RESEARCH ARTICLE

Anti-bacterial Activity of Polyethylene Oxide (PEO) against Some Pathogenic Bacteria Isolated from Human Mouth

Mohammed A. Jawad,[1*] Abed J. Kadhim[1], Saif Y. Hasan[2]

[1]*Al-Nisour University College, Baghdad, Iraq*
[2]*National University of Science And Technology, Thi-Qar, Iraq*

Received: 13th May, 2021; Revised: 21st June, 2021; Accepted: 03rd August, 2021; Available Online: 25th September, 2021

ABSTRACT

Background: Polyethylene oxide (PEO) is a non-ionic linear hydrophilic and uncrosslinked polymer available in several molecular weights. It is synthesized by ethylene oxide and has many desirable properties for drug delivery applications and antimicrobial.

Materials and Methods: In the present study, polyethylene oxide (PEO) with different concentrations (80, 40, 20, 10 µg/mL) investigates their anti-bacterial activity against two pathogenic bacteria from gram-positive *Streptococcus pyogenes* and *Staphylococcus aureus* and gram-negative *Escherichia coli* and *Enterobacter bugandensis*. The antimicrobial activity of PEO was examined by disk diffusion assay also, the minimal inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of each isolate is determined.

Results: The PEO shows powerful broad-spectrum anti-bacterial activity against tested bacteria with an increase in inhibition zone diameter that is directly proportional with the increase in PEO concentration that even exceeded the activity of selected antibiotics. The MIC of PEO ranged from 10 to 20 µg/mL, and the MBC ranged from 20 to 80 µg/mL. Other studies show that PEO strongly attached to the bacterial cells contributed to their inhibitory effect on bacterial growth formation and invasion.

Conclusion: The PEO with a suitable concentration are reduced bacterial growth significantly. It is highly recommended to use PEO as an economical alternative anti-bacterial agent, especially in treating ectopic infections without taking the risk of developing resistant bacterial strains as with antibiotics.

Keywords: Anti-bacterial activity, MIC, Polyethylene oxide (PEO).

International Journal of Pharmaceutical Quality Assurance (2021); DOI: 10.25258/ijpqa.12.3.4

How to cite this article: Jawad MA, Kadhim AJ, Hasan SY. Anti-bacterial Activity of Polyethylene Oxide (Peo) against Some Pathogenic Bacteria Isolated from Human Mouth. International Journal of Pharmaceutical Quality Assurance. 2021;12(3):187-190.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

The human oral cavity is one of the most active environments for multiple bacterial species, where they compete fiercely for space in multispecies biofilm structures. *Streptococcus*, *Lactobacillus*, *Lactococcus*, *Enterococcus*, *Staphylococcus*, *Corynebacterium*, *Veillonella*, and *Bacteroids* are the most frequent bacteria identified in the oral cavity.[1,2] *Streptococcus* and *Enterococcus* are two essential members of the oral bacteria family because they may switch from helpful microflora on the surface of the mouth cavity and oropharynx to harmful pathogens once they get access to the oral tissue and bloodstream. Dental caries, periodontitis, endocarditis, pharyngitis, pneumonia, and meningitis are only a few illnesses caused by oral bacteria. The mass of oral *Streptococcus* are gram-positive facultative anaerobes with extremely effective survival strategies include the capacity to attach to hard and soft tissues, cell-cell communication, biofilm formation, and the ability to adapt with quickly changing oral etiologies.[3]

To colonize the oral cavity, a bacterium must compete with other microorganisms. As a result, they have a lot of intra-species and inter-species communication, which helps them survive in the harsh environment of the oral cavity.[3] Production of bacteriocin is an important means of outcompeting other bacteria in this heterogeneous environment. Many gram-positive bacteria produce bacteriocins which act as toxins against other bacteria, however, the producer strain is immune to its bacteriocin due to the immunity factor.[4-6] Since the oral environment is very competitive, and it is speculated that bacterial species isolated from such environments will produce inhibitory substances against other bacteria.[7]

The PEO is a neutral, non-toxic, biocompatible, and water-soluble polymer that has found numerous applications, such

**Author for Correspondence: mohammed.a.medical.lab@nuc.edu.iq*

as in conductive composites with carbon black, cosmetology (skin creams, emulsions, personal lubricants), gene therapy, pharmaceutical products, etc.[8-13]

To enhance their favorable properties and tailor their capabilities, PEO-based graft copolymers have been investigated for their wide range of promising abilities.[11,12] These materials have found applications in nanotechnology, lithium batteries, elastomer fabrication, drug delivery systems,[9,13-17] and biomedical implants.[18,19]

This study focused on the isolation of pathogenic bacteria from the human mouth and their drug sensitivity patterns. Also, the anti-bacterial activity of PEO was studying against pathogenic bacteria isolated from the mouth to evaluate their activity.

MATERIALS AND METHODS

Bacterial Isolates

The bacterial isolates were obtained from the mouth infection patients in Teaching Hospital in Hillah, Iraq. All samples were subjected to standard bacteriological processes of culturing on blood and MacConkey's agar plates for 24–48 hours at 37°C for isolation and purification. All isolates were confirmed by Vitek 2 compact system (Biomérieux).

Solution and Media

Mueller-Hinton agar and Mueller-Hinton media were obtained from Hi-Media, Mumbai, India. PEO and DMSO were supplied from (Zhengzhou Dongyao Nano Materials Co., Ltd. China). Different antibiotic disks ciproflolothin) KF-30), Doxycyclin (DO-30), Methicillin (ME-5), Clarithromycin (CLR-15), and Novobiocin (NV-5) were purchased from (Bioanalyse, Turkey).

Anti-bacterial Activity of PEO

PEO antimicrobial activity was tested against bacteria isolated from the human mouth, two G-ve bacteria (*E. coli* and *E. bugandensis*) with two G+ve bacteria (*S. pyogenes* and *S. aureus*) maintained on nutrient agar slants. The antimicrobial activity was carried out as described by the Clinical and Laboratory Standards Institute.[20] Antibiotic sensitivity and PEO against bacteria under study are tested using a disk diffusion assay, with triplicates used in dilutions of concentration of PEO (80, 40, 20, and 10 µg/mL) in sterile deionized water. In the first step, the isolates were incubated for 15 minutes at room temperature, then incubated at 37°C overnight. Positive results were recorded when the inhibition zone was observed around the well after a period of incubation, and then, the inhibition zone diameter was measured using a digital Vernier caliper.[21]

Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) Determination[22]

The bacterial isolates were incubated at 37ºC overnight, which was used to prepare 0.5 McFarland. A total of 10 mL tube nutrient broth medium was prepared then each sample was inoculated aseptically with 1-mL of the respective bacterial suspension(about 108 CFU/mL). Four dilutions of PEO were prepared (80, 40, 20, and 10 µg/mL) in sterile deionized water, and negative control (without PEO) was used. Tests were performed in triplicates for each isolate. The inoculated sets were incubated at 37ºC overnight. After the incubation period, the visible turbidity in each tube was investigated. The lowest concentration with no turbidity is represented as the MIC for the tested strain. Tubes that showed no turbidity were cultured on nutrient agar plates and incubated at 37ºC overnight. Bacterial colonies growth was checked, and the concentration that shows no growth is represented as the MBC.

RESULTS AND DISCUSSION

The results of the current study (86 samples) revealed that predominant bacterial isolates from odontogenic infections were *S. pneumonia* (38.3%) followed by *S. aureus* (27.9%) as Gram-positive, as well as gram-negative bacteria *E. coli* (20.9) and *E. bugandensis* (12.7). The results are shown in Table 1.

The bacteria of each type were subjected to an antibiotic sensitivity test by modified Kirby-Bauer disc diffusion. Selective antibiotics are most commonly used in odontogenic infections to show their effect on different groups as shown in Figure (1–4).

Anti-bacterial Activity of PEO

PEO shows that powerful broad-spectrum anti-bacterial activity against multidrug bacteria is tested. The effects of different antibiotics on bacterial isolates were compared. Figures (1 to 4) showed that the selected antibiotics were not effective against all selected bacterial strains. PEO showed clearly inhibition zone diameter with the decrease in PEO concentration that even exceeded the activity of selected antibiotics. 10 µg/mL concentration showed the highest zone of inhibition against the test organisms, a maximum zone of inhibition of 18 mm appeared against *S. aureus* (Figure 1), and the least sensitive

Table 1: Number and percentage of bacterial isolates from odontogenic infection.

Bacterial Isolates	*Total*	*%*
Streptococcus pneumonia	33	38.3
Staphylococcus aureus	24	27.9
Escherichia coli	18	20.9
Enterobacter bugandensis	11	12.7
Total	86	100

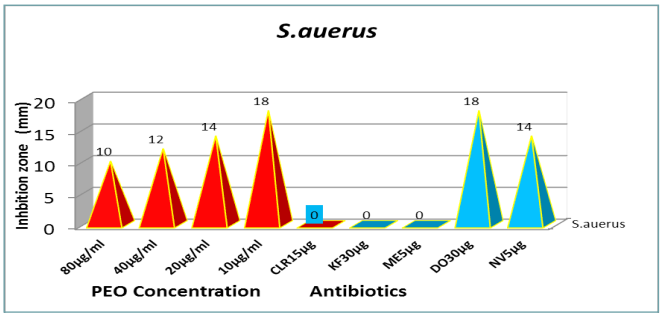

Figure 1: Anti-bacterial action of PEO on *S. aureus*

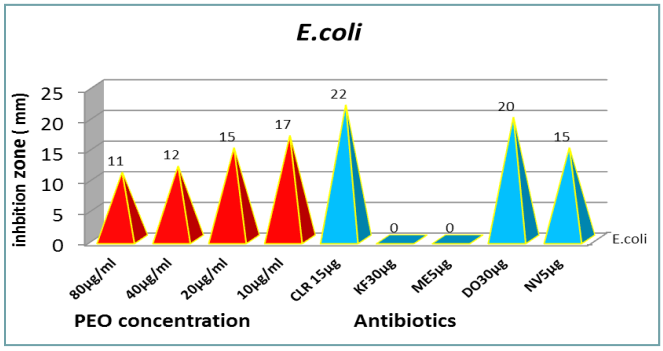

Figure 2: Anti-bacterial action of PEO on *E. coli.*

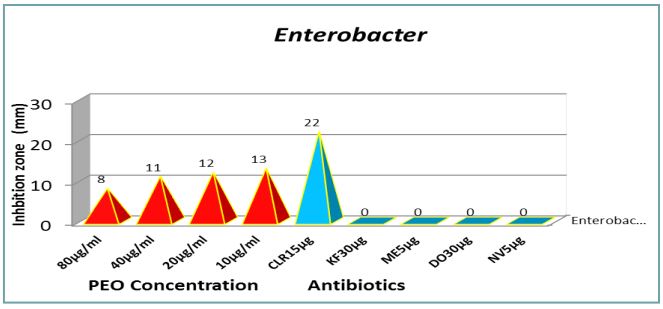

Figure 3: Anti-bacterial action of PEO on *Enterobacter*

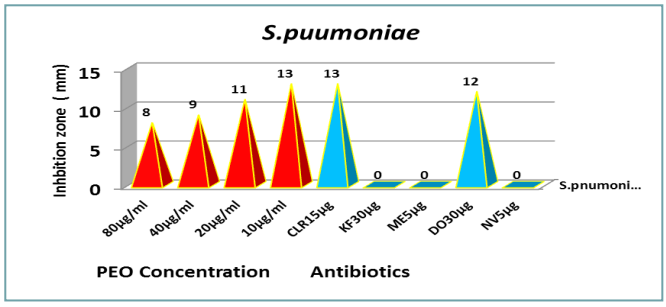

Figure 4: Anti-bacterial action of PEO on *S. pneumonia*

Table 2: MIC and MBC of PEO for some pathogenic bacteria

Bacterial isolates	*MIC*	*MBC*
Streptococcus pneumonia	10 µg/mL	20 µg/mL
Staphylococcus aureus	20 µg/mL	40 µg/mL
Escherichia coli	20 µg/mL	80 µg/mL
Enterobacter bugandensis	20 µg/mL	40 µg/mL

isolate compared with the selected antibiotics followed by *S. pneumonia* (Figure 4). The second sensitive isolate to PEO is *E. coli* (Figure 2), finally *E. bugandensis* (Figure 3).

PEO causes a sudden decline in bacterial cell membrane integrity in addition to the release of reactive oxygen species (ROS), where superoxide species is generated and contributing to the degradation of biomolecules.[23] Minimal residual disease was defined as acquired non-susceptibility to at least one agent among three or more anti-bacterial antibiotics, or categories[24] result has been agreed with the Zhang and Chen[25] shown that PEO could be inhibited the multidrug-resistant (MDR) bacteria.

Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) Determination

Table 2 shows that the MIC of PEO ranged from 10 to 20 µg/mL and the MBC ranged from 20 to 80 µg/mL where *S. aureus* showed the highest sensitivity followed by other bacteria.

CONCLUSION

This study showed that PEO has a considerable inhibitory and anti-bacterial effect on the selected pathogenic bacterial isolates from a human mouth.[26] It is highly recommended to use PEO as an economical alternative anti-bacterial agent, especially with materials that make toothpaste, mouthwashes, and dental fillings because of its effective ability to inhibit bacterial growth.

REFERENCES

1. Rogers A. Molecular Oral Microbiology. Norfolk, UK: Caister Academic Press. 2008.
2. Wang QQ, Zhang CF, Chu CH, Zhu and XF. Prevalence of Enterococcus faecalis in saliva and filled root canals of teeth associated with apical periodontitis. Int. J. Oral Sci. 2012;4:19-23.
3. Cvitkovitch DG, Li YH, and Ellen RP. Quorum sensing and biofilm formation in Streptococcal infections. J. Clin. Invest. 2003; 112:1626-1632.
4. Jack RW, Tagg JR, Ray B. Bacteriocins of grampositive bacteria. Microbiol. Rev. 1995;59:171-200.
5. Tagg JR, Dajani AS, Wannamaker LW. Bacteriocins of gram-positive bacteria. Bacteriol. Rev. 1976; 40:722-756.
6. Hossain MS, and Biswas I. SMU152 acts as an immunity protein for mutacin IV. J. Bacteriol. 2012;194: 3486-3494.
7. Weerkam P A, Bongaerts-Larik L, and Vogels GD. Bacteriocins as factors in the in vitro interaction between oral streptococci in plaque. Infect. Immun. 1977; 16:773-780.
8. Gueugnon F, Denis I, Pouliquen D, Collette F, Delatouche R, Héroguez V, Grégoire M, Bertrand P, Blanquart C. Nanoparticles Produced by Ring-Opening Metathesis Polymerization Using Norbornenyl-poly(ethylene oxide) as a Ligand-Free Generic Platform for Highly Selective *In Vivo* Tumor Targeting. Biomacromolecules. 2013;14:2396-2402.
9. Xue Z, He D, Xie X. Poly(ethylene oxide)-based electrolytes for lithium-ion batteries. J. Mater. Chem. A. 2015;3:19218-19253.
10. Quémener D, Chemtob A, Héroguez V, Gnanou Y. Synthesis of latex particles by ring-opening metathesis polymerization. Polymer.2005;46:1067-1075.
11. Bates CM, Chang AB, Momcîilovic N, Jones SC, Grubbs RH. ABA Triblock Brush Polymers: Synthesis, Self-Assembly, Conductivity, and Rheological Properties. Macromolecules 2015, 48, 4967-4973.
12. Neugebauer D. Graft copolymers with poly(ethylene oxide) segments. Polym. Int. 2007;56:1469-1498.
13. Gao AX, Liao L, Johnson JA. Synthesis of acid-labile PEG and PEG-doxorubicin-conjugate nanoparticles via Brush-First ROMP. ACS Macro. Lett. 2014;3:854-857.
14. Zhou H, Schön EM, Wang M, Glassman MJ, Liu J, Zhong M, Díaz DD, Olsen BD, Johnson JA. Crossover experiments applied to network formation reactions: Improved strategies for counting elastically inactive molecular defects in PEG gels and hyperbranched polymers. J. Am. Chem. Soc. 2014;136:9464-9470.

15. Yaqo, EA, Anaee, RA, Abdulmajeed, MH, Tomi, IHR, Kadhim, MM; Electrochemical, morphological and theoretical studies of an oxadiazole derivative as an anti-corrosive agent for kerosene reservoirs in Iraqi refineries ;Chemical Papers, (2020) 74 (6);1739-1757.
16. Al-Jbouri, AMS., Abood, FM, Hindi, NKK, Alkaim, AF; Evaluation of antimicrobial activity of the aquatic extract against bacterial isolates from periodontitis in Babylon Province, Iraq. Biochemical and Cellular Archives, (2018) 18; 1345-1350.
17. Al-Duhaidahawi, DL, Jabir, MS, Al-Amiery, AA, Moneim, AEA, Alkaim, AF, Kadhum, AAH; Novel antioxidants compounds derived from isatine Biochemical and Cellular Archives, (2018)18 (1); 709-713.
18. Radder AM, Leenders H, van Blitterswijk CA. Application of porous PEO/PBT copolymers for bone replacement. J. Biomed. Mater. Res. 1996;30:341-351.
19. Gasteier P, Reska A, Shult P, Salber J, Offenhäusse A, Moeller M, Groll J. Surface grafting of PEO-based star-shaped molecules foe bioanalytical and biomedical applications. Ma c romol. Bios ci. 20 07;7:1010 -1023.
20. Clinical and Laboratory Standards Institute.Performance Standards for Antimicrobial Susceptibility Testing; Twenty-First Informational, Supplement. CLSI Document M02-A10 and M07-A8. Texas: Clinical and Laboratory Standards Institute; 2012;3:22-44.
21. CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 26th ed. CLSI Supplement M100S. Wayne, PA: Clinical and Laboratory Standards Institute. Clinical and Laboratory Standards Institute.2006;22-45.
22. Kim SC, Lee DK. Preparation of TiO2-coated hollowglass beads and their application to the control of algalgrowth in eutrophic water. Microchem J.2005;80:227-232.
23. Zhang H, Chen G. Potent anti-bacterial activities of Ag/ TiO2 nanocomposites powders synthesized by a one-potsol-gel method. Environ Sci Technol.2009;34:2905-2910.
24. Clinical and Laboratory Standards Institute.Performance Standards for Antimicrobial Susceptibility Testing; Twenty-First Informational, Supplement. CLSI Document M02-A10 and M07-A8. Texas: Clinical and Laboratory Standards Institute. 2012;3:5678
25. Vargas KF, Borghetti RL, Moure SP, Salum FG, Cherubini K, Figueiredo MAZ. Use of polymethylmethacrylate as permanent filling agent in the jaw, mouth and face regions--implications for dental practice. Gerodontology. 2012;29:e16-e22. doi:10.1111/j.1741- 2358.2011.00479.