularly for managing bacterial infections in peri-implant disease.

Keywords Peri-implantitis, Antibacterial, Oxygen therapy, *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*

Dental implants have become a more globally preferred procedure for restoring missing teeth. They have demonstrated great long-term survival rates as a safe and effective therapeutic option for the rehabilitation of partially and fully edentulous patients[1]. However, the placement of dental implants is frequently associated with peri-implant diseases. According to a comprehensive systematic review and meta-analysis, the prevalence was estimated at 19.83% for peri-implantitis and 46.83% for peri-implant mucositis[2]. Numerous bacteria detected in peri-implantitis may vary from those often linked with periodontitis. These microorganisms include bacterial species such as *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Enterobacter aerogenes*, *Enterobacter cloace*, *Escherichia coli*, *Helicobacter pylori*, *Peptostreptococcus micra*, *Pseudomonas spp*., and fungal species such as *Candida spp*[3]. The selection of *E. coli*., *S. aureus*, and *P. aeruginosa* for this study is based on their prevalence

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as common bacteria associated with peri-implant infection, and because they are a well-established model for studying peri-implant decontamination[4–6]. *Staphylococcus aureus*, characterized as an anaerobic, facultative, gram-positive bacterium lacking sporulation capabilities, exhibits a notable affinity for titanium substrates. Conversely, *Escherichia coli*, an anaerobic facultative, gram-negative bacterium with fimbriae, relies on peritrichous flagella for motility. While typically considered a commensal, *E. coli* has emerged as an opportunistic pathogen, increasingly linked with peri-implantitis. *Pseudomonas aeruginosa* is a gram-negative, unipolar motile bacterium capable of facultative aerobic growth, though it can also survive under anaerobic conditions by utilizing nitrate. It is commonly linked to the development of peri-implantitis as it's an opportunistic pathogen[5].

Chlorhexidine (CHX), the gold standard among antimicrobial agents, has been found to be extremely effective as an adjunct to mechanical plaque control[7]. Despite its effectiveness, CHX has several limitations and potential drawbacks that must be carefully considered, especially in long-term use. The most often cited reason for discontinuation and lack of patient compliance is the extrinsic tooth staining caused by CHX[8]. The oxygen therapy (BlueM) formula is composed of Sodium perborate with specific carriers such as glycerol and cellulos, which provides controlled and slow release of oxygen[9]. The antimicrobial effect is attributed to its active components, including active oxygen, sodium perborate, and honey. According to BlueM International, the product releases reactive oxygen species (ROS) through the controlled breakdown of Sodium perborate into Hydrogen peroxide (H_2O_2) and sodium borate, both with antiseptic properties. Additionally, honey is converted to H_2O_2 by the enzyme Glucose oxidase. H_2O_2 generates ROS that disrupts bacterial cell walls, making it an effective broad-spectrum antimicrobial agent[10] Among the various available forms of Oxygen-rich agents, such as gels and mouthwashes, studies have shown favorable effects in targeting the elimination of anaerobic bacteria linked to periodontal disease and it aids in reducing periodontal pockets[11–13]. Although limited studies have explored the effects of Oxygen-rich fluid form, recent research demonstrated antibacterial efficacy against *E. faecalis*. They concluded that it can be a viable alternative to traditional irrigants, however, it was not superior to NaOCl[14].

The efficacy of Oxygen-rich fluid on the peri-implant disease related bacteria has not been investigated yet. The aim of this study, therefore, was to evaluate the direct antimicrobial efficacy of Oxygen-rich fluid against three early peri-implant colonizing bacteria, namely, *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa* bacteria.

Materials and methods

Bacterial strains and cultivation

All procedures and culture handling were performed aseptically in a Class II biological safety cabinet (SterilGARD® III Advance). The bacterial strains from the American Type Culture Collection of *Escherichia coli* (ATCC®35218), *Staphylococcus aureus* (ATCC®29213), and *Pseudomonas aeruginosa* (ATCC®27853). The bacteria were initially cultured on Brain Heart Infusion agar (BHIA) and incubated in their suitable conditions at 37 °C for 24 h. After incubation, three distinct bacterial colonies were inoculated separately into 3 mL of sterile Mueller–Hinton broth (MHB) and incubated at 37 °C for 24 h.

Treatment groups

The study included three groups based on the use or lack thereof of antibacterial agent. The first group was treated with BlueM Oxygen fluid 40 mg/L ((BlueM Europe Inc., Enkweg, Wijhe, The Netherlands) as the experimental group. The second group served as the positive control and was treated with 0.2% CHX (Avohex, Avalon pharmaceutical, Riyadh, Saudi Arabia), a standard antimicrobial agent. The third group, the negative control, consisted of sterile Mueller-Hinton broth (MHB) with bacterial inoculation and received no treatment (NT), to assess baseline conditions.

Minimum Inhibitory Concentration (MIC)

The groups were tested on a 96-well polystyrene plate (Nunc®, MicroWell™, Sigma-Aldrich, St Louis, MO, USA). The Oxygen-rich fluid and CHX were serially diluted into the wells containing MHB [Figure 1]. Minimum inhibitory concentration (MIC) was established using the microdilution technique described by the Clinical and Laboratory Standards Institute (CLSI M7-A6)[15]. Individual bacterial suspensions at an OD of 0.1 (each for *E. coli*, *S. aureus*, *and P. aeruginosa*) were added to the wells and incubated at 37 °C for 48 h. Subsequently, the Bioscreen C was used to determine the MIC by reading the absorbance of each well visually and comparing the bacterial growth with the control. OD measurements confirmed the visually recorded MIC values. All experiments, including MIC determination and the time-kill assay, were performed in triplicate. Reproducibility was ensured by conducting independent experimental repeats on separate days using freshly prepared bacterial suspensions and treatment solutions.

Time kill assay

This was done to evaluate the time-dependent effect of an Oxygen-rich agent on bacterial cells at various concentrations. The Oxygen-rich agent was tested at its MIC, as well as at 2x and 4x MIC levels for each bacterial strain. A 96-well plate was prepared, where in the first wells for each concentration category, 150 µL of Oxygen-rich agent (MIC x1, MIC x2, or MIC x4) were added. To the remaining wells 180 µL each of MHB was added. To the wells containing the treatment medium, 10 µL of each bacterial suspension at an OD of 0.1 were added. Wells containing each of the bacteria alone and wells containing MHB without bacterial inoculation were processed alongside the experimental groups to ensure the absence of contamination. To obtain the time-kill curve, the growth rate of bacterial strains was counted at different time intervals of 1, 2, 5, and 30 min. Subsequently, serial dilutions were carried out by transferring 20 µL of culture from one well to the next, mixing thoroughly between each step. After dilution, 3 × 20 µL drops (technical replicates) of each dilution were plated onto agar

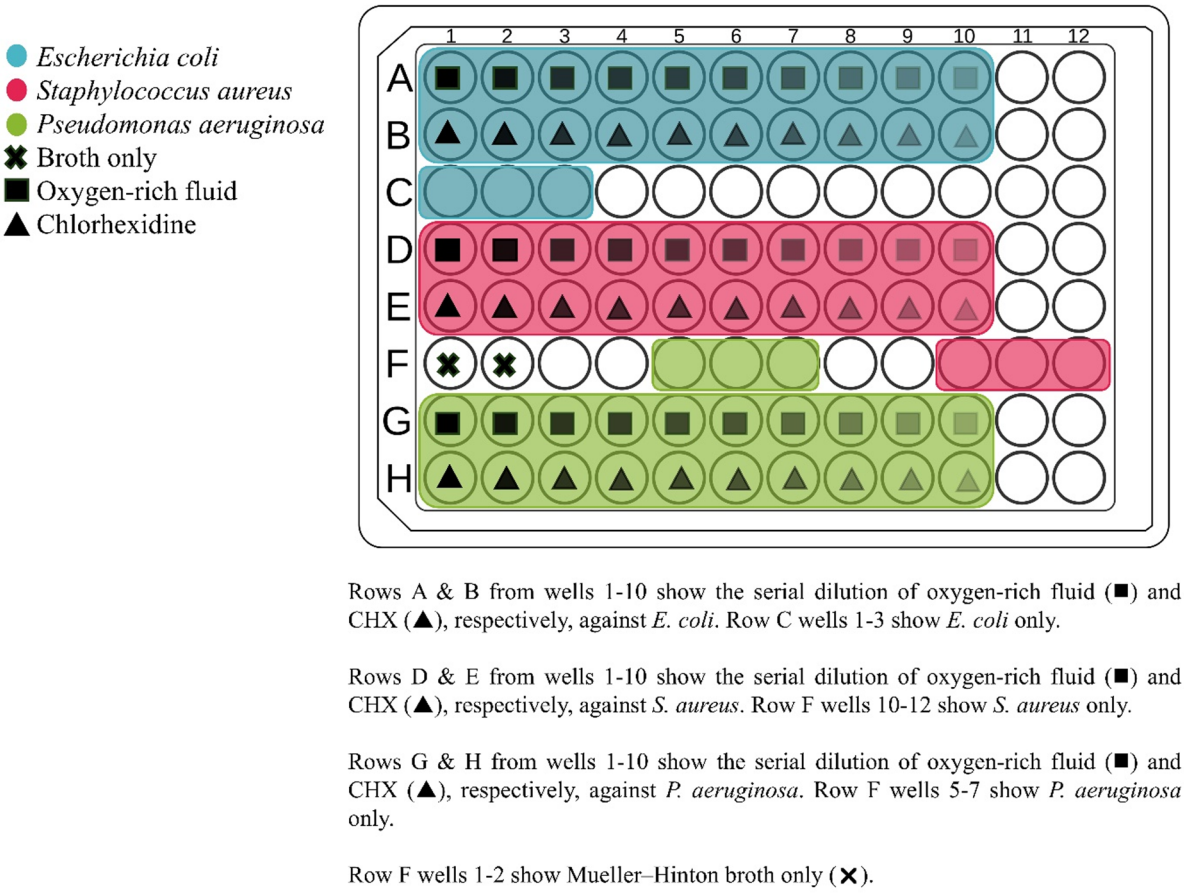


Fig. 1. Illustration of the 96-well plate used with serial microdilution of each tested group.

Bacteria	MIC
Escherichia coli	1.25
Staphylococcus aureus	2.5
Pseudomonas aeruginosa	5

Table 1. Minimum inhibitory concentration (mg/L) of oxygen-rich fluid against peri-implantitis bacteria using microdilution method.

plates. These plates were incubated at 37 °C for 18–24 h, and colonies were counted in the quadrant containing 1–50 colonies per spot[16]. The number of colony-forming units (CFU) per mL was calculated using the formula:

CFU per mL = Average number of colonies for a dilution×50×dilution factor.

Statistical analysis

All the data analysis was performed using SPSS software version 24 (IBM Corp., Armonk, NY, USA; https://www.ibm.com/products/spss-statistics). The continuous data was presented in mean and SD. The continuous data was compared between groups using ANOVA test followed by post hoc Tukey test. The within-group comparison at four-time intervals was compared using Repeated measure ANOVA test. The statistical significance was fixed at $p \leq 0.05$.

Results

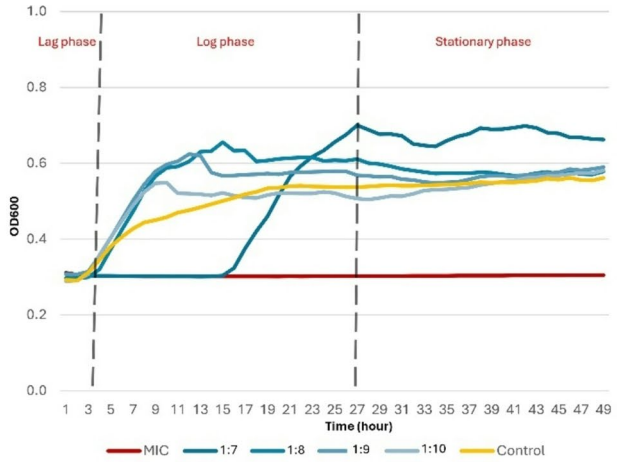
Minimum inhibitory concentration

The MIC values of the Oxygen-rich fluid against the three tested bacterial strains were determined using the broth microdilution method [Table 1]. The results revealed that *Escherichia coli* was the most susceptible bacterium, with an MIC of 1.25 mg/L, indicating the highest sensitivity to the Oxygen-rich fluid. *Staphylococcus aureus* displayed moderate susceptibility, with an MIC of 2.5 mg/L, while *Pseudomonas aeruginosa* exhibited the highest MIC of 5 mg/L, suggesting reduced sensitivity compared to the other bacterial strains. In comparison,

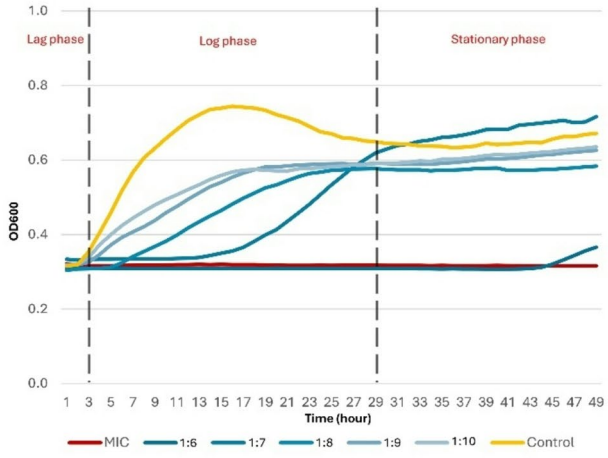
chlorhexidine exhibited no bacterial growth over 48 h for all three of the bacteria [Figure 2]. The full OD values are provided in Supplementary Table S1 online.

Time kill assay

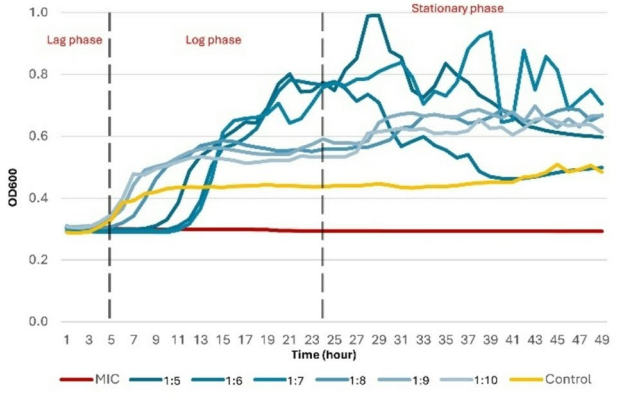
The time-kill assay was conducted to assess the antibacterial activity of Oxygen-rich fluid against *E. coli*, *S. aureus*, and *P. aeruginosa* at three treatment concentrations: MIC x1, MIC x2, and MIC x4. The effectiveness of the treatment was evaluated by measuring the $\log_{10}$(CFU per mL) at different time points (1 min, 2 min, 5 min,



(**a**) *E. coli*



(**b**) *S. aureus*



(**c**) *P. aeruginosa*

Fig. 2. Serial microdilution of oxygen-rich fluid against the bacteria. (**a**) *E. coli* the MIC was 1.25 mg/L (**b**) *S. aureus* the MIC was 2.5 mg/L (**c**) *P. aeruginosa* the MIC was 5 mg/L.

Bacteria	Time interval/concentration	Mean	SD	Minimum	Maximum
E. coli	1 min	3.823	0.192	3.640	4.023
	2 min	3.487	0.163	3.361	3.672
	5 min	3.224	0.228	2.968	3.407
	30 min	2.797	0.229	2.602	3.051
	MIC x1	3.484	0.333	3.051	3.807
	MIC x2	3.193	0.413	2.739	3.640
	MIC x4	3.320	0.580	2.602	4.023
S. aureus	1 min	3.609	0.127	3.489	3.743
	2 min	3.291	0.155	3.159	3.462
	5 min	3.092	0.187	2.978	3.309
	30 min	2.953	0.193	2.791	3.167
	MIC x1	3.384	0.186	3.167	3.598
	MIC x2	3.132	0.260	2.903	3.489
	MIC x4	3.194	0.411	2.791	3.743
P. aeruginosa	1 min	3.557	0.178	3.352	3.677
	2 min	3.064	0.076	2.978	3.119
	5 min	2.920	0.095	2.813	2.995
	30 min	2.722	0.158	2.544	2.845
	MIC x1	3.077	0.214	2.845	3.352
	MIC x2	3.053	0.403	2.778	3.643
	MIC x4	3.067	0.468	2.544	3.677

Table 2. Descriptive statistics of microbial count $\log_{10}$(CFU/ml) across exposure durations and oxygen-rich treatment concentrations.

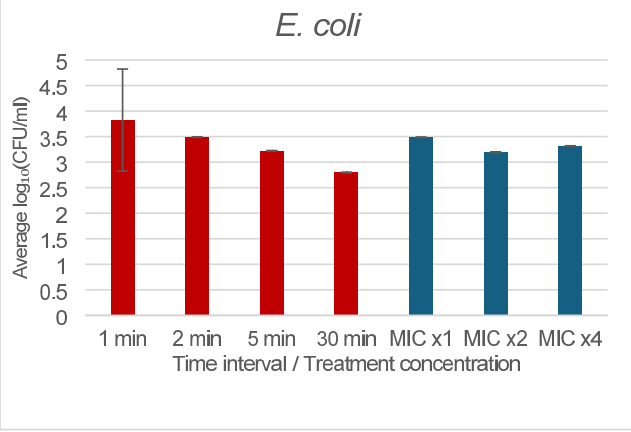


Fig. 3. Descriptive statistics of *E. coli* count $\log_{10}$((CFU/ml) across exposure durations and oxygen-rich treatment concentrations.

and 30 min) CFU represent the number of viable bacterial cells capable of forming colonies in agar, allowing easier comparison of bacterial reductions over time. Full datasets for *E. coli*, *S. aureus*, and *P. aeruginosa* are available in Supplementary Table S2 online.

E. coli

The mean microbial count of *E. coli* showed gradual reduction with time. The microbial count was 3.823, 3.487, 3.224, and 2.797 at 1 min, 2 min, 5 min, and 30 min, respectively. [Table 2] [Figure 3] The difference observed was statistically significant ($p \leq 002$). [Table 3] The post hoc pairwise comparison showed statistically significant difference except between 1 min vs. 5 min (MD = 0.599), 1 min vs. 30 min (MD = 1.026), and 2 min vs. 30 min (MD:0.69). [Table 3] The mean *E. coli* count of 3.484, 3.193, and 3.320 was observed for the treatment concentrations MICx1 MICx2, and MICx4, respectively. [Table 2] [Figure 3] However, none of the differences observed between the treatment concentrations across the three bacteria were statistically significant. [Table 4]

	E. coli			*P. aeruginosa*			*S. aureus*		
	Mean	SD	*P* value	Mean	SD	*P* value	Mean	SD	*P* value
1 min	3.823	0.192	0.002*	3.557	0.178	0.003*	3.609	0.127	0.003*
2 min	3.487	0.163		3.064	0.076		3.291	0.155	
5 min	3.224	0.228		2.920	0.095		3.092	0.187	
30 min	2.797	0.229		2.722	0.158		2.953	0.193	
df	3			3			3		
F value	20.112			16.047			16.617		

Table 3. Comparison of microbial count $\log_{10}$(CFU/ml) at different time intervals using ANOVA and post hoc tests. Repeated measure ANOVA test, *Statistical significance at $p \leq 0.05$

		MD	*P* value	MD	*P* value	MD	*P* value
		E. coli		*P. aeruginosa*		*S. aureus*	
1 min	2 min	0.336	0.178	0.493	0.065	0.318	0.090
	5 min	0.599	0.027*	0.637*	0.047*	0.518	0.061
	30 min	1.026	0.037*	0.835*	0.044*	0.656	0.05*
2 min	1 min	−0.336	0.178	−0.493	0.065	−0.318	0.090
	5 min	0.263	0.149	0.144*	0.007*	0.199	0.026*
	30 min	0.690	0.003*	0.342	0.086	0.338	0.033*
5 min	1 min	−0.599	0.027*	−0.637*	0.047*	−0.518	0.061
	2 min	−0.263	0.149	−0.144*	0.007*	−0.199	0.026*
	30 min	0.427	0.092	0.198	0.216	0.139	0.061

Table 4. Comparison of microbial count $\log_{10}$(CFU/ml) at different oxygen-rich treatment concentrations. LSD test, *Statistical significance at $p \leq 0.0$, MD: Mean difference.

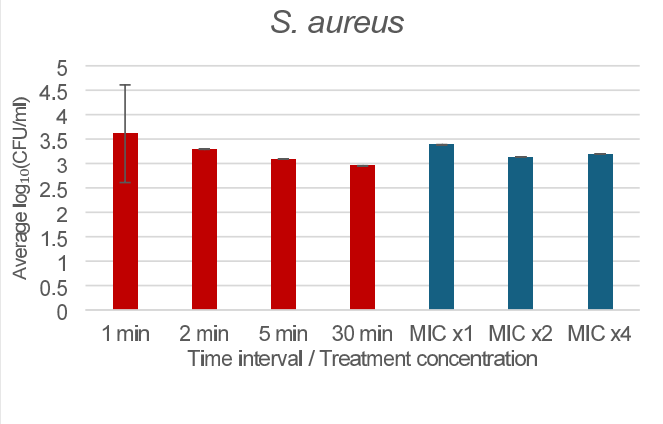


Fig. 4. Descriptive statistics of *S.aureus* count $\log_{10}$(CFU/ml) across exposure durations and oxygen-rich treatment concentrations.

S. aureus

The *S. aureus* microbial count was 3.609, 3.291, 3.092, and 2.953 at 1 min, 2 min, 5 min, and 30 min, respectively. [Table 2] [Figure 4] This shows reduction in microbial count with time progression, and this difference was statistically significant ($p = 0.003$). [Table 3] The following post hoc comparisons 1 min vs. 30 min (MD = 0.656), 2 min vs. 5 min (MD = 0.199), and 2 min vs. 30 min (MD = 0.338) showed statistically significant difference. [Table 3] The mean *S. aureus* count following treatment concentration for MICx1 was 3.384, MICx2 was 3.132, and MICx4 was 3.194 [Table 2] [Figure 4] with no statistical difference between them. [Table 4]

P. aeruginosa

The mean microbial count of *P. aeruginosa* showed reduction with time. At 1 min, 2 min, 5 min, and 30 min they were 3.557, 3.064, 2.920, and 2.722, respectively. [Table 2] [Figure 5] The difference observed in microbial count at different time intervals was statistically significant ($p = 0.003$). [Table 3] The following post hoc pairwise

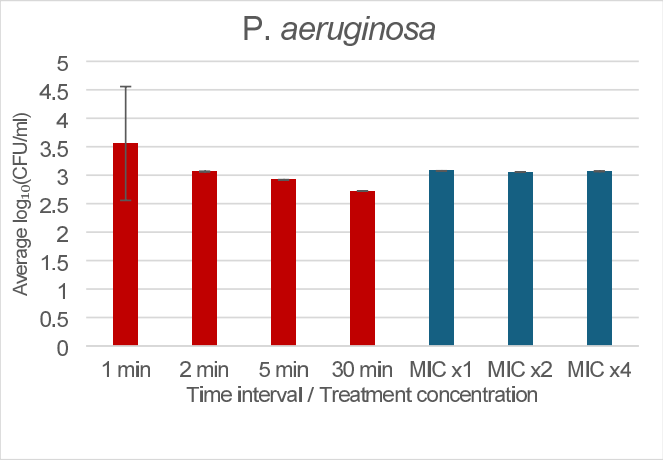


Fig. 5. Descriptive statistics of *P. aeruginosa* count $\log_{10}$ (CFU/ml) across exposure durations and oxygen-rich treatment concentrations.

comparison showed statistically significant difference except between 1 min vs. 5 min (MD = 0.637), 1 min vs. 30 min (MD = 0.835), and 2 min vs. 5 min (MD = 0.144). [Table 3] The mean *P. aeruginosa* count following treatment concentration for MICx1 was 3.077, MICx2 was 3.053, and MICx4 was 3.067. [Table 2] [Figure 5] The difference found between the treatment concentrations was statistically not significant. [Table 4]

Discussion

This study aimed to evaluate the antimicrobial activity of an Oxygen-rich fluid against *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*, with a focus on determining its minimum inhibitory concentration (MIC) and assessing its time-dependent bactericidal effects using a time-kill assay. The oxygen therapy (BlueM) is an oxygen-based therapy capable of gradual and sustained oxygen release, as it is primarily composed of glycerol, lactoferrin, cellulose, and Sodium peroxoborate. Oxygen plays a vital role in cellular metabolism, wherein it participates in various functions, such as the oxidative elimination of bacteria, the regeneration of epithelial tissue, angiogenesis, and collagen formation[17,18]. In a recent in-vitro study evaluating fibroblast cytotoxicity over 14 days comparing oxygen fluid (BlueM) therapy and CHX; oxygen-rich fluid at different concentrations significantly improved fibroblast cell viability, while also preserving the fibroblast cell morphology[19]. In contrast, CHX exposure resulted in a significant decrease in cell viability displaying a cytotoxic effects on fibroblasts[19]. The results of the MIC determination indicated that *E. coli* was the most susceptible to the Oxygen-rich fluid which is consistent with its greater sensitivity to oxidative stress. *S. aureus* displayed moderate susceptibility, while *P. aeruginosa* showed lower sensitivity to the Oxygen-rich fluid. These findings are consistent with the varying susceptibility profiles of these bacterial species, For *P. aeruginosa*, the observed results may be influenced by the bacterium's pigment production, namely, pyocyanin (blue/green), pyorubin (red/brown), and fluorescein (yellow). While pyocyanin and pyorubin are non-fluorescent, fluorescein is fluorescent. The presence of non-fluorescent pigments may interfere with optical density readings, potentially having a greater impact on the measurements[20,21]. *P. aeruginosa* is also known for its intrinsic resistance to many antimicrobial agents due to its robust efflux systems and biofilm forming capabilities[22]. While the Oxygen-rich fluid BlueM remains underexplored, a few studies have evaluated its antimicrobial effectiveness in other formulations, such as the BlueM mouthwash. A study demonstrated that the mouthwash effectively inhibited *Streptococcus mutans* biofilm formation, with MIC value of 0.005%[10]. In this study, Oxygen-rich mouthwash exhibited significant bactericidal and antibiofilm activities, even at low concentrations, which is indicative of its potential effectiveness in controlling oral biofilms, especially against cariogenic bacteria. In another similar study investigating the antimicrobial potential of Oxygenating agents, Ardox-X® mouthwash was evaluated for its efficacy against oral bacteria, including *S. aureus*. Using a serial dilution method, the MIC for *S. aureus* was determined to be 1275 mg/L[23]. Chlorhexidine (CHX) has long been considered the gold standard for antimicrobial treatment in dentistry, particularly in the management of oral biofilms and prevention of gingivitis and peri-implant infections[8]. Despite its effectiveness, CHX's broad-spectrum activity can alter the oral microbiome by reducing microbial diversity, potentially leading to an imbalance in oral flora that could promote the overgrowth of pathogenic species. This shift is characterized by an increase in the abundance of harmful bacteria like Firmicutes and Proteobacteria, and a reduction in beneficial bacteria such as Bacteroidetes and Fusobacteria[24].

The results of the time-kill assay demonstrate the time-dependent antibacterial efficiency of the Oxygen-rich fluid against *E. coli*, *S. aureus*, and *P. aeruginosa*. The bacterial counts decreased significantly across all bacterial strains over time, with the largest reduction occurring at 30 min for each of the bacterial strains. This suggests that the Oxygen-rich fluid has a sustained antimicrobial action over a longer period. The gradual reduction in bacterial growth observed indicates that the Oxygen-rich fluid continuously disrupts bacterial cell structures, likely through the generation of ROS. This is consistent with previous studies highlighting the antimicrobial

effects of Oxygen-releasing agents on bacterial cells[10,11,23]. For *P. aeruginosa*, although the reduction in bacterial count was less pronounced compared to *E. coli* and *S. aureus*, significant decreases were still observed over time.

In terms of concentration, the study found no statistically significant differences in bacterial counts between MIC x1, MIC x2, and MIC x4 for any of the bacterial strains. This can be attributed to bacterial antioxidant defenses, as enzymes that catalyze the rapid degradation of H_2O_2 into water and oxygen called "catalase" that are widely found in different bacterial species. They are known to neutralize excess ROS and limit further bacterial reduction[25]. Within the tested concentration range, the Oxygen-rich fluid is effective at reducing bacterial growth across all concentrations. The lack of significant difference between concentrations might indicate that the fluid works efficiently even at lower doses, which could be beneficial in clinical settings, where lower concentrations can reduce potential side effects while maintaining effective antimicrobial action.

According to current evidence, no prior work has evaluated this Oxygen-rich fluid against peri-implant related bacteria. This study provides a more detailed characterization of both the timing and antibacterial activity, strengthening its potential clinical relevance. The statistical analysis confirmed time-dependent effects, with significant reductions in bacterial counts between time intervals for each strain indicated that bacterial growth was most significantly reduced between the 1-minute and 30-minute time points, suggesting that prolonged exposure to the Oxygen-rich fluid resulted in a more substantial antibacterial effect. The Oxygen-rich fluid's ability to maintain its antimicrobial effectiveness across various concentrations highlights its versatility for both clinical applications and personal daily oral care. This property is especially relevant in clinical settings, such as the treatment of peri-implant tissues or post-surgical wounds, where the treatment may be diluted by oral fluids, yet it continues to effectively target pathogens.

Limitations within this study include the limited selection of bacterial strains (*E. coli*, *S. aureus*, and *P. aeruginosa*) being evaluated, which restricts a broader application of the findings to other pathogens commonly associated with peri-implantitis. Also, the study's in-vitro design does not fully replicate the complex conditions of the oral environment. The planktonic assays may overestimate clinical effectiveness compared to biofilm models. Further in-vivo studies are needed to confirm clinical applicability of the treatment. Additionally, evaluating the treatment on biofilm models would provide a more comprehensive understanding of its effectiveness. Another limitation is the absence of repeated-use assessment; while the study focused on single exposure, long-term clinical use of the Oxygen-rich fluid might be essential for managing chronic infections. Understanding its effects with repeated applications is crucial for assessing safety and efficacy over time.

Conclusion

In conclusion, the Oxygen-rich fluid exhibited significant time-dependent antibacterial activity against *E. coli*, *S. aureus*, and *P. aeruginosa*, with no substantial difference observed between concentrations, suggesting its continued action even when the concentration was diluted. The study provides evidence supporting the potential use of the Oxygen-rich fluid as an effective antimicrobial agent in peri-implant therapy, but further research including in-vivo and biofilm studies are needed to evaluate its long-term effectiveness and clinical applicability in treating complex biofilm-related infections.

Data availability

All data generated or analyzed during this study are included in this published article (and its Supplementary Information files).

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Author contributions

L.K.: conceptualization, methodology, formal analysis, investigation, visualization, data curation, project administration and writing original draft. L.K., M.A. and S.A.: review and editing. M.A. and S.A.: supervision.

Declarations

Competing interests

The authors declare no competing interests.

Additional information

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