

Duration of effect and effect of multiple doses of topical ophthalmic 0.5% proparacaine hydrochloride in clinically normal dogs

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Objective—To determine the duration of effect and the effect of multiple doses of topical ophthalmic application of 0.5% proparacaine hydrochloride on corneal sensitivity in clinically normal dogs.

Animals—8 clinically normal dogs.

Procedure—Dogs were randomly allocated to treatment order in a 2 × 2 (period × treatment) crossover study. Treatments consisted of topical application of ophthalmic 0.5% proparacaine (1 drop or 2 drops at a 1-minute interval); treatments were applied to both eyes. A Cochet-Bonnet aesthesiometer was used to determine corneal touch threshold (CTT) before corneal application, 1 and 5 minutes after corneal application, and at 5-minute intervals thereafter for 90 minutes.

Results—The CTT value before treatment differed significantly from CTT values after treatment until 45 minutes after application in the 1-drop group and until 55 minutes after application in the 2-drop group. As determined by use of the Cochet-Bonnet aesthesiometer, a significantly greater anesthetic effect was detected for the 2-drop treatment, compared with the effect for the 1-drop treatment, at 30, 35, 40, 45, 50, and 55 minutes after application. Maximal anesthetic effect lasted for 15 minutes for the 1-drop treatment and 25 minutes for the 2-drop treatment.

Conclusions and Clinical Relevance—Duration of corneal anesthetic effect induced by topical ophthalmic application of 0.5% proparacaine in dogs of this study is considerably longer than that reported elsewhere. Serial application of doses of 0.5% proparacaine increases the duration and magnitude of corneal anesthetic effects. (*Am J Vet Res* 2005;66:77–80)

Topical ophthalmic anesthetic agents have been used for many years to facilitate veterinary ophthalmic diagnostic and therapeutic procedures, including tonometry, scraping of the cornea or conjunctiva, removal of corneal sutures or foreign bodies, and intracameral injection. Topical anesthetics also can be used

in a modification of the Schirmer tear test to investigate basal tear production.^{1,2} Because of its predictability and lack of undesirable adverse effects,³ 0.5% proparacaine is a preferred topical ophthalmic anesthetic in veterinary practice.⁴

In a widely cited letter to the editor,⁵ it was reported that the duration of anesthesia induced by 0.5% proparacaine in veterinary patients is 10 to 15 minutes. However, this information appears to have been determined on the basis of clinical experience, rather than objectively determined data. It has also been stated that an additional drop applied within 1 to 2 minutes of an initial application will enhance anesthesia⁶ and that repeated doses in the form of 3 to 5 applications at 5-minute intervals will achieve complete corneal anesthesia lasting 2 hours.³

To the authors' knowledge, the duration of action of topically applied ophthalmic anesthetic agents and the effects of repeated dosing have not been objectively investigated in dogs, despite a long history of clinical use. The purpose of the study reported here was to determine the duration of effect of a single topical application and evaluate the effects of multiple doses of topical ophthalmic application of proparacaine in clinically normal dogs.

Materials and Methods

Animals—Eight dogs were used in the study. Dogs were included in the study when they were of dolichocephalic conformation and had no evidence of corneal or adnexal disease, as determined on the basis of slit-lamp biomicroscopic examination. Dogs with Schirmer tear test values of < 10 mm/min were excluded from the study. Dogs comprised 6 spayed females and 2 neutered males. Mean ± SD age was 6.1 ± 2.5 years. Breeds represented included 1 Siberian Husky, 1 Golden Retriever, 1 Labrador Retriever, 3 Labrador Retriever-crossbred dogs, and 2 mixed-breed dogs judged to be of dolichocephalic conformation. The protocol for all procedures in the study was approved by the Animal Care and Use Committee of Virginia Tech.

Treatment—The study was conducted by use of a 2 × 2 (period × treatment) crossover design with a minimum 5-day washout phase between periods. Dogs were randomly allocated to treatment order. The study was conducted during a 10-week period. For all treatments, a commercially available ophthalmic anesthetic solution^a was used that contained 0.5% proparacaine hydrochloride and inactive ingredients (0.01% benzalkonium chloride preservative, glycerin, purified water, sodium chloride, and hydrochloric acid or sodium hydroxide to adjust the pH to between 5.0 and 6.0). Treatments consisted of application of 1 drop of anesthetic or 2 drops of anesthetic applied serially at a 1-minute interval; treatments were applied to both eyes. For the 1-drop appli-

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cation, the handler applied an initial sham treatment (ie, topical treatment was simulated, but medication was not applied) followed 1 minute later by application of 1 drop of proparacaine to each eye. Time of the topical application was designated as time 0. A drop of a control solution was not applied during the sham treatment because it would have artificially increased the volume of the lacrimal lake as well as induced reflex tearing, which would have resulted indirectly in excessive dilution of the proparacaine applied 1 minute later. For the 2-drop treatment, 1 drop of proparacaine was applied to each eye, followed by a second drop in each eye 1 minute later. Time of application of the second drop was designated as time 0. Corneal sensitivity measurements began 1 minute after time 0. The same investigator (MPL) performed all treatments. To maintain consistent efficacy,⁷ the proparacaine solution was stored in a refrigerator during the study and anesthetic from the same bottle was used throughout the study.

Measurement of corneal sensitivity—A Cochet-Bonnet aesthesiometer^b was used to measure sensitivity of the central portion of the cornea. To minimize environmental influences, examinations of all dogs were conducted in the same room. Dogs were handled with minimum restraint by the same handler and were maintained in a sitting position during the procedure. To determine corneal sensitivity, the filament of the aesthesiometer was advanced slowly toward the globe and applied perpendicular to the central portion of the cornea. Pressure was increased only until a slight deflection of the filament was evident. Corneal sensitivity, defined as the corneal touch threshold (CTT), was recorded as the length of the aesthesiometer filament that induced a blink reflex on at least 3 of 5 stimulations for a specific filament length. The same investigator (MAB) performed all CTT measurements; that investigator was not aware of the treatment received by each dog.

The CTT was determined before treatment by use of an initial length of 4.0 cm for the aesthesiometer filament.⁸ When a blink reflex was not detected, the filament length was decreased in 0.5-cm increments and testing was repeated until a blink reflex was evident on at least 3 of 5 stimulations at a specific filament length. The CTT was assessed 1 and 5 minutes after topical application of the proparacaine solution and then at 5-minute intervals thereafter until 90 minutes after application. For the measurement obtained 1 minute after treatment, the initial length of the nylon filament was 4.0 cm and it was decreased sequentially, similar to the measurements obtained before treatment. For subsequent measurements, the initial length of the nylon filament was the shortest length that did not result in a positive blink reflex for at least 3 of 5 attempts during the preceding time point.^{9,10} When a blink reflex was detected for at least 3 of 5 stimulations, the length of the filament was increased by 0.5 cm and testing was repeated. The CTT after treatment was recorded as the longest filament length that elicited a blink reflex on 3 of 5 stimulations. The CTT was recorded as 0 when there was no blink reflex to stimulation for the 0.5-cm filament length.

Statistical analysis—Filament length was analyzed by use of a mixed-effects, repeated-measures ANOVA of a commercially available statistical analysis

program.^c Standardized residual plots were used to assess adequacy of the model. Significant treatment-by-time interactions were investigated by comparing the means for the 2 treatments at each time point and by comparing the mean for each treatment at each nonzero time point with the mean value for that respective treatment at time 0. Within these 2 comparisons, Bonferroni correction was performed to maintain the family-wise error rate at 0.05. Values of $P < 0.05$ were considered significant.

Results

The best estimate of corneal sensitivity was assumed to be the mean response for both eyes in each dog. When data for each eye were analyzed separately, the results were nearly identical to results obtained by use of the mean response of both eyes for each dog (ie, there were no differences in statistical outcomes). Therefore, mean values for both eyes of each dog were used for the final analysis.

Mean filament length before treatment with 1 drop of proparacaine was 1.75 cm (95% confidence limits, 1.56 and 1.94 cm). At 1, 5, 10, and 15 minutes after application of 1 drop to each eye, none of the dogs responded to the maximum stimulation (filament length of 0.5 cm). The CTT before treatment differed significantly from the CTT beginning at 1 minute and continuing through 45 minutes after topical application (Figure 1).

Mean filament length before treatment with 2 drops of proparacaine was 1.78 cm (95% confidence limits, 1.59 and 1.98 cm). At 1, 5, 10, 15, 20, and 25 minutes after application of the second drop, none of the dogs responded to the maximum stimulation (filament length of 0.5 cm). The CTT before treatment differed significantly from the CTT beginning at 1 minute and continuing through 55 minutes after topical application (Figure 1).

Pairwise comparison of mean CTT values at each time point after topical application was conducted.

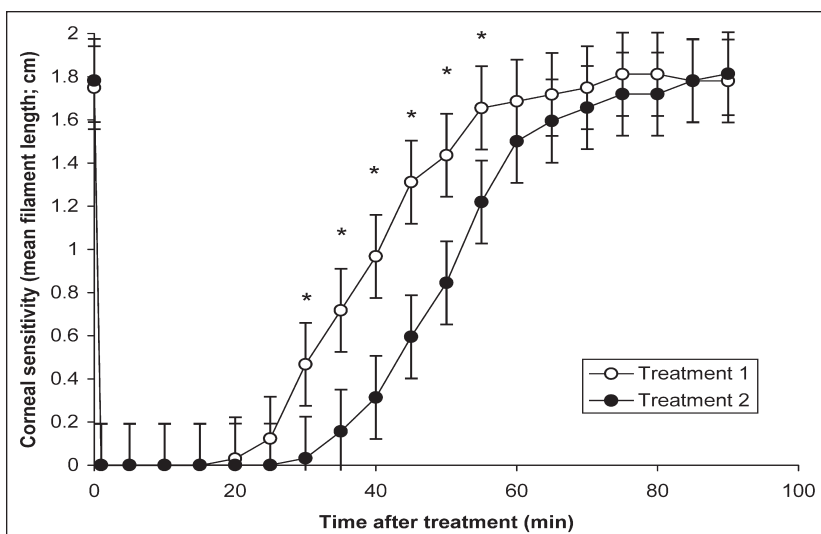


Figure 1—Mean (95% confidence limits) of the filament length for determination of corneal sensitivity in 8 dogs before and after topical ophthalmic application in both eyes of 1 drop (treatment 1) or 2 drops at a 1-minute interval (treatment 2) of 0.5% proparacaine hydrochloride. Time of application of the drop for treatment 1 and time of application of the second drop for treatment 2 were designated as time 0. *Within a time point, values differ significantly ($P < 0.05$) between treatment groups.

This analysis revealed that the effect of the 2-drop treatment was significantly greater than that of the 1-drop treatment at 30, 35, 40, 45, 50, and 55 minutes after topical application (Figure 1).

Discussion

Analysis of results of the study reported here indicates that a single application of 0.5% proparacaine in the eyes of clinically normal dogs results in a significant corneal anesthetic effect that lasts 45 minutes, with a maximal anesthetic effect that lasts approximately 15 minutes. A second drop of proparacaine applied 1 minute after the first extends the period with significant anesthetic effects to 55 minutes and results in a period of maximal anesthetic effect of 25 minutes. Onset of corneal anesthesia is rapid because no blink reflex could be elicited at 1 minute after treatment in either treatment group. The central corneal CTT measurements obtained before treatment in this study are similar to values reported^{8,11} elsewhere for clinically normal dogs.

Objective studies of the duration of the effects of proparacaine in animals are limited. In rabbits, the onset of action reportedly¹² is < 1 minute and duration of anesthesia is 63 minutes. In humans in which a cotton wisp was used as a stimulus for assessing corneal anesthesia, duration of action of 0.5% proparacaine is 10.7¹³ and 15.2 minutes.¹⁴ By use of Cochet-Bonnet aesthesiometry, duration of maximal effect of 0.5% proparacaine in humans with normal eyes is 11.7 minutes¹⁵ with a complete recovery time of 34.9¹⁵ to 45 minutes.⁹ These recovery times are similar to our findings for dogs in the 1-drop treatment group. In a comparison of 0.125%, 0.25%, and 0.5% concentrations of proparacaine, stronger concentrations increased the initial degree and duration of corneal anesthesia but a rapid recovery rate for corneal sensitivity was detected for all applied concentrations.⁹ A rapid recovery rate was also evident for both treatment groups in the study reported here (Figure 1).

In another report⁵ involving dogs, it was stated that the duration of corneal analgesia provided by topical application of 0.5% proparacaine is approximately 10 to 15 minutes. Because the methods used in that report were not described, we assume that the reported duration of effect was determined on the basis of clinical observation. Total duration of corneal anesthetic effect in the study reported here was substantially longer than for that other report.⁵ However, the duration of maximal anesthetic effect for the 1-drop treatment (approx 15 minutes) was similar to the duration of analgesic effect reported.⁵ This period probably corresponds to the time frame in which corneal anesthesia is sufficient to allow clinical manipulations of the ocular surface.

Corneal hypesthesia significantly prolongs the duration of action of 0.5% proparacaine in humans.¹⁵ Reduced corneal sensitivity has been documented¹⁶ in dogs with diabetes mellitus, and it also has been reported⁸ that dogs of mesaticephalic and brachycephalic cranial conformation have significantly lower corneal sensitivity than do dolichocephalic dogs. Only ophthalmically normal dogs of dolichocephalic confor-

mation were used in the study reported here, so it remains to be determined whether the findings of our study are applicable to dogs with other cranial conformations or afflicted with diseases that affect corneal sensitivity.

It has been stated^{3,6} that repeated topical instillation of proparacaine will enhance and prolong anesthetic effects; however, to our knowledge, objective investigation of the effect of serially applied doses of proparacaine in dogs has not been reported. In the study reported here, a second drop of anesthetic applied 1 minute after the first drop considerably prolonged the period of corneal anesthesia. This most likely was a simple reflection of increased drug concentration in corneal tissues. Proparacaine and other local anesthetics act by impeding the entrance of sodium ions into the interior of axons, thus preventing depolarization.¹⁷ The extent of sodium-channel blockade by local anesthetics is a dose-dependent phenomenon, with higher drug concentrations causing a decreased latency period for anesthetic onset and increased duration of effect.¹⁸ Repeated instillation of medication at 1-minute intervals effectively increases corneal drug concentrations,¹⁹ and there is a direct relationship between the applied concentration of topical ophthalmic anesthetics and the degree and duration of their effect in humans.^{9,20} Although many drugs applied as ophthalmic drops will induce reflex tearing that results in rapid drug loss through the nasolacrimal system, topical anesthetics dramatically reduce tear production, thereby actually enhancing drug bioavailability.²¹ The combination of these factors is likely responsible for the increased duration of effect evident in the 2-drop treatment group in our study.

The Cochet-Bonnet aesthesiometer has been widely applied in the investigation of corneal sensitivity in veterinary ophthalmology.^{8,11,16,22,23} Limitations of this instrument include subjective interpretation of filament deflection and variability in filament stiffness attributable to changes in temperature and humidity. We attempted to minimize these problems by having 1 investigator perform all tests and by performing all tests in the same physical environment. Following the onset of corneal anesthesia in our study, the aesthesiometer filament was initially set at the shortest length that did not result in a positive blink response during the preceding time point. When a blink reflex was elicited at that length, the filament was then lengthened by 0.5 cm and testing was repeated.^{9,10} By starting with a shorter filament length (a more noxious stimulus) and then increasing the length of the filament, it is possible, although unlikely, that the dogs may have become conditioned to the stimulus to some degree, thus confounding our results. The fact that corneal sensitivity by the end of the study had returned to values nearly identical to values obtained before treatment suggests that any such effect was negligible.

Unfortunately, constraints of the Cochet-Bonnet aesthesiometer prevented us from determining whether the additional dose of proparacaine achieved a higher degree of maximal corneal anesthesia. Although shortening the filament length to < 0.5 cm would have resulted in considerably more force being applied to the

corneal surface and perhaps allowed discrimination of the maximal degree of anesthesia between the 2 treatment groups, the investigators believed it likely that the rapid increase in filament stiffness as a result of shortening the filament length to < 0.5 cm would have resulted in excessive corneal trauma.²⁴ The increase in duration of maximal anesthetic effect, increase in total duration of effect, and greater anesthetic effect at 30 to 55 minutes after application for the 2-drop treatment, compared with values for the 1-drop treatment, suggest that the magnitude of maximal corneal anesthesia may have been greater with the 2-drop treatment, but we could not confirm it in our study.

The negative effect of topical anesthetics on aqueous tear production as measured by the Schirmer tear test has been established.^{1,2} Because diminished lacrimation with topical anesthetics is directly related to a reduction in corneal sensation caused by the anesthetic, it is likely that Schirmer tear test values are affected to some degree for at least 45 minutes (and perhaps even longer) after the application of proparacaine. This effect should be taken into consideration when lacrimal function is evaluated after topical administration of proparacaine. Additionally, on the basis of analysis of results of our study, we recommend that investigators wait approximately 1 hour after topical application of proparacaine before measuring corneal sensitivity to ensure accurate results.

The data reported here provide an objective reference for the duration of effect of topical ophthalmic application of 0.5% proparacaine in clinically normal dogs. The duration of effect reported here was significantly longer than that suggested in another report.⁵ Instilling a second drop of proparacaine 1 minute after the first drop results in a substantial increase in the duration of corneal anesthesia and may also increase the intensity of anesthetic effects. It must be acknowledged that although the duration for the detectable effect of a single application of proparacaine is 45 minutes as measured by Cochet-Bonnet aesthesiometry, the duration of effective corneal anesthesia required for most clinical manipulations is likely to be shorter.

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- a. Ophthetic, Allergan Inc, Irvine, Calif.
 - b. Cochet-Bonnet aesthesiometer, Luneau Ophtalmologie, Chartres Cedex, France.
 - c. SAS, version 8.02, SAS Institute Inc, Cary, NC.
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