Polyethylene Oxide(PEO) as antibacterial activity against Azo Days Bacterial Isolates with clinical application

Zaid Al-Nuaimi
University of Al-Ameed Faculty of Dentistry, Zaidalneame88@gmail.com

Mustafa A Zeinalabdeen
University of Alkafeel, College of Dentistry, Najaf, Iraq

Wahhab R Jassi
University of Alkafeel, College of Dentistry, Najaf, Iraq

Abstract

Background: Poly ethylene oxide (PEO) is a non - linear system hydrophilic and uncross linked polymer with a variety of molecular weights. Ethylene oxide is used to make it, and it offers a number of beneficial characteristics for medication delivery and antibacterial uses. PEO is effectively integrated into amorphous calcium phosphate (ACP) during its synthesis, it is predicted to influence ACP's inclination to form aggregates.

Methodology: In the present study, polyethylene oxide (PEO) with different concentration '(80, 40, 20, 10 μg/ml)' investigates their antibacterial activity, against five pathogenic bacteria isolated from azo dye (random selection from total isolates). The disk diffusion test was used to evaluate the antimicrobial activity of PEO, as In addition, each isolate's 'minimum inhibitory concentration (MIC)' and 'minimum bactericidal concentration (MBC)' were determined.

Results: PEO exhibits strong broad-spectrum antibacterial action against tested bacteria, with an inverse relationship between an increase in 'inhibition zone diameter' and a decrease in PEO concentration, and even outperforms the action of certain medicines. PEO had 'MICs of 10 to 20 g/ml and MBCs of 20 to 80 g/ml'. PEO was shown to be firmly adhered to bacterial cells in other experiments, its inhibitory impact on bacterial growth and invasion might be attributed to this factor.

Conclusion: The bacterial growth was considerably inhibited by PEO at a suitable concentration. It is strongly suggested that PEO be used as a cost-effective antibacterial agent, particularly when mixed with dyes used at home or in industries, to avoid the possibility of creating antibiotic-resistant bacterial strains.PEO has a good effect on filer and composite properties like ACP.

Keywords: *Antibacterial activity, PEO, Azo deys, Antibacterial, ACP.*

Introduction

The term azo comes from the French azote. Its discovery of dyes a watershed moment in history of the chemical industry. Azo dyes comprise one or more azo linkages and are made up of a 'diazotized amine' linked to an 'amine' or aphenol. Aromatic amines are the most important precursors for azo dyes. Azo dyes are chemical molecules with the functional group 'R-N=N-R′', with 'R and R′' generally being 'aryl'. They are azo compounds with the bond 'C-N=N-C',

which are commercially relevant. (No. 1) Azo pigments are chemically related to azo dyes and are insoluble in water and other solvents. [2,3] Although many ‘azo pigments’ are non-toxic, some are mutagenic and ‘carcinogenic’, such as ‘di-nitro aniline orange, ortho-nitroa niline orange, or pigment orange 1, 2, and 5’.[4,5] ‘Azo dyes’ are the most common and ‘versatile dyes’, accounting for more than half of all dyes made globally. ‘Azo dyes’ produced from benzidine are carcinogenic, and exposure to them has been linked to bladder cancer in the past. As a result, in the 1980s, "the most important Western industrialized countries" stopped producing benzidine azo dyes. [6]

At least 3,000 dyes were obtainable in past widely charity to color textiles, leather, some foods, coloring pens, shoes, printing inks, paints, varnish, lacquer, tattoo inks, cosmetics, hair dyes, waxes, and wood, some of which may pose a risk to human and environmental health due to their toxic properties and contamination by various organisms. [7,8] This problems of azo dyes can moving by existence many microorganisms like bacteria, fungi have been reported to eco-friendly and other may case infection disease [9]. Many of these organisms such bacteria are pathogenic and caused infection to humans in very high, especially these dyes are used daily by humans and children. Only a few aerobic bacteria, such as ‘Bacillus cereus, Bacillus subtilis, Escherichia coli, Enterococcus fecalis, Pseudomonas aeruginosa, Streptomyces cereviceae, and Candida zeylanoides’, have been discovered in azo dyes under aerobic conditions. [10-16] Dye bacteria from textile azo dyes have been isolated and described in a number of investigations [17–20]. Many investigations isolated bacteria (Bacterium firmus and Halomonas sp., respectively) and other dye bacteria (Pseudomonas aeruginosa [21] and Comamonas sp. UVS [22]. However, due to the alkaline environment, only a few studies have been done to extract and describe bacteria that have contaminated the azo dye. [23-25] This environment necessitates the existence of alkaliphilic and halophilic bacteria that can adapt and operate physiologically under such extreme circumstances. Humans are exposed to xenobiotics such as azo dyes by food, skin contact, or inhalation, and some pathogenic microorganisms found in azo dyes can cause illness. [26]

Polyethylene oxide \(PEO)’ is a non-poisonous, biocompatible, and water-solvent polymer that has a wide scope of employments, incorporating conductive composites with ‘carbon dark, cosmetology (skin creams, emulsions, individual oils)’, quality treatment, and medicinal goods [27-33]. PEO-based graft copolymers have been studied for a variety of potential characteristics, as well as ways to improve and modify their capabilities. [31,32] Nanotechnology, lithium batteries, elastomer manufacturing, drug delivery systems [28,33–36], and biomedical implants have all benefited from these materials. [37,38]. The effect of various of the molecule size dispersion, composition, and characteristics for ACP fills Furthermore, the steadiness of mechanical of amalgams produced through such ACP plasters afterward delayed submersion in water was measured. Surfactants added stomach muscle initio during the combination of ACP were relied upon to adsorb at the superficial of ACP, impacting the level of unconstrained ACP total. [39] Accumulated ACPs anticipated to scatter better in sap grids. As a result of its set up ability to lead various hydrogen holding communications and settle cations by means of numerous chelation, PEO is hydrophilic primary component present numerous surfactants, where as often as possible utilized in water-viable polymer frameworks. When PEO is effectively integrated into ACP during

its synthesis, it is predicted to influence ACP's inclination for procedure aggregates. The content of water to the precipitated ACP filler may be affected by both the PEO and surfactant additions. [37-40].

The aim of this study is to isolation and identification of bacterial contamination the different azo dye can used daily by human like lether clothes dyes, shoes dyes, childrens coloring pens may be cause toxicity and many human infection, and using PEO as antibacterial to proofing their capability to reduce azo dyes bacteria to attempt to mixing any type of polymer with azo dyes in their industry to reduce the human infection by bacteria.

MATERIALS & METHODS:

Collection of Samples

The 75 specimens were collected from the different samples of coloring dyes, dyes shoes and leather clothes dyes. Azo dye samples are collected, and the samples size included 75 azo dye samples (N=75) from lether dyes (N= 25), from shoos dye sample(N=25), and from children coloring pens (N=25) . Azo dyes collected from different locations in many house in Al-Hilla city.

Antibiotic susceptibility test:

The clinical laboratory standard Institute (CLSI) recommendations 2016 were used to evaluate antibiotic susceptibility using the Kirby-Bauer disk diffusion techniques. Nitrofurantion, Cefotaxime, Aztronam, Cephalosporins, Amoxicilin, and Mrthicillin were among the six antibiotics used to evaluate the drug resistance of five Bacillus cereus isolates (chosen at random from the total isolate). The results were expressed as the rate of resistant strains among all detected bacterial strains. MDR is defined as resistance to three or more antimicrobial classes. The bacterial strains represent MDR.

Antibacterial Properties of PEO

The antibacterial properties of PEO was evaluated against certain human pathogens that were obtained from azo dyes kept on nutritional agar slants. The Clinical and Laboratory Standards Institute's recommendations were followed while testing antimicrobial activity. [39] 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 solvent. The first step, the isolates were incubated for 15 min at room temperature, then incubated at 37°C overnight. After a time of ‘incubation’, the ‘inhibition zone’ was seen surrounding the well, A digital Vernier caliper was used to determine the width of the inhibitory zone. [40]

Preparation of Composite

Hand spatulation was used to create composite pastes by combining the BT pitch (60 mass percent) with the necessary ACP filler (40 mass percent). To eliminate the air entrained subsequent to blending, the homogenized adhesives were held below a reasonable vacuum (2.7 kPa) present moment. Satisfying the round beginnings of level Teflon casts with the pastes, covering each side of the structure in a ‘Mylar film’ notwithstanding a glass slip, then joining the assembling using coil cuts, the pastes were formed hooked on circles ‘(15.8 mm to 19.6 mm in distance across and 1.55 mm to 1.81 mm thick)’.The plates were photopolymerized by uncovering each face of the shape gathering to apparent light for 120 seconds straight.

RESULTS AND DISCUSSION:

A total 75 isolates of B. cerus were isolated and identified by stander microbiological procedure then by 'VITEK' 2. The rate of 'B.cerus' from lather clothes dyes was 25 (33.33%), The rate of B.cerus from shoes dyes was 25 (33.33%),and The rate of B.cerus from children coloring dyes was 25 (33.33%), All isolates were confirmed by Vitek 2 compact system (Biomérieux), Table (1).

Table (1): Distribution of B. cerus from different clinical sample

Source of Isolate	No. of specimens	Percentages %
Lather clothes dyes	25	33.33%
Shoes dyes	25	33.33%
Children coloring pen	25	33.33%
Total	75	100%

Antibiotic sensitivity testing was done on each kind of bacteria using a modified Kirby- Bauer disc diffusion method. To demonstrate their effect on distinct groups, selective antibiotics are often employed against 'B. cerus' infection, as indicated in the figure (1-5). [42]

Antibacterial Activity of 'PEO'

'PEO' shows that powerful broad-spectrum antibacterial movement against multidrug microscopic organisms is tried. The impacts of various anti-infection agents on bacterial detaches were looked at. The outcome in Figures (1 to 5), showed that the chose antibiotics were not effective against all isolated bacterial under study. PEO demonstrated a distinct inhibitory zone width decrease as PEO concentration dropped, which even outperformed the action of certain antibiotics. The greatest zone of inhibition against the test organisms was found at a dose of 10 g/ml, 'maximum zone' of inhibition of 19 mm appeared against B.cerus 4 Fig.(4) and the least sensitive isolate in comparison with the selected antibiotics followed by B. cerus 2 and 3 Fig.(2,3) .The second sensitive isolate to PEO is B. cerus 1 and 5 Fig.(1,5).

PEO causes a rapid loss of bacterial cell membrane integrity, as well as the production of 'reactive oxygen species (ROS)', including 'superoxide species', which contributes to biomolecule destruction.[23] Among more than three antibacterial 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.

Figure (1): Antibacterial action of on B.cerus1

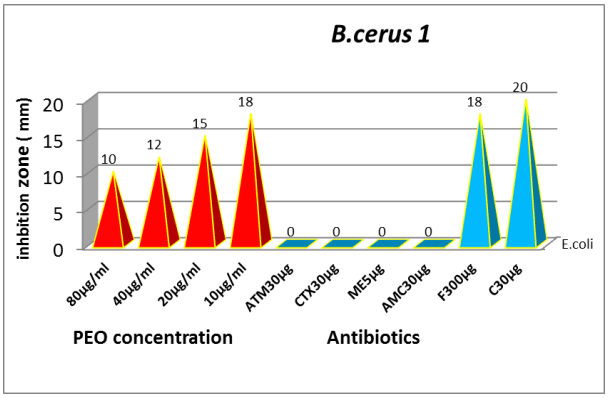


Figure (2): Antibacterial action of on B.cerus2

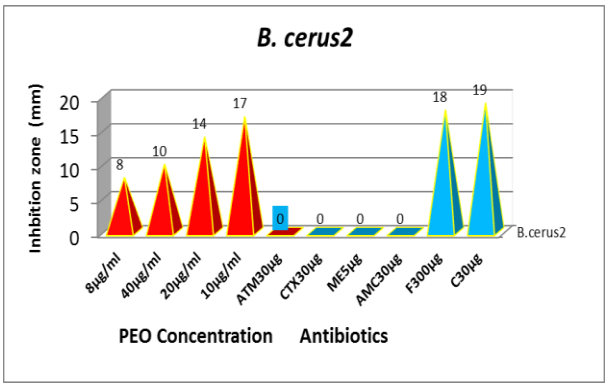


Figure (3): Antibacterial action of on B.cerus3

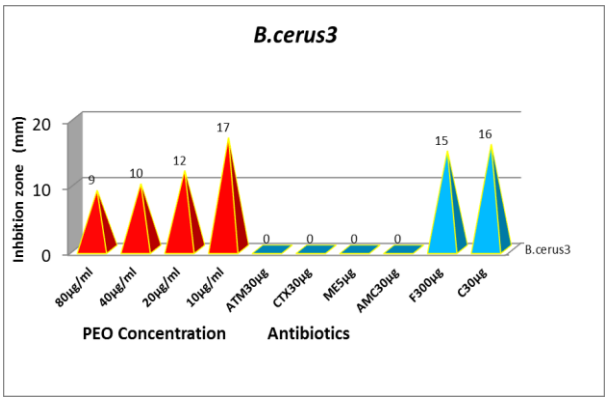


Figure (4): Antibacterial action of on B.cerus4

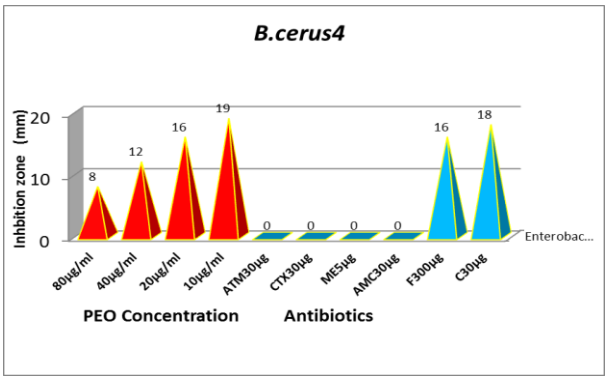
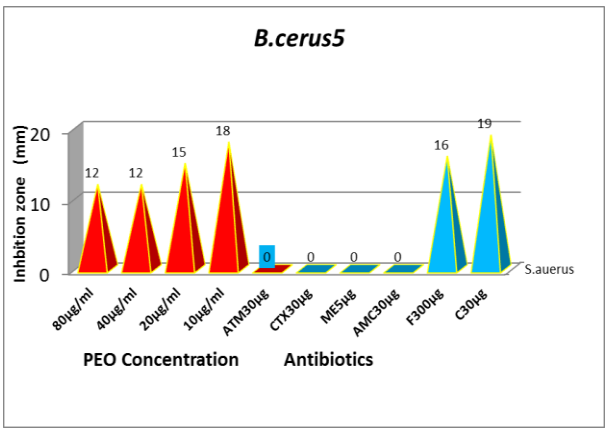


Figure (5): Antibacterial action of on B.cerus5



'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 B. cerus 4 showed the highest sensitivity followed by other bacteria. [46]

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

Bacterial Isolates	MIC	MBC
B. cerus 1	**10µg/ml**	**20 µg/ml**
B. cerus 1	**10 µg/ml**	**40 µg/ml**
B. cerus 1	**20 µg/ml**	**40 µg/ml**
B. cerus 1	**20 µg/ml**	**80 µg/ml**
B. cerus 1	**20 µg/ml**	**40 µg/ml**

The impact of different PEO, a hydrophilic, nonionic polymer, and surfactants presented stomach muscle throughout the union ACP in the design, organization, morphology, of encouraging ACP examined in this review. The speculation was associations amid ACP with surfactants as well as PEO presented at the beginning would decrease the degree of ACP conglomeration without influencing its soundness in fluid conditions, safeguarding the morphology and bioactivity of 'ACP filler as a calcium and phosphate' delivering specialist.[47] We found next to no exploratory proof to help the possibility that the substance kind of the added substance or, on account of PEO, an atomic mass reliance. The molecule size of ACP was just insignificantly diminished within the sight of the anionic surfactant. This current surfactant's horrible showing may be credited to the incomplete scattering of headgroup custodies for the relaxation of particle, a marvels regular of surfactants in direct 'alkyl tails and sulfonate, sulfate', or potentially 'carboxylate headgroups'.[48] ACP hastened within the sight of PEO, then again, looked substantially more collected (autonomous of PEO sub-atomic mass). As per reports, a "polymer connecting" system directs the total of HAP elements within the sight of

sub-atomic form polyacrylate by means restricting of this polymeric added substance's carboxylic gatherings to positive surface calcium destinations.[49]

CONCLUSION:

PEO has a significant inhibitory and antibacterial impact on selected pathogenic bacterial isolates from azo dyes, according to the findings of this investigation. It is highly recommended using PEO as an economic alternative anti-bacterial agent especially with materials that make coloring dyes, dyes shoes and leather clothes because of its effective ability to inhibit bacterial growth. When compared to the control 'Zr-ACP' composites, the surfactant- and 'PEO-ACP' composites showed no improvement in 'dry biaxial flexure strength'.

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