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Efficacy of a new comprehensive method of treatment for bacterial keratitis

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Purpose: *To assess the therapeutic efficacy of hyperbaric oxygenation (HBO) therapy and liposomal ozone-based solution (LOBS) in the comprehensive treatment of bacterial keratitis (BK).*

Material and Methods: *Ninety-eight patients (98 eyes) were treated for BK and the treatment outcomes were studied. Histories and complaints were thoroughly taken and patients underwent microbiological examination and comprehensive eye examination including visual acuity, refractomerty, pneumotonometry, biomicroscopy, fluorescein staining assessment of corneal re-epithelaliization, ophthalmoscopy, ultrasound B-scanning, and anterior-segment optical coherence tomography at admission, treatment day 10 and at one month. Patients were randomly divided into 3 clinical groups: a control group (32 patients, 32 eyes), group 1 (33 patients, 33 eyes) and group 2 (33 patients, 33 eyes). Groups were statistically comparable in terms of the number of patients, age and gender. In all three groups of patients, treatment included antiseptic (miramistin 0.1 mg/ml), antibiotics (ciprofloxacin 0.3% and tobramycin 0.3% until results of microbiological cultures became available; the most sensitive antibiotic was selected based on the results of bacterial culture), mydriatic (cyclopentolate), tear substitute with reparing agent (dexpanthenol 2% and sodium hyaluronate 0.15), and antibiotic administered parabulbarly (amikacin, 0.5 ml). In addition, patients in groups 1 and 2 were administered ten 45-minute sessions of hyperbaric 95-percent medical oxygen at 1.5 atmospheres. Moreover, patients in group 2 were administered topical LOBS.*

Results: *In group 2, inflammation attenuation was faster, and there was a reduction in the time to resolution of infiltration by 5.9 ± 0.2 days compared to the control group and by 2.7 ± 0.2 days compared to group 1. In groups 2 and 1, corneal re-epithelaliization completed 8.2 ± 0.2 days and 4.8 ± 0.2 days earlier, respectively, than in the control group ($p < 0.001$). In group 2, there was a reduction in the length of inpatient stay at the hospital of 6.6 ± 0.2 days compared to the control group and of 3.4 ± 0.2 days compared to group 1 ($p < 0.001$).*

Conclusion: *HBO therapy plus LOBS, when used as adjuncts to conventional treatment for BK, resulted in significantly faster inflammation attenuation and reduced time to resolution of infiltration, time to corneal re-epithelaliization, and length of in-patient stay at the hospital. Therefore, the new method of treatment for BK was found therapeutically effective and can be recommended for application in clinical practice.*

Keywords:

bacterial keratitis, inflammation, treatment, hyperbaric oxygenation, liposomal ozone-based solution, cornea

Introduction

Bacterial keratitis (BK) is a severe corneal disorder and the fifth most common cause of blindness. In some cases, it may result in anatomical death of the eye, leading to enucleation. The disease may be provoked by corneal microtrauma, foreign body entrapment, use of contact lenses, corneal surface disease, etc. [1]. Of concern is the fact that 55–60% of patients are of the working age. Excessive use of antibiotics in the treatment of infections and wide use and inadequate dosage of antibiotics in agriculture, fish farming and veterinary medicine have, over time, contributed to the emergence of resistant bacteria [2]. Antimicrobial resistance in relation to specific ophthalmic infections is emerging as a serious concern of ophthalmic community [3]. Acquired resistance develops gradually, leading to changes in bacteria: gene resistance to a particular antibacterial drug develops, and the progeny of the bacteria with resistance genes becomes dominant. If some bacteria in the infecting population contain an antibiotic resistance gene, they can transfer the gene to other bacteria in the population that are sensitive to the antibiotic, leading to disease progression [4].

Another cause of the formation of antibiotic resistance is the formation of biofilms, highly organized microbial communities. They may be formed by pathogenic and non-pathogenic representatives of one or several types of microorganisms. These features of the life and development of bacteria contribute to the development of high bacterial resistance to antiseptic, antibacterials and disinfectants [5, 6].

Antibiotic resistance has been given special attention in Ukraine and in the world at large: clinical protocols and regulations for the use of antibacterial medications in hospitals have been developed, and educational measures were taken to increase the awareness of these issues among medical staff and the general population. Developing new antimicrobial means with a new mechanism of action is an important strategy for the fight against the threat of antibiotic resistance.

The development of hypoxia in patients with BK results in abnormal epithelial and endothelial barrier functions, corneal edema and stromal changes.

Various oxygen therapy techniques for the correction and treatment of these conditions have been developed and improved.

Hyperbaric oxygenation (HBO) is a type of intensive therapy based on the curative features of oxygen; with oxygen delivered under hyperbaric conditions, additional oxygen is dissolved into the plasma and eye fluids, and oxygen delivery to tissues increases. The use of HBO is beneficial for the body adaptation system at all levels from the systemic to cellular to molecular, reduces hypoxia, has a bacteriostatic effect, activates reparative processes and potentiates effects of antibacterial drugs. The method is used as an adjunct to conventional treatments. Contraindications to HBO include paranasal sinus inflammation, the presence of cavities in the lungs, bilateral confluent pneumonia, severe hypertension, unstable hemodynamics, epilepsy (or other seizure disorders), claustrophobia, increased sensitivity to oxygen, and eustachian tube dysfunction [7, 8].

Ozone (O3) is an unstable gaseous substance widely occurring in nature, which is increasingly considered as a treatment option in various infections, and holds great potential for researchers and clinicians. The O3 molecule breaks down into molecular oxygen which is non-toxic to the corneal tissue, facilitates its recovery and does not cause sensitization. When bound with fatty acids present in vegetable oils, ozone forms ozonides which are stable and enable utilizing the oxidation potential of gaseous ozone. When in contact with bacteria, ozonides cause cell membrane rupture, inhibit cellular respiration and enzymatic activities, improve blood circulation and synthesis of anti-inflammatory molecules, facilitate the release of growth factors (platelet-derived growth factor (PDGF), transforming growth factor (TGF) and vascular growth factor (VEGF)), activate lipid peroxidation systems, stimulate interferon synthesis, increase phagocytosis, stimulate fibroblast and keratinoblast proliferation in tissue recovery processes, destroy and inhibit pro-inflammatory cytokines (interleukins), and oxidize prostaglandins. Contraindications include intolerance to the active substance or excipients [9-11].

The purpose of the study was to assess the therapeutic efficacy of a combination of HBO therapy and liposomal ozone-based solution (LOBS) in the comprehensive treatment of BK.

Material and Methods

Ninety-eight patients (98 eyes), aged from 18 to 82 years (mean age, 44.5 ± 1.6 years), were treated for BK and the treatment outcomes were studied. Of these patients, 55 (56.1%) were men and 43 (43.9%) were women. Informed consent was obtained from all study subjects.

In all patients, histories and complaints were thoroughly taken. In addition, all patients underwent microbiological examination and comprehensive eye examination including visual acuity, refractomerty, pneumotonometry, anterior segment slit-lamp biomicroscopy, fluorescein staining assessment of corneal re-epithelaliization, ophthalmoscopy, ultrasound B-scanning (Quantel Medical, Clermont-Ferrand, France), and anterior-segment optical coherence tomography (OCT; Optovue), at admission, treatment day 10 and at one month.

Patients were randomly divided into 3 clinical groups: a control group (32 patients, 32 eyes), group 1 (33 patients, 33 eyes) and group 2 (33 patients, 33 eyes). Groups were statistically comparable in terms of the number of patients, age and gender (Table 1).

Cultures obtained from most patients (55 patients or 56.1%) gave growth of a single gram-positive microorganism (Staphylococcus aureus, methicillin-resistant Staphylococcus aureus (MRSA), Staphylococcus epidermidis, Staphylococcus haemolyticus, Streptococcus pneumonia, Streptococcus pyogenes, or Kocuria varians). Cultures obtained from 17 patients (17.3%) gave growth of a single gram-negative microorganism (Pseudomonas aeruginosa, Esherichia coli, Klebsiellae pneumonia, or Pseudomonas putida). Cultures obtained from 7 patients (7.2%) gave growth of both gram-positive and gram-negative microor-

Table 1. Numbers and percentages of men and women and mean age by groups

Characteristic		Control group (n = 32)	Group 1 (n=33)	Group 2 (n=33)	Significance of difference between groups
Gender, n/ %	Male	17 / 53.1	20 / 60.6	18 / 54.5	$p_{\chi 2} = 0.811$
	Female	15 / 46.9	13 / 39.4	15 / 45.5	
Mean age, years, mean ± standard error of mean		46.4 ± 2.8	43.4 ± 2.6	43.8 ± 3.0	$p_F = 0.710$

Note: n, number of observations; M, mean value; m, standard error of mean; $p_{\chi 2}$, p-value as assessed by the Chi-square test; p_F, p-value as assessed by the F test

ganisms (Staphylococcus epidermidis and Staphylococcus aureus, Streptococcus pyogenes and Citrobacter braakii). No cultural growth was observed in 19 patients (19.4%).

Gram-positive pathogens were most sensitive to ciprofloxacin, levofloxacin, amikacin, tobramycin, moxifloxacin, and liposomal ozone-based solution. Gram-negative pathogens were most sensitive to ciprofloxacin, meropenem, amikacin, tobramycin, and liposomal ozone-based solution.

In all three groups of patients, in-patient treatment included antiseptic (miramistin 0.1 mg/ml, 1 drop 6 times daily), antibiotics (ciprofloxacin 0.3% 1 drop 6 times daily and tobramycin 0.3% 1 drop 6 times daily, until results of microbiological cultures became available; the most sensitive antibiotic was selected based on the results of bacterial culture), mydriatic (cyclopentolate, 1 drop twice daily), eye drops, tear substitute with reparing agent (dexpanthenol 2% and sodium hyaluronate 0.15%, 1 drop into the conjunctival sac 4 times daily), and antibiotic administered parabulbarly (amikacin, 0.5 ml once daily).

In addition, patients in groups 1 and 2 received HBO therapy (ten 45-minute sessions of hyperbaric 95-percent medical oxygen at 1.5 atmospheres in a BLKS 301M monoplace chamber).

Moreover, LOBS was instilled into the conjunctival sac (1 drop 4 times daily) in patients in group 2.

We assessed the effects of HBO therapy and liposomal ozone-based solution on longitudinal changes in the following characteristics: visual acuity, conjunctival discharge, mixed conjunctival injection, corneal edema, inflammatory infiltration, depth of the corneal ulcer, time to resolution of infiltration, re-epithelaliization time, and length of inpatient stay at the hospital.

Scores were derived from for the clinical records of patients for the convenience of analysis of changes in clinical characteristics [12].

The scale used for scoring inflammatory response consisted of the following categories: intensity of corneal edema; corneal inflammatory infiltration; depth of the corneal ulcer, mixed conjunctival injection; conjunctival discharge (before and after treatment), and corneal lesion area (before treatment).

Intensity of corneal edema was scored using the following scale: 0, no corneal edema, transparent cornea; 1, local corneal epithelial edema at the site of inflammation; 2, local edema of the epithelium and superficial stroma; and 3, local edema of the superficial and middle stroma.

Inflammatory infiltration was scored using the following scale: 0, no infiltration; 1, moderate infiltration; 2, apparent infiltration; 3, diffuse infiltration.

Depth of the corneal ulcer was scored using the following scale: 1, < 1/3 of the corneal thickness; 2, 1/3-2/3 of the corneal thickness; 3, >2/3 of the corneal thickness.

Mixed conjunctival injection was scored using the following scale: 0, physiological norm; 1, mild mixed injection; 2, moderate mixed injection; 3, marked mixed injection.

Conjunctival discharge was scored using the following scale: 0, no discharge; 1, slight conjunctival mucous discharge; 2, abundant conjunctival mucous discharge; 3, conjunctival mucopurulent discharge.

Statistica v.6.1 software (Statsoft Inc., USA; № AGAR909E415822FA) was used for statistical analysis. Data was assessed for normality using the Shapiro–Wilk test. For normally distributed data, mean, standard error of mean, standard deviation (SD) and 95% confidence interval (CI) were calculated. Paired t test was used for longitidunal analysis. Differences in mean were tested among the three groups means using a one-way analysis of variance (ANOVA) F-test and Scheffe's pairwise post hoc test. For non-normally distributed continuous variables, median and interquartile range (IQR) values were calculated, and differences were assessed by the Wikoxon W test and by the Kruskal-Wallis test followed by the Dunn post-hoc test. Contingency tables and Pearson Chi-square tests were used for group comparisons for qualitative data. A P value < 0.05 was considered statistically significant.

Results

At baseline, there was no significant difference between groups in terms of depth of the corneal ulcer, intensity of corneal edema, corneal inflammatory infiltration, corneal lesion area and location of corneal opacity before treatment ($p > 0.05$) (Table 2). Before treatment, conjunctival discharge was observed in patients of all the three groups, with 56 patients (57.1%) exhibiting abundant conjunctival mucous discharge, and 42 patients (42.9%) exhibiting conjunctival mucopurulent discharge, with no significant difference between groups ($p\chi 2 = 0.924$). After treatment, conjunctival discharge improved, with only 9 patients (9.2%; particularly, 6 patients (18.7%) in the control group, 2 patients (6.1%) in group 1 and 1 patient (3.0%) in group 2) exhibiting slight conjunctival mucous discharge. Differences in changes between groups were statistically significant (pC-1= 0.120; pC-2= 0.041 and p1-2= 0.555 as assessed by the χ^2 test, respectively) (Fig. 1). In addition, after treatment, conjunctival discharge score decreased significantly by 92.1% (from 2.41 ± 0.09 to 0.19 ± 0.07) in the control group, by 97.6% (from 2.45 ± 0.09 to 0.06 ± 0.04) in group 1, and by 98.8% (from 2.42 ± 0.09 to 0.03 ± 0.03) in group 2.

Before treatment, mixed injection was mild or moderate, with no significant difference between groups ($p\chi 2 = 0.811$), and with mean scores of 2.58 ± 0.09, 2.64 ± 0.09, and 2.56 ± 0.09 in groups 1, 2 and the control group ($p_H = 0.812$). After treatment, mixed injection improved significantly to the norm in 93.9% and 97.0% of patients in groups 1 and 2, respectively, and in 81.2 % of patients in the control group. Differences in changes between groups were statistically significant (pC-1= 0.120; pC-2= 0.041 and p1-2= 0.555 as assessed by the χ^2 test, respectively) (Table 3).

Time to resolution of infiltration with treatment in group 2 (8.27 ± 0.13 days) was 5.9 ± 0.2 days shorter,

Table 2. Characteristics of pre-treatment corneal lesions in patients with bacterial keratitis (n/ % or M ± m)

Characteristic		Control group (n = 32)	Group1 (n = 33)	Group 2 (n = 33)	Significance of difference between groups
Depth of the corneal ulcer (score)	< 1/3 of the corneal thickness (1)	15 / 46.9	14 / 42.4	11 / 33.3	p_{χ^2}=0.850
	1/3-2/3 of the corneal thickness (2)	13 / 40.6	14 / 42.4	17 / 51.5	
	> 2/3 of the corneal thickness (3)	4 / 12.5	5 / 15.2	5 / 15.2	
	Mean score	1.66±0.12 (2)	1.73±0.13 (2)	1.82±0.12 (2)	p_H=0.603
Corneal edema (score)	local corneal epithelial edema at the site of inflammation (1)	7 / 21.9	7 / 21.2	8 / 24.2	p_{χ^2}=0.919
	local edema of the epithelium and superficial stroma (2)	12 / 37.5	12 / 36.4	9 / 27.3	
	local edema of the superficial and middle stroma (3)	13 / 40.6	14 / 42.4	16 / 48.5	
	Mean score	2.19±0.14 (2)	2.21±0.13 (2)	2.24±0.14 (2)]	p_H=0.937
Inflammatory corneal infiltration (score)	moderate (1)	7 / 21.9	7 / 21.2	8 / 24.3	p_{χ^2}=0.959
	apparent (2)	16 / 50.0	17 / 51.5	14 / 42.4	
	diffuse (3)	9 / 28.1	9 / 27.3	11 / 33.3	
	Mean score	2.06±0.13 (2)	2.06±0.12 (2)	2.09±0.13 (2)	p_H=0.977
Corneal lesion area	moderate (1)	7 / 21.9	8 / 24.2	9 / 27.3	p_{χ^2}=0.980
	apparent (2)	14 / 43.7	15 / 45.5	13 / 39.4	
	diffuse (3)	11 / 34.4	10 / 30.3	11 / 33.3	
	Mean score	2.13±0.13 (2)	2.06±0.13 (2)	2.06±0.14 (2)	p_H=0.927
Location of corneal opacity	Optic zone	19 (59.4)	21 (63.6)	21 (63.6)	p_{χ^2}=0.920
	Paraoptic zone	13 (40.6)	12 (36.4)	12 (36.4)	

Note: n, number of observations; M, mean value; m, standard error of mean; p_{χ^2}, p-value as assessed by the Chi-square test; p_H, p-value as assessed by the Kruskal-Wallis test

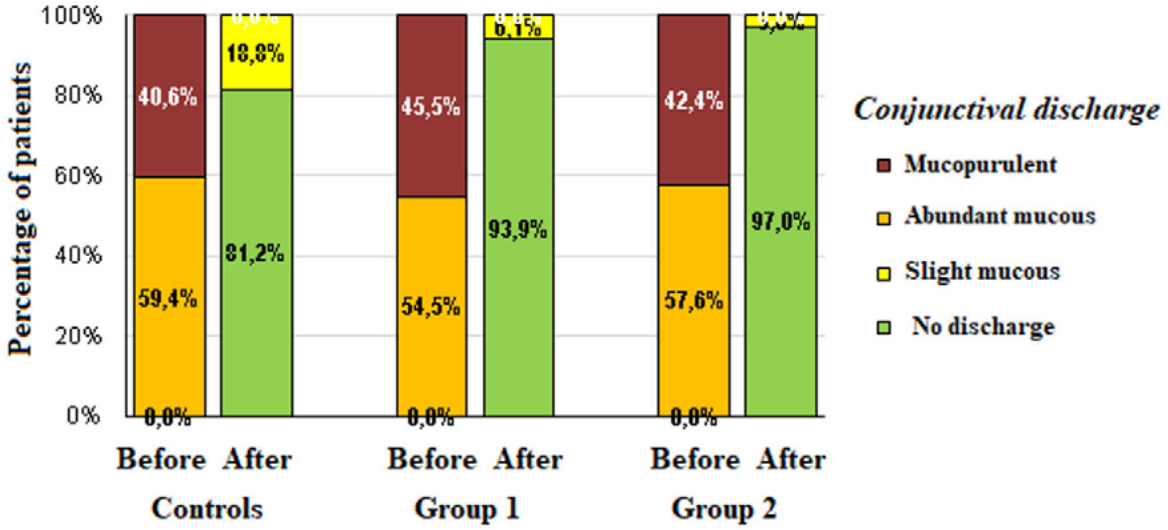

Fig. 1. Percentages of patients with slight conjunctival mucous discharge, abundant conjunctival mucous discharge, conjunctival mucopurulent discharge and no conjunctival discharge in groups of patients with bacterial keratitis before and after treatment

Table 3. Intensity (scores) of mixed conjunctival injection in patients with bacterial keratitis before and after various methods of treatment (n/ % or M ± m)

Study groups	Before or after treatment	Intensity of mixed conjunctival injection (score)				
		Physiological norm (0)	Mild (1)	Moderate (2)	Marked (3)	Mean score
Control group (n= 32)	before	-	-	14/ 43.7	18/ 56.3	2.56 ± 0.09 (3)
	after	26/ 81.2	6/ 18.8	-	-	0.19±0.07 (0)
Δ(%), p		Δ_1 = -81.2%, p_1<0.001				Δ_2 =-92.6%, p_2<0.001
Group 1 (n = 33)	before	-	-	14/ 42.4	19/ 57.6	2.58±0.09 (3)
	after	31/ 93.9	2/ 6.1	-	-	0.06±0.04 (0)
Δ(%), p		Δ_1 = -93.9%, p_1<0.001				Δ_2 =-97.7%, p_2<0.001
Group 2 (n= 33)	before	-	-	12/ 36.4	21/ 63.6	2.64±0.09 (3)
	after	32/ 97.0	1/ 3.0	-	-	0.03±0.03 (0)
Δ(%), p		Δ_1 = -97.0%, p_1<0.001				Δ_2 =-98.9%, p_2<0.001

Note: n, number of observations; M, mean value; m, standard error of mean; Δ_1 /Δ_2, percentage of change in relation to baseline; p_1, p-value as assessed by the MacNemar test; p_2, p-value as assessed by the Wikoxon W test

Table 4. Times to resolution of infiltration and start and completion of re-epithelialization and length of in-patient stay at the hospital for patients with bacterial keratitis (M ± m, days)

Characteristic	Control group (n = 32)	Group 1 (n=33)	Group 2 (n=33)	Significant of difference between groups
Time to resolution of infiltration	14.16 ± 0.13	10.94 ± 0.14	8.27 ± 0.13	p_F < 0.001
Time to start of re-pithelialization	12.78 ± 0.14	7.94 ± 0.14	6.15 ± 0.12	p_F < 0.001
Time to completion of re-epithelialization	17.63 ± 0.12	12.82±0.13	9.39±0.13	p_F < 0.001
Length of in-patient stay at the hospital	20.56 ± 0.12	17.76 ± 0.13	14.33 ± 0.14	p_F < 0.001

Note: n, number of observations; M, mean value; m, standard error of mean; p_F, p-value as assessed by the F test. Pairwise comparison differences were significant (p < 0.001).

and in group 1 (10.94 ± 0.14 days) was 3.2 ± 0.2 days shorter, compared to the control group (14.16 ± 0.13 days) (p < 0.001). Additionally, the use of both HBO therapy and liposomal ozone-based solution in the comprehensive treatment for bacterial keratitis (in group 2) enabled reducing the time to resolution of infiltration by 2.7 ± 0.2 days compared to group 1 that received HBO therapy but not liposomal ozone-based solution (p < 0.001) (Table 4).

Corneal reepithelaliization began on day 12.78 ± 0.14 in the control group, on day 7.94 ± 0.14 in group 1, and on day 6.15 ± 0.12 in group 2. In groups 1 and 2, corneal re-epithelaliization began 4.8 ± 0.2 days and 6.6 ± 0.2 days earlier, respectively, than in the control group (p < 0.001) (Fig. 1).

In groups 2 and 1, corneal re-epithelaliization completed 8.2 ± 0.2 days and 4.8 ± 0.2 days earlier, respectively, than in the control group (p < 0.001). In patients that received a combination of HBO therapy and liposomal ozone-based solution as adjunctive treatment (group 2), the period of corneal re-epithelaliization was 3.4 ± 0.2 days, and was significantly shorter than in patients in group 1 (p<0.001).

Faster inflammation attenuation and recovery of the eye with comprehensive treatment for bacterial keratitis involving both adjunctive local and systemic oxygenation enabled a significant reduction in the length of inpatient stay at the hospital. The mean length of inpatient stay at the hospital for patients in group 1 was 17.76 ± 0.13 days, which was 2.8 ± 0.2 days shorter than for the control group (p < 0.001). The use of a combination of HBO therapy and LOBS as adjunct to conventional treatment for BK enabled a mean reduction in the length of inpatient stay at the hospital of 6.6 ± 0.2 days compared to the control group (p < 0.001) and of 3.4 ± 0.2 days compared to group 1 (p < 0.001) (Table 4).

In all study groups, baseline visual acuity was low, with no significant difference between groups ($p_{\chi 2}$ = 0.868). Only four patients with corneal inflammation in the paraoptic region adjacent to the limb (Fig. 3) exhib-

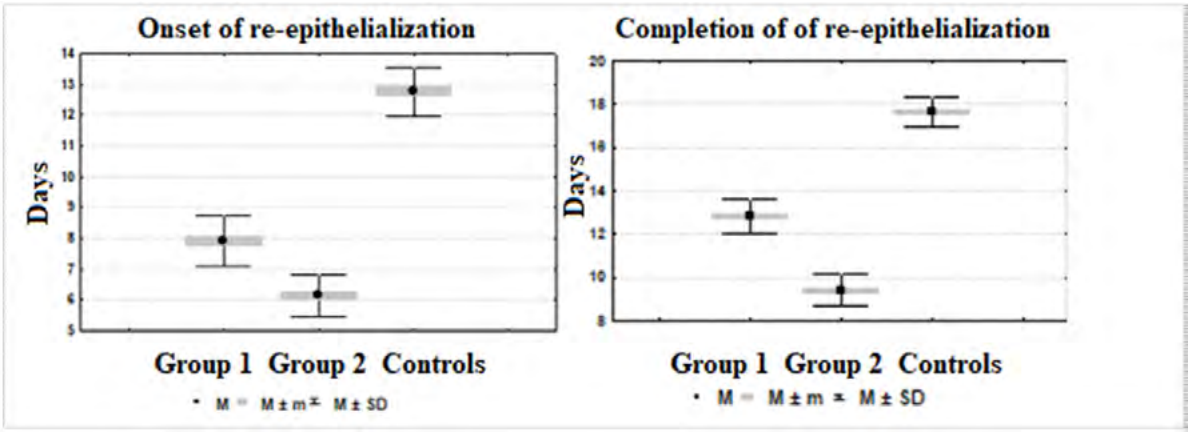

Fig. 2. Box-and-whiskers plots of time to the onset and completion of re-epithelialization for groups of patients with bacterial keratitis

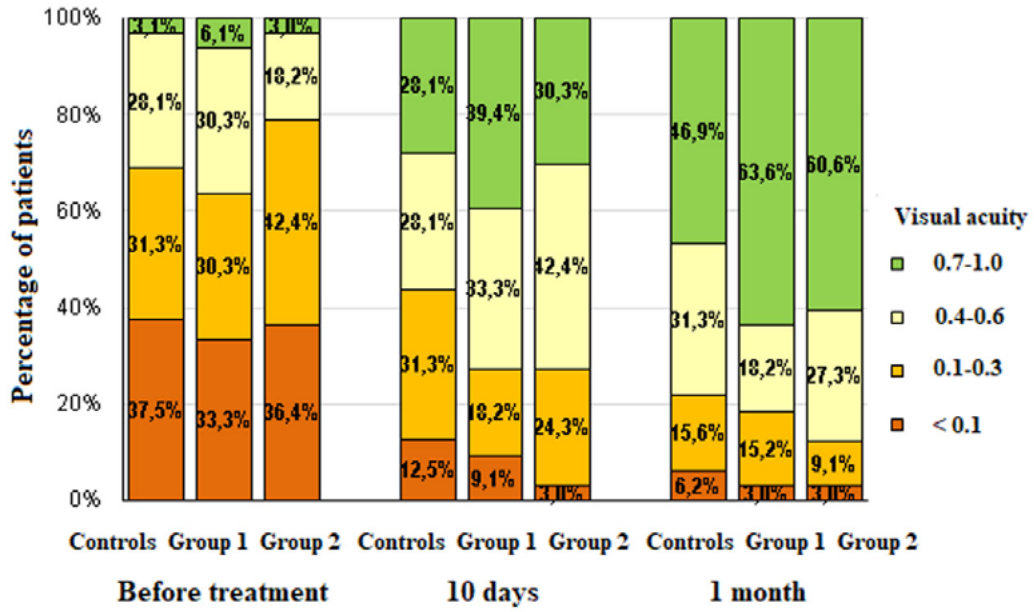

Fig. 3. Visual acuities in groups of patients with bacterial keratitis before and after treatment

ited high visual acuity (0.7–1.0) at admission. Visual acuity improved with treatment in all groups, especially in patients with a baseline visual acuity less than 0.1. The percentage of patients with a visual acuity less than 0.1 decreased from 35.7% to 8.2% at day 10 and 4.1% at the end of treatment courses ($p < 0.001$). Correspondingly, the percentage of patients with a high visual acuity (0.7–1.0) increased from 4.1% to 32.7% at day 10 and 57.1% at 1 month after initiation of treatment ($p < 0.001$). Mean visual acuity increased 2.7 times (from 0.25 ± 0.04 to 0.68 ± 0.05; $p < 0.001$) in group 1, 3.6 times (from 0.19 ± 0.04 to 0.68 ± 0.05) in group 2, and 2.7 times (from 0.23 ± 0.04 to 0.62 ± 0.05; $p < 0.001$) in the control group. There was no significant difference between the groups in visual acuity at baseline (pH = 0.717), and at 1 month after initiation of treatment (pH = 0.536). It should be noted that although initially visual acuity was lower in patients in group 2, the rate of improvement in visual acuity with treatment was higher in these patients compared to controls.

Discussion

Many publications reported on changes in the composition of microbiota and the sensitivity of microbiota to antibacterial drugs [1–3]. Biofilm formation [5, 6] and development of genetic resistance in microorganisms [4] are believed to be the most common causes of antibiotic resistance. This contributed to the search of new methods of treatment for pathogenic infections. Findings of our microbiological studies in patients with bacterial keratitis contributed to the available information on the composition of corneal microbiota and its sensitivity to antibiotics and liposomal ozone-based solution. We found gram-

positive only microbiota to be the most common (56.1% of patients), followed by gram-negative only microbiota (17.3%), and mixed microbiota (7.2%). No culture growth was observed in specimens from 19.4% of patients. Gram-positive pathogens were most sensitive to ciprofloxacin, levofloxacin, amikacin, tobramycin, moxifloxacin, and liposomal ozone-based solution. Gram-negative pathogens were most sensitive to ciprofloxacin, meropenem, amikacin, tobramycin, and liposomal ozone-based solution. HBO therapy was found to be a beneficial adjunct to conventional treatment for BK due to bioenergetic, bacteriostatic, reperative and antibacterial potentiating effects that enable improved treatment outcomes [7, 8].

LOBS has been reported to exhibit antibacterial, antiviral and antifungal activities. This was achieved through the reaction of ozone with vegetable oils which facilitated using the potential of gaseous ozone, prolonged the effect of ozone on tissues, and made the application more comfortable [9–11].

In the current study, for the first time, we found that patients with bacterial keratitis treated with conventional therapy plus both HBO therapy and liposomal ozone-based solution (group 2) exhibited reduced time to resolution of infiltration, re-epithelaliization time and length of in-patient stay at the hospital compared to those treated with conventional therapy plus HBO therapy only (group 1) and those treated with conventional therapy only (the control group) ($p < 0.001$).

In addition, visual acuity significantly improved with treatment in all groups. Moreover, although initially visual acuity was lower in patients in group 2, the rate of improvement in visual acuity with treatment was higher in these patients compared to controls.

Conclusion

HBO therapy plus liposomal ozone-based solution, when used as adjuncts to conventional treatment for bacterial keratitis, resulted in significantly faster inflammation attenuation and reduced time to resolution of infiltration, time to corneal re-epithelaliization, and length of in-patient stay at the hospital. Therefore, the new method of treatment for bacterial keratitis was found therapeutically effective and can be recommended for application in clinical practice.

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Disclosures

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Data availability statement: *Data obtained during and/or analyzed during this study can be obtained from the corresponding author upon reasonable request.*

Abbreviations: *BK, bacterial keratitis; HBO, hyperbaric oxygenation; O3, ozone*