American Journal of Animal and Veterinary Sciences

Original Research Paper

Echocardiographic Pulmonary to Left Atrial Ratio in Dogs (ePLAR): A Differential Marker of Pre- and Postcapillary Pulmonary Hypertension

[1,3]Dmitrij Oleynikov and [2]Ma Yi

[1]Research Laboratry of Bioprothesis and Cardioprotection, Almazov National Medical Research Center, IEM, Russia
[2]Laboratory of Transfusion and Efferent Therapy, Nacional'nyj Medicinskij Issledovatel'skij Centr Imeni V A Almazova, Russia
[3]Veterinary clinic "Belij Klyk", Moscow, Russian Federation, Russia

Article history
Received: 11-12-2021
Revised: 20-01-2022
Accepted: 31-01-2022

Corresponding Author:
Dmitrij Oleynikov
Research Laboratry of Bioprothesis and Cardioprotection, Almazov National Medical Research Center, IEM, Russia
Veterinary clinic "Belij Klyk", Moscow, Russian Federation, Russia
Email: wolfberg.guard@gmail.com

Abstract: Pulmonary Hypertension (PH) in dogs is a complicated syndrome that could be primary, due to idiopathic or genetic causes, or secondary due to pulmonary disease, pulmonary thromboembolism, heartworm disease, heart failure. Due to the inability of the routine use of right heart catheterization in veterinary patients, there is a lack of differential criteria between PH forms. This study was performed to verify ePLAR as a differential marker in PH forms. Analyze wide specter of echocardiography-derived markers and novel ePLAR-marker to find efficient parameters in PH differentiation and ePLAR accuracy. We studied 59 dogs of different sex, age and breed. Groups were formed according to a primary pathology: Healthy dogs (HD, n = 8); dogs with MMVD and postcapillary PH (PostPH, n = 23); dogs with MMVD and precapillary PH (PrePH, n = 28). Animals in the study were diagnosed with the primary disease by standard echocardiographic methods and algorithms. In Post PH, LA was significantly larger than in the PrePH and the control. Left vertical walls were thicker in the PrePH than in the PostPH. Left ventricle diameter was higher in the PostPH than in the control and the Pre-PH. PV was smaller in the PrePH than in the Post PH and the control ($p<0.001$ and $p<0.021$). PV/RPA in the PrePH was lower than in the control and in the PostPH ($p<0.001$). AT was lower in the PrePH and the PostPH. AT/ET ratio was higher in the control to both experimental. AT/ET was lower in the PrePH to the PostPH. RV was dilated both in the PrePH and the PostPH to control. RV wall thickness was increased in the PrePH in comparison to both control and PostPH. Significant reductionin E-wave velocity for both the PrePH and PostPH to control and PostPH; reduction in A-wave; decreased E/A. TR velocity was higher in both experimental to control, but didn't differ between each other. ePLAR increased in the PrePH and PostPH to control. In this study we found several echocardiographic parameters to differentiate Pre-and Post-PH forms: LA, LA/Ao, LV diameter, LV/RPA, IVS thickness and a novel in veterinary studies index - EPLAR.

Keywords: Canine, Pulmonary Hypertension, Echocardiography

Introduction

Pulmonary Hypertension (PH) in dogs is a complicated syndrome that could be primary, due to idiopathic or genetic causes, or secondary due to pulmonary disease, pulmonary thromboembolism, heartworm disease, heart failure. As a consequence, PH leads to right ventricular impairment and right heart failure (Galiè *et al*., 2016).

Usually, PH is subdivided into 2 groups, depending on the most affected zones of pulmonary microcirculation: Precapillary (small or resistive arterioles are altered) or postcapillary (venules are mostly affected). Also, a reactive or mixed form could develop, but it is usually a consequence of the pre-existed form of PH and additional trigger mechanisms (like acute pulmonary edema) (Reinero *et al*., 2020).

Echocardiography nowadays is the most effective and available method for the non-invasive characterization of heart structures and functional parameters in veterinary medicine.

Left ventricular function and morphology are well-studied and there are established methods to estimate left heart performance. Unfortunately, the right heart has fewer complex studies, mostly due to its specific geometry and contractile movements. Its triangular form and differentiation of outflow and inflow tracts make it difficult to estimate longitudinal and circumflex contractile properties, which are closely connected with Right Ventricle (RV)-to-Pulmonary Artery (PA) coupling. RV-to-PA coupling is one of the main markers for PH severity (Thenappan *et al*., 2016). It reflects conduit and reservoir function of the proximal pulmonary vessel, restiveness of distal part of the pulmonary circulation and is affected by the transpulmonary pressure gradient (Stenmark *et al*., 2016).

In recent ACVIM consensus in PH, we have a short list of methods to diagnose PH. Despite elucidating the fact of precapillary and postcapillary PH presence, there were limited data about the differentiation between these forms. Most of the recommendations are based on the previous history of left heart failure or right heart chambers catheterization (Reinero *et al*., 2020).

In human medicine, several studies present an echocardiography-associated method of transpulmonary gradient estimation to differentiate between forms of the PH. This method is based on the ratio of Pulmonary Arterial Pressure (PAP) and Left Atrial Pressure (LAP). PAP - was calculated from tricuspid regurgitation speed via modified Bernoulli equation and right atrial pressure. LAP - was obtained as a result of the transmitral E-wave to tissue Doppler e'-wave on mitral valve annulus. This equation could be a surrogate for transpulmonary gradient assessed by echocardiography.

In this hypothesis, we assumed the fact, that in precapillary PH, PAP will rise without LAP increase and vice versa. This study aims at elucidating the perspective of PH differentiation with calculated transpulmonary gradient (ePLAR) by echocardiography in dogs presented to the ICU with acute respiratory distress suspected of heart failure origin.

Methods

In this pilot study, we included dogs of different sexes, predominantly of age above 10 years old, with a history of pre-existent Myxomatous Mitral Valve Disease (MMVD) on stage C of ACVIM classification. The dogs were treated with standard therapy (Pimobendan, Furosemide, Spironolactone +/-ACEi).

All studied dogs were divided into three groups: A control group of dogs with MMVD but without signs of PH (n = 8); dogs with MMVD and signs of precapillary PH (n = 28); dogs with signs of postcapillary PH and MMVD (n = 23).

Inclusion Criteria

All the included dogs were taken to the intensive care unit with signs of acute respiratory distress while being on standard MMVD stage C medication. At the moment of examination, all the dogs had endocardial murmurs, harsh lung sounds, crackles.

The Control group included dogs with diagnoses of MMVD on stage C. They were treated with standard protocol and did not have signs of respiratory insufficiency, previous pulmonary disease, pulmonary hypertension and most of them were presented before routine oral cavity sanitation. In parallel, echocardiography and radiology (roentgenograms of the thoracic cavity in two projections) studies were performed to exclude subclinical forms of diseases.

The dogs that appeared in the ICU had a radiology examination to characterize the insensitivity of pulmonary edema. Then, they were placed in the oxygen camera and were treated in the standard way until clinical stabilization. After that, they underwent an echocardiographic study to estimate their heat performance.

To differentiate Precapillary (PrePH) and Postcapillary PH (PostPH), we used phenotypic markers. We assumed that all presented dogs previously had the phenotype of Left-Sided Heart Failure (LSHF): Left chambers dilation, pulmonary veins distention, preserved systolic function, diastolic dysfunction above class 2, absence of right chamber's dilatation and low velocity of tricuspid regurgitation. In cases with preserved phenotypic markers of LSHF and addition of new-onset PH findings (tricuspid regurgitation above 2,7 m/s, presence of pulmonary artery regurgitation, decreased right pulmonary artery distensibility index, right ventricle wall hypertrophy and right chambers dilation) -assumed as patients with PostPH. Meanwhile, dogs with signs of PH onset (as mentioned above), but with loss of LSHF signs were marked as PrePH. This subjective method is mentioned to find out some features closely connected with different forms of PH from the blank.

Exclusive Criteria

We excluded dogs with pre-existing PH and its therapy, congenital defects, arrhythmia, lung disease, systemic diseases affecting blood flow (arterial hypertension, Cushing's disease, pulmonary artery thromboembolism, dirofilariasis, sepsis, electrolyte abnormalities, etc.) and without previous history of MMVD.

Measurements

Echocardiography was performed in standard methods, with an estimation of chamber diameter, systolic and diastolic function of the left ventricle. Left atria and

right atria diameter were studied in long parasternal 4 chambers-view axis. Right ventricle diameter and right ventricle free wall thickness were studied in the long parasternal 4 chambers-view axis. Additional criteria were: Subjective characterization of dilated/non-dilated chambers and right wall hypertrophy presence. The longitudinal systolic function of the right heart was studied by TPASE. Systolic and diastolic diameter of the left ventricle, wall thickness and shortening fraction were studied in M-mode by the standard methods.

The velocity measurements were performed in standard methods with PW-and CW-Doppler. The pulmonary flow was studied on the right short axis: We studied anterograde flow, its phenotype, the ratio between acceleration time and effusion time and regurgitation velocity. In this projection, the ratio between PA and aorta were subjectively estimated. In the context of associated changes of the right ventricle the velocity of tricuspid regurgitation above 2,7 m/s was assumed as a PH marker. Velocity less than 2,2 m/s was declared as non-significant. The transmitral flow was studied in context of mitral regurgitation, a diastolic function of the left ventricle (velocity and ratio between E-wave and A-wave calculation). Additionally, tissue Doppler measurements were performed: S'-wave on the tricuspid valve; e'-wave on the interventricular septum and its ratio with transmitral E-wave. In parallel, we performed subjective characterization of right pulmonary artery distensibility.

Surrogate transpulmonary gradient index (ePLAR) was measured by the ratio between PAP (tricuspid regurgitation velocity as a source for modified Bernoulli equation) and LAP (transmitral E-wave to tissue Doppler e'-wave on interventricular septum ratio)

Statistics

Statistical analysis was performed by SPSS version 23.0 software (IBM Corporation, Armonk, NY) and GraphPad Prism 8.00 (GraphPad Software Inc., La Jolla, CA, USA). The comparison of group characteristics, echocardiographic indices were performed by the nonparametric Mann-Whitney test. The significance of differences in categorical variables was calculated by Fisher's exact test. All P values were 2 sided and $P<0.05$ was considered statistically significant. The correlation between variables was evaluated by Spearman's rank correlation coefficient analysis.

Results

This study included 59 dogs of a senior (9-11 years old) age, different sex and breed, predominantly of toy and small breeds.

Statistically significant weight difference was not observed ($p>0.05$, for all groups). Average heart rhythm was more rapid in group with PostPH, but did not statistically differentiate from other groups ($p>0.05$, for all groups, Table 1).

Statistical analysis elucidated significant difference in enlargement degree between PostPH and PrePH groups (Table 2) and between PostPH and control groups ($p<0.01$ and $p<0.04$). But at the same time, LA dimeter (Fig. 1) did not statistically differ between the PrePH and control group ($p>0.05$). LA to Ao ration presented the same pattern: Difference between the control group and PrePH was not statistically significant ($p>0.05$); The PostPH (group) had significantly higher ration to both PrePH and control group ($p<0.01$, for both).

There was a statistically large difference in left ventricle walls thickness between control and both PH groups ($p>0.05$). The left ventricle walls were statistically thicker in group PrePH, than in PostPH group, for both walls ($p<0.026$ and $p<0.005$; Fig. 2).

Left ventricle diastolic diameter normalized to weight (Table 2; Fig. 3) was statistically smaller in PrePH group in comparison to the control and PostPH groups ($p<0.024$ and $p<0.001$). Left ventricle diastolic diameter normalized to weight was statistically higher in PostPH group, than in the control ($p<0.001$). This pattern was preserved in left ventricle systolic diameter: LVIDs was smaller in the PrePH, than both in the control and PostPH groups ($p<0.005$, и $p<0.001$), But LVIDs did not statistically differ from the control group ($p>0.05$).

Aortic dimeter did not differ in all groups ($p>0.05$). Main pulmonary artery diameter did not statistically differ between the control and PostPH groups and between PostPH and PrePH groups ($p>0.05$). However, we found significant difference between the control and PrePH dogs: In PrePH, it was distended ($p<0.036$). Subjective estimation of the main pulmonary artery diameter showed presence of its dilatation in 100% of cases in PrePH group (28/28) and 60,6% in PostPH group (16/23).

Pulmonary vein diameter (Table 2; Fig. 4), measured from right parasternal axis view, was statistically smaller in PrePH group in comparison both to the control ($p<0.021$) and PostPH groups ($p<0.001$). In absolute units' pulmonary vein was almost twice smaller than in PostPH group. Pulmonary vein diameter was statistically smaller in the control group than in PostPH ($p<0.013$). Right pulmonary artery diameter was statistically larger than in the control group ($p<0.001$) and PostPH group ($p<0.005$). Additionally, there were no statistical difference in right pulmonary artery diameter between the control and PostPH groups ($p>0.05$). In subjective analysis of the right pulmonary artery distensibility, we used three degrees: Normal, weak and absent. In the control group, all the dogs had normal distensibility (8/8). In PostPH group, we found normal distensibility in 43,5% cases (10/23), weak in 39,1% (9/23) and was absent in 17,4% (4/23). In the PrePH, we found normal distensibility in 3,6% (1/28), weak – 39,3% (11/28) and absent – 57,1% (16/28).

The ratio of pulmonary vein to right pulmonary artery (Table 2; Fig. 4) in PrePH was significantly lower than in the control group ($p<0.001$) and in PostPH ($p<0.001$). Meanwhile, there was no difference in this parameter between the control and PostPH groups ($p>0.05$).

In pulmonary artery flow dopplerography, we estimated flow Acceleration Time (AT), Effusion Time (ET), their ratio and pulmonary regurgitation (Table 2; Fig. 5). AT was statistically lower in both experimental (PrePH and PostPH) groups ($p<0.005$ and $p<0.001$, to the control), but did not differ between each other($p>0.05$). ET did not show any difference in the control and PrePH groups ($p>0.05$). The PostPH characterized with decreasing of ET in comparison to the PrePH ($p<0.014$) and the control ($p<0.005$). AT/ET ration was higher in control group than both experimental groups ($p<0.003$ for PrePH and $p<0.043$ for PostPH). AT/ET ration was statistically lower in the PrePH in comparison to the PostPH ($p<0.005$). Despite the fact that medial velocity of pulmonary regurgitation in PrePH was higher, it did not statistically differ from PostPH.

Morphological evaluation of pulmonary flow showed that alterations in pulmonary flow were found in 89,3% (25/28) among PrePH dogs and in 60.9% (14/23) of PostPH dogs. Alterations were absent in the control group.

Right ventricle was significantly dilated both in the PrePH ($p<0.021$) and PostPH ($p<0.009$) in comparison to the control group. But intergroup comparison did not show significant differences. Right atrium was significantly dilated both in PrePH and PostPH ($p<0.001$, for both). But intergroup comparison did not show significant differences ($p>0.05$).

Right ventricle wall thickness (Table 2; Fig. 6) was significantly increased in the PrePH in contrast to both the control ($p<0.001$) and PostPH ($p<0.012$) groups. Additionally, PostPH right ventricle wall was statistically thicker than in the control group ($p<0.005$).

Right ventricle longitudinal systolic function was estimated by TAPSE. There was no significant difference between the control and PrePH, PrePH and PostPH ($p>0.05$, for both). But TAPSE was increased in PostPH group compared to the control ($p<0.04$).

The transmitral flow (Table 2; Fig. 7) showed statistically significant reduction in E-wave velocity for the PrePH both to the control and PostPH groups ($p<0.001$, for both); reduction in A-wave ($p<0.018$, both for the control and PostPH); decreased E/A ration ($p<0.001$, to PostPH). At the same time, we found statistically significant rise in E-wave velocity of PostPH dogs both to the control and PrePH ($p<0.002$ и $p<0.001$); increased A-wave in PostPH to PrePH ($p<0.018$Г); E/a ration was higher in PostPH in comparison both to the control and PrePH ($p<0.001$, for both).

The diastolic dysfunction (DD) was presented in all groups but differed in diversity. In the control group, 87,5% (7/8) of dogs had the 2nd class of diastolic dysfunction and 12,5% (1/8) – the 1st class. The PrePH group 53,6% (15/28) had the 1st degree, 32,1% (9/28) – 2nd degree, 7,1% (2/28) – 3rd degree of DD. In PostPH, 52,2% (12/23) of dogs had the 2nd degree and 47,8% (11/23) – 3rd class of DD.

Additional method to estimate diastolic function of LV was Tissue Doppler and e'-wave, in particular. This marker was statistically decreased in both experimental groups in comparison to the control ($p<0.017$ for PrePH and $p<0.006$ for PostPH). But intergroup difference was not statistically significant ($p>0.05$).

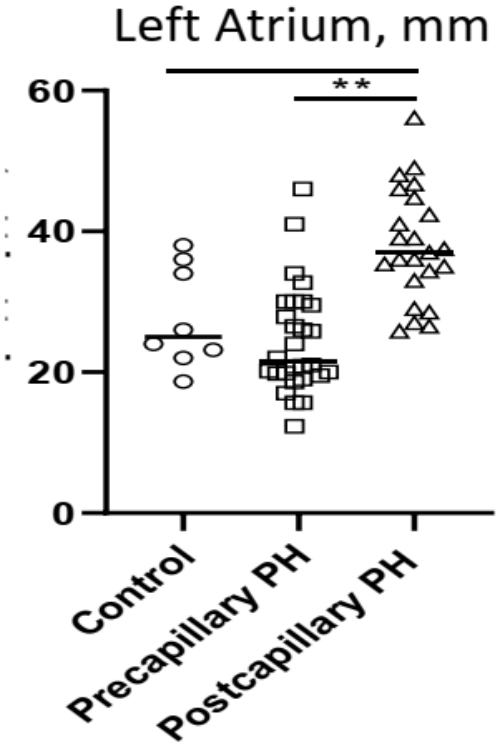

Fig. 1: Graphic of left atrium diameter diversity between groups * - <0.05; ** - <0.0001

Table 1: Weight and heart rate analyze

Indices	Control (N = 8)	Precapillary (N = 28)	Postcapillary PH (N = 23)	Differencies between control and PrePH	Differencies between control and PostPH	Differencies between PrePH and PostPH
Weight (kg)	4.3 (3.1-5.9)	3.9 (3.2-5.7)	4.5 (2.9-7.8)	>0.05	>0.05	>0.05
Heart rate, bmp	133.0 (118.5-140.8)	130.0 (110.0-159.0)	148.0 (140.0-156.0)	>0.05	0.030	>0.05

Table 2: Echocardiographic-derived measurements and indices. Complex analyze and statistics

Indices	Quality markers	Control (N = 8)	Precapillary PH (N = 28)	Postcapillary PH (N = 23)	Differencies between Control and PrePH	Differencies between Control and PostPH	Differencies between PrePH and PostPH
TAPSE (mm)		10.0 (9.6-10.6)	10.1 (9.2-12.7)	11.0 (10.0-12.8)	>0.05	0.040	>0.05
LA (mm)		25.0 (22.3-35.5)	21.5 (19.6-29.9)	37.0 (33.0-44.8)	>0.05	0.004	<0.001
Ao (mm)		13.8 (11.4-18.8)	13.8 (12.1-14.6)	13.3 (12.0-15.5)	>0.05	>0.05	>0.05
LA/Ao		1.9 (1.8-2.0)	1.6 (1.4-2.1)	2.7 (2.3-3.2)	>0.05	<0.001	<0.001
IVS (mm)		6.4 (5.5-8.2)	7.4 (6.6-8.2)	6.8 (6.0-7.4)	>0.05	>0.05	0.026
LVFW (mm)		6.1 (5.4-7.0)	6.3 (5.9-7.1)	5.5 (5.0-6.3)	>0.05	>0.05	0.005
LVIDd norm to mass		1.9 (1.8-2.0)	1.5 (1.4-1.9)	2.4 (2.2-2.5)	0.024	<0.001	<0.001
LVIDs norm to mass		1.0 (1.0-1.1)	0.8 (0.7-0.9)	1.2 (1.1-1.3)	0.005	>0.05	<0.001
MR velocity (m/s)		5.5 (5.0-5.8)	5.0 (4.5-5.5)	5.0 (4.4-5.6)	>0.05	>0.05	>0.05
MR degree N (%)	0.	0 (0%)	2 (7.1%)	0 (0%)	>0.05*	>0.05*	>0.05*
	1.	1 (12.5%)	5 (17.9%)	0 (0%)	>0.05*	>0.05*	>0.05*
	2.	6 (75.0%)	16 (57.1%)	6 (26.1%)	>0.05*	0.032*	0.046*
	3.	1 (12.5%)	5 (17.9%)	15 (65.2%)	>0.05*	0.016*	0.001*
	4.	0 (0%)	0 (0%)	2 (8.7%)	>0.05*	>0.05*	>0.05*
E (sm/s)		114.5 (98.0-123.8)	65.0 (48.0-88.8)	138.0 (120.0-164.0)	<0.001	0.002	<0.001
A (sm/s)		87.5 (79.8-95.8)	53.5 (46.0-83.0)	79.0 (60.0-100.0)	0.018	>0.05	0.018
E/A		1.3 (1.1-1.4)	0.9 (0.8-1.5)	1.8 (1.6-2.2)	>0.05	<0.001	<0.001
Diastolic dysfunction class N (%)	0 cl.	1 (12.5%)	2 (7.1%)	0 (0%)	>0.05*	>0.05*	>0.05*
	1 cl.	0 (0%)	15 (53.6%)	0 (0%)	0.011*	>0.05*	<0.001*
	2 cl.	7 (87.5%)	9 (32.1%)	12 (52.2%)	0.012*	>0.05*	>0.05*
	3 cl.	0 (0%)	2 (7.1%)	11 (47.8%)	>0.05*	0.028*	0.001*
PV (mm)		10.9 (9.7-13.0)	6.6 (4.3-11.0)	13.3 (12.5-16.1)	0.021	0.013	<0.001
RPA (mm)		5.8 (5.0-6.9)	8.2 (7.0-9.2)	6.7 (5.5-7.6)	0.001	>0.05	0.005
PV/RPA		1.9 (1.8-2.1)	0.8 (0.6-1.3)	2.1 (1.8-2.4)	<0.001	>0.05	<0.001
Right pulmonary artery dispensability N (%) (absent- 0, weak dispensability -1,	0	0 (0%)	16 (57.1%)	4 (17.4%)	0.005*	>0.05*	0.005*
normal dispensability -2)	1	0 (0%)	11 (39.3%)	9 (39.1%)	>0.05*	>0.05*	>0.05*
	2	8 (100%)	1 (3.6%)	10 (43.5%)	<0.001*	0.010*	0.001*
RV (mm)		6.9 (5.6-9.3)	11.0 (7.6-13.1)	10.5 (8.0-12.0)	0.021	0.009	>0.05
RV dilated. N, (%)		0 (0%)	16 (57.1%)	10 (43.5%)	0.005*	0.032*	>0.05*
RA (mm)		12.3 (11.9-13.2)	16.9 (14.9-25.1)	17.7 (15.1-19.0)	<0.001	<0.001	>0.05
RA dilated N, (%)		0 (0%)	26 (92.9%)	18 (78.3%)	<0.001*	<0.001*	>0.05*
RV wall (mm)		4 (3.6-4.4)	6.0 (5.4-6.8)	5.0 (4.2-6.2)	<0.001	0.005	0.012
RV wall hypertrophied N, (%)		0 (0%)	21 (75.0%)	8 (34.8%)	<0.001*	>0.05*	0.005*
PA (mm)		10.8 (10.0-11.0)	12.0 (10.7-14.3)	11.5 (10.0-13.4)	0.036	>0.05	>0.05
PA dilated N, (%)		0 (0%)	28 (100%)	16 (69.6%)	<0.001*	<0.001*	0.002*
AT (ms)		73.5 (66.8-85.8)	47.5 (33.5-53.0)	50.0 (42.0-63.0)	0.005	0.001	>0.05
ET (ms)		140.5 (123.3-153.0)	131.0 (109.3-157.5)	102.0 (92.0-121.0)	>0.05	0.005	0.014
AT/ET		0.6 (0.5-0.6)	0.4 (0.3-0.5)	0.5 (0.4-0.5)	0.003	0.043	0.005
LA regurgitation (m/s)		/	2.3 (1.5-3.5)	1.7 (1.6-2.4)	/	/	>0.05
PH by the waveform (N, %)		0 (0%)	25 (89.3%)	14 (60.9%)	<0.001*	0.004*	0.023*
TR degree (N, %)	0	4 (50%)	0 (0%)	0 (0%)	0.001*	0.002*	>0.05*
	1	4 (50%)	14 (50%)	12 (52.2%)	>0.05*	>0.05*	>0.05*
	2	0 (0%)	14 (50%)	11 (47.8%)	0.013*	0.028*	>0.05*
TR velocity (м/с)		2.5 (1.1-3.0)	3.7 (2.9-4.2)	3.0 (2.7-4.0)	0.001	0.001	>0.05
E", sm/s		10.0 (9.0-10.75)	8.0 (7.0-9.0)	8.0 (8.0-9.0)	0.017	0.006	>0.05
ePLAR		0.08 (0.0-0.24)	0.42 (0.30-0.57)	0.20 (0.16-0.23)	<0.001	>0.05	<0.001

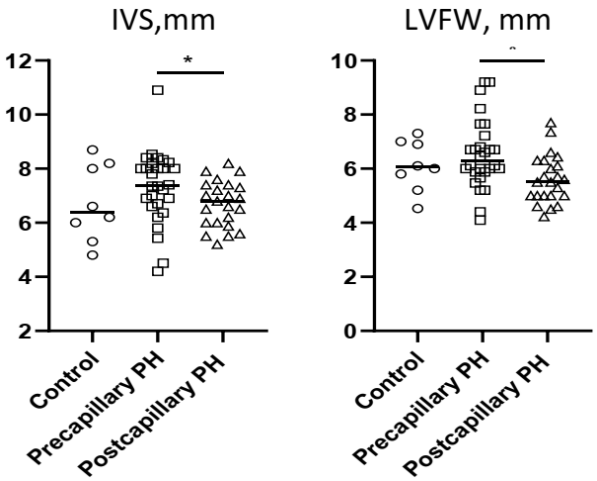

Fig. 2: Graphic of Interventricular Septum (IVS) and Left Ventricle Free Wall (LVFW) width diversity between groups. *- <0.05; ** - <0.0001

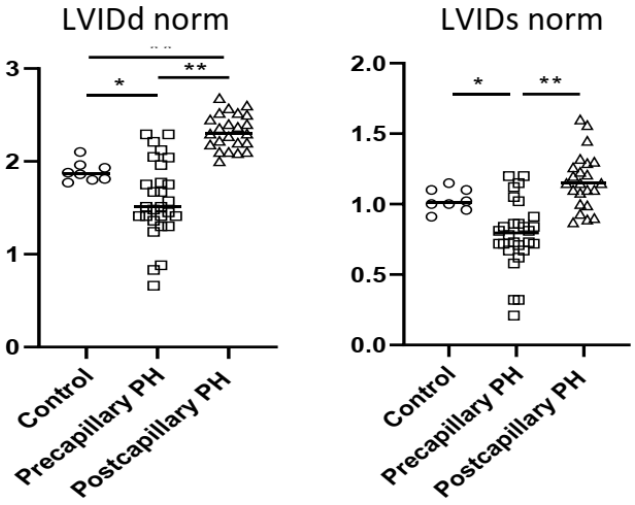

Fig.3: Graphic of left ventricle internal diastolic diameter normalized to body weight (LVIDd norm) and left ventricle internal diastolic diameter normalized to body weight (LVIDs norm) diversity between groups. * - <0.05; ** - <0.0001

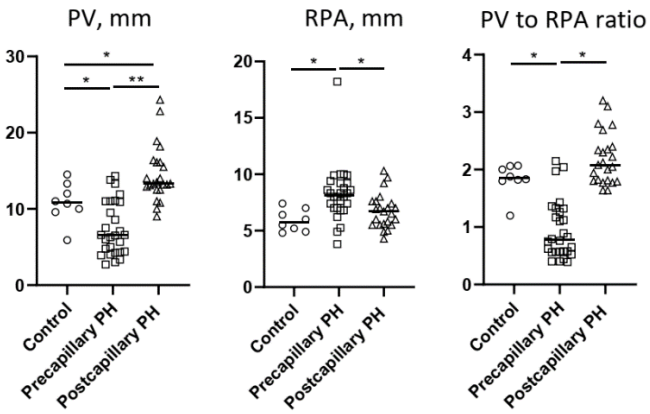

Fig.4: Graphic of Pulmonary Vein (PV), Right Pulmonary Artery (RPA) and pulmonary vein to right pulmonary artery ratio (PV to RPA) diversity between groups. *- <0.05; **- <0.0001

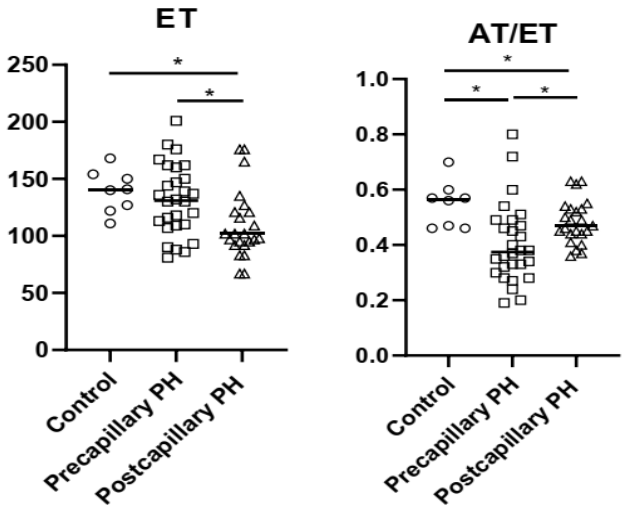

Fig. 5: Graphic pulmonary artery flow effusion time and acceleration time to effusion time ratio diversity between groups. * - <0.05; ** - <0.0001

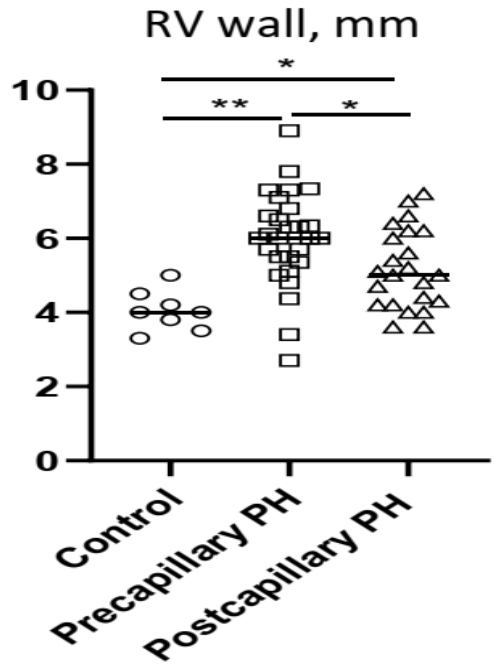

Fig.6: Graphic of right ventricle wall thickness (RV wall) diversity between groups

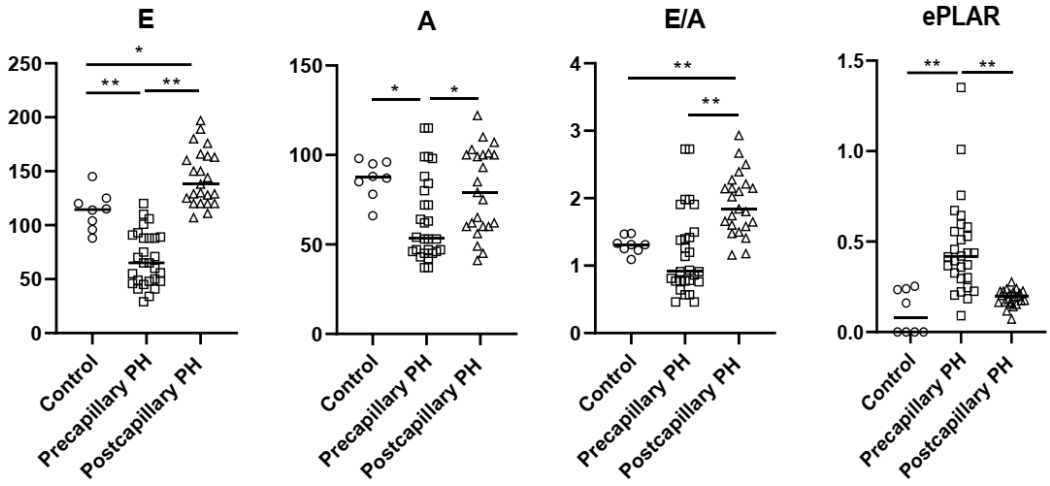

Fig.7: Graphic of mitral flow E-wave (E), A-wave (A), E-wave to A-wave ratio (E/A) and echocardiographic-derived transpulmonary gradient (ePLAR) diversity between groups.* - <0.05; ** - <0.0001

Mitral valve regurgitation (Table 2) did not statistically differ between groups (p>0.05).

Tricuspid regurgitation velocity (Table 2) was statistically higher in both experimental groups to the controls (p<0.001, for both), but did not statistically differ between each other (p>0.05).

Transpulmonary pressure gradient (ePLAR; Fig. 7) showed statistically significant increase in the PrePH in comparison to the control and PostPH (p<0.001, for both; Table 2). Statistically significant difference was not found in the control group and PostPH, due to low velocity and absence of tricuspid regurgitation in the control group (p>0.05).

Discussion

In this study, we tried to find out the value of echocardiographic markers in the differential diagnosis of different PH forms.

We found that patients with PrePH subjectively had a tendency to LV internal diameter reduction in comparison to PostPH. This fact puts us in the concept of LV blood flow reduction, due to increased Pulmonary Vessels Resistance (PVR) and additional RV pressure on IVS, which is in concordance with literature data (Reinero *et al*., 2020; Vezzosi *et al*., 2018; Vezzosi *et al*., 2018 Visser *et al*., 2015; Visser *et al*., 2018).

One of the simplest markers of PH differentiation is LA diameter and LA/Ao ratio. In this study, LA and LA/Ao ratio had differential power between PrePH and PostPH. This data is agreed with pulmonary vessels flow capacity. In the PrePH (group) we can suspect obstructive element due to high PVR, which decreases LA reservoir function, associated with internal diameter reduction, leaving exclusively conduit function to LA. The LPV/RPA ratio has high PH differential potency. This is also associated with decreased LPV (left pulmonary vein) fulling. In cases of PostPH, LA inflow is preserved, despite increased interatrial pressure and mitral valve regurgitation. However, due to the absence of vein valves there is reversal flow to pulmonary veins, leading to decreased emptying of pulmonary pool (Wright *et al*., 2021).

Transmitral flow and diastolic dysfunction had specific changes. The most important difference is based on E-wave velocity and E-to-A-wave ratio. These changes are similarly explained by decreased left chamber fulling due to increased PVR. (Wright *et al*., 2021).

Unfortunately, RV, RA diameter and RV wall thickness did not show differential abilities between PH forms, taken without left chambers changes. The same conditions are presented in PA diameter and dilatation-they were distended in both groups. But subjectively evaluated RPA distensibility could be a difference marker. We found that in 43% of cases with distended RPA but preserved collapsing, we can suspect PostPH, but in 57% of cases with dilated RPA and absence of distensibility, we could suspect PrePH. This conclusion is in concordance with observed literature data (Serrano-Parreño *et al*., 2017; Venco *et al*., 2014).

Tricuspid regurgitation velocity did not show differential potency between PH forms. This finding could be based on both preserved RV systolic function and the inability of direct PA pressure evaluation and construction of regurgitation velocity pattern of PA pressure association. Additional scepsis to isolated TR criteria of PA pressure is based on the recent study of MMVD dogs with a comparison of TR-calculated pressure and catheterization data. The findings from direct PA pressure measurement were in weak correlation with TR velocity. Also, this concept was observed in earlier studies (Menciotti *et al*., 2021; Soydan *et al*., 2015). To these days, right heart catheterization is still mostly unavailable in veterinary practice and we should base our reasonings on tricuspid valve regurgitation, but this parameter is very variable. Moreover, mentioned above studies of TR-calculated PA pressure weakness make us look closer to RPA-estimated compliance and PVR (Surkova and Kovács, 2020)

The PA flow patterns (AT/ET ratio) showed significant differential potency, but this parameter depends on many factors and in the context of unstable conditions and compensatory tachycardia, ET could be inappropriately shortened, leading to a decrease of this ratio and this marker's value reduction (Augustine *et al*., 2021; Gupta *et al*., 2021). This concept should be observed in a more specific study.

Despite many papers about TAPSE changes in PH, in this study, we did not show a significant diagnostic role of the marker. There were no pieces of evidence of a difference in TAPSE between both the controls to PrePH and PrePH to PostPH groups. TAPSE is usually evaluated as a marker of decreased RV systolic function. In this study, we did not find signs of decreased TAPSE, which could be interpreted as a still compensated stage of RV systolic performance in the presence of developed PrePH (Vezzosi *et al*., 2018; Visser, 2017). We found an increase in this marker in the PostPH to the control group, which could be explained by an adaptive systolic rise to preload and little changes in resistive pulmonary vessels and precapillary bed (Pariaut *et al*., 2012). A recent study of dogs with suspected PostPH admitted that TAPSE in dogs could be increased by volume overload and usually rise in mild and moderate forms of PH and decreased in the severe stage of PostPH (Yuchi *et al*., 2021). In our study, we worked with dogs in the acutely decompensated stage, where PostPH is enhanced mostly due to protective vasoconstriction to prevent dramatic pulmonary edema and without severe RV dysfunction.

The most promising parameter was ePLAR. Indexes calculated in this marker could reflect an interaction between PA and LA pressure. High differential power of this marker gives us an opportunity to elucidate the prevalence of PostPH or PrePH form. Unfortunately, we did not verify this data by right heart catheterization and did not correlate it with calculated transpulmonary gradient. Another limitation is an inability to verify mixed or reactive PH due to a complicated state, which could be reflected not only in the ePLAR (Galiè *et al*., 2016; Scalia *et al*., 2016).

In literature, we can find reviews with the most possible etiologies for PH development. The most important are: Left-heart diseases, pulmonary disease, cardiovascular shunts, heartworm disease, pulmonary thromboembolism, etc. These articles, present information about the prevalence of adult dogs getting affected with PH. This fact could give us a suspicion of comorbidity in PH dogs (age, breed data are similar to MMVD-affected dogs and most of them previously had a history of heart failure) with lung and heart affection (Bach *et al*., 2006; Campbell, 2007; Johnson *et al*., 1999; Kellihan and Stepien, 2010; Kellum and Stepien, 2007; Pyle *et al*., 2004a-b; Serrano-Parreño *et al*., 2017; Serres *et al*., 2007).

The retrospective study, elucidating effect on the lifespan of the dogs with PH secondary to lung disease (3rd group in ACVIM consensus), admitted that the average time of survival was about 276 days (Jaffey *et al*., 2019). Meanwhile, in another study of PH attributed to MMVD and left-heart

disease (2nd group in ACVIM consensus), the survival time was about 576 days (Borgarelli *et al*., 2015). These data show us the importance of early identification of PH form to choose the most effective treatment line.

In addition, there is a limitation in PH form prevalence in cases of unusual diseases diagnosis, such as primary lung vessels diseases (hemangiomatosis of pulmonary capillaries or veno-occlusive disease) (Reinero *et al*., 2019). These diseases could lead to confusions, with shifting between PostPH and PrePH forms. Unfortunately, we do not have specific markers of this disease, except pathohistological data (Reinero *et al*., 2019; Stenmark *et al*., 2016; Williams *et al*., 2016). The additional difficulties that arise from observed studies are associated with non-specific signs: Acute respiratory distress, markers of pulmonic edema on radiograms, rapid worsening and death in a couple of hours and age above 10,5 years. These data are very close to common signs of left-sided heart failure patients, which make it almost impossible to differentiate without specific methods (Williams *et al*., 2016).

Conclusion

In this study, we found several echocardiographic parameters, helping to differentiate Pre-and Post-PH forms: LA, LA/Ao ratio, LV diameter, LV/RPA ratio, IVS diameter, diastolic class dysfunction and RPA distensibility by quality characterization and novel in veterinary studies index - ePLAR.

Clearly, these markers should be interpreted in the context of the whole heart geometry, the most observed changes were not specific (except ePLAR). In combination, these parameters could predict the prevalence form of PH.

Acknowledgement

Special thanks to Toropova J.V., Zelinskaya I.V. for providing an opportunity to analyze studied substrates.

Author's Contributions

Oleynikov D: Collected data, provided diagnostics and treatment, coordinated the data-analysis and contributed to the writing of the manuscript.

Ma Yi: Coordinated the data-analysis and contributed to the writing of the manuscript

Ethics

The scheme of this study was ethically approved on the clinical conference.

References

Augustine, D. X. Augustine, D. X., Coates-Bradshaw, L. D., Willis, J., Harkness, A., Ring, L., Grapsa, J., Coghlan, G., Kaye, N., Oxborough, D., Robinson, S., Sandoval, J., Rana, B. S., Siva, A., Nihoyannopoulos, P., Howard, L. S., Fox, K., Bhattacharyya, S., Sharma, V., Steeds, R. P., & Mathew, T. (2021). British Society of Echocardiography Education Committee. https://researchonline.ljmu.ac.uk/id/eprint/9153/1/G11.full.pdf

Bach, J. F., Rozanski, E. A., MacGregor, J., Betkowski, J. M., & Rush, J. E. (2006). Retrospective evaluation of sildenafil citrate as a therapy for pulmonary hypertension in dogs. Journal of veterinary internal medicine, 20(5), 1132-1135. doi.org/10.1111/j.1939-1676.2006.tb00711.x

Borgarelli, M., Abbott, J., Braz-Ruivo, L., Chiavegato, D., Crosara, S., Lamb, K., ... & Haggstrom, J. (2015). Prevalence and prognostic importance of pulmonary hypertension in dogs with myxomatous mitral valve disease. Journal of Veterinary Internal Medicine, 29(2), 569-574. doi.org/10.1111/jvim.12564

Campbell, F. E. (2007). Cardiac effects of pulmonary disease. Veterinary Clinics of North America: Small Animal Practice, 37(5), 949-962. doi.org/10.1016/j.cvsm.2007.05.006

Galiè, N., Humbert, M., Vachiery, J. L., Gibbs, S., Lang, I., Torbicki, A., ... & Hoeper, M. (2016). 2015 ESC/ERS guidelines for the diagnosis and treatment of pulmonary hypertension: The joint task force for the diagnosis and treatment of pulmonary hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS): Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC), International Society for Heart and Lung Transplantation (ISHLT). European heart journal, 37(1), 67-119. doi.org/10.1093/eurheartj/ehv317

Gupta, H., Ghimire, G., & Naeije, R. (2011). The value of tools to assess pulmonary arterial hypertension. European Respiratory Review, 20(122), 222-235. doi.org/10.1183/09059180.00006911

Jaffey, J. A., Wiggen, K., Leach, S. B., Masseau, I., Girens, R. E., & Reinero, C. R. (2019). Pulmonary hypertension secondary to respiratory disease and/or hypoxia in dogs: Clinical features, diagnostic testing and survival. The Veterinary Journal, 251, 105347. doi.org/10.1016/j.tvjl.2019.105347

Johnson, L., Boon, J., & Orton, E. C. (1999). Clinical characteristics of 53 dogs with Doppler-derived evidence of pulmonary hypertension: 1992-1996. Journal of Veterinary Internal Medicine, 13(5), 440-447. doi.org/10.1111/j.1939-1676.1999.tb01461.x

Kellihan, H. B., & Stepien, R. L. (2010). Pulmonary hypertension in dogs: Diagnosis and therapy. Veterinary Clinics: Small Animal Practice, 40(4), 623-641. https://www.vetsmall.theclinics.com/article/S0195-5616(10)00039-2/fulltext

Kellum, H. B., & Stepien, R. L. (2007). Sildenafil citrate therapy in 22 dogs with pulmonary hypertension. Journal of veterinary internal medicine, 21(6), 1258-1264. doi.org/10.1111/j.1939-1676.2007.tb01947.x

Menciotti, G., Abbott, J., Aherne, M., Lahmers, S., & Borgarelli, M., (2021). Accuracy of echocardiographically estimated pulmonary artery pressure in dogs with myxomatous mitral valve disease. J. Vet. Cardiol 2021; 35, 90-100. doi.org/10.1111/j.1939-1676.2007.tb01947.x

Pariaut, R., Saelinger, C., Strickland, K. N., Beaufrère, H., Reynolds, C. A., & Vila, J. (2012). Tricuspid annular plane systolic excursion (TAPSE) in dogs: Reference values and impact of pulmonary hypertension. Journal of veterinary internal medicine, 26(5), 1148-1154. doi.org/10.1111/j.1939-1676.2012.00981.x

Pyle, R. L., Abbott, J., & MacLean, H. (2004a). Pulmonary hypertension and cardiovascular sequelae in 54 dogs. Intern J. Appl. Res. Vet. Med., 2(2), 99-109. http://www.jarvm.com/articles/Vol2Iss2/PYLEJARVMVol2No2Bweb.pdf

Pyle, R. L., King, M. D., Saunders, G. K., & Panciera, D. L. (2004b). Pulmonary thrombosis due to idiopathic main pulmonary artery disease. Veterinary medicine. https://agris.fao.org/agris-search/search.do?recordID=US201300997976

Reinero, C. R., Jutkowitz, L. A., Nelson, N., Masseau, I., Jennings, S., & Williams, K. (2019). Clinical features of canine pulmonary veno-occlusive disease and pulmonary capillary hemangiomatosis. Journal of veterinary internal medicine, 33(1), 114-123. doi.org/10.1111/jvim.15351

Reinero, C., Visser, L. C., Kellihan, H. B., Masseau, I., Rozanski, E., Clercx, C., ... & Scansen, B. A. (2020). ACVIM consensus statement guidelines for the diagnosis, classification, treatment and monitoring of pulmonary hypertension in dogs. Journal of veterinary internal medicine, 34(2), 549-573. doi.org/10.1111/jvim.15725

Scalia, G. M., Scalia, I. G., Kierle, R., Beaumont, R., Cross, D. B., Feenstra, J., ... & Platts, D. G. (2016). ePLAR-The echocardiographic Pulmonary to Left Atrial Ratio-A novel non-invasive parameter to differentiate pre-capillary and post-capillary pulmonary hypertension. International journal of cardiology, 212, 379-386. doi.org/10.1016/j.ijcard.2016.03.035

Serrano-Parreño, B., Carretón, E., Caro-Vadillo, A., Falcón-Cordón, Y., Falcón-Cordón, S., & Montoya-Alonso, J. A. (2017). Evaluation of pulmonary hypertension and clinical status in dogs with heartworm by Right Pulmonary Artery Distensibility Index and other echocardiographic parameters. Parasites & vectors, 10(1), 1-6. https://parasitesandvectors.biomedcentral.com/articles/10.1186/s13071-017-2047-2

Serres, F., Chetboul, V., Gouni, V., Tissier, R., Sampedrano, C. C., & Pouchelon, J. L. (2007). Diagnostic value of echo-Doppler and tissue Doppler imaging in dogs with pulmonary arterial hypertension. Journal of veterinary internal medicine, 21(6), 1280-1289. doi.org/10.1111/j.1939-1676.2007.tb01950.x

Soydan, L. C., Kellihan, H. B., Bates, M. L., Stepien, R. L., Consigny, D. W., Bellofiore, A., ... & Chesler, N. C. (2015). Accuracy of Doppler echocardiographic estimates of pulmonary artery pressures in a canine model of pulmonary hypertension. Journal of veterinary cardiology, 17(1), 13-24. doi.org/10.1016/j.jvc.2014.10.004

Stenmark, K. R., Krafsur, G. M., & Tuder, R. M. (2016). Pulmonary veno-occlusive disease and pulmonary hypertension in dogs: Striking similarities to the human condition. Veterinary pathology, 53(4), 707-710. doi.org/10.1177/0300985816647454

Surkova, E., & Kovács, A. (2020). Comprehensive Echocardiographic Assessment of the Right Ventricular Performance: Beyond TAPSE and Fractional Area Change. Российский кардиологический журнал, 25(S3).

Thenappan, T., Prins, K. W., Pritzker, M. R., Scandurra, J., Volmers, K., & Weir, E. K. (2016). The critical role of pulmonary arterial compliance in pulmonary hypertension. Annals of the American Thoracic Society, 13(2), 276-284. doi.org/10.1513/AnnalsATS.201509-599FR

Venco, L., Mihaylova, L., & Boon, J. A. (2014). Right Pulmonary Artery Distensibility Index (RPAD Index). A field study of an echocardiographic method to detect early development of pulmonary hypertension and its severity even in the absence of regurgitant jets for Doppler evaluation in heartworm-infected dogs. Veterinary parasitology, 206(1-2), 60-66. doi.org/10.1016/j.vetpar.2014.08.016

Vezzosi, T., Domenech, O., Costa, G., Marchesotti, F., Venco, L., Zini, E., ... & Tognetti, R. (2018). Echocardiographic evaluation of the right ventricular dimension and systolic function in dogs with pulmonary hypertension. Journal of veterinary internal medicine, 32(5), 1541-1548. doi.org/10.1111/jvim.15253

Visser, L. C. (2017). Right ventricular function: Imaging techniques. Veterinary Clinics: Small Animal Practice, 47(5), 989-1003. https://www.vetsmall.theclinics.com/article/S0195-5616(17)30037-2/fulltext

Visser, L. C., Scansen, B. A., Brown, N. V., Schober, K. E., & Bonagura, J. D. (2015). Echocardiographic assessment of right ventricular systolic function in conscious healthy dogs following a single dose of pimobendan versus atenolol. Journal of Veterinary Cardiology, 17(3), 161-172. doi.org/10.1016/j.jvc.2015.04.001

Visser, L. C., Sintov, D. J., & Oldach, M. S. (2018). Evaluation of tricuspid annular plane systolic excursion measured by two-dimensional echocardiography in healthy dogs: Repeatability, reference intervals and comparison with M-mode assessment. Journal of Veterinary Cardiology, 20(3), 165-174. doi.org/10.1016/j.jvc.2018.04.002

Williams, K., Andrie, K., Cartoceti, A., French, S., Goldsmith, D., Jennings, S., ... & Jutkowitz, A. (2016). Pulmonary veno-occlusive disease: A newly recognized cause of severe pulmonary hypertension in dogs. Veterinary Pathology, 53(4), 813-822. doi.org/10.1177/0300985815626572

Wright, S. P., Dawkins, T. G., Eves, N. D., Shave, R., Tedford, R. J., & Mak, S. (2021). Hemodynamic function of the right ventricular-pulmonary vascular-left atrial unit: Normal responses to exercise in healthy adults. American Journal of Physiology-Heart and Circulatory Physiology, 320(3), H923-H941. doi.org/10.1152/ajpheart.00720.2020

Yuchi, Y., Suzuki, R., Teshima, T., Matsumoto, H., & Koyama, H. (2021). Utility of tricuspid annular plane systolic excursion normalized by right ventricular size indices in dogs with postcapillary pulmonary hypertension. Journal of Veterinary Internal Medicine, 35(1), 107-119. doi.org/10.1111/jvim.15984