ORIGINAL ARTICLE

Effect of topical application of tetracaine on intraocular pressure in dogs: Preliminary results

Effets de l’application topique de tétracaïne sur la pression intraoculaire chez le chien : résultats préliminaires

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KEYWORDS
Intraocular pressure (IOP); Applanation tonometry; Topical anesthetics; Tetracaine; Dog

Summary
Purpose. — To evaluate the effect of topical application of tetracaine on intraocular pressure (IOP) measurement by Tonopen in dogs.

Subjects and methods. — Six healthy male Epagneul Bretons (group 1) and six healthy male black Labrador Retrievers (group 2) were examined. IOP was measured in the right eye (OD) prior to (IOP1) and 1 minute following instillation of one drop of topical tetracaine (IOP 2), and the left eye (OS) (control) prior to (IOP 3) and 1 minute following instillation of one drop of isotonic saline solution (IOP 4). Measurements were performed on two occasions: at 8:00AM and 3:00PM.

Results. — For both groups, IOP measurements were higher in the morning than in the afternoon. For group 1, IOP1 mean (SD), IOP2 mean (SD), IOP3 mean (SD) and IOP4 mean (SD) were 14.6 (2.2) mmHg, 11.3 (3.2) mmHg, 14.4 (2.2) mmHg and 13.5 (3.9) mmHg respectively, while in group 2, IOP1 mean (SD), IOP2 mean (SD), IOP3 mean (SD) and IOP4 mean (SD) were 14.2 (3.8) mmHg, 9.5 (3.7) mmHg, 13.5 (2.8) mmHg and 13.0 (3.8) mmHg respectively. For both groups at each time point, IOP 2 values were significantly lower (P < 0.007) than IOP 1 values, whereas IOP 3 and 4 values were not significantly different (P > 0.27).

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Conclusion. — This study demonstrates that topical application of tetracaine significantly lowers IOP measured by Tonopen due to a possible interaction with melanin. The potential effect of topical anesthetics should be taken in consideration when performing applanation tonometry for clinical, pharmacological and toxicological studies.

Résumé

Objectifs. — Évaluer les effets de l’application topique de tétracaine sur la mesure de la pression intraoculaire (PIO) au moyen du Tonopen, chez le chien.

Sujets et méthodes. — Six Epagneuls Bretons mâles, cliniquement normaux (groupe 1) et six Labrador Retrievers mâles à pelage noir, cliniquement normaux (groupe 2) ont été examinés. La PIO a été mesurée sur l’œil droit (OD) avant (PIO1) et une minute après instillation d’une goutte de collyre à la tétracaine (PIO2), et sur l’œil gauche OS (témoin) avant (PIO 3) et une minute après instillation d’une goutte de solution saline isotonique (PIO 4). Les mesures ont été effectuées à 8 heures et à 15 heures.

Résultats. — Pour les deux groupes, les valeurs de PIO étaient plus élevées le matin que l’après-midi. Pour le groupe 1, les valeurs de la moyenne (écart-type) de la PIO étaient de 14,6 (2,2) mmHg pour la PIO1, 11,3 (3,2) mmHg pour la PIO2, 14,4 (2,2) mmHg pour la PIO3 et 13,5 (3,9) mmHg pour la PIO4 alors que pour le groupe 2, les valeurs de la moyenne (écart-type) de la PIO étaient de 14,2 (3,8) mmHg pour la PIO1, 9,5 (3,7) mmHg pour la PIO2, 13,5 (2,8) mmHg pour la PIO3, et 13,0 (3,8) mmHg pour la PIO4. Pour les deux groupes et quelle que soit l’heure de mesure, les valeurs PIO2 étaient significativement ($p < 0,007$) inférieures aux valeurs PIO 1 alors que les valeurs PIO3 et PIO4 n’étaient pas significativement différentes ($p > 0,27$).

Conclusion. — Cette étude montre que l’application topique de tétracaine entraîne une diminution significative de la PIO mesurée avec un Tonopen, peut-être due à une interaction avec la mélanine. Les effets potentiels d’anesthésiques topiques devraient être pris en considération lors de la mesure de tonométrie par applanation pour des études cliniques, pharmacologiques et toxicologiques.

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Introduction

The estimation of intraocular pressure (IOP) is an important clinical procedure in the diagnosis and monitoring of glaucoma in animals, but also in other ocular diseases such as uveitis, and in the postoperative management of lenticular and vitreoretinal conditions [1]. The applanation tonometer is one of the most precise and trustworthy instruments for diagnosing small animal intraocular diseases [2, 3]. Because of its convenient price and its efficiency in the acquisition of clinically reliable readings in many species, the Tonopen® is currently the most widely used instrument for estimating IOP in veterinary ophthalmology [4–6] and investigative ophthalmology [7–9], while being small, light and easy to handle. According to manufacturers’ specifications, an average of three successive readings is sufficient to record IOP after application of topical anesthesia. It is important to consider that since the Tonopen® is very accurate in the normal range of IOP it tends to overestimate IOP in the low range and underestimate IOP in the high range [10]. Therefore, IOP measurements using this tonometer have variable results due to the operator and other factors including gender, age, breed, ocular diseases [11] and circadian rhythms [12, 13]. IOP is a biologic variable and diurnal variations in IOP have been documented in the dog, with higher levels in the early morning and the lowest reading in the early evening [13, 14]. These diurnal variations observed in normal dogs are enhanced in untreated primary open angle glaucoma (POAG) [14]. Finally, it has been shown in humans that local drops of anesthetics may reduce the IOP readings [15]. Interestingly and according to the author’s knowledge, there are no similar studies published about canine eyes despite the very wide use of Tonopen®.

In addition, given that the incidence of glaucoma seems to vary markedly between different dog breeds [16, 17] this study was performed to see whether there was an interbreed variability of any change in IOP measurement using Tono-Pen Vet® following corneal application of tetracaine. The present study was performed using animals chosen from two popular breeds of dogs in the South West of France: Epagneul Bretons and Labrador retrievers.

Subjects and methods

The protocol adhered to the Association for Research in Vision and Ophthalmology Statement for the Use of Animals in Ophthalmic and Vision Research. Animals were examined at the Veterinary Eye Clinic (Dr Thomas Boillot) following the consent of the owners.

Animals

Six intact white and red coated males Epagneul Bretons with light yellow/brown iris (Fig. 1a, group 1) and six black coated intact males Labrador Retrievers with dark brown

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iris (Fig. 1b, group 2) have been enrolled in this study with a mean (SD) age and weight of 3.8 (2.9) years and 15.6 (2.2) kg respectively for the Epagneul Bretons, and 3.6 (2.3) years and 34.2 (4.9) kg respectively for the Labrador Retrievers. Dogs were maintained under the same environmental conditions throughout the duration of the study.

All dogs received complete physical and ophthalmic examinations, including slit-lamp biomicroscopy (Kowa SL 15, Centravet, Lapalisse, France), applanation tonometry (Tono-pen Vet®, Centravet, Lapalisse, France) and indirect ophthalmoscopy (Heine Omega 180, Centravet, Lapalisse, France). Criteria of inclusion were an absence of systemic and ophthalmic diseases.

### Intraocular pressure measurements

All IOP measurements were performed the same day by the same investigator (T. Boillot) using the same instrument: a Tonopen Vet® tonometer. The Tonopen Vet® makes four instantaneous readings and return the average of these readings and their percentage of variation (PAV). All IOP measurements were performed without sedation and with all the dogs maintained in a stable sitting position. They were performed at the center of the cornea in order to reduce possible variations due to various corneal thicknesses [18]. When performed without topical anesthesia, the tonometer was applied very gently on the corneal surface. Some soft touches were made before to start the procedure to get the dogs used to tolerate the contact with the tonometer’s tip. Measurements consisted in three successive reliable (PAV < 5%) IOP readings. Their mean was used for further calculations. IOP was measured in four different conditions: in the right eye (OD), prior (IOP1) and 1 minute following instillation of one drop of topical anesthetic solution (Tetracaine®, Virbac, France) (IOP 2), and in the left eye (OS), prior (IOP 3) and 1 minute following instillation of one drop of isotonic saline solution (IOP 4). IOPs were measured in two occasions, at 8:00 AM and at 3:00 PM.

For each group and for each occasion, means and standard deviations were calculated and a paired t-test (P ≤ 0.05) was performed in order to determine the statistical significance.

### Results

Measurements were easily performed in all dogs and in all conditions. As indicated in Table 1A and B, irrespective of the condition and time point, mean IOP values were slightly lower in group 2 but did not yield any significant difference in comparison with group 1 (P > 0.01). As indicated in Table 1C, for both groups and for each condition, mean IOP values measured at 8:00 AM were higher in comparison with mean IOP values measured at 3:00 PM. Regardless of the time point, measurements performed without any topical application (IOP 1 and IOP 3) did not yield any significant difference (P > 0.01) in both groups. On the other hand, mean IOP values measured after topical application of tetracaine (IOP2) were significantly lower (P ≤ 0.05) than mean IOP values measured after topical application of isotonic saline solution (IOP 4). In group 1, the difference was 16% and 18% at 8:00 AM and 3:00 PM respectively, whereas in group 2, the difference was 26% and 19% at 8:00 AM and 3:00 PM respectively. As indicated in Table 2, for each group mean IOP values measured prior (IOP 3) and after (IOP 4) application of topical isotonic saline solution of isotonic saline solution did not yield any significant difference (P > 0.01) whatever the time point. Interestingly, as indicated in Table 3, for both groups and at each time point, mean IOP values measured after topical application of tetracaine (IOP 2) were significantly lower (P < 0.007) than mean IOP values measured prior topical application of tetracaine (IOP 1). In group 1, the decrease was 22% and 21% at 8:00 AM and 3:00 PM respectively, whereas in group 2 the decrease was 33% and 27% at 8:00 AM and 3:00 PM respectively. These results indicated that topical application of tetracaine induced a rapid decrease in IOP measured with Tonopen. This change seems to be independent of time and breed, however it was more prominent in black coated group, especially when measurements were performed on the morning.

### Discussion

In both groups, no significant difference has been found in the IOP baseline between OD (IOP1) and OS (IOP3). However, changes in IOP depending on time, although not significant,
Table 1  Intraocular pressure (IOP) measured with Tonopen in two breeds of dogs in two occasions. Mean (standard deviation) values and paired t-tests are presented. IOP was measured OD, prior (IOP 1) and 1 minute following instillation of one drop of topical anesthetic solution (Tetracaine®, Virbac, France) (IOP 2), and OS (control), prior (IOP 3) and 1 minute following instillation of one drop of isotonic saline solution (IOP 4). Measurements were performed in two occasions: at 8 AM and 3 PM. Group 1 (GP1) was composed with six healthy males Epagneul Bretons and group 2 (GP2) was composed with six healthy males Labrador Retrievers.

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<td>SD</td>
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<td>P</td>
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were found, which corroborates previous studies concerning influence of circadian rhythm on IOP [13,14].

The mean IOP values we measured after topical application of tetracaine (IOP2) were comparable to those obtained with the same device by other authors in previous studies with the use of tetracaine [19] or proparacaine [6,18], but were lower than values obtained with the use of proxymetacaine [17] and lidocaine [20]. This suggests that application of different local anesthetics may partly influence results in tonometry as they all induce a decrease of IOP

Table 2  Effects of saline solution on intraocular pressure (IOP) in OS (control). Mean (standard deviation) IOP values were lower on the afternoon versus morning and in group 2 versus group 1, but these effects were not statistically significant.

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<tr>
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<th>Group 1 3:00 PM</th>
<th>Group 2 8:00 AM</th>
<th>Group 2 3:00 PM</th>
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<tr>
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<td>P</td>
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<td>0.39</td>
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measurements. Interestingly, a study comparing various devices in beagle dogs such as rebound tonometer and Tonopen® reported similar effect of decrease in IOP measurement performed with a rebound tonometer after application of proparacaine [18]. This suggests that the effect of topical anaesthetic is independent of the device used to measure tonometry. This phenomenon could not be related to mechanical effects of repetitive IOP measurements because similar findings were observed in humans with the use of a non-contact tonometer less than one minute after application of various topical anesthetics as oxybuprocaine and betoxycaine [15]. Although its mechanisms remain to be determined, variations in central corneal thickness do not seemed to be associated with this effect [21].

In 2007, Taylor et al. compared IOP measurements in three different breeds of dogs with the use of Tonopen® associated with topical application of proproxytacaine and observed higher values in the group of Siberian husky (supposed to be less pigmented) than in the other groups constituted of Golden retrievers and English cocker spaniels, phenotypically comparable to Epagneul Bretons.

Notwithstanding the numerous parameters influencing IOP measurements, we hypothesize a possible effect of topical anesthetics linked with pigmentation of the iris. It is well known that a number of drugs and other chemicals are accumulated and retained for long periods in pigmented tissues due to melanin affinity [22]. Among those, xenobiotics and tetracaine, as a weak base (pKa 8.5) can bind to melanin [23]. Thus, melanin binding may significantly lower pharmacological activity [24]. Melanin binding in the iridociliary body may affect drug concentration in anterior ocular tissues and drug response [25].

In our study, black-coated Labrador Retrievers (group 2) had lower mean IOP values than Epagneul Breton (group 1). Considering the differences in IOP values between the black-coated Labrador Retrievers, we used and the Siberian Huskies enrolled in Taylor et al.’s study, it may be hypothesized that basal IOP measured with the use of tetracaine could be related to iris pigmentation. Interaction with these pigments can alter pharmacokinetic properties of the drug. Tetracaine has shown the highest affinity for melanin compared to other topical anesthetics such as proparacaine, bupivacaine and lidocaine [22]. Therefore, differences in duration of action may be expected between deeply and less pigmented eyes. Although melanin is suspected to have a lowering effect on pharmacological activity of drugs [23], this pigment could be involved in a mechanism leading to an enhancement of the effect of tetracaine in pigmented dogs.

How the interaction between melanin and tetracaine could be related to a decrease of IOP remains unknown. Still, a facilitating effect on aqueous humor outflow appears more likely than an inhibitory action on its production. The pharmacological effect of melanin is characterized by a strong uptake and a slow release. The pharmacological kinetic of its interaction with tetracaine has to be investigated to understand the effect found on IOP. Rather than variations due to the breed, it could be variation due to pigmentation. However, care must be taken because it is difficult to evaluate the concentration of melanin in the iris of both breeds. Clearly, further studies are needed for a better understanding of the interaction of topical anesthetics with pigmentation in various breeds of dog.

Our study demonstrates that tetracaine significantly lowered IOP compared to control in the Epagneul Bretons and the Labrador Retrievers. Interbreed variations in the effect of tetracaine, possibly due to interaction with melanin, have been suspected. The potential effect of topical anesthetics should be taken in consideration when performing applanation tonometry for clinical, pharmacological and toxicological studies.

Disclosure of interest

The authors declare that they have no conflict of interest concerning this article.

References

Effect of tetracaine on IOP in dog


