PAPER

Haematological and biochemical values in North American Scottish deerhounds

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Objective: Sighthounds, including deerhounds, have unique physiological traits that result in laboratory test results that may lie outside reference intervals for the general dog population. Although reference intervals for most analytes are thought to be similar among sighthounds, breed-specific reference intervals are available mainly for greyhounds. The aim of this study was to establish reference intervals for haematology and serum biochemical profiles in deerhounds.

Methods: Venous blood samples were collected from healthy deerhounds. Haematological and biochemical analytes were examined and reference intervals were established using the 5th and 95th percentiles.

Results: The reference intervals obtained from 96 dogs for platelets, reticulocytes, total thyroxine, chloride, gamma glutamyl transferase, bilirubin and glucose were lower than the general dog population. Reference intervals for mean cell volume, potassium, urea, alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase and cholesterol were higher than the general dog population. Reference intervals for eosinophils and globulin were wider than that of the general population.

Clinical Significance: These results confirm that differences in haematological and biochemical values exist in the deerhound. Some appear to be shared by all sighthounds but others may be unique to this breed.

INTRODUCTION

Scottish deerhounds are members of the sighthound family, which includes greyhounds, salukis, whippets and several other breeds (American Sighthound Field Association 2012). These breeds have unique anatomical and physiological features likely due to intentional selection for hunting by speed and sight. Consequently, it should not be surprising that laboratory test results for sighthounds often fall outside the established references intervals (RI) for the general dog population (Zaldívar-López et al. 2011).

Differences in laboratory test results are fairly well-documented in greyhounds, because of their popularity in racing, and several previous studies have determined breed-specific RIs (Porter & Canaday 1971, Steiss et al. 2000, Shiel et al. 2007a, Campora et al. 2011, Dunlop et al. 2011). For example, the ratio between

red blood cells (RBC) and plasma is higher in greyhounds than in non-sighthound breeds, reflecting a greater circulating RBC mass that presumably exists to improve oxygen delivery to tissues during times of peak demand (e.g. during a chase or race). Consequently, RIs for haematocrit (Hct), haemoglobin (Hb) concentration and RBC count are higher in greyhounds than in dogs in general. In addition, leukocyte and platelet counts are lower in greyhounds than in other dogs (Campora et al. 2011, Zaldívar-López et al. 2011). Differences have also been reported in serum concentrations of urea, globulin, calcium, creatinine (Feeman et al. 2003), total protein (TP) and albumin (Fayos et al. 2005, Dunlop et al. 2011) and serum creatine kinase (CK) activity (Porter & Canaday 1971, Steiss et al. 2000, Dunlop et al. 2011).

Haematological and serum biochemical data for Spanish greyhounds (Galgos) have recently been reported. Similar to greyhounds, Galgos had higher Hct, Hb concentration and RBC count and lower platelet counts than dogs in general (Mesa-Sanchez et al. 2012).

It was hypothesized that these differences in clinicopathological RIs might be similar in other sighthound breeds; however, limited information on clinicopathological parameters for non-greyhound sighthound breeds is available. The best studied RIs are likely the thyroid hormones, where low serum total thyroxine (T4) concentrations have been reported in several sighthound breeds (Nachreiner & Refsal 1992, Gaughan & Bruyette 2001, Hill et al. 2001, van Geffen et al. 2006, Shiel et al. 2007b, Panakova et al. 2008, Shiel et al. 2010), including the deerhound. To the authors' knowledge, low serum-free T4 concentration has been reported only in greyhounds, salukis and sloughis (Hill et al. 2001, Gaughan & Bruyette 2001, Panakova et al. 2008, Shiel et al. 2007b, Shiel et al. 2010). A recent study evaluated differences in haematological and biochemical parameters among several sighthound breeds, but not including the deerhound, and found significant differences in some values between breeds (Uhrikova et al. in press). Therefore, some of the RIs for the greyhound may apply to other sighthound breeds, but other intervals may vary between the different breeds. Thus, there is a need to establish RIs for the deerhound to accurately identify clinicopathological abnormalities in individuals of the breed.

The objective of this study was to determine RIs for haematological and serum biochemical profiles based on data generated from healthy deerhounds. The suggested deerhound-specific RIs were then compared to the RIs used by the laboratory analysers for the general dog population.

MATERIALS AND METHODS

The sample population comprised deerhounds from the USA and Canada whose owners signed a consent form to have their dogs included in the study. All dogs were considered healthy on the basis of clinical history and physical examination at the time of sample collection.

All samples were collected on May 10 and 11, 2012 at the Scottish Deerhound Club of America 2012 National Specialty show in Frankenmuth, MI (USA). Jugular or cephalic venous samples were collected in vacuum-sealed 1 mL VetCollect purple top tubes containing EDTA (IDEXX Laboratories, Inc.) for haematology, and in tubes without anticoagulant for biochemical profiles (Monoject, Sherwood). The serum was separated by centrifugation immediately after sample collection. Samples were transported refrigerated with ice packs (haematology) or frozen.

Haematology was performed within 48 hours of collection using a Pro-Cyte Dx analyser (IDEXX Laboratories) with the appropriate software settings. Blood smears were made immediately after collection and stained with Wright–Giemsa at the time of evaluation in 51 of the dogs in the study. Differential white blood cell (WBC) counts were performed manually by counting 100 nucleated cells per smear. The staining characteristics of the eosinophil granules and the presence or absence of platelet clumping were noted.

Serum samples were kept frozen at −30°C until the profiles were evaluated; all samples were analysed within 1 week of collection using a COBAS c501 chemistry analyser (Roche Diagnostics). The analytes measured were; concentrations of total T4, blood urea nitrogen (BUN), creatinine, phosphorous, total calcium, sodium, potassium, chloride, bicarbonate, cholesterol, bilirubin, TP, albumin, and glucose; and activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), gamma glutamyl transferase (GGT) and CK. This data was used to calculate anion gap, osmolality, globulin concentration and albumin:globulin (AG) ratio. Osmolality was calculated using the formula $[1\cdot86\times Na^+ +(glucose/18)+(BUN/2\cdot8)+9]$. Quantitative index values for haemolysis and lipaemia were also generated using this instrument.

Statistical analysis

Descriptive statistics and D'Agostino and Pearson normality testing of the data were obtained using GRAPHPAD PRISM 5 statistical software (GraphPad Software). All RIs were established by using the 5th and 95th percentiles, although not all data were normally distributed. Values lying outside of the interquartile range were considered outliers and were excluded from statistical evaluation.

RESULTS

The study included 96 deerhounds between 6 months and 10 years of age (mean 3·6 ±2·5 years, median 3·3 years) and included 41 males and 55 females.

The reference population for the CBC consisted of all 96 dogs included in the study. However, the instrument did not report the mean platelet volume (MPV) in 19 of the dogs, so the reference population for this value included only 77 dogs.

Results for deerhounds (mean, median and proposed RI for each parameter) are summarized in Table 1, along with RIs provided by IDEXX Laboratories for the Pro-Cyte Dx analyser (i.e. for dogs in general), and the percent of the deerhound population that was outside the non-breed-specific RI. RBC count, Hb concentration, Hct, mean cell volume (MCV), mean cell haemoglobin (MCH), red cell distribution width (RDW), and

		Reference interval (RI)				
Analyte (units)	**Deerhound Mean ±sd**	**Deerhound median**	**Deerhound**	**ProCyte Dx**	**% Deerhounds below ProCyte RI**	**% Deerhounds above ProCyte RI**
RBC (×10¹²/L)	7·54 ±0·78	7·56	6·43 to 8·94	5·65 to 8·87	0	6
Hb (g/L)	176 ±16·9	177	147 to 205	131 to 205	0	3
Hct (L/L)	53·0 ±5·3	53·3	43·6 to 62·0	37·3 to 61·7	0	5
MCV (fL)	70·4 ±2·7	70·4	66·0 to 74·5	61·6 to 73·5	0	10
MCH (pg)	23·4 ±0·8	23·5	22·0 to 24·8	21·2 to 25·9	0	0
MCHC (g/L)	332 ±7·7	331	324 to 349	320 to 379	3	0
RDW (%)	17·4 ±1·7	17·3	14·6 to 19·9	13·6 to 21·7	0	1
Ret (×10⁹/L)	25·7 ±21·9	17·0	7·39 to 77·4	10·0 to 110·0	15	0
Ret (%)	0·34 ±0·30	0·23	0·10 to 1·11	N/A	N/A	N/A
PLT (×10⁹/L)	144 ±66	145	36·9 to 270	148 to 484	53	0
MPV (fL)	10·9 ±0·8	11·0	9·3 to 12·5	8·7 to 13·2	0	0
WBC (x10⁹/L)	8·78 ±2·58	8·50	5·54 to 13·80	5·05 to 16·76	3	1
Neutr (×10⁹/L)	5·61 ±2·03	5·15	3·22 to 10·30	2·95 to 11·64	1	2
Lymph (×10⁹/L)	1·93 ±0·74	1·78	1·09 to 3·43	1·05 to 5·10	3	0
Mono (×10⁹/L)	0·56 ±0·22	0·54	0·27 to 0·92	0·16 to 1·12	0	2
Eos (×10⁹/L)	0·63 ±0·50	0·60	0·01 to 1·43	0·06 to 1·23	16	9
Baso (×10⁹/L)	0·04 ±0·05	0·02	0·00 to 0·13	0·00 to 0·10	0	9

Table 1. Deerhound-specific and non-breed-specific haematology reference intervals

RBC erythrocyte count, Hb haemoglobin concentration, Hct haematocrit, MCV mean corpuscular volume, MCH mean corpuscular haemoglobin, MCHC mean corpuscular haemoglobin concentration, RDW red cell distribution width, Ret reticulocyte, PLT platelet count, MPV mean platelet volume, WBC total leukocyte count, Neutr neutrophil count, Lymph lymphocyte count, Mono monocyte count, Eos eosinophil count, Baso basophil count

MPV had a Gaussian distribution. The other analytes were not normally distributed.

When the deerhound RIs are compared to the non-breed-specific RIs, the majority of the haematology results were similar to the generic intervals, although narrower, as expected when evaluating a subpopulation of patients with similar characteristics. The major exception is the interval for platelets, where both the upper and lower limits for the deerhounds were lower than the generic limits; >50% of the population in this study was below the non-breed-specific RI. Both the upper and lower limits of the RI were higher for MCV. The reticulocyte count RI was lower for both the lower and upper limits. The RI for eosinophils was wider than the generic interval for the instrument, with 15·6% of the population below the non-breed-specific lower limit and 9·4% of the population above the upper limit (Table 1).

Of the 96 samples, 51 blood smears were evaluated; 12 dogs had variable degrees of platelet clumping present. In addition, macroplatelets were noted in 6 dogs. Seventeen dogs had vacuolated (grey) eosinophils, 5 dogs had vacuolated eosinophil granules with a red-orange inclusion in the center, and 29 dogs had normal staining eosinophils (Fig 1). Manual differential cell counts were similar to those generated by the ProCyte Dx, with the exception of eosinophil counts in some dogs. In 29 dogs, eosinophil numbers were underestimated by the analyser. In 13 of these dogs, the values reported by the analyser were approximately half of those based on the manual count; 10 of these dogs had vacuolated (grey) eosinophils. However, in each case where the ProCyte Dx significantly under-recovered eosinophils, the digitally displayed dot plot of cellular events demonstrated an obvious misidentification of eosinophils, which indicated a manual differential was required. In each case, the digital events associated with the eosinophils were in close proximity to the digital events associated with the neutrophils and they were misclassified as neutrophils. Figure 1 shows representative dot plots and eosinophil granule morphology in three deerhounds.

Serum chemistry analysis

Because of the small sample volume in some of the dogs, the reference population for biochemical profiles included only 86 of the dogs. The population consisted of only 85 dogs for ALP and GGT and 81 dogs for CK activities after elimination of outliers. Only 77 dogs were evaluated for total T4 concentration because of insufficient sample volume.

Results for deerhounds (mean, median, and proposed RI for each parameter) are summarized in Table 2, along with non-breed-specific RIs used for dogs in general at The Ohio State University Veterinary Medical Center, and the percent of the deerhound population that were outside the non-breed-specific RI. Total T4, potassium, anion gap, bicarbonate, cholesterol, TP, and AG ratio had a Gaussian distribution. The rest of the analytes were not normally distributed.

For most of the serum chemistry parameters, the RI was similar for deerhounds and for dogs in general. The lower limit for T4 and calcium were lower than the generic lower limit; >60% of the Deerhounds were below the non-breed-specific RI for calcium. Both the upper and lower limits of the reference interval were higher for BUN, ALT, AST, ALP, and cholesterol in the deerhounds. The upper limit for potassium was above the generic limit. The glucose, chloride, GGT, and bilirubin RIs were lower for both the lower and upper limits. The RI for globulin was wider than the generic interval for the instrument, with 29·1% of the population below the non-breed-specific lower limit and 12·5% of the population above the upper limit (Table 2).

Six samples had moderate haemolysis. For the instrument used, moderate haemolysis may affect ALP, ALT, AST, CK, total bilirubin, potassium, and GGT. Mild haemolysis was detected in 11 samples; according to the manufacturer, this degree of haemolysis would have only minor influence on the analytes investigated. All of these samples were included as none were identified as outliers. No samples showed more than mild lipaemia.
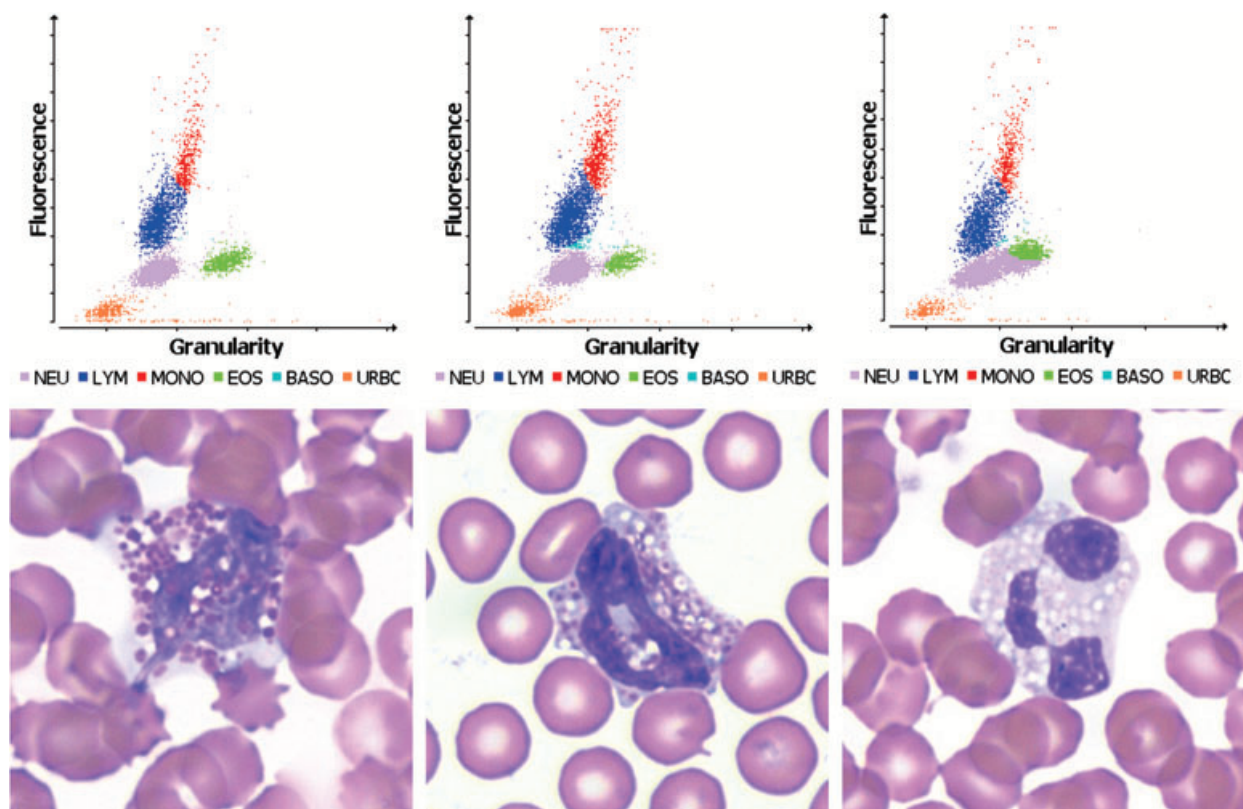
FIG 1. Representative dot plots and eosinophil granule morphology in three Deerhounds. The first dog had normal staining granules and similar eosinophil differential counts (7% on manual count, 4·9% on ProCyte Dx). The second had vacuolated eosinophils with an inclusion and also had similar differential counts (8% on manual count, 4·6% on ProCyte Dx). The last dog had grey eosinophils; the eosinophils were underestimated by the analyser (21% on manual count, 11·8% on ProCyte Dx) as evidenced by the dot plot of cellular events, which demonstrated a misidentification of some eosinophils and neutrophils. The eosinophil cloud is depicted in green, and the neutrophil cloud in lavender

Table 2. Deerhound-specific and non-breed-specific biochemical profile reference intervals

	Reference interval (RI)					
Analyte (units)	Deerhound Mean ±sd	Deerhound Median	Deerhound	OSU non-breed-Specific	% Deerhounds below normal RI	% Deerhounds above normal RI
T4 (nmol/L)	15·4 ±6·69	14·2	4·50 to 28·4	6·4 to 27·0	5	5
BUN (mmol/L)	6·21 ±1·45	6·07	4·05 to 8·80	1·8 to 7·1	0	19
Creatinine (µmol/L)	94·6 ±15·9	97·2	70·7 to 123·8	53 to 141	0	1
Phosphorous (mmol/L)	1·62 ±0·36	1·55	1·18 to 2·42	1·0 to 2·6	0	1
Ca (mmol/L)	2·16 ±0·40	2·1	1·56 to 2·79	2·3 to 2·9	63	0
Na (mmol/L)	150·1 ±3·01	149	147·4 to 158·3	143 to 153	0	7
K (mmol/L)	5·05 ±0·46	5·13	4·21 to 5·82	4·2 to 5·4	3	17
Cl (mmol/L)	110·3 ±2·98	110	106·1 to 116·0	109 to 120	27	2
Anion Gap (mmol/L)	22·4 ±2·03	22	19·0 to 25·0	15 to 25	0	3
Osmolality (mOsm/kg)	295·5 ±29·7	297	293·0 to 316·3	285 to 304	1	6
Bicarbonate (mmol/L)	22·4 ±1·47	22·3	20·14 to 25·13	16 to 25	0	4
ALT (U/L)	50·3 ±25·0	43·5	25·35 to 107·6	10 to 55	0	22
AST (U/L)	30·4 ±14·3	26·5	17·35 to 62·75	12 to 40	0	10
ALP (U/L)	72·6 ±47·7	60·0	30·0 to 198·2	15 to 120	1	10
GGT (U/L)	3·04 ±1·87	3·0	1·0 to 6·7	2 to 8	17	2
CK (U/L)	150·1 ±69·5	134	72·0 to 286·3	50 to 400	0	6
Cholesterol (mmol/L)	6·81 ±1·41	6·76	4·25 to 9·04	2·0 to 8·2	0	14
Bilirubin (µmol/L)	2·57 ±1·03	2·39	1·20 to 4·62	1·7 to 6·8	15	0
Total Protein (g/L)	60·8 ±5·16	61·0	52·4 to 72·0	51 to 71	1	4
Albumin (g/L)	37·1 ±2·80	37·0	31·4 to 41·0	29 to 42	0	0
Globulin (g/L)	23·7 ±4·78	23·0	17·4 to 32·7	22 to 29	29	13
AG Ratio	1·62 ±0·32	1·70	1·1 to 2·1	0·8 to 2·2	1	2
Glucose (mmol/L)	4·34 ±0·69	4·44	3·13 to 5·34	4·27 to 6·99	35	0

T4 thyroxine, BUN blood urea nitrogen, Ca total calcium, Na sodium, K potassium, Cl chloride, ALT alanine aminotransferase, AST aspartate aminotransferase, ALP alkaline phosphatase, GGT gamma glutamyl transferase, CK creatine kinase, AG Ratio albumin:globulin ratio

DISCUSSION

Although the sample sizes in this study did not meet the recommended numbers of individuals for establishing RIs (i.e. >120) (Clinical and Laboratory Standards Institute 2008, Geffre *et al.* 2009), these intervals can be used as a guide for interpreting the haematological and biochemical values in the deerhound. A larger population would be ideal; however, the deerhound is less common in the USA than in the UK, where it originated. It is estimated that there are approximately 1200 deerhounds in North America (J. Dillberger, personal communication, 2012); therefore, this study evaluated roughly 8% of the total population.

Parameters such as Hct, MCV, mean corpuscular (MCHC), lymphocyte count, reticulocyte count, TP, ALP, phosphorus and calcium can be affected by age. Two dogs in this study were approximately 10 months old at the time of sampling and four others were between 6 and 9 months of age. However, haematocrit, MCV, MCHC, lymphocyte and reticulocyte counts, TP, and phosphorous were within the RI for all six dogs. Four had calcium concentrations slightly below the reference interval and two had slightly elevated ALP activity, which may have a minor effect on the RIs proposed in this study.

The idiosyncrasies in the deerhound may be evolutionary. In the 15th century, these dogs closely resembled the English greyhound and the Irish wolfhound. Their ancestors were greyhound-type dogs known for hunting and referred to as Scotch greyhounds, rough greyhounds, Irish greyhounds and Highland deerhounds. These dogs were popular in the 16th and 17th centuries, but by the 19th century the deerhound's popularity had decreased because of changes in the style of hunting favoured. With much smaller numbers, the few remaining enthusiasts crossbred deerhounds with breeds such as the bloodhound and borzoi to improve size and vigour. It was not until later in the 19th century that the deerhound would become a recognized breed in both the UK and the USA (Barrett 1998). In addition, recent sequencing of the dog genome has allowed breeds to be organized into an evolutionary hierarchy with four primary groups. One of these groups, consisting of "herding" breeds, includes the deerhound, greyhound, Irish wolfhound and whippet (Wayne & Ostrander 2007). Because of the apparent shared ancestry of the deerhound and the greyhound, it is not surprising that these breeds may share some physiological and clinicopathological idiosyncrasies.

There were a large number of dogs in this study with low platelet counts. This has previously been reported in greyhounds, Galgos, whippets and several other sighthound breeds (Campora *et al.* 2011, Mesa-Sanchez *et al.* 2012, Uhrikova *et al.* in press, Zaldívar-López *et al.* 2011). Deerhound platelets may be "hyperaggregable", as in the greyhound (Zaldívar-López *et al.* 2011), resulting in platelet clumping after sample collection. Of the 51 deerhound blood smears, platelet clumping was noted in 12 dogs, making this a likely cause; although, platelet clumping correlated with low platelet numbers on the ProCyte Dx in only some of these dogs. Alternatively, one study reported a negative correlation between HCT and platelet count in greyhounds (Shiel *et al.*

2007a). While the majority of the deerhounds had Hct values within the non-breed-specific RI, approximately 70% of the dogs had an Hct >50 L/L; deerhounds had a mean Hct of 53 L/L, while greyhounds have been reported to have a slightly higher mean Hct (59 L/L in one study (Campora *et al.* 2011), and 56·2 L/L in another (Uhrikova *et al.* in press). The inverse correlation between Hct and platelet numbers may be explained by the bipotential stem cell theory: cells in the bone marrow differentiate into either red cell precursors or megakaryocytes (i.e. platelet precursors), and an increase in production of one of them would lead to a decrease in the other (McDonald & Sullivan 1993). Antibody coating of platelets is another potential cause for low platelet counts; however, one study found normal concentrations of platelet-bound immunoglobulin in greyhounds (Santoro *et al.* 2007).

The high MCV found in the deerhound (mean 70·4 fL) has also been reported in the greyhound (mean 74·5 fL) (Campora *et al.* 2011, Zaldívar-López *et al.* 2011). This may be because of a shorter erythrocyte life span and increased immature cells resulting from accelerated removal from circulation. Although the erythrocyte life span has not been measured in the deerhound, one study reported that it was significantly shorter in greyhounds (Novinger *et al.* 1996); however, the reticulocyte numbers in the present study were within the RI for the instrument (Table 1), thus making chronic regeneration an unlikely explanation for the high MCV in deerhounds. In addition, for parameters that measure the ratio between RBC and plasma (i.e. RBC count, Hb concentration and Hct), the mean for deerhounds appear to be in the upper end of the RI for dogs. However, the mean MCHC for the deerhound appears to be in the lower end of the RI for dogs. Taken together, these results suggest that deerhounds have a relatively greater circulating RBC mass than do dogs in general, partly because deerhounds tend to have a greater number of circulating RBCs and partly because those RBCs tend to be larger than in other breeds. However, this is offset by a tendency towards a slightly lower average Hb concentration in each deerhound RBC (MCHC). Alternatively, the increased MCV and decreased MCHC may be artefacts because of sample age, as it took up to 48 hours to process some of the samples. However, the consistency of these values suggest that this had only very minor affects on these results.

The wider reference interval for eosinophils is likely due to the presence of eosinophils with no visible staining of the cytoplasmic granules ("vacuolated" or "grey" eosinophils). Grey eosinophils were noted in 22 of the 51 deerhound blood smears. Grey eosinophils have also been reported in greyhounds, Italian greyhounds and whippets (Iazbik & Couto 2005, Giori *et al.* 2011, Zaldívar-López *et al.* 2011). These eosinophils are not associated with clinical disease and do not represent functional changes (Iazbik & Couto 2005, Giori *et al.* 2011). Proposed causes include alteration in basic proteins in the granules or a decrease in pH of the granules, resulting in less binding of the eosin stain (Iazbik & Couto 2005, Zaldívar-López *et al.* 2011). The clinical relevance of the grey eosinophils is the ability or lack thereof of automated analysers to correctly estimate eosinophil counts. One study found that two separate automated analysers significantly

underestimated eosinophil counts in dogs with grey eosinophils when compared to manual counts; however, the analysers were consistent with manual counts in dogs with normal staining eosinophils (Giori *et al*. 2011). Some of the deerhounds in this study had eosinophil counts that appeared to be underestimated by the analyser when compared to manual cell counts and this was more frequent in dogs with grey eosinophils; however, further studies on eosinophil morphology and its effect on automated analysers are needed to determine if this is clinically relevant. As previously stated, it should be noted that in each case where the ProCyte Dx significantly under-recovered eosinophils, the digitally displayed dot plot of cellular events demonstrated a misidentification of eosinophils. In each case, the digital events associated with the eosinophils were in close proximity to the digital events associated with the neutrophils and they were misclassified as neutrophils (Fig 1). The data collected in this particular study is being used by IDEXX Laboratories, Inc. in an attempt to develop an algorithm that will be able to accurately identify all eosinophils of the deerhound as well as other sighthound breeds.

The RI for total T4 in the deerhounds is consistent with reports of low total T4 concentration in sighthounds in several other studies, although the upper limit for the deerhound was slightly higher than the generic interval. Therefore, this low T4 concentration could be a characteristic shared by all sighthound breeds to some degree; however, it has not been associated with clinical signs of hypothyroidism or non-thyroidal illness in greyhounds, salukis, whippets, sloughis and Irish wolfhounds (Nachreiner & Refsal 1992, van Geffen *et al*. 2006, Shiel *et al*. 2007b, Panakova *et al*. 2008, Shiel *et al*. 2010). Low plasma T4 concentrations have also been reported in sled dogs used in endurance races such as the Iditarod (Lee *et al*. 2004), potentially implying that race training may be a cause. However, several studies on racing dogs suggest that training does not result in low T4 concentrations, although immediately after a sprint, T4 may be increased in greyhounds (Hill *et al*. 2001) and immediately after sled dogs finish an endurance race T4 was further decreased (Lee *et al*. 2004). Shiel *et al*. (2007b) reported that training had no influence on T4 concentration in greyhounds.

Alternatively, because of the fact that a subset of dogs had high cholesterol concentrations (Table 2), and because free T4 and canine thyroid stimulating hormone (TSH) were not evaluated, some of the deerhounds may have been truly hypothyroid. However, of the dogs with T4 concentrations below 6·4 nmol/L, none had cholesterol concentrations above the RI for the general dog population. Therefore, it is likely that low T4 concentrations are a physiological variation of sighthounds and should not be used alone to support a diagnosis of hypothyroidism. Misdiagnosis of hypothyroidism in sighthounds may be fairly common because of this variation. In one study on diagnosis of hypothyroidism in sighthounds, diagnosis was made on the basis of low total T4 or low total T3 alone in 70·1% of cases; however, only 16·3% of these dogs had additional abnormalities suggestive of hypothyroidism (Shiel *et al*. 2007a,b).

Although only 13·5% of the individuals had cholesterol concentrations above the RI for our laboratory, 60·9% were above 6·5 mmol/L. In comparison, the mean cholesterol concentration in greyhounds was 3·9 mmol/L (Zaldívar-López *et al*. 2011), which is lower than in deerhounds (6·81 mmol/L). As previously discussed, this may have been the result of hypothyroidism in some of the dogs. Because of the lack of concurrent abnormalities suggesting disease, idiopathic hypercholesterolaemia is a more likely cause and may reflect a breed-specific variation.

High ALT activity has been reported in several sighthound breeds (Uhrikova *et al*. in press, Zaldívar-López *et al*. 2011), and was especially high in Italian greyhounds in one study (Uhrikova *et al*. in press). One hypothesis in greyhounds is that high ALT activity is due to muscular dystrophy and necrosis because of large muscle mass (Zaldívar-López *et al*. 2011). However, the deerhound, similar to some of the other sighthounds, does not have the large muscle mass of the greyhound. The mean ALT in the deerhound (50·3 U/L) was lower than that of the greyhound (64·8 U/L) (Dunlop *et al*. 2011). This difference in values may be due to the use of a different instrument to analyse serum chemistry. Haemolysis and lipaemia may also artefactually increase ALT, but only one of the dogs with mild to moderate haemolysis had an increased ALT. Another possibility is liver disease; however, the deerhounds in this study and the sighthounds in other studies were considered clinically healthy on the basis of physical examination. Deerhounds also had high ALP and AST activities; however, these increases were of lesser magnitude than the increase in ALT. Mildly increased ALP activity has also been reported in the Pharaoh hound, but was considered clinically irrelevant (Uhrikova *et al*. in press).

Low total calcium concentration has also been reported in several sighthound breeds, including the greyhound; this has been linked to low phosphorus concentrations in some studies (Uhrikova *et al*. in press, Zaldívar-López *et al*. 2011). Potential causes are vitamin D deficiency, decreased dietary calcium intake, decreased renal reabsorption of calcium, or others. Yet, these are not likely to be present in the deerhound considering the phosphorous concentration was within the non-breed-specific reference intervals in 99% of the population, and the majority of the population in this study was below the non-breed-specific RI for calcium. Four dogs with low calcium concentrations were below 1 year of age; however, this was not considered a relevant cause of the decreased lower limit for the calcium RI because of the large percent of the adult population below the non-breed-specific RI. Hypoalbuminaemia can also result in decreased total calcium concentration, but none of the deerhounds had albumin concentrations outside the non-breed-specific reference interval. Therefore this is likely to be a breed-specific variation. Measuring ionized calcium in deerhounds may shed additional light on this issue.

In conclusion, there may be clinicopathological similarities shared by all sighthound breeds that are not found in other breeds. However, there also appears to be laboratory values that are unique to the deerhound, such as the high serum cholesterol and low total calcium concentrations. Because of these possible breed-specific variations, further studies are needed to justify applying greyhound and deerhound-specific RIs to other sighthound breeds. This is essential to accurately diagnose and manage medical conditions with laboratory testing in sighthounds.

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