Japanese Journal of Veterinary Research 68(1): 13-20, 2020

JJVR

REGULAR PAPER Internal Medicine

Indocyanine green angiography findings with Collie eye anomaly in Hokkaido dogs.

Arisa Masuko[1)], Seiya Maehara[1,*)], Miri Hayashi[1)], Reiko Kato[1)], Arisa Shimode[1)], Osamu Yamato[2)] and Tetsuya Nakade[1)]

[1)] Department of Small Animal Clinical Sciences, School of Veterinary Medicine, Rakuno Gakuen University, 582 Bunkyodai Midori-machi, Ebetsu, Hokkaido 069-8501, Japan

[2)] Laboratory of Clinical Pathology, Department of Veterinary Clinical Sciences, Faculty of Agriculture, Kagoshima University, 1-21-24 Kohrimoto, Kagoshima 890-0065, Japan

Received for publication, March 5, 2019; accepted, September 26, 2019

Abstract

Collie eye anomaly (CEA) is an inherited, congenital ocular disorder caused by a defective mesodermal differentiation in the posterior segment of the eye. Major ocular finding of CEA is abnormalities of choroidal vessels, that is choroidal hypoplasia. Indocyanine green angiography (IA) is one of the useful ocular examination to observe choroidal vessels in both human and dogs. The purpose of this study was to evaluate IA with CEA in Hokkaido dogs, which is one of the traditional Japanese breed and natural monument in Japan. Ten Hokkaido dogs that had been carried out genetic tests in advance were included in this study. Dogs included in this study had ophthalmic examination, such as menace response, dazzle reflex, direct and indirect pupillary light reflex, slit-lamp biomicroscopy, simple funduscopy, and IA. According to the result of genetic tests, they were classified as 8 affected and 2 carrier dogs. Simple funduscopy revealed choroidal hypoplasia bilaterally and temporal or dorsotemporal area to the optic disc in all affected dogs. With IA, we could observe the abnormalities of choroidal vessels not only at the area coincided with choroidal hypoplasia with simple funduscopy but also at the area detected normal with simple funduscopy in affected dogs. No abnormalities on fundus were observed with both simple funduscopy and IA in all carrier dogs. In conclusion, it was revealed that choroidal hypoplasia in CEA Hokkaido dogs was existed also in the area that could not be observed with simple funduscopy.

Key Words: choroidal hypoplasia, Collie eye anomaly, Hokkaido dog, indocyanine green angiography

Introduction

Collie eye anomaly (CEA) is a congenital, hereditary ocular disorder that is bilateral, non-progressive and not related to sex, coat color, or type of coat[1-4,6,13-15)]. In 2007, a 7799 base pairs deletion in the fourth intron of the Non-homologous end-joining factor 1 (NHEJ1) gene has been reported[16)]. This disease affects many breeds both Collie-related and non-Collie breeds[1-4,6,11,13-15)]. In addition, Hokkaido dog, which is one of the traditional Japanese breed and natural

* Corresponding author: Seiya Maehara, School of Veterinary Medicine, Rakuno Gakuen University, 582 Bunkyodai Midori-machi, Ebetsu, Hokkaido 069-8501, Japan
Phone: +81-11-388-4853. Fax: +81-11-388-4853. E-mail: seiya-m@rakuno.ac.jp
doi: 10.14943/jjvr.68.1.9

monument in Japan, has also been reported that the known CEA-associated mutation was detected[10]. Clinically, most major ocular lesion associated with CEA is choroidal hypoplasia in the region temporal or dorsotemporal area to the optic disc, and other clinical features are coloboma of the optic disc, retinal detachment, intraocular hemorrhage, tortuous retinal vessels, retinal dysplasia, and microphthalmia[1-4,6]. It is desirable to make a diagnosis to perform ophthalmic examinations including slit-lamp biomicroscopy and simple funduscopy in conjunction with genetic test in predilection breeds.

In ocular fundus of dogs, it is impossible to evaluate the choroid accurately that is obstructed by the shielding effect of the tapetum and retinal pigment epithelium (RPE) containing pigment. In other words, we could not observe the choroid with simple funduscopy if it is masked by the tapetum and pigment in the RPE, and not evaluate that area in detail. Generally, the choroidal hypoplasia in CEA dogs is diagnosed with simple funduscopy, but it has not been known whether choroidal hypoplasia exists in the area that cannot be observed with simple funduscopy due to the tapetum or RPE. We considered that there may be the choroidal hypoplasia in the area which cannot be observed with just simple funduscopy in CEA dogs.

The previous studies suggested that indocyanine green angiography (IA) is used to visualize the choroidal vessels not only in human ophthalmology but also veterinary ophthalmology including dogs and cats[9,16]. It has also been reported that IA employing an infrared-sensitive charged coupled device (CCD) provides to identify the whole vasculature of the choroid in normal dogs and cats[9,16]. There have been no reports to evaluate IA for clinical study in dogs, including CEA. We also considered that IA could evaluate the choroidal vessels and existing of choroidal hypoplasia in invisible area with simple funduscopy in CEA dogs.

The purpose of this study was to evaluate choroid with IA and clarify whether choroidal hypoplasia could be observed or not in the area which cannot be observed with simple funduscopy in CEA Hokkaido dogs.

Materials and Methods

Animals: Ten Hokkaido dogs, which had been carried out genetic tests in advance, and came to Rakuno Gakuen University Animal Medical Center, were included in this study. They were 5 males and 5 females. The age range was 4 months to 9 years old and their body weight was 10.2 to 23.4 kg. We have permission from their owners to publish their profile and ocular findings in this study.

Genetic tests: The DNA samples were genotyped for the presence of the mutant allele in the NHEJ1 gene that has been previously reported as the mutation of choroidal hypoplasia[12]. The procedure of genetic test was performed according to the methods described by previous reports[8,10]. Dogs were classified as affected which has two copies of the mutant allele is present, carrier which has the mutation in single copy, and normal in the absence of mutant allele.

Ophthalmic examinations: Ophthalmic examinations which included menace response, dazzle reflex, direct and indirect pupillary light reflex, slit-lamp biomicroscopy (SL-7, Kowa, Nagoya, Japan), and simple funduscopy (TRC-50IX, Topcon, Tokyo, Japan) were performed in all dogs. Slit-lamp biomicroscopy and simple funduscopy were conducted after mydriasis with 0.5% tropicamide and 0.5% phenylephrine hydrochloride (Mydrin-P, Santen, Osaka, Japan).

Indocyanine green angiography: We performed IA in the same methods as the previous reports by Wakaiki and Hayashi[9,16]. For IA, we used the aforementioned fundus camera fitted with an infrared-sensitive CCD, and the fundus camera had an 805-nm excitation filter and an 835-nm

Table 1. Signalment and the results of CEA genotype, simple funduscopy and Indocyanine green angiography

Case	Gender	Age	Genotype	Area of CH with simple funduscopy		Area of CH with IA	
				R	L	R	L
1	F	4m	Affected	Temporal	Temporal	Temporal	Temporal
2	F	1y	Affected	Dorsotemporal	Dorsotemporal	Dorsotemporal	Dorsotemporal
3	F	2y	Affected	Temporal	Undetermined*	Temporal	Undetermined*
4	F	7y	Affected	Temporal	Temporal	Temporal	Temporal
5	F	9y	Affected	Temporal	Temporal	Temporal and Dorsotemporal	Temporal and Dorsotemporal
6	M	1y	Affected	Temporal	Temporal	Temporal	Temporal
7	M	3y	Affected	Temporal	Temporal	Temporal	Temporal
8	M	7y	Affected	Temporal	Temporal	Temporal and Dorsal	Temporal and Dorsal
9	M	5y	Carrier	Normal	Normal	Normal	Normal
10	M	9y	Carrier	Normal	Undetermined*	Normal	Undetermined*

CH: Choroidal hypoplasia, IA: Indocyanine green angiography,
F: Female, M: Male, R: Right eye, L: Left eye
*: could not be observed fundus because of severe lens opacity

Choroidal hypoplasia with IA was found in the same area as that with simple funduscpy in most cases. While in Case 5 and 8, choroidal hypoplasia was found with IA also in another area where not seen with simple funduscopy.

barrier. Digital fundus photographs taken with the IMAGEnet 2000 (Topcon, Tokyo, Japan) system were recorded on a HDD recorder. All dogs were inserted an intravenous catheter (Surflo F&F, TERUMO, Tokyo, Japan) in the cephalic vein prior to examination. Indocyanine green angiography was conducted with the dogs in a sternal position under sedation with a combination of 0.01 mg/kg medetomidine (Domitor, Zenoaq, Tokyo, Japan), 0.15 mg/kg midazolam (Dormicam, Astellas, Tokyo, Japan), and 0.025 mg/kg butorphanol (Vetorphale, Meiji Seika, Tokyo, Japan), injected intravenously. Indocyanine green (ICG) dye (Diagnogreen, Daiichi, Tokyo, Japan) was injected (1.0 mg/kg body weight) as a bolus through the intravenous catheter followed by a 5-mL saline flush. The timer was started immediately after administration of the ICG dye, and sequential photographs (30 images/sec.) were recorded on HDD for approximately 40 min. From the onset of angiography, the infrared light intensity of the fundus camera was set to approximately 20 mW/cm^2 (the highest setting), and the intensity was adjusted to match luminance if fluorescence increased excessively; this was a precaution to maintain clear visualization on the angiograms. We recorded photographs about 15 minutes post-injection of ICG dye. After completing IA of each eye, we used the recorded HDD recorder for detailed observation and measurement of the ICG angiogram. To reverse from sedation after the procedure, we injected 0.05 mg/kg atipamezole (Atipame, Kyoritsu, Tokyo, Japan) into all dogs intravenously.

Results

According to the result of the genetic tests, 8 (3 males and 5 females) were affected and 2 (all males) were carrier dogs. There were no normal dogs in this study.

Signalment of all cases and the results of CEA genotype, simple funduscopy and IA findings were shown in Table 1. In ophthalmic examinations, although menace response of 2 eyes (2 dogs) was negative due to severe lens opacity, dazzle reflex and direct and indirect pupillary

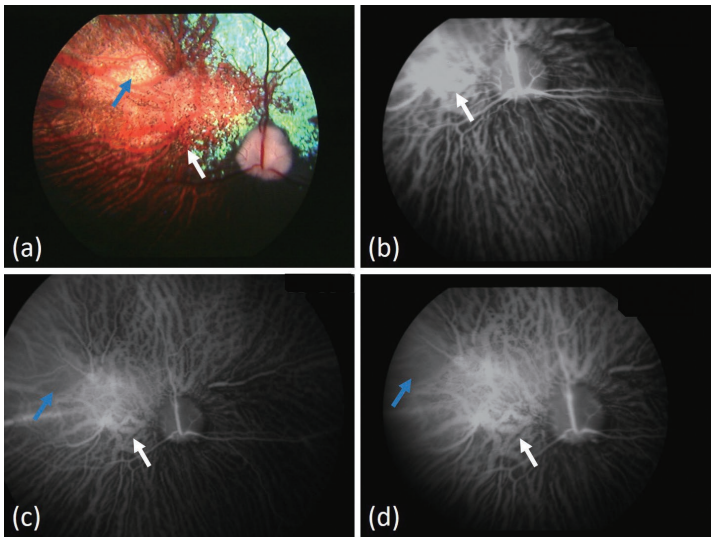

Fig. 1. Simple funduscopy and IA of right eye in affected female, 1 year old Hokkaido dog (Case 2).
Irregular choroidal vessels (white arrow) showing choroidal hypoplasia and white sclera (blue arrow) were observed in the dorsotemporal area of the optic disc with simple funduscopy (a). In IA (b-d), tortuous, irregular choroidal vessels permeating the temporal area to the optic disc (white arrow) were contrasted, and gradually decreased contrast level at the same area. Since there were no choroidal vessels at the area where the white sclera was found with simple funduscopy, defects of fluorescence were observed (blue arrow; c, d).

(a) Simple funduscopy, (b)30 seconds, (c)9 minutes, (d)11 minutes after injection of ICG in IA, respectively

light reflex in all dogs were positive, respectively. From the result of slit-lamp biomicroscopy, mild lens opacity was seen in 15 eyes (9 dogs) such as Fig. 2(a) and severe lens opacity in 2 eyes (2 dogs). No abnormalities in the anterior ocular segment were observed except for lens opacity in all eyes.

We could perform simple funduscopy with 18 eyes except for 1 eye of affected dog and 1 eye of career dog, which was difficult to examine the fundus in detail because of severe lens opacity. We showed simple funduscopic photographs in 3 affected dogs in Fig. 1(a), 2(b), and 3(a), respectively. With simple funduscopy, irregular choroidal vessels showing choroidal hypoplasia were observed in the temporal or dorsotemporal area of the optic disc in all affected dogs, and this is as well as previous reports of choroidal hypoplasia in CEA dogs[1-4,6]. Other abnormalities in the fundus, such as posterior polar coloboma, retinal detachment, were not observed in affected dogs. In carrier dogs, on the other hand, no abnormalities in the posterior ocular segment including choroidal hypoplasia were observed.

Indocyanine green angiography could be carried out in all dogs. In this study, all dogs have no side effects, complications, and sequelae with IA using near-infrared light. We showed IA photographs in 3 affected dogs in Fig. 1(b-d), 2(c-e), and 3(b-d), respectively. In the early phase (about 30 seconds to 2 minutes after injection of ICG), tortuous, irregular choroid blood vessels permeating the temporal or dorsotemporal area to the optic disc were contrasted in all affected dogs. The same area as the early phase was discovered to have decreased contrast in the middle phase (about 4 minutes to 10 minutes after injection of ICG). Finally, the late phase (about 11 minutes to 15 minutes after injection of ICG) allowed us to recognize that the same area of decreased contrast observed in the middle phase remained at a decreased contrast level.

In 2 affected dogs (Case 5 and 8), we could observe the areas that appeared no abnormality under simple funduscopy showed irregularly contrasted with IA. Although choroidal vessels could not be observed in the dorsotemporal area

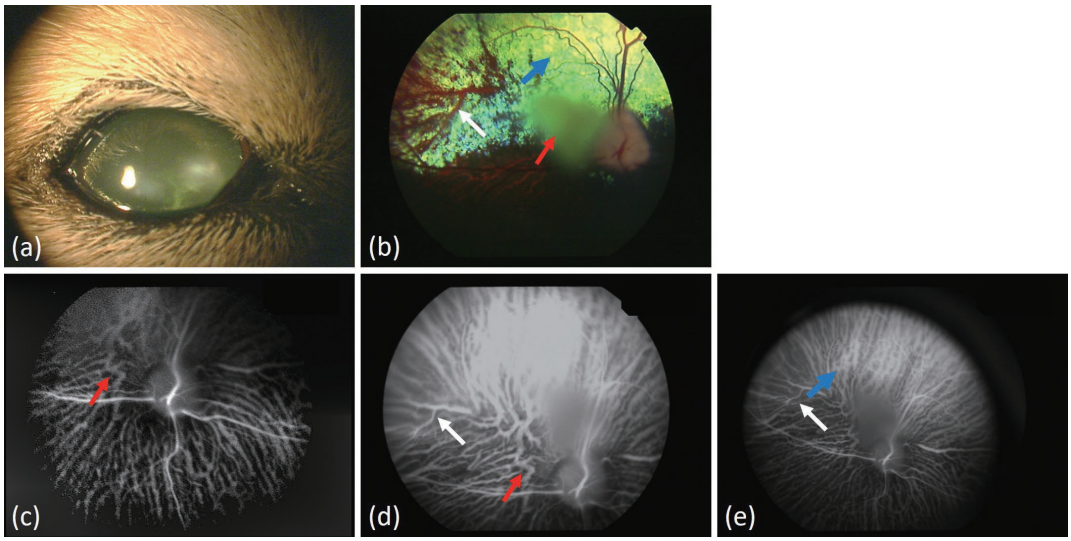

Fig. 2. Slit-lamp biomicroscopy, simple funduscopy and IA of right eye in affected female, 9 years old Hokkaido dog (Case 5).
Mild, partial opacity is observed at posterior cortex of lens (a). Irregular choroidal vessels (white arrow) showing choroidal hypoplasia were observed in the temporal area of the optic disc with simple funduscopy (b). The area of white opacity (red arrow) in the temporal area of near the optic disc was shown lens opacity. Tortuous, wider diameter of abnormal choroidal vessels which could not be observed with simple funduscopy because of lens opacity, could be observed with IA (red arrow; c, d). As time went on, tortuous, irregular choroidal vessels permeating the temporal area to the optic disc (white arrow; d) were contrasted, and gradually decreased contrast level at the same area (e). In addition, the dorsotemporal area to the optic disc that appeared no abnormality under simple funduscopy showed irregularly contrasted (blue arrow; e).

(a) Slit-lamp biomicroscopy, (b) simple funduscopy, (c)2 minutes, (d)8 minutes, (e)12 minutes after injection of ICG in IA, respectively

to the optic disc with simple funduscopy (Fig. 2(b) and 3(a)) because of masking by the tapetum, we observed choroidal hypoplasia in this area with IA in the late phase (Fig. 2(e) and 3 (d)).

Coincidentally, choroidal vessels of eyes which could not be observed with simple funduscopy because of mild, partial lens opacity, could be observed with IA (Fig. 2). We detected choroidal hypoplasia in temporal area to the optic disc for 1 eye with mild lens opacity of 1 affected dog, and no abnormalities of fundus were detected for 1 eye with mild lens opacity of 1 carrier dog. In contrast, we could not observe fundus with IA in 2 eyes of 2 affected dogs which had severe and whole lens opacity, respetively. It also could be observed that the area where the white sclera was found in simple funduscopy had defects of fluorescence in 1 affected dog. No abnormalities were observed even with IA in any phases in all carrier dogs.

Discussion

This is the first report that evaluating IA for dogs with CEA. Indocyanine green angiography revealed choroidal vessels which meandered through in the region coincided with choroidal hypoplasia, and it became clear that choroidal hypoplasia in CEA Hokkaido dogs also could be observed with IA in the area that could not be observed with simple funduscopy.

We revealed that the area of choroidal hypoplasia with IA is wider than that observed with simple funduscopy in CEA Hokkaido dogs. In dog puppies, as they grow older, developing the RPE and tapetum in the ocular fundus, so it is impossible or difficult to evaluate the choroidal vessels accurately with simple funduscopy. If mild lesions with choroidal hypoplasia observed at about 5-7 weeks of age dogs with CEA, they may be masked by the pigment in the RPE,

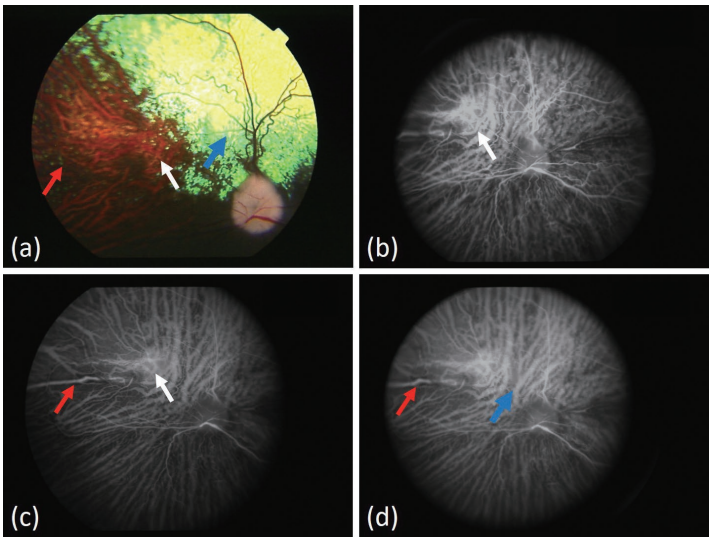

Fig. 3. Simple funduscopy and IA of right eye in affected male, 7 years old Hokkaido dog (Case 8).
Irregular choroidal vessels (white arrow) showing choroidal hypoplasia was observed in the temporal area of the optic disc in simple funduscopy (a). In IA, tortuous, irregular choroidal vessel permeating the temporal area to the optic disc (white arrow) were contrasted, and gradually decreased contrast level at the same area (b, c). Another irregular choroidal vessel which could not be detected with simple funduscopy because of the retinal pigment epithelium was observed in the temporal area of the optic disc over time (red arrow; c). In addition, the dorsal area to the optic disc that appeared no abnormality under simple funduscopy showed irregularly contrasted (blue arrow; d).

(a) Simple funduscopy, (b)38 seconds, (c)6 minutes, (d)14 minutes after injection of ICG in IA, respectively

the so-called "go normal" phenomenon[3,6,7]. It may lead examiners to wrongly recognize CEA affected dog as normal fundus. To evaluate choroidal vessels, IA is one of the useful ocular examination in both human ophthalmology and veterinary ophthalmology including dogs and cats[9,16]. Indocyanine green dye binds more than 98% to blood proteins, absorbs the near-infrared light, and readily penetrates the RPE[5,17]. According to these properties, choroidal vessels can be accurately visualized with IA than simple funduscopy. In the present study, as a matter of the fact, it is considered that a part of abnormality which was revealed in IA was appeared normal in simple funduscopy in 2 of 8 affected dogs because of masking by development of the tapetal layer or by pigmentation of the RPE. It is recommended that dog puppies of highly susceptible breeds of CEA be perform ocular examination before choroidal hypoplasia is masked[3,6,7]. In the present study, it was suggested that performing IA can be recognized choroidal vessels of even older dogs with CEA-affected which have apparently normal findings by simple funduscopy.

We recorded photographs of IA about 15 minutes after injection of ICG dye and as a result we found that the best time to observe choroidal hypoplasia is about 1 to 2 minutes after injection of ICG. The contrast level of choroidal vessels decreased gradually after that, so it would be better to observe in early phase. However, in the early phase, it is difficult to evaluate choroidal vessels accurately in some cases because around choroidal vessels become fluorescent. In the late phase, it is relatively easy to evaluate choroidal vessels because only choroidal vessels become highly fluorescent in that phase. That is why we can detect the differences of findings between simple funduscopy and IA in only late phase. Since abnormalities of choroidal vessels in another area may be found after the contrast decreased, it is desirable to keep observation until about 15 minutes after injection of ICG dye when the fluorescence becomes diffuse. In addition,

although the both No.5 and No.8 cases that we detect the different findings between simple funduscopy and IA were old age, we consider there is no difference with age. The tapetum and RPE develop until several months after birth[3,6,7]. If a few weeks old dogs that has not developed yet the tapetum and RPE, we can observe choroidal hypoplasia with only simple funduscopic, even without IA. However, all dogs in this study were adult dogs that have already developed the tapetum and RPE in their fundus, so we couldn't observe the choroid with simple funduscopy because the choroid is masked by the tapetum and pigment in the RPE.

In 2 carrier dogs, no abnormalities of choroidal vessels were observed with not only simple funduscopy but also IA. It is a well-known fact that in CEA, an autosomal recessive inheritance, choroidal hypoplasia is not observed with simple funduscopy in carrier dogs, which is the heterozygous animal[12]. In mature dogs, however, if choroidal hypoplasia is masked by the RPE and tapetum, it cannot be observed accurately with only simple funduscopy and it may be insufficient for diagnosis of CEA. In the present study, we could observe choroidal hypoplasia with IA in 2 affected dogs that appeared no abnormality under simple funduscopy. There was a possibility that choroidal hypoplasia was observed with IA even in carrier dogs that appeared normal under simple funduscopy, but we could not observe choroidal hypoplasia with IA in carrier dogs in this study. It was demonstrated that no choroidal hypoplasia was observed with IA in carrier dogs.

Although many cases have lens opacity in this study, we coincidentally found that some of the eyes which had mild lens opacity could be detected the choroidal vessels behind the opacity with IA. According to the property that ICG dye absorbs near-infrared light and emits peaks in the near-infrared spectrum[5,17], we used a fundus camera fitted with an infrared-sensitive CCD. The near-infrared light penetrates not only the RPE but also other ocular structures including lens. In the dogs which had mild lens opacity, we might have detected the choroidal vessels with IA, which was not seen with simple funduscopy, since it does not have much influence on reaching near-infrared light to the choroid. However, the eyes which had severe or entire lens opacity could not be detected any structures of fundus. As the progression of lens opacity may affect the absorption, reflection and scattering of near-infrared light, it might have been impossible to detect with IA the choroidal vessels of dogs which had severe lens opacity as a result that near-infrared light did not reach the choroid normally.

There are some of the limitations in this study. We should originally be compared affected dogs with genetically normal dogs, but unfortunately, there was no normal dogs in this study. In recent years, Hokkaido dogs are kept as domestic dog, but it is fewer than other Japanese dogs such as Shiba and Akita dog, also designated as a national monument in Japan. For that reason, there may be cases in which inbreeding is carried out to preserve the number of species, so normal Hokkaido dogs are very few possibilities. It is necessary to further investigate if normal Hokkaido dogs are found in the near future. As another limitation, we cannot compare the findings of IA with other breeds since it has not evaluated IA for dogs with CEA in Hokkaido dogs as well as other breeds so far. It seems that we need to investigate whether choroidal hypoplasia could be observed or not in the area which cannot be observed with simple funduscopy in even other CEA dog breeds, like CEA Hokkaido dogs in this study. Moreover, the decrease of the contrast level of the choroidal vessels in the middle and late phase with IA is subjectively. In IA, it does not generally represent the contrast level numerically in human, also dogs and cats[9,16]. Therefore, we need to careful observe the decrease of the contrast level of the choroidal vessels when we conduct IA.

In this study, as we observed fundus with IA in CEA Hokkaido dogs, all dogs in this study have no side effects, complications, and sequelae with IA using near-infrared light. In previous

study, there are no clinical problems by IA using an infrared-sensitive CCD in normal dogs and cats[9,16]. Therefore, we consider IA using near-infrared light a safe examination. It was indicated that IA is a one of the methods for detecting choroidal hypoplasia in CEA Hokkaido dogs and the area of choroidal hypoplasia in CEA Hokkaido dogs might be wider than the area of observed with simple funduscopy in some cases. As a diagnosis of CEA, it may be helpful to carry out IA in addition to simple funduscopy and genetic tests.

References

1) Barnett KC, Stades FC. Collie eye anomaly in the Shetland sheepdog in the Netherlands. J Small Anim Pract 20, 321-329, 1979.
2) Bedford PG. Collie eye anomaly in the border collie. Vet Rec 111, 34-35, 1982.
3) Bedford PG. Collie eye anomaly in the United Kingdom. Vet Rec 111, 263-270, 1982.
4) Bedford PG. Collie eye anomaly in the Lancashire heeler. Vet Rec 143, 354-356, 1998.
5) Bischoff PM, Flower RW. Ten years experience with choroidal angiography using indocyanine green dye: a new routine examination or an epilogue? Doc Ophthalmol 60, 235-291, 1985.
6) Bjerkås E. Collie eye anomaly in the rough collie in Norway. J Small Anim Pract 32, 89-92, 1991.
7) Buening G, Erhardt G. Influences on the frequency of estimated Collie eye anomaly (CEA) in Collies and Shelties in preventive examination-results of a breeding club organization. Kleintierpraxis 47, 407-413, 2002.
8) Chang HS, Mizukami K, Yabuki A, Hossain MA, Rahman MM, Uddin MM, Arai T, Yamato O. A novel rapid genotyping technique for Collie eye anomaly: SYBR Green-based real-time polymerase chain reaction method applicable to blood and saliva specimens on Flinders Technology Associates filter paper. J Vet Diagn Invest 22, 708-715, 2010.
9) Hayashi M, Maehara S, Ito Y, Yamashita K, Kubo A, Nakade T. Comparison between indocyanine green angiography and fluorescein angiography in normal cats. Vet Ophthalmol 20, 310-315, 2017.
10) Mizukami K, Chang HS, Ota M, Yabuki A, Hossain MA, Rahman MM, Uddin MM, Yamato O. Collie eye anomaly in Hokkaido dogs: case study. Vet Ophthalmol 15, 128-132, 2012.
11) Munyard KA, Sherry CR, Sherry L. A retrospective evaluation of congenital ocular defects in Australian Shepherd dogs in Australia. Vet Ophthalmol 10, 19-22, 2007.
12) Parker HG, Kukekova AV, Akey DT, Goldstein O, Kirkness EF, Baysac KC, Mosher DS, Aguirre GD, Acland GM, Ostrander EA. Breed relationships facilitate fine-mapping studies: a 7.8-kb deletion cosegregates with Collie eye anomaly across multiple dog breeds. Genome Res 17, 1562-1571, 2007.
13) Roberts SR. The Collie eye anomaly. J Am Vet Med Assoc 155, 859-865, 1969.
14) Rubin LF, Nelson E, Sharp CA. Collie eye anomaly in Australian Shepherd Dogs. Prog Vet Comp Ophthalmol 1, 105-108, 1991.
15) Stades FC, Barnett KC. Collie eye anomaly in collies in the Netherlands. Vet Quart 3, 66-73, 1981.
16) Wakaiki S, Maehara S, Abe R, Tsuzuki K, Igarashi O, Saito A, Itoh N, Yamashita K, Izumisawa Y. Indocyanine green angiography for examining the normal ocular fundus in dogs. J Vet Med Sci 69, 465-470, 2007.
17) Yoneya S. A new approach for studying the retinal and choroidal circulation. J Jpn Ophthalmol Soc 108, 836-861, 2004.