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Original article

INVESTIGATING THE DIAGNOSTIC ACCURACY OF VERTEBRAL HEART SCORE, VERTEBRAL LEFT ATRIUM SCORE AND THORACIC WIDTH MEASUREMENT IN CATS WITH SUSPECTED CARDIAC PROBLEMS

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Summary

Ozdil, B., T. M. Parlak & M. Arican, 2024. Investigating the diagnostic accuracy of vertebral heart score, vertebral left atrium score and thoracic width measurement in cats with suspected cardiac problems. *Bulg. J. Vet. Med.* (online first).

This study aimed to evaluate vertebral heart score (VHS) in terms of sensitivity and accuracy of radiographic heart examination for early diagnosis of cardiac diseases in cats and to compare vertebral left atrium score (VLAS), N-terminal pro-B-type natriuretic peptide (Nt-proBNP) with thoracic width measurement (TWM). Thirty cats were used. Ten of these cases were used as the control group. The remaining 20 cases formed the group with suspected heart problems. The VHS, VLAS and thoracic width were measured by radiographic examination. Blood was drawn and Nt-proBNP concentrations were assayed in all animals. As a result, VHS was above the reference range in 14 cases. For VLAS measurements, 7 cases had values above the reference range. TDM was above the normal reference range in 5 cases from the control group. There was no statistically significant correlation between VHS and VLAS (P=0.07), VHS and TWM (P=0.06), VHS and Nt-proBNP (P=0.05). However, in cases with high VHS, VLAS, TWM, and Nt-proBNP values, elevated levels were observed in 50% of the cases compared to the reference values. However, radiographically measured VHS, VLAS, and TWM may represent independent measurements. No statistically significant relationship was found between them. It would be beneficial to conduct mroe extensive studies by considering the breed, age and weight in cats with suspected heart problems.

Key words: cardiomegaly, cat, Nt-proBNP

INTRODUCTION

Although echocardiography is superior in the evaluation of the heart because of its practicality, radiography is still used in the diagnosis of heart problems and is considered an important part of heart disease diagnosis and management in dogs and cats. However, studies on its use in heart problems of cats are limited. Heart disease is common in cats and affects 10–15% of feline population (Buchanan, 1999; Payne

et al., 2015; Santiago *et al.*, 2020; Kittleson & Côté, 2021). It is a serious medical condition that requires emergency care in cats. The most common cardiac diseases in cats include pathologies such as hypertrophic cardiomyopathy, restrictive cardiomyopathy, and unclassified cardiomyopathy. Congenital heart diseases in cats can be inherited (Silverstein & Hopper, 2015). Acquired heart disease, often called adult-onset heart disease, occurs in middle-aged and older cats due to wear and tear on the heart. It can also be caused by an injury or infection. In some cases, adult-onset heart disease develops as a secondary problem while the primary problem originates from another part of the body, such as the thyroid gland. In some cats, heart disease presents with a thrombus manifested in a painful and paralysing condition. This situation is attributed to the progression of developing thrombosis in the heart and occlusion of the caudal part of the aorta. This cuts off the blood flow to the cats' hind legs. Emergency care is needed immediately. Cats with structural heart disease will develop recurrent signs of congestive heart failure over time and will require lifelong medical treatment. The survival expectancy in cats with congestive heart disease is 6–12 months. In these cases, regular cardiac examination should be performed. For this reason, most importantly, it is necessary to solve problems before cats get heart disease. In this respect, it is necessary to evaluate heart diseases through imaging and biochemical parameters (Laudhittirut *et al.*, 2020).

Echocardiography is the best method for diagnosing heart disease. Although precise, it requires experience and equipment. For this purpose, three basic methods are used for the diagnosis of heart diseases in cats: cardiac biomarkers measurements with radiography, thoracic-focused assessment, and sonography for trauma (Laudhittirut *et al.*, 2020). The vertebral heart scale (VHS) is considered an objective assessment of the size of the heart silhouette. With this method, the long and short axis of the heart are marked on lateral thoracic radiographs and compared with the thoracic spine (Buchanan & Bucheler, 1995; Guglielmini *et al.*, 2009). Thus, early diagnosis of heart problems can be made. The VHS method may not be able to distinguish between cats with and without heart disease in assessing heart size. The VHS threshold for cats with heart disease is 8 (Guglielmini *et al.*, 2014). Cats with VHS >9.3 are extremely prone to developing heart disease. However, cats with VHS 8–9.3 are classified in the gray zone and further investigation is required to diagnose heart disease (Sleeper *et al.*, 2013; Fucharoen *et al.*, 2017). Increases in vertebral left atrium score (VLAS) are associated with increased left atrium (LA) size. Thoracic width measurement (TWM) can also be done on thoracic radiographs. A TWM of 66% is accepted as normal heart size (Bodh *et al.*, 2016; Puccinelli *et al.*, 2021).

Laboratory tests are also important in the evaluation of heart disease. Common tests include serum biochemistry, complete blood count, urinalysis, effusion analysis, and blood culture tests (Boswood, 2009). Cardiac biomarkers, such as N-terminal Pro-B-type natriuretic peptide (NT-proBNP) and the renin-angiotensin-aldosterone system, can be also employed (Connolly *et al.*, 2009; Boswood, 2009; Sergeeva & Cristoffels, 2013; Liquori *et al.*, 2014). The Pro-B-type natriuretic peptide (proBNP) is a substance released from the heart muscle when the heart is under strain or stress (Weber & Hamm,

2006). It is generally accepted that the detection of radiographic cardiomegaly is associated with heart disease. Heart conditions such as congenital heart disease, dilated cardiomyopathy, and congestive heart failure may result in radiographic cardiomegaly (Ware, 2013).

This study aimed to evaluate VHS in terms of sensitivity and accuracy of radiographic heart examination in the early diagnosis of cardiac diseases in cats and to compare the values of VLAS, blood NT-proBNP with thoracic width measurement.

MATERIALS AND METHODS

Thirty cats of various age, gender, breed and weight were used. Ten of these cases were used as control group. The control group had no heart problems. The remaining 20 cases formed the suspected heart problem group. Their breed, age, gender and weight distribution is shown in Table 1. In the suspected heart disease group, history of restlessness and weakness was present in 17 cases (85%). In 6 cases, exercise intolerance was detected (30%). In 5 cases dyspnea was detected (25%). History of chronic cough was reported in 17 cases (85%). Anorexia was present in all cases (100%). The study was carried out with the permission of Selcuk University, Faculty of Veterinary Medicine, Animal Experiments Local Ethics Committee 2022/18 of 24.02.2022.

VHS was measured using the right lateral projection. The carina (bifurcation of trachea) of the trachea was found on the radiographic image, then a straight line was drawn from here to the apex of the heart, corresponding to the long axis of the heart. A second line was drawn between the widest points of the heart perpendicularly to the first line: the short axis of the heart. Then, the 4th thoracic vertebra was identified and lines showing the long and short heart axes were placed cranially; finally the VHS was determined by counting the number of vertebrae in the area covered by the lines (Fig. 1). The threshold value of VHS for cats with heart disease is 8 vertebrae (de Oliveira *et al.*, 2014; Guglielmini *et al.*, 2014; Fucharoen *et al.*, 2017).

Table 1. Breed, age, gender and weight distribution of the cases

Cases	Breed	Gender	Weight, kg	Age, years
Control 1	British Shorthair	♂	3.5	2
Control 2	Tabby	♀	2.1	1
Control 3	Ankara	♂	4	1
Control 4	Coiled	♀	3.3	3
Control 5	Tabby	♀	2.6	4
Control 6	Tabby	♂	5.1	4
Control 7	British Shorthair	♀	3.2	2
Control 8	Tabby	♂	2.2	1
Control 9	Tabby	♂	3.6	3
Control 10	Coiled	♂	3	3
1	Tabby	♀	3.1	13
2	British Shorthair	♂	3.6	5
3	Tabby	♀	3.9	6
4	British Shorthair	♂	3	5
5	Ankara	♂	3.5	4
6	Bombay	♀	2.8	7
7	Coiled	♂	3.8	6
8	Tabby	♀	2.6	5
9	Tabby	♀	2.9	8
10	Scottish Fold	♂	3	5
11	Tabby	♂	4	7
12	Tabby	♂	6.5	5
13	Tabby	♀	3	6
14	British Shorthair	♀	2.3	5
15	Tabby	♂	3.8	6
16	Tuxedo	♂	3.5	8
17	Tabby	♀	3	7
18	Tabby	♂	4	5
19	Tabby	♀	2.6	3
20	Tabby	♀	3	4

The VLAS measures the LA size on a right lateral projection. First, a line was drawn from the centre of the most ventral part of the carina to the most caudal part of the left atrium, dorsal side of the caudal vena cava. Similar to the VHS measurement, a second line of equal length was drawn starting from the cranial edge of T4 parallel to the ventral and immediately caudally of the vertebral column. Finally, the VLAS value was calculated by counting the number of thoracic vertebrae along the length of the line (Fig. 2). The reference range was accepted as 1.2 to 1.8 vertebrae.

All radiographs were measured in the inspiratory phase. The noses of the animals were slightly pinched or stimulated with a feather to capture the inspiratory phase. Thoracic width was measured in dorsoventral view. First, a line was drawn at the widest point of the heart silhouette. In the second step, the width of the thoracic cavity was measured by making a second measurement line from the point closest to the first line and parallel to it. The thoracic width was calculated by dividing the value obtained in the first measurement by the value obtained in the second measurement (Fig. 3). The thoracic width measurement value was high in more than 55–65% of the cases with heart diseases (Bodh *et al.*, 2016).

Blood was taken from v. *cephalica antebrachi* into a heparin-free injector and transferred to a BD Vacutainer tube. Then, it was centrifuged on centrifuge device (Tubes Cence TDZ4-WS, China) at 5000 rpm for 5 min and blood serum was separated. The separated serum was

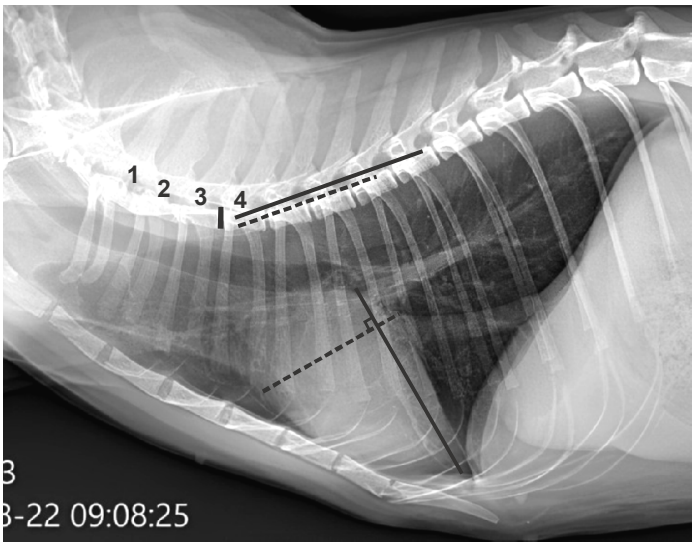

Fig. 1. Measurement of VHS on right lateral radiograph in cats. The carina (bifurcation of trachea) of the trachea was found on the radiographic image, then a line was drawn from here to the apex of the heart, corresponding to the long heart axis (solid black line). A second line was drawn between the widest points of the heart perpendicularly to the first line: the short heart axis (dotted black line). Then, the 4th thoracic vertebra was identified and lines of long and short heart axes were placed cranially; finally the VHS was determined by counting the number of vertebrae in the area covered by the lines (long axis 5 vertebrae; short axis 3.75 vertebrae; total 8.75 vertebrae).

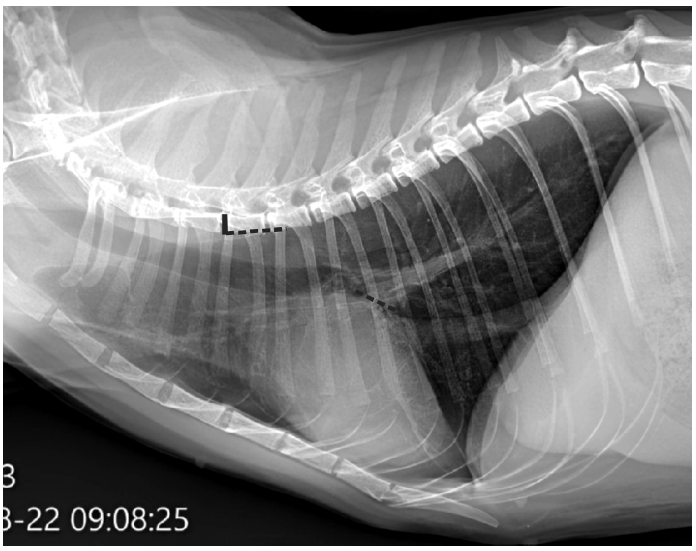

Fig. 2. Measurement of VLAS on right lateral radiograph in cats. First, a line was drawn from the centre of the most ventral part of the carina to the most caudal part of the left atrium, dorsal side of the caudal vena cava. A second line of equal length was drawn starting from the cranial edge of T4 parallel to the ventral and immediately caudally of the vertebral column.

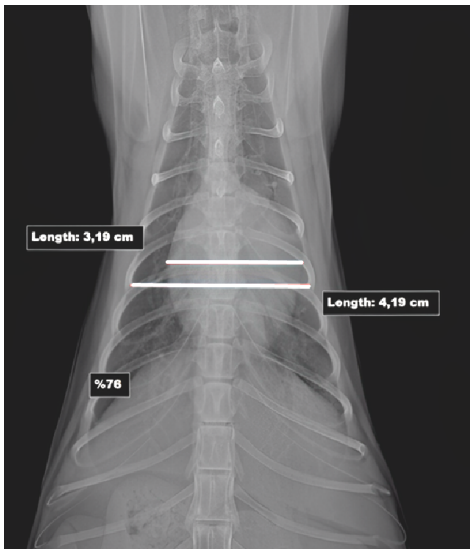

Fig. 3. Thoracic width measurement. First, a line was drawn at the widest point of the heart silhouette. In the second step, the width of the thoracic cavity was measured by making a second measurement line from the point closest to the first line and parallel to it.

transferred to an Eppendorf tube and stored at −20 °C until the day of measurement. Cat N-terminal pro-brain natriuretic peptide, NT-proBNP ELISA KIT (E0000cat; Bioassay Technology Laboratory, China) was used. Samples were read in MWGt LambdaScan 200 ELISA device (China). NT-pro-BNP levels greater than >440 pmol/L was indicative for heart disease (Connolly *et al.*, 2009).

Statistical analysis

The chi-square test and Ficher's exacts test were used for analysis of VHS-VLAS, VHS-TD, VHS-Nt-proBNP associations.

RESULTS

A history of restlessness, weakness and exercise intolerance were reported in cats. Shortness of breath was detected in 5 (25%) cats (cases 2, 8, 9, 10, 11). Chronic cough was reported for 17 (85%) cases

Table 2. Data from radiographic evaluation of VHS, VLAS, TWM and serum NT-proBNP analysis

Cases	Vertebral heart scale (VHS)	Vertebral left atrium score (VLAS)	Thoracic width measurement (TWM %)	N-terminal pro-B-type natriuretic peptide (NT-proBNP, pmol/L)
Control 1	7.6	1.2	55	430
Control 2	7.5	1.5	71	540
Control 3	7	1.8	61	394
Control 4	7.2	1.8	53	350
Control 5	7.5	1.1	69	350
Control 6	7	1.1	52	420
Control 7	7.6	1.4	58	370
Control 8	7.5	2	67	550
Control 9	8	1.5	69	420
Control 10	8	1.6	67	520
1	7.5	1.5	80	420
2	8.2	1.8	75	426
3	8.3	2	65	312
4	8.1	1.8	58	326
5	8.2	1.5	73	428
6	8.5	1.9	68	637
7	8.2	1.8	69	421
8	9	1.8	66	484
9	9	1.9	75	472
10	9	1.8	73	361
11	8.8	1.6	76	418
12	8	1.9	66	536
13	8	1.5	66	368
14	8.3	1.2	71	451
15	9	1.5	60	513
16	8	2	68	481
17	8.2	1.6	63	431
18	8.5	2	66	546
19	8	1.5	55	483
20	8.3	2	76	451

(all cats except cases 2, 12, 13). Anorexia was present in all cases (100%).

Radiographic evaluation of VHS, VLAS, TWM and serum NT-proBNP analysis in cats is given in Table 2. All of the cats in the control group were found to be within the normal reference range (Ref. value: 6.7-8.1). The vertebral heart score measurements of the cases with suspected heart disease were 19 cases above 8 vertebrae detected. It was determined that the VHS value was high in 95% of the cats in the suspected group.

Seven cats in the control group had VLAS within the normal reference range of 1.2–1.8 (Table 2). The parameter was 1.73±0.3 (mean ± SD) vertebrae in the group with suspected heart problems. In three cases VLAS value was 1.8-2. In cats with heart diseases VLAS was determined above the reference range in 7 cases, and in 5 cases, VLAS was at the upper limit.

VLAS values were high in 35% of the cats with suspected heart diseases.

TWM percentage in cats is given in Table 2. It was above the reference value in five animals from the control group (> 55–65%). In 14 cases with suspected heart disease TWM was higher than the reference value. In cats with heart problems, the thoracic width measurement value was high in 70% of the cases.

In the control group, three cats had serum NT-proBNP above the normal reference value (440 pmol/L). NT-proBNP value exceeded the normal reference value in 10 patients with suspected heart disease, e.g. NT-proBNP value was high in 50% of the cases.

There was no statistically significant relationship between VHS and VLAS (P=0.07), VHS and TWM (P=0.06), VHS and Nt-proBNP (P>0.05). However, the levels of VLAS, TWM and Nt-proBNP were simultaneously higher than the reference value in 50% of the cats in the cases with a high level of VHS. In the control group, where there was only 1 case of high VHS, the rest of the values were the same. However, no clinical problem was observed in this case.

DISCUSSION

Although the cats with heart disease in this study were 50% male and 50% female, the reported incidence is generally higher in male cats (Payne *et al.*, 2015; Saunders, 2021). In this study, the mean ± SD VHS in cats was 7.49 ± 0.5 vertebrae in the control cases. It was determined as 8.35 ± 0.5 vertebrae in cats with heart problems. VHS in this study was consistent with the literature findings in cases used for control purposes and in findings with suspected heart problems (Sleeper *et al.*, 2013; Fucharoen *et al.*, 2017). Cats with VHS >9.3 have been reported to be extremely susceptible to developing heart disease. Nevertheless, cats exhibiting VHS values between 8–9.3 have been documented to indicate cardiac pathology (Sleeper *et al.*, 2013; Fucharoen *et al.*, 2017). Guglielmini *et al.* (2015) showed in their study that the VHS in cats was 7.56±0.54 and de Oliveira *et al.* (2014) – 7.61±0.34 vertebrae. Some conditions may complicate the interpretation of radiograms; for example, respiratory tract disease, non-cardiogenic pleural effusion, heartworm, radiological position, pericardial fat, heartbeat and respiratory phases (Benigni *et al.*, 2009; Sleeper *et al.*, 2013; de Oliveira *et al.*, 2014; Guglielmini *et al.*, 2014).

For these reasons, the evaluation of VHS may not always yield results. However, among the cases under study with suspected heart disease, 19 cases revealed discrepancies in the measurements of the vertebral heart score pertaining to 8 vertebrae. The VHS value was high in 95% of the cats in the suspicious group. The variation of VHS value in different cat breeds was very close. However, Persian cats have an average VHS of 8.16, which is high for brachycephalic cats although there are not many studies. In another study conducted on 59 healthy cats, 41% had heart disease and the most common cause was hypertrophic cardiomyopathy (Pemberton *et al.*, 2000); only 52% of apparently healthy cats with VHS >8 were diagnosed with cardiomyopathy. This finding may support the use of echocardiography for the definitive diagnosis of heart disease in cats with a VHS of 8 to 9.3. Measurement of plasma NT-proBNP, a peptide released in response to a cardiac problem, can be used to distinguish between cardiac and non-cardiac causes of respiratory distress. Cats with cardiac

causes have higher levels of NT-proBNP (Connolly *et al.*, 2009; Wess *et al.*, 2009). NT-proBNP concentration values >150 pmol/L were abnormal and suggestive of congestive heart failure in cats with respiratory symptoms. This test can be used to confirm the cause of shortness of breath. In our study, the reference value recommended by the kit was >440 pmol/L. In 10 cases with a VHS value of 8 and above, NT-proBNP concentrations were higher than >440 pmol/L, comparable to other studies. This scenario demonstrated a corresponding value in 50% of the 20 cases afflicted with cardiac issues.

In the available literature, no research was found to discuss VLAS measurements in cats. This project is the first study in cats in this sense, providing a reference value for future studies with more cases. The mean ± SD VLAS in the control group was 1.5±0.3 vertebrae. It was determined as 1.73±0.3 vertebrae in the group with suspected heart problem. However, there was no statistically significant relationship between VHS and VLAS (P=0.07), VHS and TWM (P=0.06), VHS and Nt-proBNP (P>0.05). However, in 50% of cats with high VHS, similar to VLAS, TWM and Nt-proBNP values were above the reference value. While VHS was elevated in only one case within the control group, other values yielded similar outcomes. However, no clinical problem was observed in this case. In a canine study, comparable findings between the VLAS measurements and echocardiography data were observed (Keene *et al.*, 2019), suggesting that a VLAS >2.3 vertebra could be used as a radiographic indicator of LA enlargement in dogs (Keene *et al.*, 2019). It is important to study more cats with heart disease in relation to breed and age in order to get complete results. In order to interpret these values correctly, they should be specific according to race and age classification.

Diagnosis of occult heart disease and heart failure by NT-proBNP testing has been reported (Connolly *et al.*, 2008; Fox *et al.*, 2011; Singletary *et al.*, 2012; Oyama *et al.*, 2013). In most, but not all cats, the NT-proBNP test has a high sensitivity and specificity for detection (Connolly *et al.*, 2008; Hsu *et al.*, 2009; Wess *et al.*, 2009; Fox *et al.*, 2011; Hezzel, 2016). A study involving 227 cats, assessed by 12 distinct cardiologists, stands out with the most extensive case pool demonstrating the effectiveness of NT-pro BNP. According to the findings obtained from echocardiographic examination, the NT-proBNP test was 91.2% specific and 85.8% sensitive in detecting underlying heart disease (Fox *et al.*, 2011). These results gave a positive predictive value of 90.7% and a negative predictive value of 86.7%. Therefore, in a positive cat, the clinician can be 90.7% sure that underlying heart disease is present and 86.7% sure that heart disease is not present in a negative test cat. In this study, the mean NT-proBNP value was found to be 434.4 pmol/L in the 10 cats from the control group without heart problems. The mean NT-proBNP value of the group with suspected heart disease was 448.25 pmol/L. The NT-proBNP value can be used as a reference in the detection of heart disease. However, the test is semi-quantitative and has some limitations. Non-cardiac diseases such as severe respiratory or renal diseases (creatinine level >2.8 mg/dL) may increase the likelihood of false positive results (Connolly *et al.*, 2008). Elevations in NT-proBNP levels may be observed in cats with severe arrhythmia, pulmonary hypertension, systemic hypertension, and hyperthyroidism (Sangster *et*

al., 2014). Connolly *et al.* (2009) showed median, 95% confidence intervals and range of serum NT-proBNP concentrations in the RD (respiratory distress)-CHF group (n=33) of 523 fmol/mL (95% CI 437.2–612.3; range 95–982), and in the RD-NC (non-cardiac) group (n=41): 45 fmol/mL (95% CI 54.03–115.29; range 6-394). In their study of 41 healthy cats and 33 cats with respiratory and cardiac failure, serum NT-proBNP concentrations were 45–614 pmol/mL in healthy cats and 523–1690 pmol/mL in patients. In their study on 167 cats with acute dyspnea, Fox *et al.* (2009) found heart failure in 101 cats and only respiratory problems in 66 cats. They stated that there was a significant difference between serum NT-proBNP levels in cats diagnosed with heart failure and those diagnosed with respiratory tract disease.

Furthermore, because the natriuretic peptide system operates constitutively along a physiological continuum of activity, circulating NT-proBNP concentrations in cardiac patients often overlap to some extent with clinically normal animals, particularly animals with mild or subclinical heart disease. NT-proBNP appears to be useful in assisting clinicians in assessing the risk or likelihood of detecting disease in cats with suspected heart disease. However, not NT-proBNP test, but echocardiography remains the gold standard in the diagnosis of heart disease. Research has shown that the results of the NT-proBNP test should be interpreted in conjunction with other findings from physical examination, medical history, echocardiography, chest radiography, ECG, arterial blood pressure measurement, and other clinicopathological tests (Maisel *et al.*, 2008; Singletary *et al.* 2012). Results of isolated measurements of NT-proBNP recorded in a subclinically affected dog or cat should not be used as an empirical basis for initiating therapy.

The study's limitation was the relatively small sample size of cats, which restricted the comprehensive assessment of the results within a broader context. In addition, a wide age range in cats was used in this study. Depending on the age range of cats, thoracic radiographs may reveal different heart shapes (Guglielmini & Diana, 2015) which may also affect the results of the study. Echocardiography was not used in this study because in cats with heart disease, especially in case of congestive heart failure problems, may occur during electrocardiography under sedation. Besides all this, further diagnostic methods, such as echocardiography, may explore whether medical treatment is necessary to prevent heart disease or alternatively, future clinical manifestations that will affect anaesthesia protocols for procedures.

CONCLUSION

The results of the study suggest that the measurement of VHS is radiographically inexpensive and easy to implement for cats. Case selection and evaluation are important in the radiologic evaluation of VHS. Radiographic measurements can be used to accurately diagnose advanced cardiomegaly problems. The reliability of radiographic measurements should be evaluated for the diagnosis of heart disease, especially in the early stage of heart disease or in mild heart disease. The radiologically obtained vertebral heart score (VHS), vertebral left atrial size (VLAS), and thoracic width measurements did not demonstrate any correlations with each other. In this study, no statistically significant relationship was found between these parameters. Nevertheless, a parallel rela-

tionship was noted between vertebral heart score (VHS) and vertebral left atrial size (VLAS), as well as between thoracic width measurement (TWM) and N-terminal pro-B-type natriuretic peptide (NT-proBNP) in 50% of the cases. In order to determine the reference ranges of VHS, VLAS, TWM, it will be useful to perform measurement for each breed and age. Serum NT-proBNP appears to be useful in helping clinicians assess the risk or likelihood of disease in cats with suspected heart disease. The NT-proBNP test is not the gold standard for diagnosing heart disease. VLAS has not been extensively studied in cats so it would be beneficial to perform more studies in this direction. It would be beneficial to conduct more studies in cats with suspected heart problems, taking into account breed, age and weight. All methods used in this study are designed for use as a first-line diagnosis; therefore, echocardiography is still needed to confirm the type of heart disease in cats.

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