Shiraz University

IJVR

ISSN: 1728-1997 (Print)
ISSN: 2252-0589 (Online)

Vol. 26 No. 4 Ser. No. 93 2025

IRANIAN JOURNAL OF VETERINARY RESEARCH

Original Article

Evaluation of normal ophthalmic parameters and fundoscopic images along with the comparison of two methods of A-mode ocular ultrasonographic biometry and intraocular pressure in healthy working canines

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10.22099/ijvr.2025.49696.7451

(Received 15 Jun 2024; revised version 16 Jul 2025; accepted 03 Aug 2025)

Abstract

Background: Working canines play a very important role in assisting forces by uncovering evidences that might be missed by the human eye. To date, there is no published data on normal ophthalmological parameters for working dogs. **Aims:** This study aims to document normal reference ranges of ophthalmological parameters and blood pressure (BP). Additionally, the study compared the two methods of intraocular pressure (IOP) measurement and A-scan biometry, and assess correlations between IOP to both systolic blood pressure and axial globe length. **Methods:** The study was conducted on 18 dogs of Sashastra Seema Bal (SSB), Alwar (Rajasthan), aged 3.5-6 years, which were divided into three breed-specific groups, i.e., group 1 (Belgian Shepherd), group 2 (German Shepherd) and group 3 (Labrador Retriever) with six animals each. Schirmer's tear test strips were used to perform the Schirmer's tear test (STT). To measure intraocular pressure (IOP), rebound tonometry (Tonovet) and indentation tonometry (Schiotz) were used. A-scan ultrasonography was performed by contact and immersion method. Fundoscopic images were recorded using Fundus-On-Phone equipment. **Results:** STT values revealed non-significant differences among groups, sexes, and eyes. The cumulative mean reflected a higher reading by the rebound tonometer than the indentation tonometer. In all the three groups, a significantly longer anterior chamber depth and axial globe length were observed in the immersion method. Axial globe length was longest in the Labrador breed, followed by German Shepherd and Belgian Shepherd. Systolic and diastolic pressure were significantly different among groups. **Conclusion:** These breed-specific various ophthalmic parameters of working canines will be useful in the clinical diagnosis of various ophthalmic affections.

Key words: A-mode ultrasonography, Fundoscopy, Intraocular pressure, Tonometry

Introduction

The eye is an important organ of the body and many systemic diseases may be diagnosed by their ophthalmic manifestations. For obtaining an accurate diagnosis, apart from a thorough medical history and complete physical examination, ophthalmic examination plays an important role.

The precorneal tear film is vital for normal corneal health. Schirmer's tear test, a quantitative measurement of tear production is an important diagnostic test. It measures the aqueous component of tears and is useful for diagnosing "dry eye" or keratoconjunctivitis sicca (KCS) condition. Normal values of the Schirmer's tear test in dog ranges between 15-25 mm/min (Brooks, 2005).

Intraocular pressure (IOP) is a balance between the aqueous humour production and outflow, and it helps to diagnose low or high-pressure conditions like uveitis and glaucoma, respectively (Bauer and Ambros, 2016). For diagnosis and follow-up of ocular diseases, m easurement of IOP is very important. Increased IOP triggers degeneration of the optic nerve and the malfunction of retinal ganglion cells, which results in permanent blindness in chronic glaucoma (Park *et al*., 2011). In clinical cases, indentation tonometer, applanation tonometer, and rebound tonometer are noninvasive tonometers (Shim *et al*., 2021). TonoVet, a portable rebound tonometer, is based on return-bounce motion after its impact on the cornea. Schiotz tonometer, relies on gravity and must be placed vertically upon the cornea. The depth of indentation is reflected by the movement of the lever on a scale.

IOP is correlated with systemic blood pressure. Hypertension may harm the eye as a target organ damage (TOD) and can relatively increase the risk of visual field deficits (Klein *et al*., 2005).

Ocular ultrasonography is a non-invasive, real-time, rapid diagnostic imaging technique that allows the evaluation of internal structures of the eye when posterior segment of the eye is not visible due to ocular opacities (Ganesan and Chandrasekhara, 2018). In the present scenario, ocular ultrasonography is one of the fundamental pre-operative diagnostic and prognostic tools to predict the visual outcome of animals, based on the structural integrity of intraocular tissues (Williams, 2004). Clinical indications for ocular ultrasonography can be either foreign body investigation, ocular traumatic conditions, lens dislocation, intraocular hemorrhage, retinal detachment, or any opacity making ophthalmoscopic examination difficult (Andrade *et al*., 2021).

Ocular ultrasonographic biometry is very useful in determining the intraocular prosthetic size and calculating the dioptric power of the intraocular lens in cataract surgery (Kobashigawa *et al*., 2015). A-mode is more accurate than B-mode in detecting small lesions of the eye (Audu *et al*., 2017). In A-scan ultrasound, sound waves from a 10 MHz ultrasound transducer are projected through the_eye and then echoes are obtained as tall spikes of high amplitude on the display. Reflection happens at the junction of two different densities, such as the interface between the aqueous humour and the anterior lens capsule. Measurement of axial globe length by A-scan is commonly performed using either applanation (contact) or immersion technique (Trivedi and Wilson, 2011).

Blood pressure is categorized as systolic blood pressure (SBP) and diastolic blood pressure (DBP) and can be measured by indirect and direct methods. In veterinary practice Doppler ultrasonic and oscillometric methods are used as indirect, non-invasive procedures (Wehner *et al*., 2008). Blood pressure has a direct effect on the functioning of organs. Systemic hypertension, i.e., constantly high arterial blood pressure is responsible for certain systemic diseases causing devastating damage to vital organs and even eyes (LeBlanc *et al*., 2011).

Examination of the fundus is significant in visual impairment conditions and for differential diagnosis of systemic infectious diseases, vascular disorders, hypertension, or central nervous system disease. The eye is the only location in body where central nervous system and vascular system can be seen directly. Besides, due to the high blood flow of choroid, the eye is very vulnerable to blood-borne infectious and neoplastic diseases (Wilkie, 2001).

Fundoscopic images can be recorded using Fundus-On-Phone equipment. The canine fundus typically exhibits wide variation. Tapetal fundus that is roughly triangular in shape may appear green, blue, orange, yellow or even absent in color-dilute dogs. The optic disc is normally white in color and its location is generally at the junction of the tapetum and non-tapetum, but may be above or below it. Familiarity with normal variations is of utmost importance for diagnosing conditions of intraocular hemorrhages, retinal detachment, sudden acquired retinal degeneration syndrome (SARDs), or any inherited retinal degeneration (Smith, 2014).

In India, a dearth of documented literature regarding ophthalmic study in working canines is available. Therefore, the study was designed to standardize ocular ultrasonographic biometry, tonometry, STT, etc. in ophthalmologically healthy force dogs of various breeds. Besides, the purpose of the study was also to compare two methods of intraocular pressure measurements and A-scan ocular biometry and to investigate correlations between systolic blood pressure and intraocular pressure, and axial globe length and intraocular pressure.

Materials and Methods

The present study was carried out on 18 force dogs of Dog Training and Breeding Center, SSB, Dera, Alwar (Rajasthan), aged 3.5-6 years and weighing 34.17±0.36 kg. The study was conducted with the limited availability of dog breeds and number of dogs at Sashastra Seema Bal (SSB), Alwar (Rajasthan). Dogs were divided into three breed-specific groups, i.e., group 1 (Belgian Shepherd), group 2 (German Shepherd), and group 3 (Labrador Retriever) with six animals (three male and three female) each.

Schirmer's tear test (STT)

STT was performed with STT strips, the round end of which was bent and positioned in the lower conjunctival sac at the junction of the middle and lateral third of the lower eyelid. The strip was left for 1 min and wetness readings in millimeters were measured (Brooks, 2005).

Intraocular pressure (IOP)

Two methods, rebound tonometry and indentation tonometry were used to measure intraocular pressure in animals of all groups. Measurements of rebound tonometry and indentation tonometry in both eyes were obtained using iCare Tonovet Plus and Schiotz tonometer, respectively. Rebound tonometry was performed first and thereafter topical anaesthetic, 0.5% proparacaine drops were instilled. One minute after instillation of topical anaesthetic, indentation tonometry was performed. The animal was restrained with its head elevated and eyelids carefully retracted without placing pressure on the globe. Schiotz tonometer was placed vertically and centrally on the corneal surface with no weight in place (the plunger weighs 5.5 g). The depth of indentation was reflected by the movement of the lever on a scale. Three readings on the scale were taken and readings were converted to the IOP via calibration table and an average of them was taken.

A-scan ultrasonography

A-scan ultrasonography was performed using Suoer MLabs A-scan Machine using a 10 MHz A-scan probe and immersion (scleral) shell (Fig. 1). Two methods, i.e., contact and immersion method, were used to obtain ocular biometry. The contact method was followed by the immersion method. In the contact method, the animal was positioned in sternal recumbency, and after topical anaesthesia the probe was gently touched directly at the center of the cornea. In the immersion method, the probe was fixed in an immersion shell, and fluid was infused into the shell through tubing fitted with a 5 ml syringe of saline after positioning the animal in dorsal recumbency (Trivedi and Wilson, 2011).

Fig. 1: (**A**) SuoerMlabs A-scan machine, (**B**) 10 MHz A Scan probe, and (**C**) Immersion (Scleral) shell with probe

Blood pressure

Systolic and diastolic blood pressure was recorded using an oscillometric automated device (Dr. Morepen^{TM}, India). Dogs were positioned in lateral recumbency and kept calm and motionless. The cuff was applied above the carpus in the forelimb. Five measurements were taken, and an average of three nearby values of systolic and diastolic blood pressure was recorded.

Fundoscopic examination

Fundoscopic images were recorded using Remidio Fundus-On-Phone equipment for the animals in all groups (Fig. 2). Fundoscopic images were taken in a dark room after pupil dilation with instillation of mydriatic eye drop 1% tropicamide. Fundoscopic images were evaluated for the color of the tapetal and non-tapetal areas, the junction of tapetal and non-tapetal areas, and the location, color, shape, and degree of myelination of the optic disc.

Fig. 2: Fundus on Phone (FOP) used to capture fundus images after pupil dilatation

Statistical analysis

Using SPSS version 20 software, data were analyzed using ANOVA. Two methods of IOP and A-scan were analyzed using a paired t-test. The correlation coefficient was derived using the Pearson method (P≤0.05). The assumptions of the t-test were: the sample size was small, samples were drawn randomly, data were normally distributed, and all groups had approximately equal variance.

Results

STT

There was a non-significant difference among groups, sexes, and eyes. The mean STT values in males and females were 22.56±0.32 and 22.44±0.32, respectively. Mean STT values in groups 1, 2, and 3 are depicted in Table 1.

Intraocular pressure

In all groups, there was a highly significant difference between intraocular pressure measured by two methods. A non-significant difference was observed among groups, sexes, and eyes. The cumulative mean reflected a higher IOP reading by the rebound tonometer than the indentation tonometer. Intraocular pressure with the TonoVet tonometer ranged from 17 to 22 mmHg,

Table 1: Mean±SE values of STT, IOPt, IOPs, systolic pressure, and diastolic pressure in three breeds

Group	STT (mm)	IOP (mm Hg)			BP (mm Hg)	
		IOPt	IOPs	Mean IOP	SP	DP
BSD	23.08±0.39	19.75±0.45^{a}	18.90±0.36^{b}	19.29±0.28	146.11±0.93^{3}	84.78±0.66^{3}
GSD	22.17±0.39	19.58±0.38^{a}	18.72±0.32^{b}	19.15±0.28	132.22±0.93^{2}	79.83±0.66^{2}
Labrador	22.25±0.39	18.92±0.23^{a}	18.28±0.26^{b}	18.60±0.28	128.50±0.93^{1}	67.28±0.66^{1}

Values having different alphabetical superscripts in rows differ significantly (P<0.05) in their respective parameter, and values having different numerical superscripts in columns differ significantly (P<0.05) in their respective parameter. BSD: Belgian Shepherd, GSD: German Shepherd, STT: Schirmer's tear test, IOP: Intraocular pressure, IOPt: IOP by Tonovet, IOPs: IOP by Schiotz, BP: Blood Pressure, SP: Systolic Pressure, and DP: Diastolic pressure with a mean value of 19.39±0.22 mmHg, while pressure measured with the Schiotz tonometer varied in individual dogs from 14.60 to 20.60 mmHg, with a mean value of 18.63±0.23 mmHg. The mean values for intraocular pressure obtained using the two methods were statistically different, but this difference was only 0.76 mmHg, with values higher for the TonoVet tonometer (Table 1). The mean IOP values in males and females were 19.10±0.23 and 18.93±0.22, respectively. The left and right eye had mean IOP values of 19.05±0.22 and 18.97±0.23, respectively.

A-scan ultrasonography

In the contact method, five amplitude peaks of probe and cornea, anterior lens capsule, posterior lens capsule, retina and sclera were seen (Fig. 3). In the immersion method as probe and cornea were no longer in contact, six peaks were displayed. As the corneal spike was separated from the probe spike, it reflected as double peaks that represented the epithelial and endothelial layers of the cornea. In the immersion method, a significantly longer anterior chamber depth (ACD) and axial globe length (AGL) were observed within all the three groups with non-significant differences in lens thickness and vitreous chamber depth by the two methods. The overall mean value of all groups revealed a significant difference between anterior chamber depth by two methods. Differences in_anterior chamber depth and axial globe length by both the methods ranged from 0.08-0.31 mm and 0.07-0.37 mm, respectively. Among the groups, there was a significant difference regarding anterior chamber depth, lens thickness (LT), vitreous chamber depth (VCD) and axial globe length. The mean values for axial globe length in groups 1, 2, and 3 were 21.03±.09, 21.69±.09 and 21.73±.09 mm, respectively; this reveals longer axial globe length in Labrador followed by German Shepherd and Belgian Shepherd (Table 2). The overall mean value for sex revealed a longer anterior chamber depth, lens thickness and axial globe length in males as compared with females. Mean values of ACD, LT, VCD, and AGL in males and females were 4.06±0.04, 7.38±0.03, 10.18±0.04, 21.62±0.08; 3.84±0.03, 7.29±0.02, 10.24±0.04, 21.35±0.07, respectively. No significant difference was observed in all the parameters between the left and right eyes.

Blood pressure

Systolic and diastolic blood pressure were significantly different among groups with higher values in Belgian Shepherd group followed by German Shepherd and Labrador Retriever group. No significant difference was observed between the sexes of all groups (Table 1). Mean values of systolic and diastolic pressure in males and females were 135.70±0.75, 77.81±0.54 and 135.52±0.75, 76.78±0.54, respectively.

Correlation between systolic blood pressure and intraocular pressure (SBP:IOP) and axial globe length and intraocular pressure (AGL:IOP)

A weak, non-significant positive correlation was observed between systolic blood pressure and intraocular pressure (r=+0.15, P= 0.55). Similarly, a weak, non-significant negative correlation was found between axial globe length and intraocular pressure (r=-0.19, P= 0.44). In both cases, the correlations did not reach statistical significance, indicating that the data do not provide sufficient evidence to support a meaningful linear relationship between these variables (Figs. 4a and b).

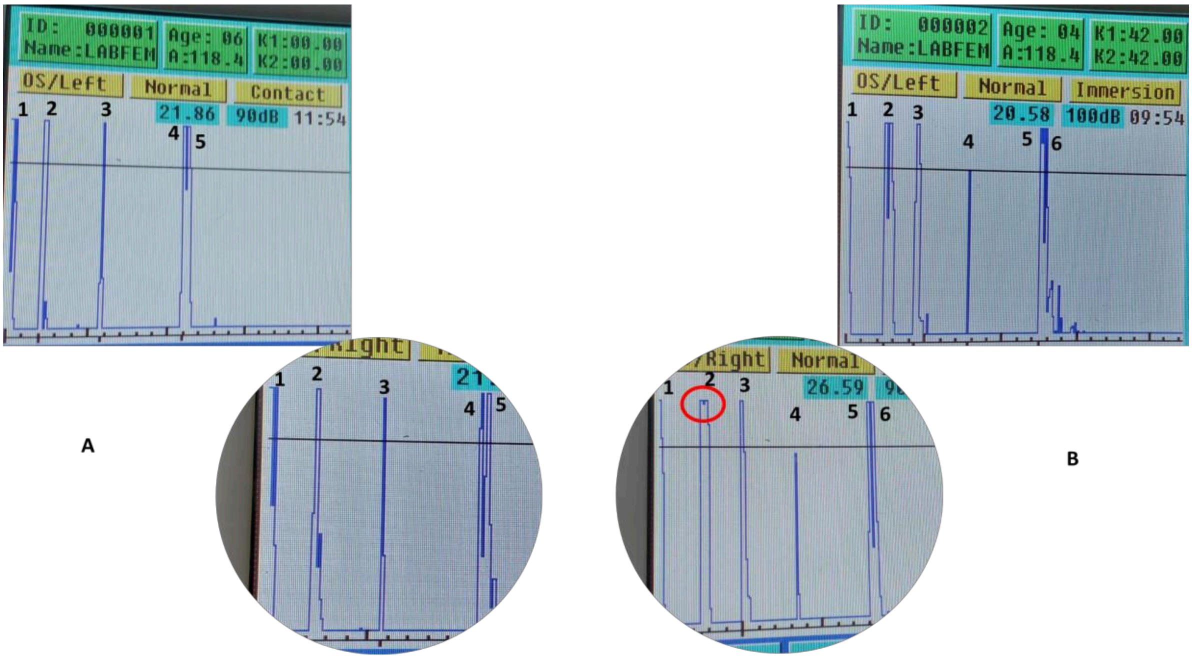

Fig. 3: (**A**) Applanation method: Five tall spikes indicate: (1) Probe and Cornea, (2) Anterior lens Capsule (ALC), (3) Posterior Lens Capsule (PLC), (4) Retina, and (5) Sclera, (**B**) Immersion method: Six tall spikes indicate: (1) Probe, (2) Cornea, (3) ALC, (4) PLC, (5) Retina, and (6) Sclera. In immersion method corneal spike is seen with double peaks representing epithelium and endothelium

Table 2: Mean±SE values of ACD, LT, VCD, and AGL by contact and immersion A-scan method in three breeds

Group	ACD			LT			VCD			AGL		
	ACD_c	ACD_i	Mean	LT_c	LT_i	Mean	VCD_c	VCD_i	Mean	AGL_c	AGL_i	Mean
BSD	3.96 ± 0.07^{a}	$4.05 \pm 0.07^{b*}$	4.01 ± 0.05^{2}	7.25 ± 0.02	7.26 ± 0.02	7.26 ± 0.04^{2}	9.76 ± 0.04	9.77 ± 0.04	9.76 ± 0.05^{1}	20.98 ± 0.07^{a}	$21.07 \pm 0.06^{b*}$	21.03 ± 0.09^{1}
GSD	4.02 ± 0.05^{a}	$4.23 \pm 0.04^{b*}$	4.13 ± 0.04^{2}	7.00 ± 0.04	6.99 ± 0.04	7.00 ± 0.03^{1}	10.57 ± 0.05	10.56 ± 0.05	10.56 ± 0.05^{3}	21.59 ± 0.04^{a}	$21.78 \pm 0.03^{b*}$	21.69 ± 0.10^{2}
Labrador	3.62 ± 0.08^{a}	$3.80 \pm 0.10^{b*}$	3.70 ± 0.05^{1}	7.76 ± 0.08	7.75 ± 0.07	7.75 ± 0.04^{3}	10.31 ± 0.11	10.29 ± 0.10	10.30 ± 0.05^{2}	21.63 ± 0.22^{a}	$21.82 \pm 0.23^{b*}$	21.73 ± 0.09^{2}
Mean	3.87 ± 0.04^{a}	4.03 ± 0.04^{b}	3.95 ± 0.04	7.34 ± 0.02	7.33 ± 0.03	7.34 ± 0.03	10.21 ± 0.04	10.20 ± 0.04	10.21 ± 0.05	21.40 ± 0.07	21.56 ± 0.08	21.48 ± 0.09

Values in mm, values having different alphabetical superscripts in rows differ significantly in their respective parameter ($P<0.05$), values having different numerical superscripts in columns differ significantly ($P<0.05$), and values in row highly significant from the1st value ($P<0.01$). BSD: Belgian Shepherd, GSD: German Shepherd, ACD: Anterior Chamber Depth, LT: Lens thickness, VCD: Vitreous Chamber Depth, AGL: Axial Globe Length, c: Contact method, and i: Immersion method

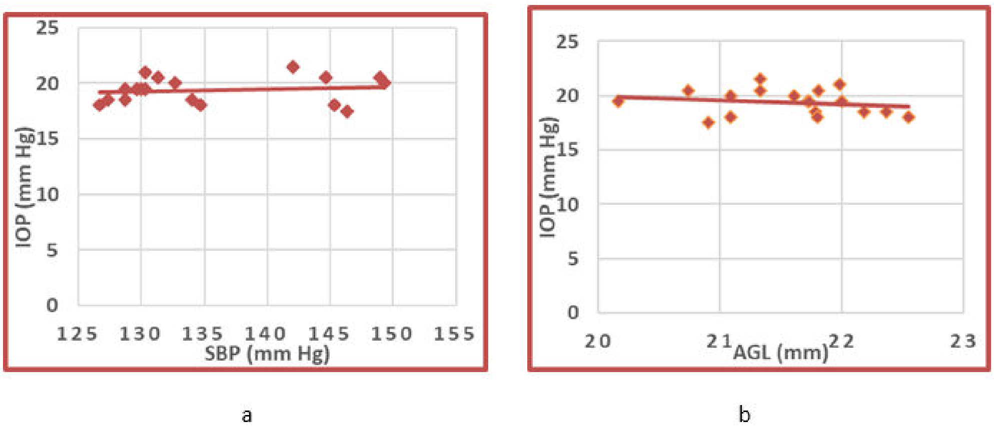

Fig. 4: (**a**) A weak, non-significant positive correlation (r=+0.15, P=0.55) between systolic blood pressure and intraocular pressure, and (**b**) A weak, non-significant negative correlation (r=-0.19, P=0.44) between axial globe length and intraocular pressure

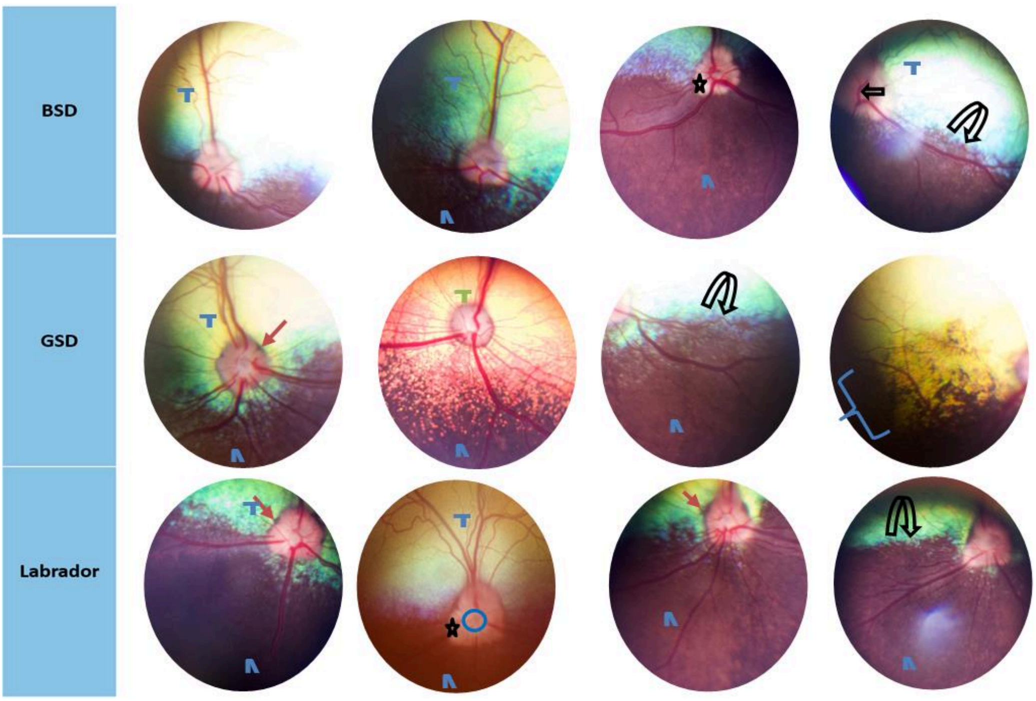

Fig. 5: Fundoscopic images depicting variability in colour of tapetum (T) and non-tapetum (N) area, location, size and shape of optic disc, appearance of tapetal-nontapetal junction. (star shape) Note excessive myelination around optic disc, (arrow) "C" shaped venous anastomosis, (curved arrow) sharp tapetal-nontapetal junction in short-haired dogs, (left brace) Feathered or scalloped tapetal-nontapetal junction in long-haired dogs, (red arrow) Conus, and (blue circle) Optic cup in centre of optic disc

Fundoscopic examination

The ocular fundus showed enormous variations in normal ophthalmoscopic appearance. Holangiotic retinal vascular pattern with 3 or 4 retinal veins that emerged from the optic disc at "C" shaped venous anastomosis and 17-20 thinner, lighter in color and more tortuous retinal arteries that traversed over the entire fundus was noticed. Tapetum lucidum color showed a vast variety with yellow-green-blue (YGB) as the most common color (38.88%, n=7) followed by green-blue (27.77%, n=5), green (16.66%, n=3) and others (16.66%, n=3) (yellow-blue/yellow/orange/- 1 each). The non-tapetal area was noticed as dark brown to black with different shades of red color. Tapetal and non-tapetal junctions were found feathered (scalloped) in long-haired German Shepherd dogs, while in short-haired German Shepherds, Belgian Shepherds, and Labrador dogs it was a sharp border. The shape of optic discs varied from triangular (61.11%, n=11) being the most common to polygonal (22.22%, n=4) and round (16.66%, n=3) shape. A variable degree of myelination around the optic disc gave optic disc pink to white color. Pinkish white (50.00%, n=09) was the most common color found followed by pink (44.44%, n=08) and white (5.55%, n=1). The optic disc was predominantly located at the junction of the tapetal and non-tapetal area (72.22% of eyes), followed by the tapetal area (27.77% of eyes). Choroidal pigment or conus around the optic disc was also visible in fundoscopic images of some dogs (Fig. 5).

Discussion

Schirmer's tear test measures the aqueous portion of the tear film, and is important in the diagnosis of KCS condition. Tear secretion occurs as a reflex action involving sensory nerve endings of the trigeminal nerve and depends on the secretory potential of lacrimal glands (Conrady *et al*., 2016). Mean STT values were non-significantly different among the three breeds and were 23.08±0.39, 22.17±0.39, and 22.25±0.39 in BSD, GSD, and Labrador, respectively. STT results were satisfactory with values reported by Hamor *et al*. (2000) in different breeds. These documented breed-specific STT reference values will help clinically in diagnosing cases of existing keratoconjunctivitis sicca or dry eye. Additionally, regular STT test will help in cases that are progressing towards dry eye. Prompt treatment in such cases will prevent further progression of the condition and will preserve the ocular health.

Early diagnosis of abnormalities in IOP causing glaucoma and uveitis in dogs is important in managing ocular diseases and proper vision (Andrade *et al*., 2012). IOP can be measured by invasive or non-invasive methods. Tonometry, a non-invasive technique can be performed with three types of tonometers based on different principles of indentation, applanation and rebound (Kulualp *et al*., 2018). The present study compared the intraocular pressure measurements in three breeds obtained with two tonometers, Schiotz and TonoVet Plus along with clinical efficacy and user-friendly benefits. The results of intraocular pressure with two types of tonometers in the study were within normal limits and the mean difference was only 0.76 mmHg, but it was statistically significant due to small sample size. The findings of the study were similar to the results quoted by Shim *et al*. (2021) and Wrzesniewska *et al*. (2018) with only 0.69 mmHg lower and 1.29 mmHg higher IOP, respectively by Tonovet Plus. The findings with the Schiotz tonometer differ by values cited by Wrzesniewska *et al*. (2018) with 2.33 mm Hg higher in the present study.

Schiotz works on the principle of indentation of the globe that is affected by the IOP (Estrovich *et al*., 2015). Schiotz tonometer even the oldest instrument, still remains in use with animals (Wrzesniewska *et al*., 2018). It is a simple and inexpensive device that provides reliable measurements. However, due to cumbersome assembly, the need for sterilization, difficulty in positioning animals, and the availability of newer user-friendly devices, its use is decreasing in routine patients. TonoVet Plus, based on the rebound principle, has the advantage of providing more reliable and faster measurements with no requirement for topical anesthesia. Additionally, its small and lightweight probe ensures minimal corneal contact. Small diameter probe head also facilitates selective IOP measurement on healthy cornea sparing damaged areas in focal corneal diseases. It is more sensitive to hypertensive conditions of the eye and assists in early detection and treatment of canine glaucoma in clinical cases (Spiessen *et al*., 2015). Measurement of IOP with rebound tonometer is well tolerated and minimal stress-inducing in dogs. Due to its smaller and sensitive tip, it is helpful in IOP assessment after corneal or intraocular surgery and in small eyes. This hand-held, portable tonometer ensures no user error as only the measurement button is pressed with no touching of the globe by the user (Leiva *et al*., 2006). Thus, rebound tonometry is a comparatively more accurate, newer and easier technique for non-invasively IOP measurement in clinical cases. These documented breed-specific IOP reference values by both tonometers will help clinically in timely diagnosis of increased or decreased intraocular pressure cases. As glaucoma in dogs is a neurodegenerative disease, and can damage all layers of the neuroretina. Thus, prompt diagnosis and treatment will help prevent further ocular damage and vision as well.

A-scan ultrasonography was performed for ocular biometry and achieved by contact method and immersion method. In the contact method, the probe was touched directly at the center of the cornea that might cause corneal compression resulting in a shallow anterior chamber and a shorter axial length. In the immersion method, probe was fixed in an immersion shell, and fluid was infused into the shell through tubing fitted with a 5 ml syringe of saline which served to couple sound waves. In this method, as the probe and cornea were no longer in contact, it eliminated corneal compression and was found superior to contact biometry in terms of accuracy. Anterior chamber depth was significantly higher in the immersion method as compared with the contact method. In both methods, lens thickness and vitreous chamber depth remained unaffected. Thus, axial globe length in both methods was affected by anterior chamber depth only, which was significantly higher in the immersion method. Due to better reproducibility and higher accuracy, the immersion method can be considered as a better method as compared with contact method for axial length measurement. The overall mean value for axial globe length in all groups revealed longer axial globe length in Labrador followed by German Shepherd and Belgian Shepherd. This could be due to the fact that each breed has different skull conformation that affects ocular biometry.

These ocular biometric values are of enormous importance in diagnosing ocular changes, correct selection of ocular prosthesis size, and intraocular lens power in cataract surgery (Sampaio *et al*., 2002). Hamidzada and Osuobeni (1999) found A-mode ultrasonography more accurate than B-mode in measuring intraocular dimensions. Their findings suggested that as compared with A-mode values, B-mode overestimates corneal thickness and anterior chamber depth, while it underestimates lens thickness, vitreous chamber depth and axial length. Consequently, they stated that A-mode ultrasonography is the procedure of choice in ocular biometry while B-mode ultrasonography is used principally for diagnostic purposes. Literature regarding the use of A-scan alone and the comparison of two methods in veterinary ophthalmic practice is very scarce. Though, sample size was small in this study, our study would serve as a reference for future scope of A-scan and its implications in veterinary practice with a large sample size. These documented breed-specific ocular biometric reference values will help clinically in determining the echoic spikes for the biometric evaluation of cataractous lens and selection of dioptric power for cataract surgery. Biometry will further help in diagnosing stage of cataract as mature cataract has higher dimensions compared with immature cataract and diabetic cataract has increased diameter as compared with the other types of cataracts.

Systolic and diastolic blood pressure was significantly different among groups, with higher values in Belgian Shepherds followed by German Shepherds and Labrador Retrievers. These values were in accordance with ACVIM (American College of Veterinary Internal Medicine, 2024) guidelines (https://www.vethdo.com). LeBlanc *et al*. (2011) found that ocular lesions were common in dogs with systemic hypertension and included hyphema, retinal hemorrhage, tortuous vessels, retinal detachment and subretinal edema. They further stated that dogs suffering from hypertension or any diseases related to hypertension should be monitored carefully for the confirmation of ocular damage, and hypertension should also be clinically ruled out in dogs with typical ocular lesions. The ophthalmic findings and blood pressure measurements must be interpreted collectively with clinico-pathologic findings and treatment should be planned accordingly. Similarly, Klein *et al*. (2005) (95% CI: 0.16 to 0.27) found that IOP is directly associated with changes in systemic blood pressure. Alterations in SBP result in changes in aqueous humour production, due to increased capillary pressure in the ciliary body resulting in increased IOP. Besides, blood pressure affects episcleral venous pressure that regulates aqueous flow across trabecular meshwork into Schlemm's canal. In the present study, a weak, non-significant positive correlation was observed between systolic blood pressure and intraocular pressure (r=+0.15, P=0.55). This finding was contrary to the above findings, as the correlation did not reach statistical significance, indicating no linear relationship between these variables. This could be due to fact that the study was conducted on healthy dogs, representing values within the normal range. Boillot *et al*. (2014) found an inverse correlation between IOP and AGL (high IOP and low AGL) and suggested that smaller AGL may render dogs more vulnerable to the development of a high IOP. Contrary to this, in the present study, a weak, non-significant negative correlation was found between axial globe length and intraocular pressure (r=-0.19, P=0.44). Non-significant correlation represents no direct relationship between these variables. Therefore, no definitive conclusion can be drawn regarding their association in this study population.

The fundoscopic examination is a very crucial part of the ophthalmic examination as many diseases may be diagnosed or even ruled out by observing any abnormality in the fundus. Canine fundus exhibits a huge variability in its normal appearance, even each individual of a single breed shows a wide variety. Expertise in these variations is of utmost importance to differentiate normal from abnormal to arrive at a final diagnosis. None of the dogs of all three breeds showed any abnormality during fundus examination and findings were nearly similar to the normal pattern illustrated for dogs. There was a quite normal variation with regard to the color of the tapetum, location, size, and shape of the optic disc, retinal vasculature, and appearance of the tapetal and non-tapetal junction. The most common color of tapetum lucidum was yellow-green-blue followed by green-blue, green and others (yellow-blue/yellow/orange). The findings of tapetal color were different from colors studied by Sini *et al*. (2017) who found green-blue color as the most followed by yellow-green-blue, green, and combinations of colors. The location of the optic disc was approximately similar to the findings of Sini *et al*. (2017) who found optic disc mostly at the junction of the tapetal to non-tapetal fundus (50.00%, n=35) followed by tapetal area (18.57%, n=13) and in the non-tapetal fundus in various breeds of dogs. Variation in non-tapetal area coloring depends on the degree of pigmentation in the choroid and also in retinal pigment epithelium (RPE) (Sini *et al*., 2017). Proficiency in these documented normal variations will certainly clinically helpful in judging any abnormality of hyper-reflective or hypo-reflective conditions, retinal degeneration, retinal detachment, Sudden Acquired Retinal Degeneration

Syndrome (SARDS), coloboma, etc. in dogs. Knowledge of reference values for each breed helps greatly in the diagnosis of ophthalmic diseases. The findings of the present study may remedy data deficit with regard to normal ophthalmic parameters and ocular biometry indices in these breeds of working canines, and may prove useful in diagnosing ophthalmologic abnormalities.

The study was conducted on 18 force dogs to document normal reference ranges of various ophthalmological parameters and blood pressure. Additionally, the two methods of IOP measurement and A-scan biometry were compared. During the study, breed-specific various ophthalmic parameters were documented. TonoVet tonometer provided higher values than Schiotz tonometer but within the normal range. The immersion method exhibited better reproducibility and higher accuracy than the contact method. A weak positive correlation between SBP and IOP and a weak negative correlation between AGL and IOP was found. The study was limited to three working dog breeds (Belgian Shepherd, German Shepherd, and Labrador Retriever). Future research can be expanded to include a wider range of dog breeds with a large sample size to document more comprehensive various normal ophthalmological parameters. These documented breed-specific reference values will be useful in the timely clinical diagnosis of various ophthalmic affections in working canines. Prompt diagnosis and their treatment will help in preventing further ocular damage and vision as well.

Acknowledgements

We are thankful to DUVASU University, Mathura for the availability of instruments necessary to execute the study. We are also grateful to the Veterinary staff of SSB, Rajasthan for providing assistance in performing the ophthalmic examination of dogs.

Conflict of interest

The authors declare that they do not have any relevant financial or non-financial interests.

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