New Developments in Vision Research

Selected Antimicrobial Activity of Topical Ophthalmic Anesthetics

Margaret M. Reynolds[1], Kerryl E. Greenwood-Quaintance[2,3], Robin Patel[2,3], and Jose S. Pulido[1,4,5]

[1] Department of Ophthalmology, Mayo Clinic, Rochester, MN, USA
[2] Department of Laboratory Medicine, Mayo Clinic, Rochester, MN, USA
[3] Department of Infectious Disease, Mayo Clinic, Rochester, MN, USA
[4] Department of Molecular Medicine, Mayo Clinic, Rochester, MN, USA
[5] VRS Foundation Board Member, Mayo Clinic, Rochester, MN, USA

Correspondence: Jose S. Pulido, Mayo Clinic, Department of Ophthalmology and Department of Molecular Medicine, 200 First Street, SW, Rochester, MN 55905, USA. e-mail: pulido.jose@mayo.edu

Purpose: Endophthalmitis is a rare complication of intravitreal injection (IVI). It is recommended that povidone-iodine be the last agent applied before IVI. Patients have reported povidone-iodine application to be the most bothersome part of IVIs. Topical anesthetics have been demonstrated to have antibacterial effects. This study compared the minimum inhibitory concentration (MIC) of topical anesthetic eye drops (proparacaine 0.5%, tetracaine 0.5%, lidocaine 2.0%) and the antiseptic, 5.0% povidone-iodine, against two organisms causing endophthalmitis after IVI.

Methods: Minimum inhibitory concentration values of topical anesthetics, povidone-iodine, preservative benzalkonium chloride (0.01%), and saline control were determined using five isolates of each *Staphylococcus epidermidis* and viridans group *Streptococcus* species (VGS). A broth microdilution technique was used with serial dilutions.

Results: Lidocaine (8.53×10^{-5} mol/mL) had MICs of 4.27 to 8.53×10^{-5} mol/mL, and tetracaine (1.89×10^{-5} mol/mL) had MICs of 9.45×10^{-6} mol/mL for all isolates. Proparacaine (1.7×10^{-5} mol/mL) had MICs of 1.32 to 5.3×10^{-7} and 4.25×10^{-6} mol/mL for *S. epidermidis* and VGS, respectively). Benzalkonium chloride (3.52×10^{-7} mol/mL) had MICs of 1.86×10^{-9} to 1.1×10^{-8} and 4.40×10^{-8} mol/mL for *S. epidermidis* and VGS, respectively. Povidone-iodine (1.37×10^{-4} mol/mL) had MICs of 2.14 to 4.28×10^{-6} and 8.56×10^{-6} mol/mL for *S. epidermidis* and VGS, respectively.

Conclusion: Proparacaine was the anesthetic with the lowest MICs, lower than that of povidone-iodine. Benzalkonium chloride had lower MICs than proparacaine. All tested anesthetics and povidone-iodine inhibited growth of *S. epidermidis* and VGS at commercially available concentrations.

Translational Relevance: For certain patients, it could be possible to use topical anesthetic after povidone-iodine for comfort without inhibiting and perhaps contributing additional antimicrobial benefit.

Introduction

Intravitreal injection (IVI) is one of the most common procedures performed by ophthalmologists. More than 2.3 million injections were recorded in 2012 in the United States alone.[1] The number of IVIs is anticipated to continue to grow. Endophthalmitis is the most feared complication of intravitreal injection. To prevent endophthalmitis, antisepsis is attempted before injections using 5.0% povidone-iodine (Beta-dine).[2] Topical anesthetic also is given to patients before injections for their comfort. Commonly used anesthetics include proparacaine 0.5% or tetracaine 0.5% drops, as well as subconjunctival lidocaine 2.0%. Interestingly, these anesthetics have been demonstrated to have antibacterial properties even against bacteria that cause post-IVI endophthalmitis (Table 1). The current expert guidelines for IVI stress the importance of 5.0% povidone-iodine use. Povidone-iodine should be the last agent applied before injection; therefore, it should be the first agent applied

Table 1. Anesthetics Have Been Previously Demonstrated to Have Effects Against Bacteria that Cause Endophthalmitis after Intravitreal Injection

	Coagulase-Negative Staphylococci	VGS	B. cereus	S. aureus	MRSA
Lidocaine	+ [7]	+ [7]	+ [25]	+ [26, 27]	+ [28]
Proparacaine	−	−	−	+ [19]	−
Tetracaine	+ [6]	−	−	+ [19]	−

+, data have been demonstrated to be effective against bacteria; −, denotes no data rather than that they are ineffective against bacteria.

for antisepsis and reapplied after topical anesthetic use.[2]

The most common bacteria isolated in postinjection endophthalmitis have been demonstrated to be coagulase-negative staphylococci followed by *Streptococcus* species, most commonly the viridans group streptococci (VGS) *S. salivarius* and *S. mitis*.[3,4] Other bacteria, including *Bacillus cereus* and *Staphylococcus aureus,* also have been implicated.[3,4] Proparacaine, tetracaine, and lidocaine have been demonstrated to have antibacterial properties against *S. epidermidis* and VGS.[5–7] We determined the minimum inhibitory concentration (MIC) values of these three topical anesthetics, preservatives, and povidone-iodine against *S. epidermidis* and VGS using a broth microdilution technique. We also analyzed whether the topical anesthetics have synergistic or antagonistic effects with povidone-iodine. These results would provide information whether it could be possible to use the topical anesthetic after the povidone-iodine for patient comfort with perhaps additional antimicrobial benefit.

Methods

Five clinical isolates of *S. epidermidis* and five clinical isolates of biofilm-forming VGS (four *S. mitis* and one *S. mutans*) were studied. The clinical isolates were from biofilm-associated infections. Isolates were stored at −80°C in the Mayo Clinic Infectious Diseases Research Laboratory.

Three commonly used and commercially available anesthetic eye drops were tested: tetracaine 0.5% (preservative-free; Alcon, Fort Worth, TX), proparacaine 0.5% (preservative, benzalkonium chloride 0.01%; Akorn Pharmaceuticals, Lake Forest, IL), and lidocaine 2.0% (preservative-free; Hospira, Lake Forest, IL). These anesthetics were analyzed for their effect on bacterial growth in vitro. They were tested alone and in combination with 5.0% povidone-iodine (Alcon). Also, povidone-iodine 5.0% was tested alone. The preservative, benzalkonium chloride 0.01% (Nature's Tears; Rugby Laboratories, Livonia, MI), also was tested as a control. Finally, saline eye drops (Unisol 4; Alcon) were used as a control.

Three to five isolated bacterial colonies grown on sheep blood agar plates were used to inoculate tryptic soy broth (*S. epidermidis*) or Todd-Hewitt broth (VGS), which were incubated to exponential phase growth, adjusted to a 0.5 McFarland standard (1.5×10^7 colony-forming units [CFU]/mL), and diluted in Cation-adjusted Mueller Hinton broth (CAMHB; Becton, Dickinson France SAS, Le Pont-De-Claix, France) to generate a final concentration of 1.5×10^5 CFU/mL.

Stock solutions of proparacaine and tetracaine were diluted in CAMHB to obtain a working suspension of 0.5%. Lidocaine was diluted in CAMHB to obtain a working suspension of 2.0%, and benzalkonium chloride was diluted in CAMHB to a working suspension of 0.01%. Finally, povidone-iodine solution was diluted in CAMHB to obtain working suspensions of 5.0%.

Broth microdilution assays were performed according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.[8] Briefly, serial dilutions of the stock suspensions were made in 50 μL of CAMHB with 50 μL of CAMHB alone as a growth control (with 2.5% lysed horse blood added for VGS); 50 μL of each bacterial suspension were added to the wells and incubated for 24 hours at 37°C (with 5% CO_2 for VGS). Saline eye drops were used as a negative control solvent. The MIC was determined to be the lowest concentration demonstrating no visible growth.[9]

Synergy versus antagonism for drug combinations was determined using a previously reported method.[10,11] Briefly, MICs were used to determine the
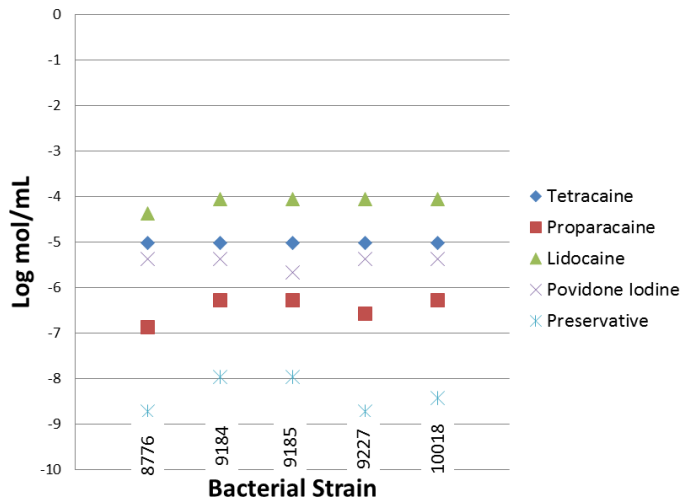
Figure 1. Minimum inhibitory concentration values of topical anesthetics against *S. epidermidis*.

Figure 2. Minimum inhibitory concentrations of topical anesthetics against VGS.

fractional inhibitory concentration (FIC) as follows:

$$FIC_{drug\,A} = \frac{MIC_{drug\,A} + MIC_{drug\,B}}{MIC_{drug\,A}}$$

$$FIC_{drug\,B} = \frac{MIC_{drug\,B} + MIC_{drug\,A}}{MIC_{drug\,B}}$$

$$FIC = FIC_A + FIC_B$$

Fractional inhibitory concentrations were interpreted according to previously accepted criteria as follows: ≤0.5, synergy; 0.5 to 1.0, additivity; 1.0 to 4.0, indifference; and ≥4, antagonism.[10–12]

Results

Of the topical anesthetics, proparacaine had the lowest MICs for *S. epidermidis*, ranging from 1.32 to 5.3×10^{-7} mol/mL, a 1/128 to 1/32 dilution of its commercially available form; MICs for VGS strains were 4.25×10^{-6} mol/mL, a 1/4 dilution of its commercially available form. Povidone-iodine had MICs for *S. epidermidis* ranging from 2.14 to 4.28×10^{-6} mol/mL, a 1/64 to 1/32 dilution of its commercially available form; MICs for VGS were 8.56×10^{-6} mol/mL, a 1/16 dilution of its commercially available form. The tetracaine MIC was 9.45×10^{-6} mol/mL for all strains (1/2 dilution of its commercially available form) of all isolates studied. Finally, lidocaine had the highest MICs, with MICs for *S. epidermidis*, ranging from 4.27 to 8.53×10^{-5} mol/mL (1/2–1 dilution of its commercially available form);
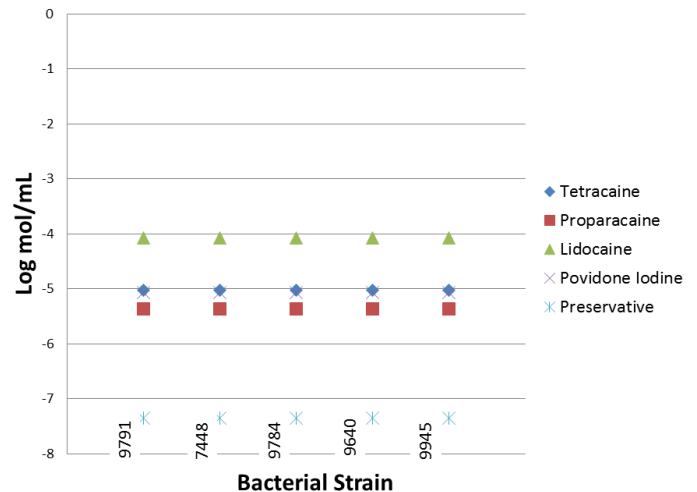
MICs for VGS were 8.53×10^{-5} mol/mL, which is 2.0% of its commercially available concentration. The benzalkonium chloride MIC was 1.86×10^{-9} to 1.1×10^{-8} mol/mL (1/16 to 1/128 dilution of its commercially available form) for *S. epidermidis* and 4.40×10^{-8} mol/mL (1/8 dilution of its commercially available form) for all isolates of VGS studied. Normal saline did not inhibit bacterial strains at any concentration (Figs. 1, 2; Table 2).

Synergy for all study isolates was assessed using povidone-iodine at 25% and 50% of its MIC (Table 3). Lidocaine with povidone-iodine demonstrated antagonism for 60% of *S. epidermidis* at 50% povidone-iodine's MIC and 80% at 25% povidone-iodine's MIC. Lidocaine demonstrated antagonism for 80% of VGS isolates studied. Proparacaine with povidone-iodine demonstrated synergy against all VGS studied. Benzalkonium chloride with povidone-iodine demonstrated synergy against 80% of *S. epidermidis* isolates studied.

Discussion

Intravitreal injections are one of the most common procedures performed by ophthalmologists. As indications for IVI expand and the population with conditions treated with IVI grows, it is beneficial to know how to prevent complications, such as endophthalmitis. Antiseptic has been demonstrated to be an important part of IVI as it decreases the number of cases of endophthalmitis.[13,14] Our results demonstrated that the tested topical anesthetics studied inhibited bacterial growth at their clinically used concentra-

Table 2. Comparison of the MIC Required to Inhibit Bacterial Growth and the Concentration of the Clinically Used Formulations

Drug	MIC *S. epidermidis*, mol/mL	MIC VGS Species, mol/mL	Formulation, mol/mL
Tetracaine	9.45×10^{-6}	9.45×10^{-6}	1.89×10^{-5}
Proparacaine	$1.32–5.3 \times 10^{-7}$	4.25×10^{-6}	1.7×10^{-5}
Lidocaine	$4.27–8.53 \times 10^{-5}$	8.53×10^{-5}	8.53×10^{-5}
Povidone-iodine	$2.14–4.28 \times 10^{-6}$	8.56×10^{-6}	1.37×10^{-4}
Benzalkonium chloride	1.86×10^{-9} -1.1×10^{-8}	4.40×10^{-8}	3.52×10^{-7}

tions. Proparacaine and benzalkonium chloride had lower MICs than the other tested topical anesthetics as well as povidone-iodine.

The incidence of post-IVI endophthalmitis has been reported to be 0.021% to 0.09%.[3,15] The most common bacteria isolated post-IVI have been reported by McCannel et al.[4] to be coagulase-negative staphylococci (65%); *Streptococcus* species, most commonly the VGS *S. salivarius* and *S. mitis* (31%);

and *Bacillus cereus* (4%). Dossarps et al.[3] found the most common bacteria isolated to be coagulase-negative staphylococci (78%), *S. aureus* (9%) and VGS (4%).[3] Because they are most commonly isolated in injection-associated endophthalmitis, coagulase-negative staphylococci and VGS were used in this study. Future studies with *S. aureus* would be worthwhile.

The use of povidone-iodine before IVI is supported

Table 3. Synergy Versus Antagonism for Topical Anesthetics Combined with Povidone-Iodine for *S. epidermidis* and VGS as Determined by FIC Values, Where ≤0.5, Synergy; 0.5 to 1.0, Additivity; 1.0 to 4.0, Indifference; and ≥4, Antagonism

Agent	FIC Range	Number of Tested Isolates			
		Synergy, FIC ≤ 0.5	Additivity, FIC 0.5–1.0	Indifference, FIC 1.0–4.0	Antagonism, FIC ≥ 4.0
Tetracaine					
S. epidermidis + 1/2 MIC PI (n = 5)	0.7–1.3	0	20%	80%	0
S. epidermidis + 1/4 MIC PI (n = 5)	1.3–2.1	0	0	100%	0
VGS + 1/2 MIC PI (n = 5)	0.45	100%	0	0	0
VGS + 1/4 MIC PI (n = 5)	0.2–1.8	20%	60%	20%	0
Lidocaine					
S. epidermidis + 1/2 MIC PI (n = 5)	3.5–7.4	0	0	40%	60%
S. epidermidis + 1/4 MIC PI (n = 5)	3.5–7.4	0	0	20%	80%
VGS + 1/2 MIC PI (n = 5)	3.7–7.4	0	0	20%	80%
VGS + 1/4 MIC PI (n = 5)	3.7–7.4	0	0	20%	80%
Proparacaine					
S. epidermidis + ½ PI (n = 5)	0.3–1.0	20%	60%	20%	0
S. epidermidis 1/4 MIC + PI (n = 5)	0.5–2.5	0	20%	80%	0
VGS + 1/2 PI (n = 5)	0.02–0.4	100%	0	0	0
VGS + 1/4 MIC PI (n = 5)	0.02–0.2	100%	0	0	0
Benzalkonium chloride					
S. epidermidis + 1/2 PI (n = 5)	0.2–0.5	80%	20%	0	0
S. epidermidis + 1/4 MIC PI (n = 5)	0.3–1.0	80%	0	20%	0
VGS + 1/2 MIC PI (n = 5)	0.5	0	100%	0	0
VGS + 1/4 MIC PI (n = 5)	1	0	0	100%	0

PI, povidone-iodine.

by a number of studies.[13,14] Apt et al.[16] demonstrated that one drop of povidone-iodine administered preoperatively reduced bacterial colonies in conjunctival cultures by 91% versus 33% for control eyes. Isenberg demonstrated that preoperative administration of povidone-iodine was as effective as neomycin, polymyxin B, and gramicidin (Neosporin; Johnson & Johnson, New Brunswick, NJ) ophthalmic solution given three consecutive days before surgery in reducing conjunctival cultures.[13] Finally, in a study by Speaker and Menikoff,[17] including 8000 patients undergoing cataract surgery, those treated with povidone-iodine had a lower rate of endophthalmitis (0.06%) compared to controls (0.24%, $P < 0.03$). Overall, povidone-iodine is nontoxic to the eye in the appropriate concentrations, has a broad antimicrobial spectrum (including bacteria, fungi, protozoa, and viruses), has a low propensity to select for bacterial resistance, is inexpensive, and is widely available.[18]

The antibacterial properties of topical anesthetics reported by this study are consistent with previous results. A study on the effects of topical anesthetics on bacteria using a disk diffusion technique studied proparacaine and tetracaine at 0.5, 0.25, and 0.125% concentrations found that tetracaine inhibited *S. aureus* growth at 5000 µg/mL and *Pseudomonas aeruginosa* at 2500 to 5000 µg/mL, and that proparacaine inhibited the growth of *S. aureus* at 1250 µg/mL, and inhibited *P. aeruginosa* growth at 2500 µg/mL.[19] Notably, a 2000-disk diffusion study demonstrated conflicting results with neither proparacaine nor tetracaine inhibiting *S. aureus* or *P. aeruginosa*.[20] Proparacaine has been shown to reduce the number of culture-positive eyes, with 4 of 40 conjunctival swabs being culture-positive after proparacaine, versus 12 of 36 eyes being culture-positive after receiving control solution, containing only preservative.[5] A study using broth microdilution demonstrated that tetracaine inhibited strains of *S. epidermidis* at a concentration of 625 µg/mL.[6] Finally, 2.0% lidocaine has been demonstrated to have rapid bactericidal effects against *S. epidermidis* and VGS.[7] The same study found that when patients were treated with subconjunctival injection of 2.0% lidocaine, 0 of 6853 patients experienced endophthalmitis versus 8 of 8189 ($P = 0.03$) treated with other topical anesthetics.[7] The mechanism by which topical anesthetics act as antimicrobials has been studied; they are thought to disrupt bacterial cell membranes causing permeability and lysis.[21,22] Finally, benzalkonium chloride has antimicrobial effects, with an in vitro study showing that benzalkonium chloride inhibits *S. aureus*

growth.[20] In summary, tetracaine, lidocaine, and proparacaine have been demonstrated to have antibacterial effects against organisms causing endophthalmitis. Tetracaine has been demonstrated to have effects against coagulase-negative staphylococci and *S. aureus*. Lidocaine has effects against coagulase-negative staphylococci, *S. aureus,* and VGS. Proparacaine has effects against *S. aureus* (Table 1).

This study compared the efficacy of tetracaine, lidocaine, proparacaine, and benzalkonium chloride with the standard of care, povidone-iodine; all studied topical anesthetics inhibited bacterial growth at clinically used concentrations. Interestingly, proparacaine and benzalkonium chloride had MICs lower than those of povidone-iodine. Additionally, benzalkonium chloride had a lower MIC than did proparacaine. Proparacaine has been demonstrated to have antimicrobial effects independent of benzalkonium chloride. The effects of proparacaine with benzalkonium chloride 0.01% versus benzalkonium chloride 0.01% in aqueous solution have been tested on conjunctival flora,[5] with findings that proparacaine reduced the number of positive conjunctival cultures compared to control (10% vs. 33% positive, respectively). With this in mind, it is likely that proparacaine has antimicrobial effects independent of benzalkonium chloride.

Does the observation that proparacaine has a lower MIC than povidone-iodine have clinical significance? A study comparing conjunctival cultures of patients treated with lidocaine, proparacaine, and tetracaine alone and with povidone-iodine may be informative from an antimicrobial standpoint. From the perspective of patient experience, recent studies have demonstrated great patient dissatisfaction with the application of povidone-iodine. In a study by van Asten et al.,[23] more patients reported povidone-iodine application than intravitreal injection to be the most bothersome part of the intravitreal injection process. Currently, topical anesthetics before the povidone-iodine are not sufficient to prevent the discomfort of povidone-iodine; therefore, in such patients, the use of proparacaine following the povidone-iodine may be a reasonable alternative.

Fractional inhibitory concentration also was studied to determine the impact of the tested agents and povidone-iodine on each other. Lidocaine demonstrated evidence of antagonism with povidone-iodine for *S. epidermidis* and VGS. It would be worthwhile to repeat the study to clarify these results. An ideal topical anesthetic would not decrease the efficacy of povidone-iodine. On the other hand, proparacaine

with povidone-iodine demonstrated synergy against all VGS studied. Benzalkonium chloride with povidone-iodine demonstrated synergy against 80% of *S. epidermidis* isolates studied. Overall, it was difficult to draw conclusions from these results. A study comparing the effect of 5% povidone-iodine with and without 4% lidocaine gel on bacterial colony counts on blood agar plates inoculated with *S. epidermidis*, *S. aureus*, *P. aeruginosa*, and *H. influenzae* demonstrated that when 4% lidocaine gel was applied before application with PI, there was decreased antimicrobial activity of PI.[24] To our knowledge, our findings are the first using the FIC to compare synergy versus antagonism of PI with proparacaine, tetracaine, lidocaine, and benzalkonium chloride. It also is the first to analyze the effects of proparacaine, tetracaine, and benzalkonium chloride in combination with lidocaine.

Our study has several limitations. First, it was conducted in vitro using a broth microdilution, which is different from the situation in vivo. This limitation may be particularly significant for film-forming bacteria. For conclusions surrounding topical anesthetic use to be clinically significant, in vivo studies are necessary. Additional studies using other types of bacteria, especially those derived from IVI-associated endophthalmitis, as well as further strains of *S. epidermidis* and VGS, are merited. It also is important to note that it can be difficult to draw conclusions about MIC values for topical medications. Minimum inhibitory concentration values may not reflect the toxicity of the agent on the host cell. It has been postulated that topical anesthetics act on bacterial cell membranes, so it is possible that the observed effects may generalize to other bacteria, and the associated antimicrobial effects may be quickly acting.[21,22]

The current expert guidelines for IVI stress the importance of 5.0% povidone-iodine use. The guidelines recommend that povidone-iodine should be the last agent applied before performing injections, and physicians should be careful as the anesthetic may interfere with the contact of povidone-iodine to the conjunctival surface. Therefore, the guidelines recommend applying povidone-iodine before and after topical anesthetic. Our results suggested that, while it may not be necessary in all patients, if the patient has irritation following the application of the povidone-iodine, the reapplication of tetracaine or lidocaine afterwards will not be detrimental to the patient.

Acknowledgments

References

1. Williams GA. IVT Injections: health policy implications. *Rev Ophthalmol*. 2014;21:62–64.
2. Avery RL, Bakri SJ, Blumenkranz MS, et al. Intravitreal injection technique and monitoring: updated guidelines of an expert panel. *Retina*. 2014;34(suppl 12):S1–S18.
3. Dossarps D, Bron AM, Koehrer P, Aho-Glele LS, Creuzot-Garcher C. Endophthalmitis after intravitreal injections: incidence, presentation, management, and visual outcome. *Am J Ophthalmol*. 2015;160:17–25.
4. McCannel CA. Meta-analysis of endophthalmitis after intravitreal injection of anti-vascular endothelial growth factor agents: causative organisms and possible prevention strategies. *Retina*. 2011; 31:654–661.
5. Oguz H, Oguz E, Karadede S, Aslan G. The antibacterial effect of topical anesthetic proparacaine on conjunctival flora. *Int Ophthalmol*. 1999; 23:117–120.
6. Pelosini L, Treffene S, Hollick EJ. Antibacterial activity of preservative-free topical anesthetic drops in current use in ophthalmology departments. *Cornea*. 2009;28:58–61.
7. Tustin A, Kim SJ, Chomsky A, Hubbard GB III, Sheng J. Antibacterial properties of 2% lidocaine and reduced rate of endophthalmitis after intravitreal injection. *Retina*. 2014;34:935–942.
8. Cockerill I, Franklin R, Wikler MA, Alder J, et al. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard*, 9th ed., v. 32. Wayne, PA: Clinical and Laboratory Standards Institute; 2012.

translational vision science & technology

9. Chusri S, Tongrod S, Chokpaisarn J, Limsuwan S, Voravuthikunchai SP. Antagonistic interactions of "Ya-Sa-Marn-Phlae" ethanol extract in combination with topical antiseptics against clinical isolates of staphylococcus aureus. *Biomed Res Int*. 2014;2014:867603.

10. Sueke H, Kaye SB, Neal T, et al. An in vitro investigation of synergy or antagonism between antimicrobial combinations against isolates from bacterial keratitis. *Invest Ophthalmol Vis Sci*. 2010;51:4151–4155.

11. Eliopoulos GM, Eliopoulos CT. Antibiotic combinations: should they be tested? *Clin Microbiol Rev*. 1988;1:139–156.

12. Moody JA, Gerding DN, Peterson LR. evaluation of ciprofloxacin's synergism with other agents by multiple in vitro methods. *Am J Med*. 1987;82:44–54.

13. Isenberg SJ, Apt L, Yoshimori R, Khwarg S. Chemical preparation of the eye in ophthalmic surgery. IV. Comparison of povidone-iodine on the conjunctiva with a prophylactic antibiotic. *Arch Ophthalmol*. 1985;103:1340–1342.

14. Apt L, Isenberg SJ, Yoshimori R. Antimicrobial preparation of the eye for surgery. *J Hosp Infect*. 1985;6(suppl A):163–172.

15. Day S, Acquah K, Mruthyunjaya P, et al. Ocular complications after anti-vascular endothelial growth factor therapy in Medicare patients with age-related macular degeneration. *Am J Ophthalmol*. 2011;152:266–272.

16. Apt L, Isenberg S, Yoshimori R, Paez JH. Chemical preparation of the eye in ophthalmic surgery. III. effect of povidone-iodine on the conjunctiva. *Arch Ophthalmol*. 1984;102:728–729.

17. Speaker MG, Menikoff JA. Prophylaxis of endophthalmitis with topical povidone-iodine. *Ophthalmology*. 1991;98:1769–1775.

18. Isenberg SJ. The ocular application of povidone-iodine. *Community Eye Health*. 2003;16:30–31.

19. Mullin GS, Rubinfeld RS. The antibacterial activity of topical anesthetics. *Cornea*. 1997;16:662–665.

20. Dantas PE, Uesugui E, Nishiwaki-Dantas MC, Mimica LJ. Antibacterial activity of anesthetic solutions and preservatives: an in vitro comparative study. *Cornea*. 2000;19:353–354.

21. Johnson SM, Saint John BE, Dine AP. Local anesthetics as antimicrobial agents: a review. *Surg Infect (Larchmt)*. 2008;9:205–213.

22. Leung YW, Rawal BD. Mechanism of action of tetracaine hydrochloride against pseudomonas aeruginosa. *J Infect Dis*. 1977;136:679–683.

23. van Asten F, van Middendorp H, Verkerk S, et al. Are intravitreal injections with ultrathin 33-g needles less painful than the commonly used 30-G needles? *Retina*. 2015;35:1778–1785.

24. Boden JH, Myers ML, Lee T, Bushley DM, Torres MF. Effect of lidocaine gel on povidone-iodine antisepsis and microbial survival. *J Cataract Refract Surg*. 2008;34:1773–1775.

25. Wachowski I, Jolly DT, Hrazdil J, et al. The growth of microorganisms in propofol and mixtures of propofol and lidocaine. *Anesth Analg*. 1999;88:209–212.

26. Noda H, Saionji K, Miyazaki T. [Antibacterial activity of local anesthetics]. *Masui*. 1990;39:994–1001.

27. Sakuragi T, Ishino H, Dan K. Bactericidal activity of clinically used local anesthetics on staphylococcus aureus. *Reg Anesth*. 1996;21:239–242.

28. Parr AM, Zoutman DE, Davidson JS. Antimicrobial activity of lidocaine against bacteria associated with nosocomial wound infection. *Ann Plast Surg*. 1999;43:239–245.