Intraocular pressure and Schirmer tear test values in maned wolf (*Chrysocyon brachyurus*)[1]

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ABSTRACT.- Honsho C.S., Jorge A.T., Oliveira L.T., Paulino-Junior D., Mattos-Junior E., Nishimura L.T. & Dias W.O. 2016. **Intraocular pressure and Schirmer tear test values in maned wolf (*Chrysocyon brachyurus*).** *Pesquisa Veterinária Brasileira 36(9):919-923*. Curso de Pós-Graduação de Medicina Veterinária, University of Franca, Parque Universitário, Avenida Dr. Armando Salles Oliveira 201, Franca, SP 14404-600, Brazil. E-mail: cristiane.honsho@unifran.edu.br

The purpose of this study was to establish baseline data on lacrimal quantity (STT-1) and intraocular pressure (IOP) in captive maned wolves. Ten healthy adult maned wolves were contained with a snare pole and muzzle and kept in decubitus of the left side. STT-1 measurement was performed on the lateral third of the lower conjunctival sac for one minute. The cornea was desensitized and intraocular pressure was measured with an tonopen. Average STT-1 in both eyes was 11±5mm.min^{-1}, with no statistical difference between the left and right eye (p=0.960). Average IOP in both eyes was 20±6mmHg, with no statistical difference between the left and right eye (p=0.836). Average STT-1 was lower than, and IOP was the same as normal levels found in dogs. There was no statistical difference in the age of the animals, and STT-1 and IOP values. In the present paper, average maned wolf STT-1 levels were lower compared with those found in dogs, while the IOP was the same in maned wolves as in dogs. Due to the increased incidence in providing emergency care for maned wolf victims of road kill and fires, determination reference values of ocular parameters may improve the correct diagnosis and treatment of the disease.

INDEX TERMS: Intraocular pressure, Schirmer tear test, maned wolf, *Chrysocyon brachyurus,* ophthalmology, tonometry, eye.

RESUMO.- [**Pressão intraocular e teste lacrimal de Schirmer em lobos-guará (*Chrysocyon brachyurus*).**] O objetivo deste estudo foi estabelecer dados de referência sobre a produção lacrimal (STT-1) e pressão intraocular (PIO) em lobos-guará em cativeiro. Foram utilizados 10 lobos-guará, saudáveis e adultos. Os animais foram contidos com cambão e mordaça e mantidos em decúbito lateral esquerdo. O TLS foi realizado no terço médio da pálpebra inferior, durante um minuto. A córnea foi dessensibilizada e a PIO mensurada com tonopen. A média do TLS-1 dos olhos direitos e esquerdo foi 11±5mm/min, não havendo diferença significativa entre os olhos (p=0,960). A média da PIO dos olhos direitos e esquerdo foi 20±6mmHg, não observando diferença entre os olhos direitos e esquerdos (p=0,836). Média STT-1 foi menor do que, e PIO foi o mesmo que os níveis normais em cães. Não houve diferença estatística na idade dos animais e valores STT-1 e da PIO. No presente trabalho, os níveis médios de guará STT-1 foram mais baixos em comparação com as observadas nos cães, enquanto que a pressão intraocular foi a mesma nos lobos guará como em cães. Devido ao aumento da incidência na prestação de cuidados de emergência para vítimas de lobos-guará atropelamentos e incêndios, determinar os valores de referência dos parâmetros oculares podem melhorar o diagnóstico correto e tratamento de doenças.

TERMOS DE INDEXAÇÃO: Oftalmologia Veterinária, pressão intraocular, Lobo-guará, animal de cativeiro, olho.

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INTRODUCTION

Baseline ocular data in healthy animals is essential to establishing normal ranges. Due to the immense variation in these values across species, it is impossible to extrapolate between wild and exotic species (Grundo et al. 2011). Lacrimal production and intraocular pressure values have not been clarified in maned wolf and since these suffer from environmental interference, it is important to establish specific values by region.

The maned wolf (*Chrysocyon brachyurus* Illiger, 1815) (Fig.1) is the largest canine that inhabits the South American savannahs (Teodoro et al. 2012) of Brazil, Paraguay, Bolivia, Argentina, Uruguay and Peru (Rodden et al. 2008). Solitary and territorial, it has historically been considered the most typical animal of the Brazilian *cerrado.* With the passage of time, there has been a decline in the population due to habitat loss caused by deforestation, road accidents, hunting, illegal trade (Santos et al. 2003, Rodden et al. 2008) and popular belief that hold parts of maned wolf bodies (e.g. the eyes) serve as amulets and provide protection (Diniz et al. 1999). For these reasons, it is categorized as vulnerable on the Brazilian list of threatened species (MMA 2003) (Swenson et al. 2012) and near threatened according to the IUCN (2008) (Rodden et al. 2008, Swenson et al. 2012).

Given that the literature contains sparse reference to ocular parameters in maned wolf, the objective of this study was to establish the lacrimal quantity and intraocular pressure of captive animals from the Companhia Brasileira de Metalurgia e Mineração's (CBMM) conservation breeding center located in Araxá, Minas Gerais, Brazil, at an altitude of 1,095m.

Fig.1. A male adult maned wolf from Companhia Brasileira de Metalurgia e Mineração's (CBMM) Wild Fauna Conservation Breeding Center located in Araxá, Minas Gerais, Brazil.

MATERIALS AND METHODS

The study complies with the ethical criteria of the Association for Research in Vision and Ophthalmology (ARVO 2013), as well as CBMM and the Committee for the Ethical Use of animals of the Franca University, as registered under protocol 031/13 approved on June 17, 2013.

Animals Ten healthy maned wolves, female (n=4) and male (n=6), aged between 15 and 148 months without ocular disease. Examinations were carried out in October 2013.

Physical restrain. CBMM's workers contained the animals with the assistance of a snare pole (Ketch All Pole, San Luis Obispo, CA, USA) and muzzles. The maned wolves were placed in a left lateral position so that the pole would only exert pressure if there were an attempt to escape.

Ocular measurements. Following physical restrain, the Schirmer tear test (STT-1) was performed on the lateral third of the lower conjunctival sac applying sterile commercial strips (Tear Flo®, Contacare Ophthalmics & Diagnostics, India) for one minute. Then, during the same procedure, the cornea was desensitized with proxymetacaine eye drops (Anestalcon® - Alcon S/A, Brazil). To measure intraocular pressure, three consecutive measurements were taken in each eye using an applanation tonometer (Tonopen Avia® - Reichert Inc., USA) with significance set at less than 5%. The device was positioned perpendicular to the cornea, lightly touching the central region. All the measurements were gathered between 9:00 am and 4:00 pm by the same veterinary ophthalmologist and, the head of wolves positioned in the same position in all measurements. Prior to the procedure, the device was calibrated for each animal.

Statistical analysis. The average IOP and STT-1 from the right and left eyes were compared using the unpaired t test (GraphPad Prism v. 5). Pearson's correlation coefficient was used to evaluate the correlation between IOP and STT-1 in relation to age. The level of significance was defined as 5%.

RESULTS

No animal presented signs of ocular disease. There was no statistical difference in the STT-1 values between the right and left eyes (p=0.960), with the average being 11±5mm/min. This variable had an inverse relationship to age (r=-0.139) (Fig.2).

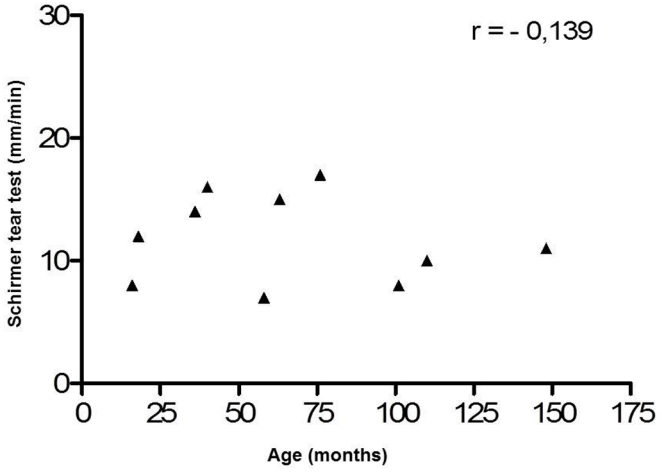

Fig.2. Correlation between individual tear production as measured by the Schimer tear test and age in 10 maned wolves. The variables show an inverse relationship, with little or no probability of differences based on age (p=0.701).

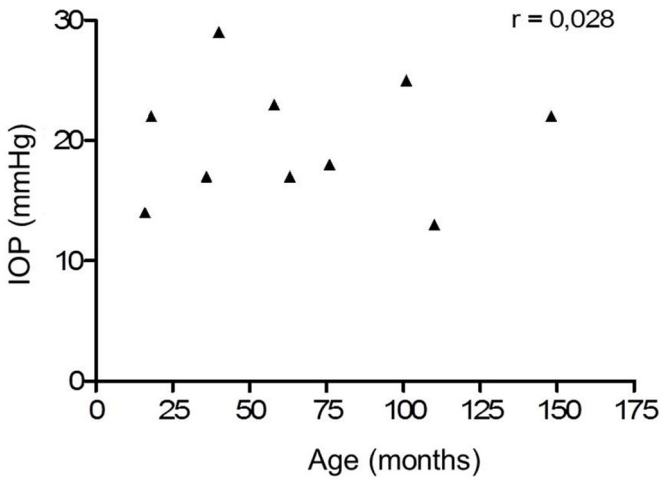

Fig.3. Correlation between individual IOP values and age in 10 maned wolves. The variables exhibit the same trend, with little or no probability of aged-based alterations (p=0.937).

The average IOP of the right and left eyes was 20 ± 6 mmHg, with no significant difference between the two (p=0.836). No correlations were found between IOP and age and between Schirmer tear test results and age trend (r=0.028) (Fig.3).

DISCUSSIONS AND CONCLUSIONS

Knowing the normal ranges of ocular morphophysiology is essential to the correct diagnosis of ocular disorders in different species (Oriá et al. 2015), even though at times alterations can be seen even within the same family. Thus, establishing reference values is necessary to avoid diagnostic and treatment errors (Ofri et al. 2002). Maned wolves are susceptible to parasitic, viral and bacterial illness in both wild and captive settings (Diniz et al. 1999). Several studies have been published in the literature on the habitat, behavior, anatomy and physiology of maned wolves. Work at CBMM's breeding center with maned wolves has reported the presence of urolithiasis in four animals (Fernandes & Marcolino 2007), a comparison of serum hormone levels (Maia et al. 2008), seasonal aspects of male reproductive physiology[2] and aspects of reproductive physiology and stress in captivity (Spercoscki 2013).

Systemic anesthetic agents can alter STT-1 and IOP values (Ghaffari et al. 2011). In dogs, the decrease in IOP occurs due to the relaxation of the extraocular muscles and the drop in episcleral blood pressure (Ofri 2002, Ofri et al. 2002). However, sedating wild animals often makes ocular examination impossible (Ofri et al. 2002, Ghaffari et al. 2011). Since the animals in the present study are adapted to being handled by breeding center workers, it was decided to contain them physically in a standardized fashion in order to obtain real data related to tear quantity and IOP.

Standardizing the lateral position on the left side facilitated containment and physical restrain. Dogs maintained in left-side position did not present alterations in IOP levels in either eye (Honsho et al. 2004), however another study demonstrated a 2-3 mmHg difference in the IOP value depending on the decubitus studied (dorsal, ventral, seated) (Broadwater et al. 2010). Furthermore, the authors relate the importance of maintaining the animals in the same position prior to measuring IOP and always using the same position to annul the effects that body position may have on the obtained results (Broadwater et al. 2008).

STT-1 measures the basal and reflex production of the lacrimal film (Broadwater et al. 2010, Trbolova et al. 2012). The values are different across species due to diverse habitats, variations in the size of the conjunctival sac and modification on the ocular surface. A 1-2mm/min decrease in some species may not provoke a significant difference, but in others species it may have serious implications (Ofri et al. 2002).

In the present study, the average STT-1 levels were lower than the normal values seen in dogs (Gelatt et al. 1975). In 2012 the STT-1 in a single maned wolf with a prolapsed gland in the lower eyelid was 15mmHg (right eye) and 12mmHg (left eye) (Morales et al. 2012), which is within the normal range found in this study. The normal STT-1 level in wild dog (*Cerdacyon thous*), 13.97±2.46mm/min in both eyes, was lower than maned wolf values (Renzo et al. 2014).

There is great variation in the STT-1 levels across species: goats 15.8±5.7mm/min (Broadwater et al. 2007), Sanjabi sheep, 18.52±2.55mm/min (Ghaffari et al. 2011), pigs 15.6±3.7mm/min (Trbolova & Ghaffari, 2012), capybara 14.97±4.66mm/min (Montiani-Ferreira et al. 2008) and broad-snouted caiman (3.4±1.2mm/min) (Oriá et al. 2015). In male ferrets it was 4.80±0.88mmHg and in females 5.72±1.50mmHg (Montiani-Ferreira et al. 2006). Guinea pigs have measurements of 0.36±1.09mm/min and this low level is thought to be the result of not having reflex secretions due to the low corneal sensitivity or limited lacrimal reflex (Trost et al. 2007). Findings in llama (*Lama glama*) were similar to those of domestic ruminants, such as sheep and goat (15.8±5.7mm/min), with the left eye measuring 17.6±1.2mm/min and the right eye 17.1±1.3mm/min (Trbolova et al. 2012).

There are reports that circadian rhythm affects STT-1 (Hartley et al. 2006, Piccione et al. 2009) and IOP by altering the level of the adrinocorticotropic hormone (Ofri 2002). In dogs, the daily STT-1 variation was 0.7 mm/min (Hartley et al. 2006), although a different study reported a variation of 2 mm/min (Piccione et al. 2009). The difference in the STT-1 was not seen as relevant in the diagnosis of dry keratoconjuntivitis and thereby did not require controlling time of day (Hartley et al. 2006). Given this, and also to prevent alterations resulting from the stress caused by physical restrain several times in the same day, the choice was made to measure the maned wolf only during the day.

Even though there were variables with inverse relationships in the present study, there was no difference related to the age of the animal and STT-1 values. In dogs, there was a decrease of 0.4 mm seen for each year of life (Hartley et al. 2006). Lacrimal production was significantly altered based on age, weight and sex in dogs, and after nine-to-ten weeks of age, there was an increase of 0.15mm/min for each day of life of the animal and of 0.84 mm/min for each kilogram of weight (Broadwater et al. 2010).

To avoid external influences on the results and discordance with what has been reported in the literature (Hartley et al. 2006), a single lot of STT was used to prevent alterations in the absorption and porosity of the paper. In addition, the test was always performed by the same veterinary ophthalmologist during the same time of year to standardize the humidity and ambient temperature.

Ocular factors that contribute to IOP across species result from anatomical and physiological differences in ciliary body, aqueous humor drainage, corneal curvature and rigidity (Ofri et al. 2002, Ribeiro et al. 2010), extraocular muscular tone, eyelid closure and muscular retraction of the eyeball (Ribeiro *et al.* 2010). Other factors that can affect IOP include age, race, weight, season and circadian cycle (Ofri 2002). Variables such as season, circadian cycle and body position were standardized in order to limit oscillation in the IOP values.

Reports in the literature related interference of the circadian cycle on IOP in Saanen goats (Ribeiro et al. 2010) and cats (Sole et al. 2007). An increase in IOP is related to the animal's active period and researchers observed an IOP peak during the night in cats (Sole et al. 2007). In the present study, IOP measurements were not carried out at night in maned wolf to avoid the interference of stress related to containing and physical restrain.

All the animals were muzzled and constrained in a loose pole snare that did not apply pressure to the cervical region. In dogs, the pressure exerted on the jugular vein was confirmed to increase IOP, possibly due to venous stasis, provoking a resistance to aqueous humor drainage because of the increased pressure in the episcleral vessels, or the elevated volume of blood in the choroid (Klein et al. 2011). In the present study, efforts were made to prevent the snare from pressuring the cervical region since this was described in the literature to lead to elevated IOP levels in maned wolves compared to dogs (Ferreira et al. 1996), nevertheless, this situation may have influenced the IOP values.

The precision of IOP values is verified by using a gauge in the anterior chamber (Ofri et al. 2002), although applanantion tonometry is the most commonly applied method due to ease of handling, cost, portability and the fact that it can be used in any species. For these reasons, a tonopen was employed for maned wolves.

In lions, age influenced IOP, with increased levels seen in animals over 20 months of age, up to 40 months of age, there were decreases in IOP values (Ofri et al. 2008). In a study conducted with Labrador dogs with average ages of 45 and 409 days, a difference of 2 mmHg was seen in the IOP, however there was no clinical manifestation of this difference (Mughannam et al. 2004). In the present study, IOP and age presented the same relationship with little or no probability of differences.

Comparing normal IOP values between canines, such as dog, wild dog (*Cerdocyon thous*) and maned wolf, there were differences in the evaluated values, with IOP in maned wolf being higher than that of a young Brazilian fauna wild dog (8.9±1.75mmHg) (Renzo et al. 2014), and the same normal range of domestic dogs (Maggs et al. 2013). The only report described in the literature related to IOP was in seven maned wolves in the Curitiba, Brazil zoo who had levels of 25.6±4.2mmHg in both eyes (Ferreira et al. 1996).

In the maned wolf there were no differences in IOP values in the left and right eyes, contrary to the 2 mmHg difference between eyes reported in the Labrador dog study. The authors of that paper did not provide a logical explanation for the occurrence nor were there clinical manifestations in the eyeball (Mughannam et al. 2004).

Average IOP levels of several species have been reported in the literature, including hedgehog (20.1±4.0mmHg) (Ghaffari et al. 2011), goat (*Capra hicus*) (10.8±1.7mmHg) (Broadwater et al. 2007), Sanjabi sheep (9.60±2.87mmHg LE, 9.15±2.36mmHg RE) (Ghaffari et al. 2011), llama (*Lama glama*) and alpacas (*Lama pacos*) 16.55±3.55mmHg (Nuhsbaum et al. 2000), rhinoceros (32mmHg) (Ofri et al. 2002), capybara (16.47±4.28 mmHg) (Montiani-Ferreira et al. 2008), ferret (14.5±3.2mmHg) (Montiani-Ferreira et al. 2006), zebra (29.47±3.43mmHg), *Arabian oryx* (11.76±3.43mmHg) (Ofri et al. 1998) and broad-snouted caiman (12.9±3.2mmHg) (Oriá et al. 2015).

In the present work, average maned wolf STT-1 levels were lower compared to those seen in dogs, while the IOP was the same in maned wolves as in dogs. It was possible to carry out the study using only mechanical restraint and topical anesthesia however new studies are needed to standardize normal ranges during different seasons and regions of the animals' range.

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