Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
DOI: 10.29261/pakvetj/2019.002

RESEARCH ARTICLE

Ultrasound Biomicroscopy Studies to Evaluate Ciliary Cleft Parameters in Healthy Eyes of American Cocker Spaniels

Kwang-Hun Choi[1], Seoung-Woo Choo[2], Soon-Wuk Jeong[1] and Joon-Young Kim[2*]

[1]Department of Veterinary Surgery, College of Veterinary Medicine, Konkuk University, 120 Neungdong-ro, Gwangjin-gu, Seoul, Korea, 05029; [2]Veterinary Medical Teaching Hospital, Konkuk University, 120 Neungdong-ro, Gwangjin-gu, Seoul, Korea, 05029
*Corresponding author: canvet@hanafos.com

ARTICLE HISTORY (17-194)

Received: June 05, 2017
Revised: October 26, 2018
Accepted: October 27, 2018
Published online: January 24, 2019

Key words:
American cocker spaniel
Ciliary cleft area
Intraocular pressure
Iridocorneal angle
PACG
Ultrasound biomicroscopy

A B S T R A C T

The purpose of the present study was to quantify ciliary cleft parameters in the healthy eyes of American Cocker Spaniels (ACSs) and assess their correlations with intraocular pressure (IOP) using ultrasound biomicroscopy (UBM) studies. We evaluated 26 healthy eyes from 13 ACSs. Iridocorneal angle (ICA), ciliary cleft area (CCA), ciliary cleft width (CCW), and angle-opening distance (AOD) were measured. All UBM studies were performed without pupil dilatation in both eyes of each dog. The IOPs of all eyes were measured using applanation tonometry. The mean IOP, ICA, CCA, CCW, and AOD values were 15.92±3.21 mmHg, 13.94±1.96°, 0.32±0.03 mm^2, 0.33±0.06 mm, and 0.45±0.10 mm, respectively, for the entire study sample; 16.63±3.81 mmHg, 13.51±1.90°, 0.32±0.02 mm^2, 0.32±0.06 mm, and 0.40±0.07 mm, respectively, in female dogs; and 14.80±1.48 mmHg, 14.63±1.96°, 0.35±0.07 mm^2, 0.32±0.03 mm, and 0.53±0.09 mm, respectively, in male dogs. IOP had significant negative correlations with CCA and AOD in all eyes. There were no significant differences in IOP between male and female dogs, although there were significant differences in AOD. Our results suggest that IOP is correlated with CCA and AOD in ACSs.

To Cite This Article: Choi KH, Choo SW, Jeong SW and Kim JY, 2019. Ultrasound biomicroscopy studies to evaluate ciliary cleft parameters in healthy eyes of American cocker spaniels. Pak Vet J, 39(1): 66-70. http://dx.doi.org/10.29261/pakvetj/2019.002

INTRODUCTION

The ciliary cleft is a virtual space containing the uveal trabecular meshwork. It is defined anteriorly by the pectinate ligament, externally by the sclera, posteriorly by muscle and the ciliary body, and internally by the iris root and pars plicata of the ciliary body (Pizzirani and Gong, 2015). Ciliary cleft parameters correlate with its anatomical structure and include the iridocorneal angle (ICA), angle-opening distance (AOD), ciliary cleft width (CCW), and ciliary cleft area (CCA).

Abnormalities in the ICA are considered to be risk factors for canine primary angle-closure glaucoma (PACG). Gonioscopy is a classical method for evaluation of the ICA width and anatomy and the degree of pectinate ligament dysplasia (Kato *et al.*, 2006) However, gonioscopic examination has limitations (Renwick, 2014). First, it cannot be used to quantify abnormal structures in the ciliary cleft region (Hasegawa and Kawata, 2015). Second, gonioscopic analysis of the angle is not always accurate, and ICA may occasionally appear narrow or closed if the iris root is close to the corneosclera (Gibson *et al.*, 1998). Third, grading systems based on gonioscopy have failed to accurately predict glaucoma prior to onset (Gibson *et al.*, 1998). Fourth, an opaque or edematous cornea prevents observation of the anterior part of the ciliary cleft by gonioscopy (Gibson *et al.*, 1998).

Noninvasive, microscopic visualization of structures between living tissues *in vivo* is a goal of many imaging techniques (Bentley *et al.*, 2003). Recently, high-frequency ultrasound probes with frequencies ranging from 20 MHz (high-resolution ultrasound) to 60 MHz (ultrasound biomicroscopy [UBM]) have been developed. These probes allow imaging at resolutions of approximately 50 to 80 µm (Bentley *et al.*, 2003). Tissue penetration with high-frequency probes is limited to 5 to 10 mm (Bentley *et al.*, 2003). This depth is adequate for assessments of anterior segment structures. UBM provides images of anterior chamber structures with a resolution higher that that provided by high-resolution ultrasound

(HRU) (Park *et al*., 2015; Gao *et al*., 2018). Furthermore, it facilitates the quantitative analysis of micromeasurements. Although the penetration depth of UBM probes is currently limited to 4 to 5 mm, the image resolution obtained allows the approximate discrimination of structures about 50 µm in size (Pavlin, 1995; Gibson *et al.*, 1998; Shi *et al*., 2018). UBM may be used to produce information regarding pathogenesis and prognosis and can be performed on opaque or edematous corneas (Gibson *et al.*, 1998; Martin, 2018). UBM thus compensates for the limitations of gonioscopy (Gibson *et al.*, 1998; Hasegawa and Kawata, 2015) and is expected to be clinically useful for the elucidation of unknown pathophysiologies, selection of accurate treatment approaches, and improvement of prognosis in patients with ocular diseases (Hasegawa and Kawata, 2015; Yan *et al*., 2018).

American cocker spaniels (ACSs) are prone to PACG (Gelatt and MacKay, 2004), and increased IOP is an important indicator of glaucoma in dogs (Pizzirani and Gong, 2015). To the best of our knowledge, no studies have quantified ICA, AOD, CCW, and CCA in the healthy eyes of ACSs and correlated them with IOP. The purpose of the present study was to quantify these parameters in healthy eyes from ACSs and to assess their correlations with IOP in UBM studies. The findings will also provide reference ranges for ciliary cleft parameters for healthy eyes of ACSs.

MATERIALS AND METHODS

We analyzed UBM data obtained from ACSs without eye abnormalities that visited two animal hospitals (Woosung Animal Hospital and the Veterinary Medical Teaching Hospital of Konkuk University) to undergo medical examinations between July 2013 and January 2014. In total, 26 healthy eyes from ACSs (eight female and five male dogs) were prospectively reviewed. To exclude differences in raw UBM values caused by the inclusion of different breeds with varied ocular sizes, we selected ACSs with healthy eyes for the present study. Cases that were not included in initial records were excluded. We obtained the sexes of the dogs from the initial records. All animals underwent routine ophthalmological (menace response, dazzle reflex, pupillary light reflex, Schimer tear test, IOP, direct ophthalmoscopy examination, and slit lamp biomicroscopy [Hawk Eye, Dioptrix, France] examinations). No abnormalities were noted in these examinations. In addition, IOP was measured using applanation tonometry (TONO-Pen VET™, Reichert Technologies, USA) and recorded.

For measurements of ICA, AOD, CCW, and CCA, all animals were manually restrained in a standing position and subjected to UBM (MD-320WD1101; MEDA Co., Ltd; China) in a room with standard lighting conditions. The eyelids were manually held open while avoiding pressure on the globe. Measurements of ICA, AOD, CCW, and CCA were obtained without pupil dilation using a UBM (MD-320WD1101; MEDA Co., Ltd; China). We used a 50 MHz UBM probe held perpendicular to the limbus at the 12 o'clock position and section planes were obtained along the vertical meridian. The following four parameters were estimated using UBM measurement software.

A. ICA measurement: The ICA represents the peripheral circumference of the anterior chamber, where the cornea, sclera, and base of the iris converge (Pizzirani and Gong, 2015) (Fig. 1).

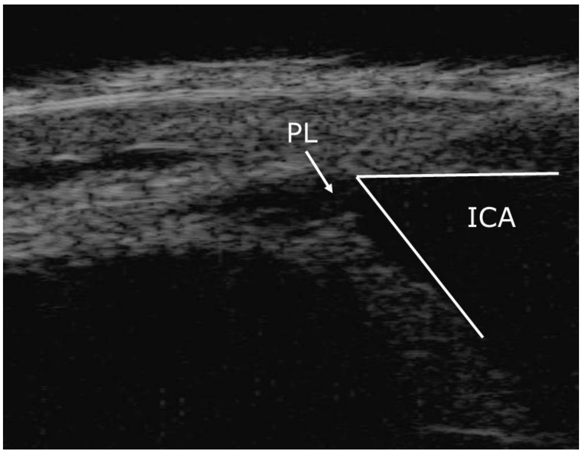

Fig. 1: Measurement of the iridocorneal angle (ICA) using ultrasound biomicroscopy in the healthy eye of an American cocker spaniel. The ICA represents the peripheral circumference of the anterior chamber, where the cornea, sclera, and base of the iris converge. PL: pectinate ligament.

B. AOD measurement: The AOD was calculated as the perpendicular distance measured from the end of Descemet's membrane to the anterior iris surface (Tsai *et al.*, 2012) (Fig. 2).

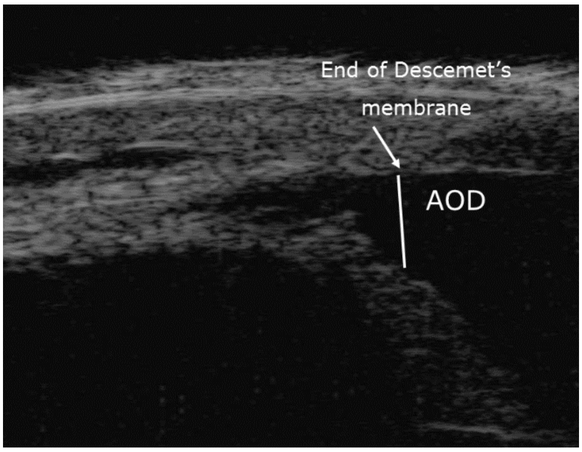

Fig. 2: Measurement of the angle-opening distance (AOD) using ultrasound biomicroscopy in the healthy eye of an American cocker spaniel. The AOD was calculated as the perpendicular distance measured from the end of Descemet's membrane to the anterior iris surface.

C. CCW measurement: CCW was measured from the superior surface of the root of the iris to the inner surface of the sclera on a perpendicular line (Hasegawa and Kawata, 2015) (Fig. 3).

D. CCA measurement: CCA was measured as the area surrounded by CCW, the line tracing the inner scleral side of the ciliary cleft from the inner surface of the sclera to the angle recess, and the line tracing the superior side of the root of the iris from the angle recess to the superior surface of the root of the iris (Hasegawa and Kawata, 2015) (Fig. 3).

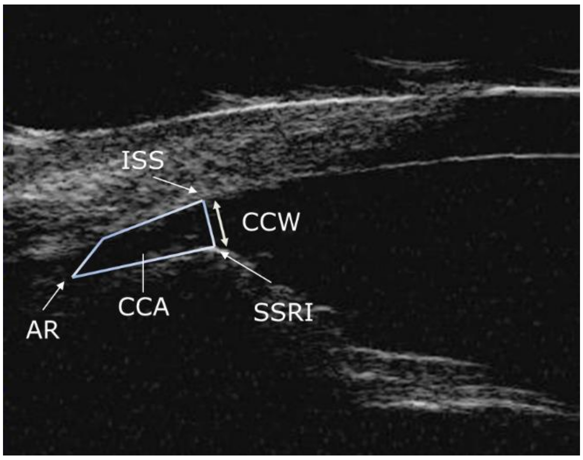

Fig. 3: Measurements of the ciliary cleft width (CCW) and ciliary cleft area (CCA) using ultrasound biomicroscopy in the healthy eye of an American cocker spaniel. CCW was measured from the superior surface of the root of the iris to the inner surface of the sclera on a perpendicular line. CCA was measured as the area surrounded by CCW, the line along the inner scleral side of the ciliary cleft from the inner surface of the sclera to the angle recess, and the line along the superior side of the root of the iris from the angle recess to the superior surface of the root of the iris. ISS: inner surface of the sclera, AR: angle recess, SSRI: superior surface of the root of the iris.

Spearman's correlation coefficients were determined to assess the correlations between IOP and ICA, AOD, CCW and CCA. The Mann-Whitney U test was used to assess differences in values between males and female dogs. All statistical analyses were performed using SPSS ver.22.0 software for Windows (SPSS, Inc.; Chicago, IL, USA). P-values <0.05 were considered statistically significant.

RESULTS

The mean age of the 13 dogs was 7.09 years (SD, 3.67; range, 0.4-14 years). Four dogs were castrated males, one was an intact male, seven were spayed females, and one was an intact female. The mean body weight was 12.28 kg (SD, 1.32; range, 10-14.5 kg).

Tables 1, 2, and 3 show IOP and UBM measurements for all dogs, female dogs, and male dogs, respectively. IOP had no significant correlations with ICA or CCW (P>0.05, Figs. 4A and 4B). However, it showed significant negative correlations with CCA and AOD (P<0.001, Figs. 4C and 4D). There were significant differences in AOD values between male and female dogs (P<0.01). None of the other parameters had sex-related differences (P>0.05).

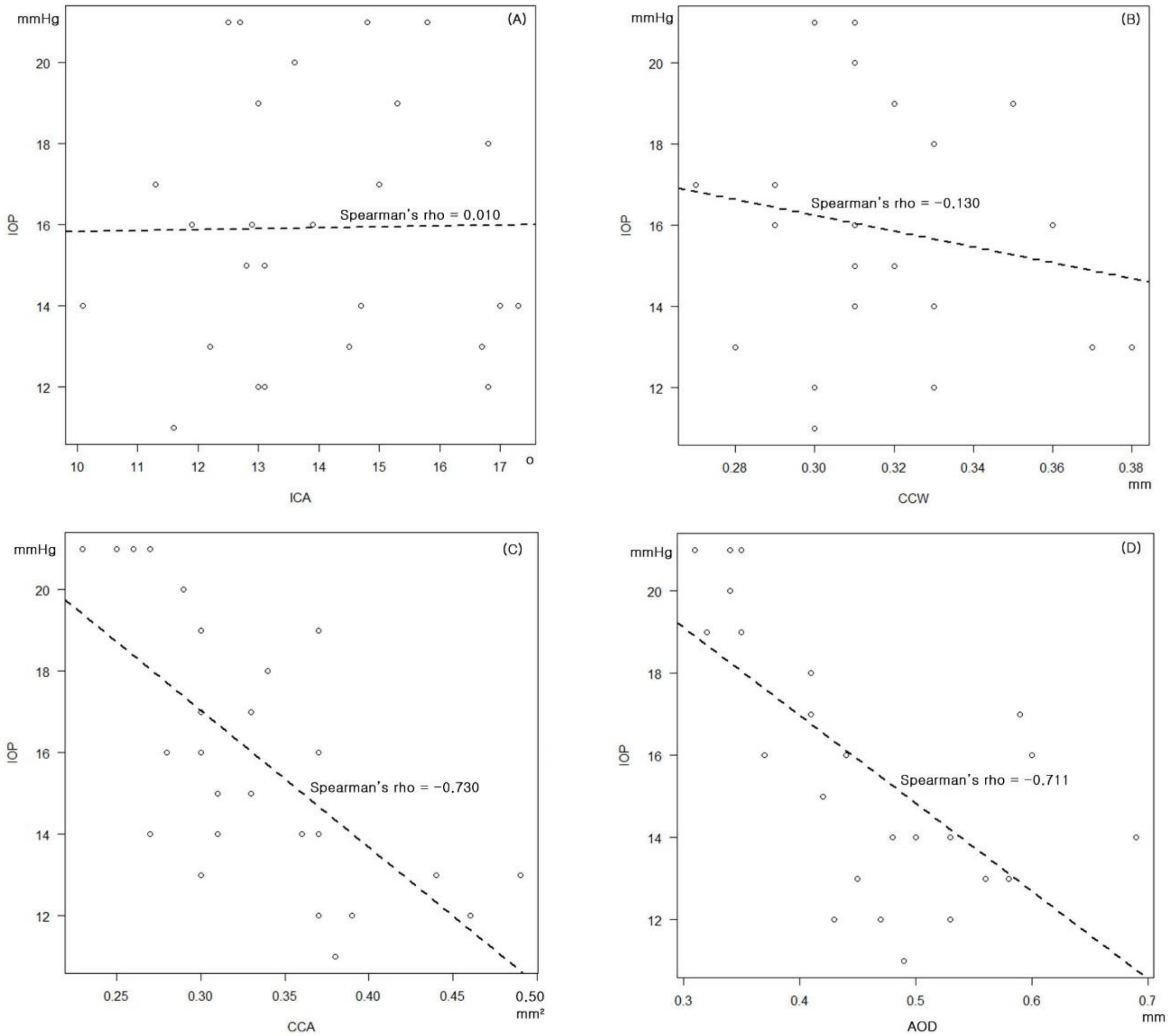

Fig. 4: Statistical correlations between intraocular pressure (IOP) values and ciliary cleft parameters measured using ultrasound biomicroscopy in healthy eyes of American cocker spaniels. (A) There is no significant correlation between IOP and the iridocorneal angle (ICA, P>0.05). (B) There is no significant correlation between IOP and the ciliary cleft width (CCW, P>0.05). (C) There is a significant negative correlation between IOP and the ciliary cleft area (CCA, P<0.001). (D) There is a significant negative correlation between IOP and the angle-opening distance (AOD, P<0.001).

Table 1: IOP, CCA, CCW, and AOD values for healthy eyes from 13 American cocker spaniels (26 eyes)

Measurements	Mean	SD	Range
IOP (mmHg)	15.92	3.22	11-21
CCA (mm^2)	0.34	0.07	0.23-0.46
CCW (mm)	0.32	0.03	0.27-0.38
AOD (mm)	0.45	0.10	0.31-0.69
ICA (°)	13.99	1.99	10.10-17.30

IOP: intraocular pressure, CCA: ciliary cleft area, CCW: ciliary cleft width, AOD: angle-opening distance, ICA: iridocorneal angle. IOP was measured using TONO-Pen VET™ and CCA, CCW, and AOD were measured using ultrasound biomicroscopy.

Table 2: IOP, CCA, CCW, and AOD values for healthy eyes from eight female American cocker spaniels (16 eyes)

Measurements	Mean	SD	Range
IOP (mmHg)	16.63	3.81	11-21
CCA (mm^2)	0.32	0.06	0.25-0.46
CCW (mm)	0.32	0.03	0.27-0.38
AOD (mm)	0.40	0.07	0.31-0.53
ICA (°)	13.51	1.90	10.10-16.80

IOP: intraocular pressure, CCA: ciliary cleft area, CCW: ciliary cleft width, AOD: angle-opening distance, ICA: iridocorneal angle. IOP was measured using TONO-Pen VET™ and CCA, CCW, and AOD were measured using ultrasound biomicroscopy.

Table 3: IOP, CCA, CCW, and AOD values for healthy eyes from five male American cocker spaniels (10 eyes)

Measurements	Mean	SD	Range
IOP (mmHg)	14.80	1.48	13-17
CCA (mm^2)	0.35	0.07	0.27-0.49
CCW (mm)	0.32	0.03	0.27-0.38
AOD (mm)	0.53	0.09	0.41-0.69
ICA (°)	14.63	1.96	11.30-17.30

IOP: intraocular pressure, CCA: ciliary cleft area, CCW: ciliary cleft width, AOD: angle-opening distance, ICA: iridocorneal angle. IOP was measured using TONO-Pen VET™ and CCA, CCW, and AOD were measured using ultrasound biomicroscopy.

DISCUSSION

In the present study, we quantified ciliary cleft parameters for healthy eyes of ACSs and assessed their correlations with IOP using UBM. In dogs, an elevated IOP is a principal risk factor for glaucoma (Gelatt *et al.*, 2013; Pizzirani, 2015). The ciliary cleft, which contains the trabecular meshwork, is associated with outflow of the aqueous humor, and the aqueous humor contributes to the maintenance of IOP (Pizzirani and Gong, 2015). Gonioscopy is generally used to evaluate ICA, which is a ciliary cleft parameter. However, it cannot be used to identify abnormalities in the deep ciliary cleft region and can only evaluate ICA (Gibson *et al.*, 1998). Gibson compared gonioscopy and UBM measurements for the estimation of ICA in dogs and found no significant differences between the two modalities (Gibson *et al.*, 1998). However, they found that gonioscopy cannot be used to measure ICA in the presence of corneal edema or increased corneal opacity, while UBM findings are not affected by these abnormalities (Gibson *et al.*, 1998; Ishikawa 2007). Therefore, UBM is considered a more useful tool than gonioscopy (Gibson *et al.*, 1998; Rose *et al.*, 2008; Hasegawa and Kawata, 2013).

The other ciliary cleft parameters, namely AOD, CCW, and CCA, are associated with its anatomic structure. In other words, if the ciliary cleft collapses, then these parameters also change (Sihota *et al.*, 2005; Hasegawa and Kawata, 2015; Park *et al.*, 2018). Inadequate assessment of structural changes within the ciliary cleft may prevent the early diagnosis of canine primary glaucoma (Hasegawa and Kawata, 2015). Therefore, evaluation of ciliary cleft parameters in healthy eyes of dogs using UBM is important for the early diagnosis of primary glaucoma (Hasegawa and Kawata, 2015).

In the present study, we found no significant correlation between IOP and ICA. The canine trabecular meshwork is located within the recess of the ciliary cleft, an anatomy different location than its location in the human eye. Therefore, IOP may not be influenced by ICA narrowing (Kwak *et al.*, 2016) in dogs. Nevertheless, even in Rüfer's study, which included 390 healthy white human volunteers (242 men and 148 women), no correlation was found between IOP and ICA (Rüfer, *et al.*, 2010).

We found no significant correlation between CCW and IOP. Dulaurent *et al.* (2012) used UBM to evaluate changes in the anterior segment after the instillation of a mydriatic agent in normotensive dogs and found that the central CCW did not change with a decrease in IOP. CCW may be independent of the geometric angle formed by the iris and cornea and does not affect the outflow of the aqueous humor, as the trabecular meshwork is located within the recess of the ciliary cleft in dogs (Dulaurent *et al.*, 2012).

In a previous study, CCA was significantly smaller in the study group with high IOP than in the group with normal IOP (Boillot *et al.*, 2014; Hasegawa and Kawata, 2015). The ciliary cleft contains the uveal trabecular meshwork, which is a spongiform cobweb-like tissue defined by irregular trabecular beams (Pizzirani and Gong, 2015). The empty spaces between the beams are called Fontana's spaces (Pizzirani and Gong, 2015). This structure plays an important role in regulating outflow and IOP (Pizzirani and Gong, 2015). The CCA includes this structure. Therefore, it was considered that IOP is influenced by CCA, including that for normal Fontana's spaces. Even in the present study, there was a significant negative correlation between CCA and IOP.

We also found a significant negative correlation between IOP and AOD in the present study. This finding is consistent with that in a previous human study, which demonstrated that the mean IOP decreased with an increase in the mean AOD and that there was a significant correlation between these measures (Huang *et al.*, 2012). The opening of the ciliary cleft is an important structure for the prediction of responsiveness to medical therapies. This opening was found to be larger in dogs that responded to medical therapy than in dogs that were unresponsive (Hasegawa and Kawata, 2015).

Our study had several limitations. These included a small sample size, limitation of UBM examination sites on the eye, the use of a two-dimensional measurement tool, and the animals' cooperation. However, we believe that the major limitation was that we were unable to measure the ciliary cleft over the entire 360°. The aqueous humor flows in a three-dimensional space. This is why it is necessary to measure the entire 360° of the ciliary cleft. However, there is currently no instrument that provides three-dimensional measurements of the flow of the aqueous humor. UBM is a two-dimensional measurement tool that is used to measure all 360° of the ciliary cleft to overcome this limitation. However, this procedure cannot be performed without anesthesia, and most pet owners are against the use of anesthesia. Further studies should include three-dimensional measurements of aqueous humor flow. In conclusion, the results of the present study suggest that IOP correlates with CCA and AOD in healthy eyes of ACSs and can be used as a risk indicator for PACG in these animals.

Author contribution: KHC carried out the survey and drafted the manuscript. SWC helped with patient management. SWJ participated in the design of the study and reviewed the manuscript. JYK designed and carried out the survey and drafted the manuscript. All authors read and approved the final manuscript.

REFERENCES

Bentley E, Miller PE and Diehl KA, 2003. Use of high-resolution ultrasound as a diagnostic tool in veterinary ophthalmology. J Am Vet Med Assoc 223:1617-22.

Boillot T, Rosolen SG, Dulaurent T, *et al*., 2014. Determination of morphological, biometric and biochemical susceptibilities in healthy Eurasier dogs with suspected inherited glaucoma. PLoS One 9: e111873.

Dulaurent T, Goulle F, Dulaurent A, *et al*., 2012. Effect of mydriasis induced by topical instillations of 0.5% tropicamide on the anterior segment in normotensive dogs using ultrasound biomicroscopy. Vet Ophthal 15 Suppl 1:8-13.

Gao K, Li F, Li Y, *et al*., 2018. Anterior choroidal thickness increased in primary open-angle glaucoma and primary angle-closure disease eyes evidenced by ultrasound biomicroscopy and SS-OCT. Invest Ophthalmol Vis Sci 59:1270-7.

Gelatt KN, Brooks DE and Kallberg ME, 2013. The canine glaucomas. In: Essentials of Veterinary Ophthalmology. (Gelatt KN, ed). John Wiley & Sons: USA, pp:155-87.

Gelatt KN and MacKay EO, 2004. Prevalence of the breed-related glaucomas in pure-bred dogs in North America. Vet Ophthal 7:97-111.

Gibson T, Roberts S, Severin G, *et al*., 1998. Comparison of gonioscopy and ultrasound biomicroscopy for evaluating the iridocorneal angle in dogs. J Am Vet Med Assoc 213:635-8.

Hasegawa T and Kawata M, 2013. Evaluation of the distance between Schwalbe's line and the anterior lens capsule as a parameter for the correction of ultrasound biomicroscopic val ues of the canine iridocorneal angle. Vet Ophthal 16:169-74

Hasegawa T and Kawata M, 2015. Ultrasound biomicroscopic findings of the iridocorneal angle in live healthy and glaucomatous dogs. J Vet Med Sci 77:1625.

Huang G, Gonzalez E, Lee R, *et al*., 2012. Association of biometric factors with anterior chamber angle widening and intraocular pressure reduction after uneventful phacoemulsification for cataract. J Cataract Refract Surg 38:108-16.

Ishikawa H, 2007. Anterior segment imaging for glaucoma: OCT or UBM? Br J Ophthalmol 91: 1420-1

Kato K, Sasaki N, Matsunaga S, *et al*., 2006. Incidence of canine glaucoma with goniodysplasia in Japan: a retrospective study. J Vet Med Sci 68:853-8.

Kwak J, Kang S, Lee ER, *et al*., 2017. Effect of preservative-free tafluprost on intraocular pressure, pupil diameter, and anterior segment structures in normal canine eyes. Vet Ophthal 20:34-9.

Martin R, 2018. Cornea and anterior eye assessment with slit lamp biomicroscopy, specular microscopy, confocal microscopy, and ultrasound biomicroscopy. Indian J Ophthalmol 66:195-201.

Park SW, Kang SM, Lim JG, *et al*., 2018. Effects of prostaglandin-mediated and cholinergicmediated miosis on morphology of the ciliary cleft region in dogs. Am J Vet Res 79:980-5.

Pavlin C, 1995. Practical application of ultrasound biomicroscopy. Can J Ophthalmol 30:225-9.

Pizzirani S and Gong H, 2015. Functional anatomy of the outflow facilities. Vet Clin North Am Small Anim Pract 45:1101-26.

Pizzirani S, 2015. Difinition, classification, and pathophysiology of canine glaucoma. Vet Clin North Am Small Anim Pract 45:1127-57.

Renwick P, 2014. Glaucoma. In: BSAVA Manual of canine and feline ophthalmology. 3rd Ed, (Gould D and McLellan GJ, ed). British Small Animal Veterinary Association: gloucester, The United Kingdom pp:274-82.

Rose MD, Mattoon JS, Gemensky-Metzler AJ *et al*., 2008. Ultrasound biomicroscopy of the iridocorneal angle of the eye before and after phacoemulsification and intraocular lens implantation in dogs. Am J Vet Res 69:279-88.

Rüfer F, Schröder A, Klettner A, *et al*., 2010. Anterior chamber depth and iridocorneal angle in healthy White subjects: effects of age, gender and refraction. Acta Ophthalmol 88:885-90.

Shi M, Han X, Zhang J, *et al*., 2018. Comparison of 25 MHz and 50 MHz ultrasound biomicroscopy for imaging of the lens and its related diseases. Int J Ophthalmol 11:1152-7.

Sihota R, Dada T, Gupta R, *et al*., 2005. Ultrasound biomicroscopy in the subtypes of primary angle closure glaucoma. J Glaucoma 14:387-91.

Tsai S, Bentley E, Miller PE, *et al*., 2012. Gender differences in iridocorneal angle morphology: a potential explanation for the female predisposition to primary angle closure glaucoma in dogs. Vet Ophthal 15:60-3.

Yan S, Tang G, Zhang H, *et al*., 2018. Ultrasound biomicroscopy characteristics of late capsular block syndrome before and after treatment. JCRS Online Case Reports https://doi.org/10.1016/j.jcro.2018.08.002.