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Evaluation of red cell distribution width in dogs with pulmonary hypertension

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Running Title: Red cell distribution width in dogs with pulmonary hypertension

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Abstract

Objectives: To compare red cell distribution width (RDW) between dogs with different causes of pulmonary hypertension (PH) and a control dog population to determine whether RDW was correlated with severity of PH as measured by echocardiography. A further aim was to determine the prognostic significance of increased RDW for dogs with PH.

Animals: Forty-four client-owned dogs with PH and 79 control dogs presented to a single tertiary referral institution.

Methods: Signalment, clinical pathological and echocardiographic data were obtained retrospectively from the medical records of dogs with PH, and RDW measured on a Cell-Dyn 3500 was compared between dogs with pre- and post-capillary PH and a control population. Referring veterinary surgeons were contacted for follow-up information and Kaplan-Meier analysis was conducted to investigate differences in survival time between affected dogs with different RDW values.

Results: The RDW was significantly greater in dogs with pre-capillary PH compared to control dogs. There was no difference in median survival times between dogs with PH divided according to RDW values. The RDW was positively correlated with mean corpuscular volume and haematocrit in dogs with PH, but did not correlate with echocardiographic variables.

Conclusions: An association was found between dogs with PH and increased RDW; however there was considerable overlap in values between control dogs and dogs with PH. The RDW was not associated with survival in this study.

Keywords: *Angiostrongylus vasorum*, canine, erythrocyte, tricuspid regurgitation, sildenafil

Abbreviations

cTnI	cardiac troponin I
IQR	inter-quartile range
LA:Ao	ratio of left atrial to aortic root diameter
NT-proBNP	N-terminal pro-brain natriuretic peptide
PH	pulmonary hypertension
PR	pulmonic regurgitation
PRPG	peak diastolic pulmonic regurgitant pressure gradient
RDW	red cell distribution width
ROC	receiver operator characteristic
TR	tricuspid regurgitation
TRPG	peak systolic tricuspid regurgitant pressure gradient

Introduction

Red cell distribution width (RDW) is a measure of degree of anisocytosis in the erythrocyte population and is expressed as the coefficient of variation of the erythrocyte size distribution data.[a] The RDW is regularly reported by modern haematology analysers.[1-3] A number of recent publications have identified RDW as an independent predictor of outcome in a wide range of different human diseases.[4-9] The ability of RDW to predict outcome independently in human patients with pre- and post-capillary pulmonary hypertension (PH) has been widely investigated and found to be clinically useful.[10-15]

Pulmonary hypertension is recognised in dogs with increased frequency due to growing access to Doppler echocardiography enabling non-invasive measurement of tricuspid and pulmonic insufficiency jet velocities, which are surrogate measures of systolic and diastolic pulmonary artery pressure gradients, respectively.[16] Pulmonary hypertension can be classified as pre-capillary or post-capillary, or a five point classification system can be used based on the pathological process underlying the PH. These five categories of PH include pulmonary arterial hypertension associated with parasite infestation or congenital systemic-to-pulmonary shunts, PH due to left-sided cardiac disease, PH related to diseases of the pulmonary parenchyma, PH resulting from thromboembolic events involving the pulmonary vasculature, and miscellaneous causes.[17,18]

A number of studies have explored the diagnostic utility of circulating biomarkers, including natriuretic peptides and cardiac troponin I (cTnI), in dogs with pre- and post-capillary PH.[19-22] To date, the relationship between PH and RDW in dogs has been described in a single study, which suggested that RDW was increased in dogs with pre-capillary PH when compared to controls.[b] There remains a paucity of information regarding the diagnostic and prognostic utility of RDW in dogs with PH due to a variety of causes.

The aims of the study were to determine if RDW differed between dogs with pre- and post-capillary PH and clinically normal dogs, to ascertain if RDW is associated with severity of PH determined by Doppler echocardiography, and to evaluate the prognostic value of a single measurement of RDW at time of presentation in dogs with PH.

Animals, materials and methods

Selection of cases and controls

The computerised medical record system of a tertiary referral hospital was searched to identify dogs that had a diagnosis of PH between February 2008 and February 2012. Cases were selected if a complete medical history and physical examination were available, routine diagnostic samples were submitted for complete haematology and serum biochemistry analysis at the same visit, and a full echocardiographic examination was also performed to confirm the presence of PH. Other diagnostic procedures were instituted based on the presenting signs and initial assessment. Cases were excluded if the haematocrit value was below the lower reference limit, if the complete medical record was not available for review, or if the dog had concurrent systemic disease including cancer, diabetes mellitus, portosystemic shunt or had undergone surgical procedures or blood transfusion within the previous 90 days.

A control group of dogs was also identified, that was comprised of blood donor dogs and clinical cases treated at the same institution over the same time period. All dogs underwent complete physical examination and had blood samples submitted for complete haematology and serum biochemistry analysis either as part of clinical investigations or prior to blood donation. Blood donors were included if they had not donated blood for at least 90 days prior to submission of blood samples, if there were no abnormalities on physical examination and no reported health problems, and if blood sample results were unremarkable.

Blood sample results were considered to be unremarkable if the values for all parameters were within the laboratory reference intervals, or, if outside these intervals, were judged to be clinically unimportant by two of the authors (DJC, AH). Clinical cases were included if their clinical signs were not associated with systemic inflammatory or infectious diseases and if their blood sample findings were unremarkable, as described above. These cases largely consisted of dogs presented for investigation of orthopaedic or ophthalmological problems. Clinical cases were excluded if they had undergone surgical procedures or blood transfusion prior to sampling or had clinical or historical evidence of haemorrhage within the previous 90 days.

Data collection

Data relating to signalment, clinical presentation, and blood sample results were extracted from the medical records of each case with PH. Referring veterinary surgeons were contacted by telephone to obtain follow-up data in January 2014 using a protocol that conformed to good research practice policy at the institution. The endpoint of the study was death, categorised as cardiorespiratory or non-cardiac. Cardiorespiratory causes included death or euthanasia due to clinical signs of congestive heart failure, worsening breathlessness, signs attributable to polycythaemia or sudden death unrelated to known systemic disease or trauma. Non-cardiac causes were death or euthanasia following clinical signs not attributable to cardiorespiratory disease (e.g. immune mediated disease, trauma, and neoplasia other than pulmonary neoplasia). The end of the study period was February 2014.

Red cell distribution width

The RDW was measured using a single haematological analyser[a] previously validated for canine haematology as part of a routine haematology analysis[c] as previously reported.[3]

Erythrocyte histograms, blood smears, or blood smear reports were reviewed where available, and individuals were excluded if histogram separation appeared poor or if platelet clumps were identified. Blood samples were processed within 24 hours of collection at the same site as the hospital, and serum biochemical analyses were conducted with the same analyser in all cases.[d]

Echocardiography

All patients with PH underwent complete echocardiographic examination[e] and all echocardiographic measurements were made off-line[f] by one observer (DJC). Standard two-dimensional views were obtained[23] and Doppler studies were also completed using a 2.5-5.5 or 5.5-7.5 MHz transducer. Dogs were positioned in left and right lateral recumbency with echocardiography performed on the dependent side. For Doppler evaluation, the right- or left-sided view that allowed for optimal alignment of the continuous wave Doppler interrogation beam through the regurgitant flow across the tricuspid valve and/or pulmonic valve was used to measure instantaneous peak systolic tricuspid regurgitation (TR) or peak diastolic pulmonic regurgitation (PR) velocities. Pulmonic stenosis was excluded by confirming normal valvular anatomy and mobility on two-dimensional echocardiography and identifying laminar pulmonic flow profile via pulsed-wave Doppler echocardiography with peak pulmonary artery flow velocities less than 1.5 m/s. Doppler flow interrogations of TR and PR jets provided estimates of systolic and diastolic pulmonary artery pressure respectively, allowing diagnosis of PH and estimation of severity. A peak systolic TR flow velocity $\geq$ 2.8 m/s or a peak diastolic PR flow velocity $\geq$ 2.2 m/s was considered compatible with PH.[16] The modified Bernoulli equation was applied to the peak systolic TR and peak diastolic PR flow velocity to estimate systolic and diastolic pulmonary artery pressure.

155 **Statistical analysis**

156 All statistical analyses were performed using a commercially available software package[g] and
157 an alpha value of 0.05 was used throughout. Variables were assessed for normality by visual
158 inspection of histograms and using Shapiro-Wilks tests. Parametrically and non-
159 parametrically distributed variables were compared using Student's t test or Mann-Whitney *U*
160 test, respectively. Categorical variables were compared using Fisher's exact test or Chi square
161 test. Data from parametric variables were presented as mean (± standard error), and those
162 from non-parametric data were presented as median (± inter-quartile range (IQR)). Pearson
163 correlation coefficients were calculated to determine whether RDW was correlated with age,
164 haematocrit, mean corpuscular volume (MCV), total white blood cell count, platelet count,
165 serum creatinine concentration, TR pressure gradient (TRPG), PR pressure gradient (PRPG)
166 or ratio of left atrial to aortic root diameter (LA:Ao) in dogs with PH.

167 For the purpose of further analysis, cases with PH were divided into those with post-
168 capillary hypertension (due to left-sided cardiac disease) or pre-capillary hypertension (due to
169 primary respiratory disease, idiopathic PH, or pulmonary vascular disease). Cases with PH
170 were also divided according to the severity of the PH. Dogs with a peak systolic TRPG
171 regurgitation pressure gradient of 31-50 mmHg were considered to have mild PH, dogs with
172 a gradient of 51-75 mmHg were considered to have moderate PH and dogs with a gradient
173 greater than 76 mmHg were considered to have severe PH.[20]

174 To determine whether RDW was a useful predictor of moderate or severe PH (TRPG
175 > 50 mmHg), a receiver operator characteristic (ROC) curve was constructed using RDW
176 values. The area under the curve was determined, and a suitable cut off value was chosen to
177 estimate sensitivity and specificity values.

178 Dogs with PH were divided into balanced terciles according to RDW values (lowest
179 to 15.7, 15.8 to 17.3 and 17.4 to highest). Average survival times were compared between

these groups using Kaplan-Meier product limit estimates and log-rank test. This analysis was also conducted separately for dogs with known cardiorespiratory death. Dogs that were still alive at the study end point were censored. Similar survival analyses were also conducted to compare dogs with pre- and post-capillary PH and according to the severity of PH.

Results

Study populations

Forty-four dogs were diagnosed with PH during the study period. The median age of affected dogs was 9.0 years (IQR, 4.3-11.4). The group was comprised of 18 intact males, 9 neutered males, 4 intact females and 13 neutered females. Twenty-five different breeds were represented, and the 5 most common were Jack Russell terrier (n=5), cross-breed (n=4), Border Collie (n=3), West Highland White terrier (n=3), and Yorkshire terrier (n=3).

The control group was composed of 79 dogs, of which 42 were blood donors and 37 were dogs presented for problem investigation. The median age of this group was 2.9 years (IQR, 2.0–6.0), which was significantly lower than the median age of the dogs with PH ($P < 0.001$). This group was composed of 18 intact males, 30 neutered males, 7 intact females and 24 neutered females. Twenty-six different breeds were represented, and the most common were cross-breeds (n=17), Labrador retrievers (n=13), Golden retrievers (n=7), German Shepherd dogs (n=6), and English springer spaniels (n=4).

Pulmonary hypertension

All of the dogs diagnosed with PH had systolic TR flow velocities > 2.8 m/s, and PR flow velocities were > 2.2 m/s in 24/44 dogs in which it was present. The median TRPG in dogs with PH was 71.0 mmHg (IQR, 51.1-98.7; range, 37.5-238.8). Eleven dogs had mild PH, 14

had moderate PH, and 19 had severe PH. Among the 24 dogs for which measurements were available, the median PRPG was 38.1 mmHg (IQR, 33.8-49.5; range, 14.4-63.4).

Twenty-eight dogs had pre-capillary PH caused by *Angiostrongylus vasorum* infestation (n=7), primary PH with no detected underlying cause (n=7), idiopathic pulmonary fibrosis (n=5), suspected or confirmed pulmonary thromboembolism (n=3), other parenchymal pulmonary disease (n=2), systemic-to-pulmonary shunt (n=4) including 2 dogs with right-to-left shunting patent ductus arteriosus. Sixteen dogs had post-capillary PH, caused by degenerative mitral valve disease (n=12), dilated cardiomyopathy (n=3), and aortic stenosis with systolic failure (n=1). The LA/Ao was significantly greater in dogs with post-capillary PH compared to dogs with pre-capillary PH, and TRPG, PRPG and total white blood cell count were all significantly greater in dogs with pre-capillary PH (Table 1).

Once the diagnosis was established, dogs with PH were treated with one or more of the following drugs at the discretion of the attending clinician: angiotensin converting enzyme inhibitor (n=44), pimobendan (n=24), furosemide (n=16), fenbendazole (n=10), sildenafil (n=7), spironolactone (n=7), and atenolol (n=1).

Red cell distribution width

The median RDW in control dogs was 15.7% (IQR, 15.0-16.4; range, 13.5-18.3), which was significantly lower than in dogs with pre-capillary PH (16.3%; IQR, 15.6-17.9; range, 12.7-19.6; $P = 0.008$) but not post-capillary PH (16.9; IQR, 15.0-17.6; range, 14.5-21.1; $P = 0.063$). There was no difference in RDW between dogs with pre- and post-capillary PH ($P = 0.714$). Distributions of RDW in each group are shown in Figure 1.

There was no difference in RDW between dogs with mild, moderate or severe PH (Fig. 2, $P = 0.384$). The area under the ROC curve constructed using RDW values for prediction of a TRPG value greater than 50 mmHg was 0.612 (95% confidence interval,

0.429–0.794) (Fig. 3). Using a cut-off value of 15.9%, RDW had a sensitivity of 60.6% and specificity of 45.5% for differentiation of mild from moderate or severe PH. The RDW was significantly correlated with mean corpuscular volume ($P < 0.001$) and haematocrit ($P = 0.045$) in dogs with PH but was not associated with LA:Ao, TRPG, PRPG, total white blood cell count, platelet count or serum creatinine concentration (Table 2).

Survival times

There was no significant difference in median survival times between dogs with PH divided into terciles according to RDW values ($P = 0.071$), though there was a trend for greater mortality with higher RDW values (Fig. 4A). Survival curves for dogs with PH stratified according to cause of PH and severity of peak systolic TR are shown in Figures 4B and 4C, respectively. There was no difference in survival between these groups ($P = 0.948$ and $P = 0.622$, respectively).

Discussion

This study demonstrates that RDW differs significantly between dogs with pre-capillary PH and control dogs, but not between control dogs and those with post-capillary PH. There was no difference in survival between dogs grouped according to RDW values, and RDW was not a useful predictor of the severity of PH. The RDW was correlated with MCV and haematocrit in dogs with PH.

The findings presented here are in agreement with retrospective and prospective studies in human patients, which indicate that RDW can provide additional diagnostic information in patients with PH.[24] In a recent study in dogs, RDW was significantly increased with pre-capillary PH compared to controls, but not in dogs with post-capillary PH,[b] similar to the findings of this study. There was no difference in RDW between healthy dogs and

254 those with either compensated or decompensated heart failure due to degenerative mitral
255 valve disease.[25]

256 The reasons for alteration in RDW in patients with PH remain unclear. Because RDW
257 is an indicator of anisocytosis, it varies with iron deficiency anaemia, and iron status appears
258 to be essential in the regulation of pulmonary vascular tone.[26,27] In human patients, iron
259 deficiency has been identified in idiopathic PH.[28,29] In heart failure patients with post-
260 capillary PH, RDW is thought to be affected by chronic inflammation, renal dysfunction,
261 altered erythropoiesis, and oxidative and nutritional stress.[9,14,30] It is therefore likely that
262 RDW is influenced by numerous factors involved in the pathogenesis of PH.
263 *Angiostrongylus vasorum* infestation is known to cause variable effects on erythroid
264 parameters,[31,32] though, to the authors' knowledge, RDW values have not been assessed in
265 infected dogs.

266 Significant correlations between RDW and serum urea and creatinine concentration
267 were previously reported in dogs with PH.[b] No such correlation was detected in the present
268 study, but the relationship between RDW and serum urea and creatinine concentration in
269 human patients with PH is contradictory, with one study showing a significant correlation
270 with serum urea and a strong trend towards significance with serum creatinine[15] whereas in
271 other studies no such correlation was identified.[13,14] Interestingly, in contrast to studies in
272 human patients[10,12] but in line with the canine study by Poser,[b] we found no significant
273 correlation between RDW and peak TR or PR jet velocity, which may reflect differences in
274 sample size between canine studies and the larger investigations in human patients. To date,
275 the only circulating biomarker shown to be associated with peak TR gradient in dogs with PH
276 is N-terminal pro-brain natriuretic peptide (NT-proBNP).[20] The RDW was found to be
277 significantly correlated with haematocrit and mean corpuscular volume in dogs with PH, as
278 was also found in dogs with degenerative valve disease.[25]

The RDW has been shown to predict outcome in human patients with pre- and post-capillary PH in numerous studies.[10-15] In the present study however, RDW did not provide clinically useful prognostic information in dogs with PH. This result may reflect the small sample size of affected dogs, particularly as there was a trend towards increased mortality with higher RDW values as observed in people,[15] or the influence of treatments such as furosemide, sildenafil and pimobendan instituted following diagnosis of PH. Studies in humans indicate incremental increases in serial RDW measurements are associated with decreased survival in heart failure[33,34] therefore serial RDW measurements in dogs may be an area of future study.

A number of publications have explored the diagnostic utility of circulating biomarkers including natriuretic peptides and cardiac troponin I (cTnI) in dogs with pre- and post-capillary PH.[19-22] One report indicated that measurement of circulating NT-proBNP but not N-terminal pro-atrial natriuretic peptide (NT-proANP) or cTnI was able to stratify dogs with pre-capillary PH into mild, moderate and severe.[20] In an experimental canine model of pre-capillary PH, circulating NT-proANP and NT-proBNP could identify dogs with moderate and severe but not mild PH.[19] In the present study, a trend was identified with increasing RDW values in dogs with moderate and severe PH, but this did not reach significance which may reflect the relatively small number of cases in each group.

In a study of 162 human patients of which 62% had pulmonary arterial hypertension, RDW was found to be independently associated with death in patients with severe PH and performed better as a prognostic indicator than NT-proBNP.[15] The RDW was also shown to add significant prognostic value to measurements of NT-proBNP in human patients with idiopathic pulmonary hypertension.[14] Unfortunately, in the retrospective report presented here, NT-proBNP data was not available and it was not possible to compare the ability of the two biomarkers to identify dogs with PH and predict survival.

The present study is the first to indicate that RDW, a simple parameter routinely measured as part of a haematological profile, may be useful in the diagnosis of dogs with PH with a number of different aetiologies. The sensitivity and specificity of RDW in the diagnosis of PH are based on the performance of the parameter in the whole study sample. It is clear for our results that some overlap in RDW values occurs between the control population and dogs with pre-capillary PH, which suggests that RDW may not be a useful parameter for determining the disease status of single individuals. The performance of a screening test depends not only on its intrinsic ability to identify affected individuals but also on the prevalence of disease within the population. In this retrospective study, populations were preselected on the basis of the presence of confirmed PH or control dogs representing groups with 100% or 0% disease prevalence, respectively. Under these circumstances, a screening test is likely to exhibit superior performance than would the same test used in a more heterogeneous population encountered in the clinic. Furthermore, RDW may be increased due to other pathological processes, so this indicator may also lack specificity in clinical settings. Further studies will be required to determine whether RDW values may contribute useful prognostic information, as they do in humans.

This study has a number of limitations as a result of its retrospective nature. Due to the strict inclusion criteria, the study population of affected animals was small which reduces statistical power. The study was not designed to assess the influence of different treatments for dog with pre- and post-capillary PH on survival or RDW. Affected dogs with PH were significantly older than the control population. An age related increase in red blood cell membrane stability and RDW has been reported in women[35] but no information is available for dogs. The RDW was assessed as a single prognostic factor in the present study as the small sample size and low event (mortality) rate within the study period precluded more sophisticated analysis, such as Cox proportional hazards regression. Future studies of a larger

size will be required to establish whether the prognostic value of this parameter is altered after other variables have been accounted for. The retrospective nature of the study meant that patient and control RDW values were obtained from a Cell-Dyn 3500, although the majority of the human literature refers to more recent haematology analyser models. Prospective studies using newer generation analysers may provide better discrimination for RDW value prognostication.

The results of this study show that RDW differed significantly between dogs with pre-capillary PH and control dogs, but RDW was not a significant prognostic marker in dogs with PH. Further studies will be required to determine whether RDW has prognostic significance in larger groups of dogs with PH, or in dogs with other forms of cardiorespiratory disease.

Footnotes

[a] Cell-Dyn 3500 System Operators Manual, Abbott Laboratories, Abbott Park, Illinois, USA..

[b] Poser H, Mazzotta E, Menciotti G, Contiero B, Baron Toaldo M, Guglielmini C. Red blood cell distribution width in dogs with pre-capillary and post-capillary pulmonary hypertension. Poster presented at the 23rd ECVIM-CA Congress, Liverpool, UK, September 2013.

[c] RVC Diagnostic Laboratories, Royal Veterinary College, London, UK.

[d] ILab600 Instrumentation Laboratory, Werfen Life Group, Barcelona, Spain.

[e] GE Vivid 7, GE Healthcare, Hatfield, UK

[f] GE EchoPAC PC, GE Healthcare, Hatfield, UK.

[g] IBM SPSS Statistics for Windows, Version 20.0, Released 2011, IBM Corp. Armonk, NY, USA.

Conflict of Interest: The authors declare no conflict of interest.

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Figure Legends

Figure 1: Red cell distribution width values in control dogs and dogs with pre-capillary or post-capillary pulmonary hypertension. Lines represent the median for each group.

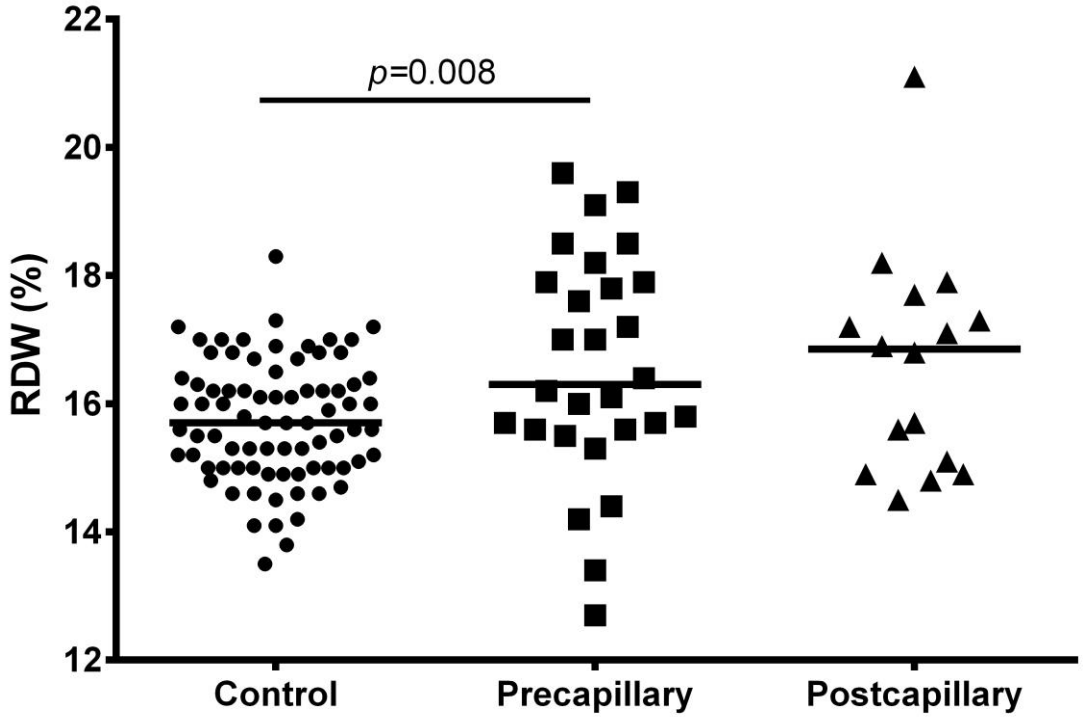

Figure 2: Red cell distribution width values in control dogs and dogs with mild, moderate and severe pulmonary hypertension defined by peak systolic tricuspid regurgitant pressure gradient. Lines represent the median for each group.

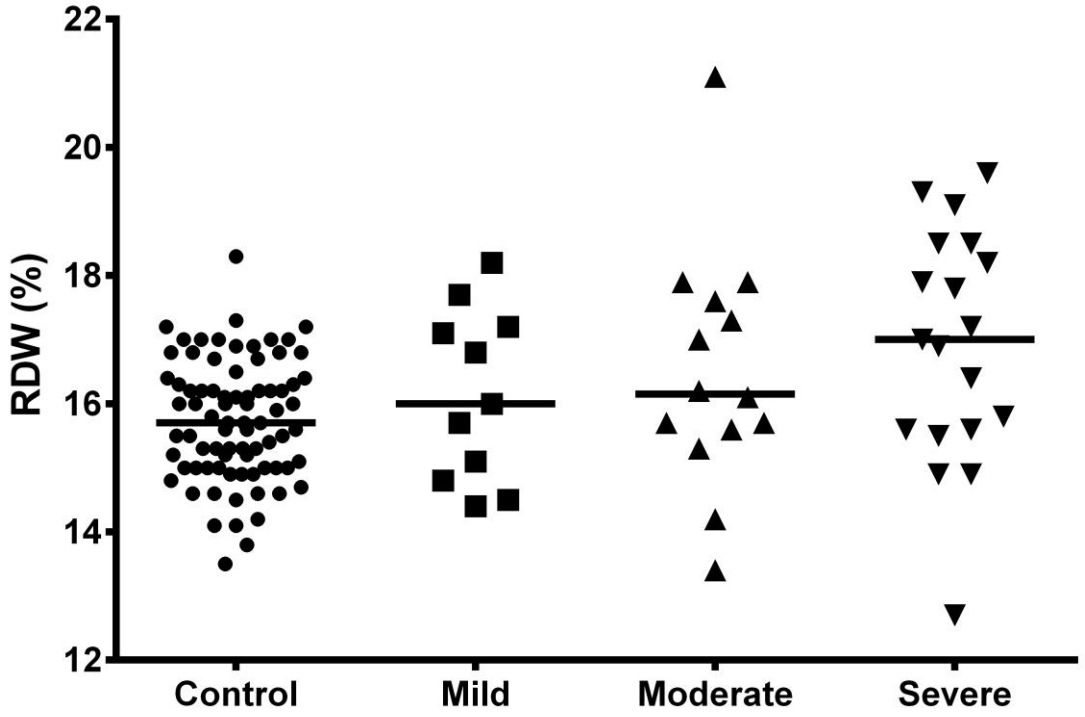

Figure 3: Receiver operator characteristic curve constructed to determine whether red cell distribution width values can predict mild (peak systolic tricuspid regurgitant pressure gradient < 50 mmHg) or moderate to severe (peak systolic tricuspid regurgitant pressure gradient ≥ 50 mmHg) pulmonary hypertension.

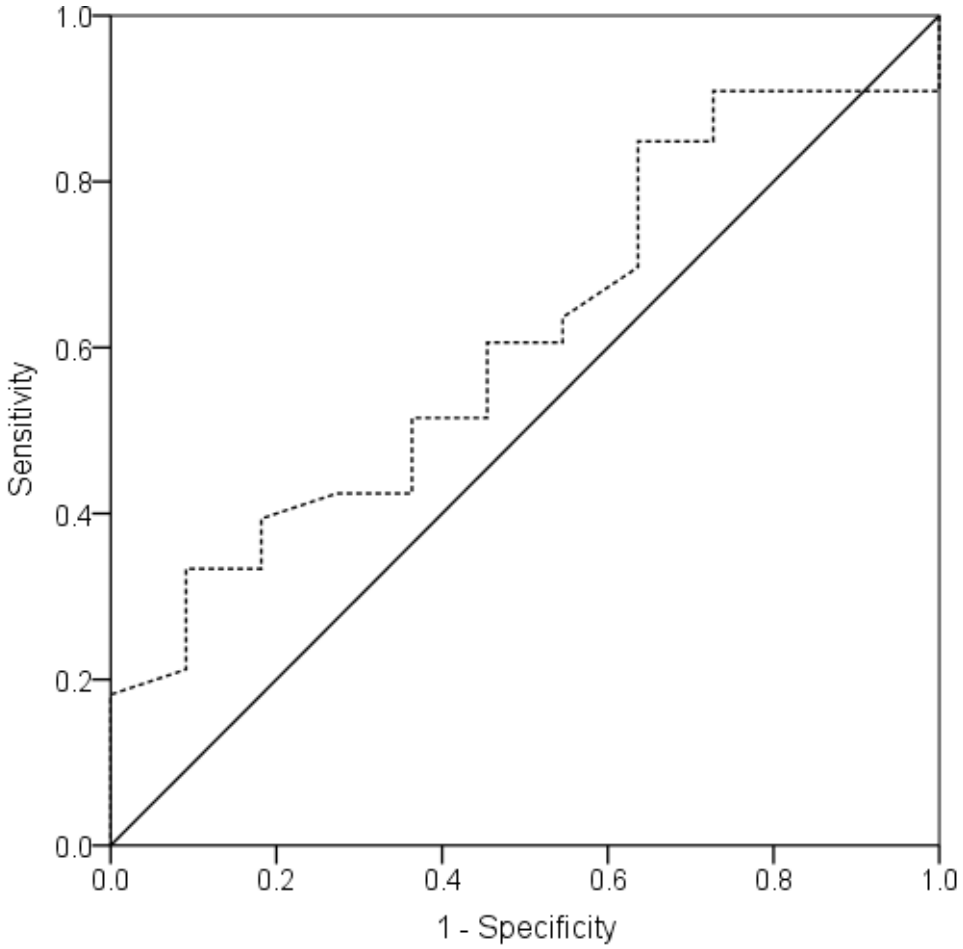

Figure 4: Kaplan-Meier survival curve in dogs with (A) pulmonary hypertension divided into terciles according to red cell distribution width values: lowest value to 15.7 (solid line), 15.8 to17.3 (dotted line), 17.4 to highest (dashed line), (B) pre-capillary pulmonary hypertension (dotted line) or post-capillary pulmonary hypertension (solid line), and (C) mild (solid line), moderate (dotted line) and severe pulmonary hypertension (dashed line). Tick marks indicate censored cases.

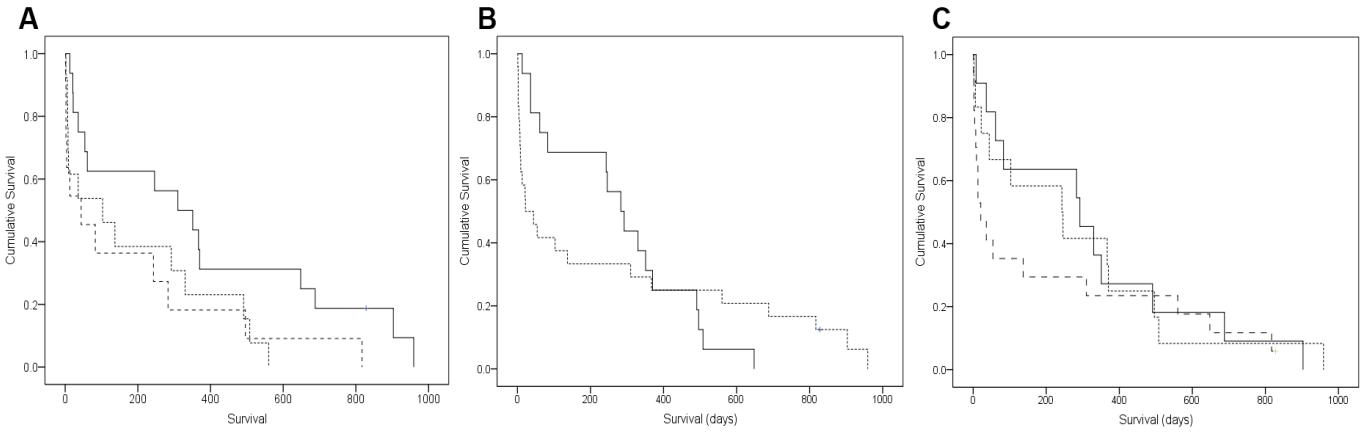

Table 1: Echocardiographic, haematological and biochemical variables in dogs with pre-capillary and post-capillary pulmonary hypertension and control dogs. P values represent comparison between dogs with pre-capillary and post-capillary pulmonary hypertension.

Parameter	Pre-capillary PH[a] (median, IQR[b])	Post-capillary PH[a] (median, IQR[b])	*P* value	Control
LA:Ao[c] ratio	1.33 (1.13-1.55)	2.21 (1.16-1.54)	<0.001	
TRPG[d] (mmHg)	80.4 (56.3-108.8)	52.5 (43.2-70.8)	0.001	
PRPG[e] (mmHg)	40.1 (34.9-52.1)	33.9 (22.3-38.5)	0.040	
RDW[f] (%)	16.3 (15.6-17.9)	16.9 (15.0-17.6)	0.714	15.6 (15.0-16.2)
Haematocrit (%)	43.7 (39.0–49.5)	47.6 (43.7–50.9)	0.205	46.8 (44.5-50.0)
MCV[g] (µl)	69.1 (66.8–71.1)	70.7 (68.7–71.7)	0.317	71.5 (70.1-73.8)
WBCC[h] (x10^9/l)	16.2 (11.2-19.8)	10.6 (8.3-12.8)	0.010	9.3 (8.2-10.4)
Platelet count (x10^9/l)	302 (187-377)	290 (227-519)	0.317	229 (193-260)
Serum creatinine concentration (µmol/l)	93.0 (81.0-120.5)	108.5 (90.8-120.0)	0.432	104.0 (95.0-119.0)

PH, pulmonary hypertension; IQR, inter-quartile range; LA/Ao, ratio of left atrial to aortic root diameter; TRPG, peak systolic tricuspid regurgitation pressure gradient; PRPG, peak diastolic pulmonic regurgitation pressure gradient; RDW, red cell distribution width; MCV, mean corpuscular volume; WBCC, total white blood cell count.

510 Table 2: Results of correlations between red cell distribution width and clinical pathological

511 variables in dogs with pulmonary hypertension.

Variable	Correlation coefficient	P value
Age (years)	-0.249	0.103
LA:Ao[a]	0.054	0.777
TRPG[b]	0.113	0.465
PRPG[c]	-0.020	0.927
Haematocrit	0.304	0.045
MCV[d]	-0.664	<0.001
WBCC[e]	-0.066	0.668
Platelet count	-0.268	0.079
Serum creatinine concentration	0.147	0.385

512 LA/Ao, ratio of left atrial to aortic root diameter; TRPG, peak systolic tricuspid regurgitation

513 pressure gradient; PRPG, peak diastolic pulmonic regurgitation pressure gradient; RDW, red

514 cell distribution width; MCV, mean corpuscular volume; WBCC, total white blood cell

515 count.