

## Rectification of Width and Area of the Ciliary Cleft in Dogs

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**ABSTRACT.** The width and area of the ciliary cleft (CCW and CCA) according to ultrasound biomicroscopy was rectified to compare dogs of different body sizes/weights, including toy breed (TBG), small breed (SBG), medium breed (MBG) and large/giant breed groups (LGBG). A linear correlation was detected between the natural log of body weight and the ocular axial length (OAL) reflecting the ocular size (OS;  $r=0.81$ ,  $p<0.001$ ). A significant positive correlation was found between OAL and the distance of the Schwalbe's line to the anterior lenticular capsule (SLD;  $r=0.87$ ,  $p<0.001$ ), suggesting that SLD could rectify canine OS. Rectified CCW (r-CCW) and CCA (r-CCA) were calculated using SLD and fixed SLD as the SLD mean (SLDM). The SLDM was 2.55 mm in this study, and r-CCW and r-CCA were calculated as  $r\text{-CCW} = \text{CCW} \times (2.55/\text{SLD})$  and  $r\text{-CCA} = \text{CCA} \times (2.55/\text{SLD})^2$ . The CCW values of TBG, SBG and MBG were narrower than those of LGBG ( $p<0.05$ ). There were significant statistical differences in CCA between all groups other than TBG and SBG ( $p<0.05$ ). There were no significant differences in r-CCW and r-CCA in any of the groups. In addition, CCW/SLD did not show significant changes in dogs with different body sizes/weights. These results suggested that r-CCW, r-CCA and CCW/SLD can be used for comparison between dogs of different body sizes/weights.

**KEY WORDS:** area and width, canine, ciliary cleft, rectification, ultrasound biomicroscopy.

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Since structural abnormalities of the iridocorneal angle (ICA) can result in glaucoma including persistent mesodermal bands/pectinate ligament dysplasia and a narrow/closed angle [2, 3], gonioscopy is one of the essential examinations in the diagnosis and monitoring of glaucomatous eyes [2, 3, 5, 8]. However, gonioscopy is a subjective test, and interpretation errors occur among examining ophthalmologists [10]. In addition, gonioscopic examination only provides information for the anterior aspect of the ICA [12]. Hence, the entire structures of the ICA cannot be assessed by examination using gonioscopy.

Ultrasound biomicroscopy (UBM) has been recently introduced in veterinary ophthalmology as a diagnostic tool for detailed and quantitative evaluation of the anterior segment structures, including the entire structures of the ICA in animals [1, 4, 7, 13]. UBM is also used for assessment of abnormalities in ocular structures; many abnormalities can be detected in glaucomatous eyes, such as closure of the ciliary cleft (CC) and significant decrease of the width and area of the CC (CCW and CCA). In general, it would be difficult to compare the results of CCW and CCA measured by UBM between dogs because the measured values would vary by the canine ocular size (OS) due to differences in body size/weight [6]. Hence, CCW and CCA obtained from an individual UBM image should be rectified for comparison between animals with different body sizes/weights. In this study, rectification of CCW and CCA as measured by an

individual UBM image was assessed between dogs of different body sizes/weights.

### MATERIALS AND METHODS

**Animals:** In this study, 200 eyes of 100 dogs without ophthalmic disorders that were presented to Osaka Prefecture University Veterinary Medical Center during 2007–2009 for examination and treatment of medical or surgical diseases were used. All dogs examined received general ophthalmologic examinations to rule out glaucoma and uveitis.

The dogs were divided into 4 groups according to body weight (BW); <4 kg [toy breed group (TBG): 6 Yorkshire Terriers, 5 Chihuahuas, 3 Pomeranians, 2 Toy Poodles, 1 Maltese and 1 mongrel]; 4–8 kg [small breed group (SBG): 12 Miniature Dachshunds, 7 Shih Tzus and 6 Papillons]; 8–20 kg [medium breed group (MBG): 19 Beagles, 7 Shiba Inus, 5 American Cocker Spaniels, 3 Pembroke Welsh Corgis, 3 Shetland Sheepdogs, 2 Miniature Schnauzers and 2 mongrels]; and >20 kg [large/giant breed group (LGBG): 6 Labrador Retrievers, 4 Golden Retrievers, 2 Doberman Pinschers, 1 Afghan Hound, 1 Bernese Mountain Dog, 1 Irish Setter and 1 Newfoundland].

**Examination procedure:** Both eyes of each dog were examined by UBM by using a UD-1000 with 10-MHz and 40-MHz transducer probes (Tomey Corporation, Nagoya, Japan). Unanesthetized or unsedated dogs were examined with manual restraint in sternal recumbency, while anesthetized/sedated cases were examined in lateral recumbency. After topical administration of 0.4% oxybuprocaine hydrochloride (Santen, Osaka, Japan), hydroxyethylcellulose (Senjyu, Osaka, Japan) was applied to the eyes to protect the

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cornea and sclera from the hypoallergenic ultrasound gel (Parker Laboratories, Fairfield, NJ, U.S.A.). All images of anterior segment structures containing the ICA were obtained with 40-MHz probes and hypoallergenic ultrasound gel in the longitudinal position, which meant that the scan plane was perpendicular to the limbus or corneal surface. Three to 5 images were sequentially obtained from the 10 to 3 o'clock positions, which were the easiest positions to access in the dogs. The 10-MHz probe was used only for assessment of the ocular axial length (OAL). After UBM examination, the examined eyes were washed with saline (Otsuka, Tokyo, Japan) and treated with 0.3% ofloxacin (Santen, Osaka, Japan). The following 8 parameters were evaluated: (1) OAL, measured from the corneal epithelial surface to the inner surface of the retina on the equatorial plane (Fig. 1-A); (2) the anterior chamber depth (ACD), measured from the posterior aspect of the Descemet's membrane (PADM) to the anterior lenticular capsule (ALC) (Fig.

1-B); (3) the distance of the Schwalbe's line (borderline of the cornea and sclera) to the ALC (SLD) (Fig. 1-C); (4) the corneal thickness at the limbus (CT), measured from the corneal epithelial surface to the PADM at the limbus (Fig. 1-C); (5) CCW (the width of the ciliary cleft), measured from the superior surface of the root of the iris to the inner surface of the sclera on a perpendicular line (Fig. 1-C); (6) CCA (the area of the ciliary cleft), calculated by the formula for a trapezoid as the average of the depth of CC (CCD)  $\times$  CCW =  $(\text{CCD}_1 + \text{CCD}_2) \times 1/2 \times \text{CCW}$  (Fig. 1-C); (7) the ratio of CCW and SLD (CCW/SLD); and (8) the ratio of CCW and CT (CCW/CT).

*Statistical analysis:* Each parameter was measured from an obtained image in the examined dogs, and then the mean (standard deviation; SD) was calculated. The results obtained were evaluated by statistical analysis (Statcel 2nd ed.; OMS Publishing Co., Tokyo, Japan). Correlation coefficients were calculated with Spearman's correlation coefficient.

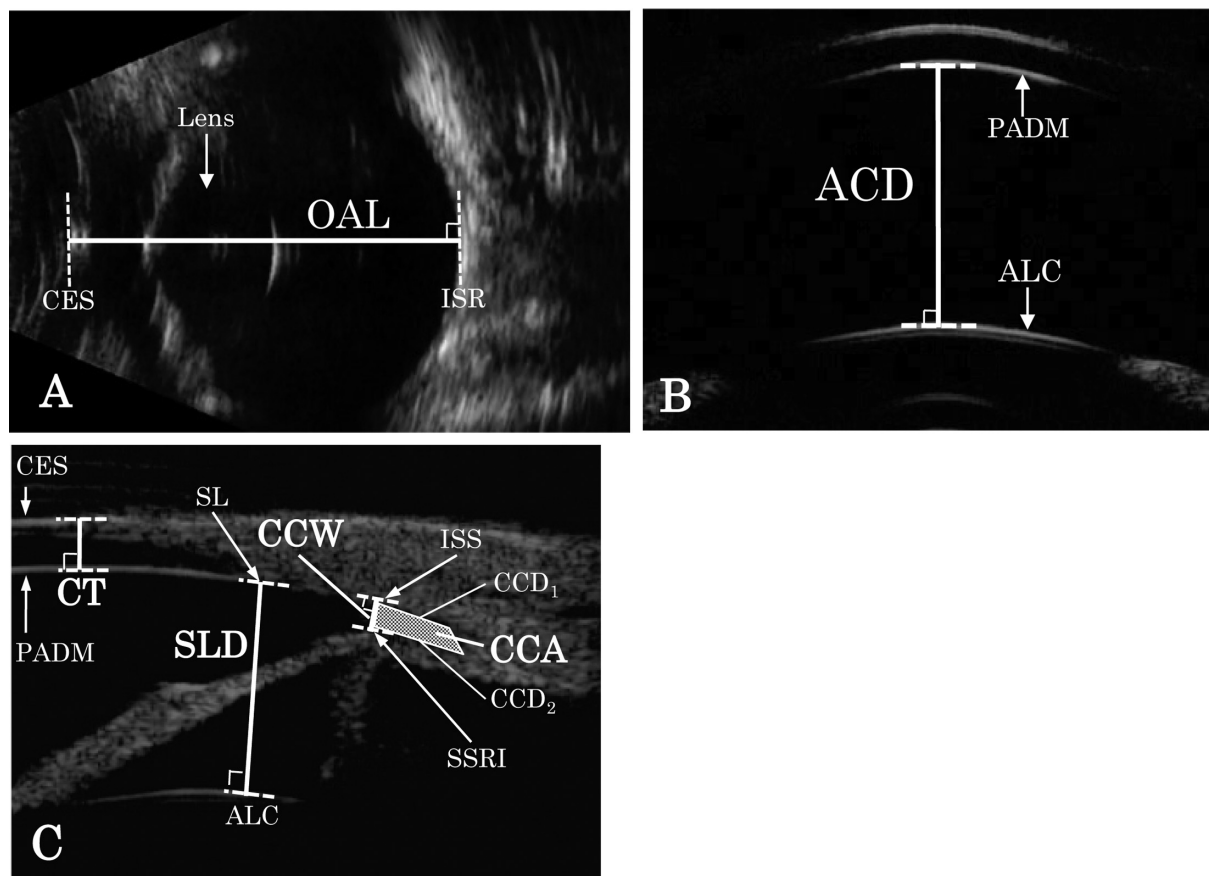


Fig. 1. Typical images of ultrasound of the eye (A), the anterior chamber (B) and iridocorneal angle (C) in a dog. The ocular axial length (OAL) was measured from the corneal epithelial surface (CES) to the inner surface of the retina (ISR) on the equatorial plane. The anterior chamber depth (ACD) was measured from the posterior aspect to the Descemet's membrane (PADM) to the anterior lenticular capsule (ALC). SLD was measured as the distance of the Schwalbe's line (SL; the borderline of the cornea and sclera) to the ALC. The corneal thickness at the limbus (CT) was measured from the CES to the PADM at the limbus. The width of the ciliary cleft (CCW) was measured from the superior surface of the root of the iris (SSRI) to the inner surface of the sclera (ISS) on a perpendicular line. The area of the ciliary cleft (CCA) was calculated by the following formula for a trapezoid: average of the depth of the ciliary cleft (CCD)  $\times$  CCW =  $(\text{CCD}_1 + \text{CCD}_2) \times 1/2 \times \text{CCW}$  (white dotted area).

Table 1. Results for each parameter measured in toy breed group (TBG), small breed group (SBG), medium breed group (MBG), and large/giant breed group (LGBG)

	TBG (n=36)	SBG (n=50)	MBG (n=82)	LGBG (n=32)
BW (kg)	2.82 ± 0.72 <sup>a,b,c</sup>	6.08 ± 1.66 <sup>a,d,e</sup>	11.10 ± 2.78 <sup>b,d,f</sup>	29.90 ± 7.38 <sup>c,e,f</sup>
OAL (mm)	17.86 ± 0.81 <sup>a,b,c</sup>	19.86 ± 0.89 <sup>a,d,e</sup>	20.59 ± 0.89 <sup>b,d,f</sup>	21.93 ± 0.47 <sup>c,e,f</sup>
ACD (mm)	2.93 ± 0.28 <sup>a,b,c</sup>	3.27 ± 0.42 <sup>a,d,e</sup>	3.48 ± 0.31 <sup>b,d,f</sup>	3.98 ± 0.28 <sup>c,e,f</sup>
SLD (mm)	2.19 ± 0.18 <sup>a,b,c</sup>	2.48 ± 0.23 <sup>a,d,e</sup>	2.48 ± 0.09 <sup>b,d,f</sup>	2.93 ± 0.18 <sup>c,e,f</sup>
CT (mm)	0.65 ± 0.06 <sup>a,b</sup>	0.71 ± 0.10 <sup>c</sup>	0.73 ± 0.92 <sup>a</sup>	0.77 ± 0.06 <sup>b,c</sup>
CCW (mm)	0.20 ± 0.08 <sup>a</sup>	0.25 ± 0.10 <sup>b</sup>	0.25 ± 0.09 <sup>c</sup>	0.31 ± 0.07 <sup>a,b,c</sup>
Rectified CCW (mm)	0.24 ± 0.09	0.26 ± 0.10	0.24 ± 0.08	0.27 ± 0.07
CCA (mm <sup>2</sup> )	0.12 ± 0.04 <sup>a,b</sup>	0.15 ± 0.05 <sup>c,d</sup>	0.19 ± 0.06 <sup>a,c,e</sup>	0.25 ± 0.08 <sup>b,d,e</sup>
Rectified CCA (mm <sup>2</sup> )	0.15 ± 0.05	0.15 ± 0.06	0.18 ± 0.06	0.19 ± 0.07
CCW/SLD	0.09 ± 0.04	0.10 ± 0.04	0.09 ± 0.03	0.11 ± 0.03
CCW/CT	0.31 ± 0.12 <sup>a</sup>	0.37 ± 0.17	0.34 ± 0.13	0.41 ± 0.11 <sup>a</sup>

Data are represented as means ± SD. Mean values within a row with the same superscripts are significantly different ( $p < 0.05$ ).

cient by the rank test. Comparison of non-rectified and rectified values was evaluated by multiple comparisons with Shaffer's test. A  $p$ -value less than 0.05 was considered to be statistically significant.

## RESULTS

The results for each parameter in TBG, SBG, MBG and LGBG are shown in Table 1. The correlation between BW and OAL was initially examined to demonstrate that differences of OS depended on canine body size/weight. To account for the non-linear relationship between BW and OAL, the BW data were transformed to natural logarithm. A significantly linear correlation was detected between these 2 variables ( $r = 0.81$ ,  $p < 0.001$ ; Fig. 2). Since OAL could not be measured with the high-frequency ultrasound probe (40 MHz), another parameter considered to be in good agreement with OAL was examined. The correlations between OAL and ACD, SLD and CT were  $r = 0.80$  ( $p < 0.001$ ),  $r = 0.87$  ( $p < 0.001$ ) and  $r = 0.44$  ( $p < 0.001$ ), respectively (Fig. 3). A significant positive correlation was found between OAL and SLD, suggesting that SLD could be used to rectify OAL to reflect OS. The rectified CCW ( $r$ -CCW) was recalculated with the SLD obtained from the UBM images and the optional fixed SLD (OFS), such as the mean of the SLDs of the examined dogs as follows:  $r$ -CCW = CCW × (OFS/SLD). The CCW/SLD was also evaluated. The rectified CCA ( $r$ -CCA) was recalculated using the following formula:  $r$ -CCA = CCA × (OFS/SLD)<sup>2</sup>. In this study, the SLD mean was 2.55 mm. The  $r$ -CCW and  $r$ -CCA were calculated as  $r$ -CCW = CCW × (2.55/SLD) and  $r$ -CCA = CCA × (2.55/SLD)<sup>2</sup>. The means (± SD) of CCW and CCA in TBG, SBG, MBG, and LGBG were 0.20 ± 0.08 mm and 0.12 ± 0.04 mm<sup>2</sup>, 0.25 ± 0.10 mm and 0.15 ± 0.05 mm<sup>2</sup>, 0.25 ± 0.09 mm and 0.19 ± 0.06 mm<sup>2</sup>, and 0.31 ± 0.07 mm and 0.25 ± 0.08 mm<sup>2</sup>, respectively. The CCW values of TBG, SBG and MBG were narrower than those of LGBG (TBG vs. LGBG,  $p < 0.001$ ; SBG vs. LGBG,  $p = 0.03$ ; MBG vs. LGBG,  $p = 0.01$ ; Table 1). There were significant statistical

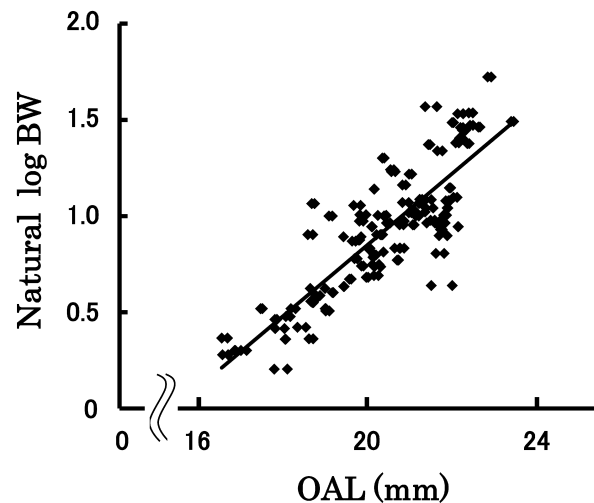


Fig. 2. Correlation between the ocular axial length (OAL) and the natural log-transformed body weight (BW). This figure shows the linear relationship of BW and OAL after natural logarithmic transformation. The correlation coefficient in all dogs was  $r = 0.81$  ( $p < 0.001$ ).

differences in CCA between all the groups except for TBG and SBG (Table 1). The  $r$ -CCW and  $r$ -CCA in TBG, SBG, MBG, and LGBG were 0.24 ± 0.09 mm and 0.15 ± 0.05 mm<sup>2</sup>, 0.26 ± 0.10 mm and 0.15 ± 0.06 mm<sup>2</sup>, 0.24 ± 0.08 mm and 0.18 ± 0.06 mm<sup>2</sup>, and 0.27 ± 0.07 mm and 0.19 ± 0.07 mm<sup>2</sup>, respectively. The CCW/SLD and CCW/CT in TBG, SBG, MBG, and LGBG were 0.09 ± 0.04 and 0.31 ± 0.12, 0.10 ± 0.04 and 0.37 ± 0.17, 0.09 ± 0.03 and 0.34 ± 0.13, and 0.11 ± 0.03 and 0.41 ± 0.11, respectively. There were no significant statistical differences in  $r$ -CCW,  $r$ -CCA and CCW/SLD in any of the groups (Table 1).

## DISCUSSION

In glaucomatous eyes, many structural abnormalities can be detected by UBM examination including CC closure and

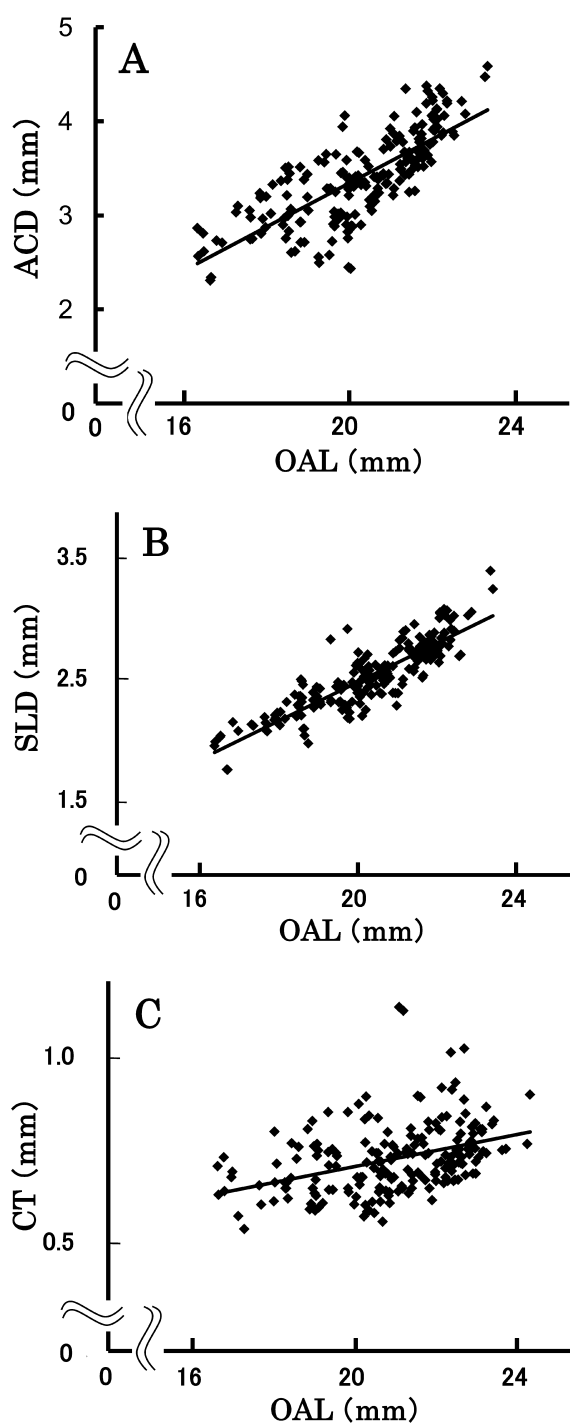


Fig. 3. Correlation between ocular axial length (OAL) and the anterior chamber depth (ACD; A), the distance of the Schwalbe's line to the anterior lens capsule (SLD; B) and the corneal thickness at the limbus (CT; C) in the dogs. The correlations between OAL and ACD, SLD and CT were  $r=0.80$  ( $p<0.001$ ),  $r=0.87$  ( $p<0.001$ ) and  $r=0.44$  ( $p<0.001$ ), respectively.

significant decrease of CCW and CCA [11]. CC abnormalities are also involved in the pathogenesis and/or onset of glaucoma [3]. Therefore, detailed and quantitative CC evaluations are important to not only diagnose glaucoma but also monitor therapeutic effects in animals suffering from the disease. Although CCW and CCA can be evaluated quantitatively by UBM in each animal, misdiagnosis could be made when the results of CCW and CCA obtained from UBM images are used. In the present study, the measured CCW values of TBG, SBG and MBG were narrower than those of LGBG (Table 1). In addition, there were significant differences in CCA between all groups except TBG and SBG (Table 1). This phenomenon was attributed to OS variation due to differences in body size/weight [6] because the examined dogs in all the groups had normotensive eyes with no ophthalmologic abnormalities including the ICA and CC. Therefore, some variables such as CCW and CCA obtained from UBM images should be rectified for comparison between individual animals of different body sizes/weights.

Since OAL generally reflects OS [9], the correlation between OAL and BW was initially assessed in this study. A significantly linear correlation was detected between OAL and the natural log of BW (Fig. 2), suggesting that the differences in OS depended on the canine body size/weight. It would be possible to compare values between dogs of different body sizes/weights if the OAL could be fixed. The OAL data was obtained solely from an image examined with a 10-MHz probe, and it could not be observed on the same UBM images of the ICA used for determining CCA, which were examined only with the high-frequency 40-MHz ultrasound probe. Hence, an optimal alternate parameter should be identified for rectifying CCA on the same UBM images of the ICA. ACD was measured with the high-frequency 40-MHz ultrasound probe, though the parameter could not be evaluated on the same UBM images used for determining CCA, indicating that ACD was not a good candidate for rectifying CCA. Although the CT and SLD could be observed on the same UBM imaged used for determining CCA, the correlation between OAL and CT was poor ( $r=0.44$ ). A significant positive correlation was found between OAL and SLD ( $r=0.87$ ), suggesting that SLD could be used to rectify CCA. The CCA could be rectified by setting the fixed SLD as the OFS. In this situation, CCW and CCD are converted by the following formulae: converted CCW ( $c\text{-CCW}$ ) =  $\text{CCW} \times (\text{OFS}/\text{SLD})$  and converted CCD ( $c\text{-CCD}$ ) =  $\text{CCD} \times (\text{OFS}/\text{SLD})$ . The  $r\text{-CCA}$  formula was derived as follows:

$$\begin{aligned} r\text{-CCA} &= (c\text{-CCD}_1 + c\text{-CCD}_2) \times 1/2 \times c\text{-CCW} \\ &= \text{CCA} \times (\text{OFS}/\text{SLD})^2. \end{aligned}$$

CCW was also rectified by using the SLD and OFS as in the case of CCA. The formula for  $r\text{-CCW}$  was as follows:  $r\text{-CCW} = \text{CCW} \times (\text{OFS}/\text{SLD})$ . The CCW/SLD and CCW/CT were evaluated as additional parameters of CCW because they could be calculated using non-rectified or non-con-

verted values.

The CCW values of TBG, SBG and MBG were narrower than those of LGBG, and there were significant statistical differences in CCA between all groups except TBG and SBG (Table 1). However, there were no significant statistical differences in r-CCW and r-CCA in TBG, SBG, MBG or LGBG (Table 1). These results suggested that the CCW and CCA obtained from the UBM images could be rectified for comparison between the examined dogs. The ratio of CCW and SLD did not show significant changes in dogs of different body size/weights. The formula for rectification of CCW and CCA and the CCW/SLD cannot be used for dogs with lens luxation or aphakia because the SLD cannot be determined. There were breeds predisposed to primary angle closure glaucoma (PACG) in the dogs examined, although the examined dogs belonging to breeds with PACG had no ocular diseases, normotensive eyes and no abnormalities of ICA structures that were evaluated using UBM. Therefore, the dogs were not excluded from the examination in this study. The rectified formula presented here might not be perfect. Improvements in the formula for rectification of CCW and CCA are needed via evaluation of more clinical cases, although the formulae for r-CCW and r-CCA and the CCW/SLD presented here are clinically available for comparison of the CCW and CCA obtained from UBM images between dogs of different body sizes/weights.

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