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Clinico-diagnostic evaluation of cardiac diseases in dogs using cardiac biomarkers: A two-years study

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Abstract

The current study assessed changes in hematological, biochemical, and cardiac biomarkers in dogs with heart conditions (n = 52) compared to healthy controls (n = 6). Mild decreases in hemoglobin, packed cell volume, and erythrocyte count were found by hematological analysis, suggesting mild normocytic, normochromic anemia, which is frequently linked to chronic heart disease. A significant leukocytosis characterized by neutrophilia and lymphopenia reflected a stress leukogram, suggestive of systemic stress, hypoxia, or secondary infections. Biochemical assessment showed markedly elevated blood urea nitrogen and mildly increased serum creatinine levels in diseased dogs, indicating prerenal azotemia and possible cardiorenal syndrome. Serum albumin concentrations were significantly reduced, consistent with hemodilution, hepatic congestion, or chronic disease-associated malnutrition, whereas liver enzyme activities (ALT, AST) showed no significant changes. Cardiac biomarker evaluation demonstrated that cardiac troponin-I (cTnI) was undetectable in all healthy dogs but elevated in 70% of cardiac cases, reflecting myocardial injury. NT-proBNP concentrations were significantly increased in affected dogs, indicating cardiac wall stress and neurohormonal activation. Overall, the findings highlight that canine cardiac diseases are associated with mild anemia, stress-related leukocytic changes, azotemia, hypoalbuminemia, and significant elevations in cardiac biomarkers. These alterations, while not disease-specific individually, provide valuable diagnostic and prognostic insight when interpreted alongside clinical, electrocardiographic, and echocardiographic evaluations.

Keywords: Cardiac biomarkers, hematobiochemical, myocardial injury

1. Introduction

Cardiac diseases are a major health issue in dogs, contributing substantially to illness and mortality, particularly in middle-aged and senior animals. Both congenital and acquired heart conditions gradually reduce the efficiency of the heart and can lead to extensive systemic disturbances due to disrupted hemodynamics and stimulation of neurohormonal mechanisms (Smith *et al.*, 2015) [1]. Although extensive research and documentation on canine cardiac diseases exist worldwide, information from India remains limited. This scarcity may be attributed to low owner awareness and inadequate diagnostic facilities. (Devi *et al.*, 2009) [2]. Traditional tests such as biochemical, hematological, and coagulation profiles; urine and feces analysis; blood culture; and effusion analysis may show alterations in cardiovascular diseases but are generally not specific. Increasing attention has therefore been directed toward newer and more specific biomarkers capable of providing clinically relevant information about myocardial injury for use in the investigation and monitoring of patients with cardiac disease. In recent years, hematologic biomarkers such as cardiac troponins and natriuretic peptides have been extensively studied, and innovative research has also explored fecal proteomics and microbiota changes in dogs with cardiac disease. Interpretation of laboratory tests must always be integrated with clinical evolution, clinical signs, and other diagnostic modalities to achieve a comprehensive multimodal assessment of cardiac pathology and its potential complications (Gavazza *et al.*, 2020) [3]. Despite the growing body of knowledge, studies focusing on the incidence and clinicopathological features of canine cardiac diseases under Indian conditions, particularly in south Gujarat, are limited. Considering the regional variations in breed predispositions and management practices, such investigations are essential. The present study was therefore undertaken to evaluate the incidence of cardiac diseases and to investigate their haematobiochemical, and biomarker

changes in dogs presented to the Veterinary Clinical Complex, Kamdhenu University, Navsari, South Gujarat.

2. Materials and Methods

The present study was conducted from 2021 to 2022 in the Department of Veterinary Medicine in collaboration with the Veterinary Clinical Complex (VCC), College of Veterinary Science and Animal Husbandry, Kamdhenu university, Navsari, Gujarat, India. Out of 5566 dogs presented during this period, 52 dogs of different breeds, ages, and sexes with clinical suspicion of cardiac disease were included in the study, while six apparently healthy dogs brought for routine vaccination were selected as controls after obtaining informed owner consent. For laboratory investigations, blood samples were obtained aseptically from cephalic vein. Samples collected in EDTA tubes were used for hematological analysis, while those in plain vials were used for serum biochemical assays. Hematological values, including hemoglobin (Hb), packed cell volume (PCV), total erythrocyte count (RBC), total leukocyte count (TLC), and differential leukocyte count, were determined using an automated hematology analyzer (MEK-6420, Nihon Kohden India). Biochemical parameters, such as blood urea nitrogen (BUN), creatinine, total protein, albumin, alanine aminotransferase (ALT), and aspartate aminotransferase (AST), were analyzed using commercial kits and a semi-automated biochemical analyzer (Microlab-300, Q-line Biotech, India) (Kaneko *et al.*, 2008) [4]. Cardiac biomarkers were also evaluated. Serum cardiac troponin-I (cTnI) was assessed using a qualitative rapid test kit and confirmed by enzyme-linked immunosorbent assay (ELISA) where necessary. Plasma concentrations of N-terminal pro-B-type natriuretic peptide (NT-proBNP) were measured with a canine-specific ELISA kit according to the manufacturer's protocol. These markers were chosen for their established diagnostic accuracy in identifying myocardial damage and cardiac stress (Oyama & Singletary, 2010) [7].

3. Results and Discussion

3.1 Hematological Parameters in Cardiac Diseases

The hematological profile of dogs with cardiac diseases (n = 52) was compared to that of healthy controls (n = 6), and the mean values of the parameters are presented in the table. A marginal reduction in hemoglobin (10.79±0.28 g/dl) and packed cell volume (33.18±0.94%) was observed in affected dogs compared to controls (11.57±0.48 g/dl and 36.49±1.03%, respectively). Similarly, the RBC count was slightly lower in diseased dogs (5.23±0.09 ×10⁶/cumm) than in controls (5.85±0.18 ×10⁶/cumm). These findings point toward mild anemia, a common occurrence in chronic cardiac disease, often linked to reduced renal perfusion, decreased erythropoietin production, and fluid retention-induced hemodilution (Gavazza *et al.*, 2020; Martinelli *E. et al.*, 2016) [2, 5]. Comparable reductions in red cell indices have also been documented in canine cardiac insufficiency (Hoque, M., *et al.*, 2019) [6]. The total leukocyte count (TLC) was significantly higher in diseased dogs (14301±936/cumm) compared to controls (11066±912/cumm). Differential leukocyte analysis revealed a marked neutrophilia (74.75±1.34%) along with lymphopenia (19.78±1.23%), whereas eosinophils and monocytes showed minor decreases of limited diagnostic significance. The neutrophilic response coupled with

lymphocyte reduction reflects a stress leukogram, which may arise due to chronic heart failure, systemic hypoxia, or secondary infections (Hamilton-Elliott, *j et al.* 2018) [17]. Similar leukocyte alterations have been reported in dogs with cardiac disorders (Hoque, M., *et al.*, 2019) [6].

3.2 Biochemical Parameters in Cardiac Diseases

The biochemical parameters of dogs with cardiac diseases (n = 52) were compared to healthy controls (n = 6), and the mean values are presented in the table. A significant elevation in blood urea nitrogen (BUN) was observed in diseased dogs (44.39±4.28 mg/dl) compared to controls (25.66±2.37 mg/dl). Serum creatinine levels also showed a slight rise in affected dogs (0.72±0.06 mg/dl) relative to controls (0.61±0.05 mg/dl). These alterations are suggestive of prerenal azotemia, likely resulting from reduced renal blood flow and secondary renal involvement in advanced cardiac disease, commonly referred to as cardiorenal syndrome. Comparable findings of azotemia in dogs with congestive heart failure have been previously documented (Tidholm and Johnson, 1996; Dhanya Pai *et al.*, 2012; Deepa *et al.*, 2012 and Indhu *et al.*, 2019) [13, 14, 15, 16]. Total serum protein concentrations did not differ significantly between diseased (6.17±0.13 g/dl) and control dogs (6.35±0.24 g/dl). However, a notable reduction in serum albumin was observed in cardiac cases (1.65±0.05 g/dl) compared to controls (2.97±0.06 g/dl). The hypoalbuminemia may be attributed to hemodilution from fluid overload, protein losses associated with congestive hepatopathy, or poor nutritional status secondary to chronic illness (Patowary *et al.*, 2022) [12]. This finding is clinically important since reduced albumin contributes to edema and effusion formation by lowering plasma oncotic pressure. The activities of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) did not differ significantly between groups. ALT values averaged 44.46±3.45 IU/L in diseased dogs and 47.17±4.61 IU/L in controls, while AST levels were almost identical (27.03±1.31 vs. 27.17±4.80 IU/L). This suggests that hepatic enzyme leakage was not a consistent feature in the present study, although other investigations have reported elevated transaminases in dogs with right-sided congestive heart failure due to hepatic congestion (Boon, 2011) [11].

3.3 Cardiac Biomarkers in Cardiac Diseases

The evaluation of cardiac biomarkers demonstrated marked variations between dogs with cardiac disease (n = 52) and healthy controls (n = 6). Cardiac troponin-I (cTnI), a sensitive and specific indicator of myocardial injury, was undetectable in all control animals, whereas 70% of the diseased dogs showed positive results. The elevation of cTnI reflects myocyte injury and the release of structural proteins into circulation, consistent with earlier observations (Oyama & Sisson, 2004; Sleeper *et al.*, 2001) [9, 10]. Similarly, N-terminal pro B-type natriuretic peptide (NT-proBNP) concentrations were significantly increased (p<0.001) in affected dogs (527.46±29.38 pg/ml) compared to controls (226.34±32.95 pg/ml). N-terminal pro-B-type natriuretic peptide (NT-proBNP) measurement has become an important diagnostic tool in veterinary cardiology over the past decade. NT-proBNP levels are significantly elevated in dogs with mitral valve disease and dilated cardiomyopathy, with the highest concentrations typically observed in those that progress to congestive heart failure. This biomarker is

especially valuable in differentiating congestive heart failure from primary respiratory tract diseases when evaluating dogs presenting with respiratory signs (Boswood *et al.*, 2008). The concentration of NT-proBNP in the blood correlates strongly with the severity of cardiac disease and

helps predict the likelihood of future complications. Growing evidence indicates that NT-proBNP is a valuable tool for diagnosing cardiac disorders in dogs, assessing disease severity, and providing prognostic information in affected patients (Oyama & Singletary, 2010)^[7].

Table 1: Hematological, Biochemical, and Cardiac Biomarker Parameters in Dogs with Cardiac Diseases

Sr. No.	Parameter	Control (n = 6)	Cardiac Diseases (n = 52)
Hematological Parameters			
1	Hemoglobin (g/dl)	11.57±0.48	10.79±0.28
2	Packed Cell Volume (%)	36.49±1.03	33.18±0.94
3	RBC (×10 ⁶ /cumm)	5.85±0.18	5.23±0.09
4	WBC (×10 ³ /cumm)	11066±912	14301±936
5	Neutrophils (%)	70.66±0.71	74.75±1.34
6	Lymphocytes (%)	22.50±0.76	19.78±1.23
7	Eosinophils (%)	2.30±0.33	2.11±0.13
8	Monocytes (%)	4.50±0.56	3.32±0.26
Biochemical Parameters			
1	Blood Urea Nitrogen (mg/dl)	25.66±2.37	44.39±4.28
2	Creatinine (mg/dl)	0.61±0.05	0.72±0.06
3	Total Protein (g/dl)	6.35±0.24	6.17±0.13
4	Albumin (g/dl)	2.97±0.06*	1.65±0.05*
5	ALT (IU/L)	47.17±4.61	44.46±3.45
6	AST (IU/L)	27.17±4.80	27.03±1.31
* indicate significant difference at <0.001			
Cardiac Biomarkers			
1	Cardiac Troponin-I (ng/ml)	All Negative	70% Positive
2	NT-proBNP (pg/ml)	226.34±32.95*	527.46±29.38*
* Significant difference at <i>p</i> <0.001			

4. Conclusion

The present study highlights the significant prevalence of cardiac diseases in dogs presented to the Veterinary Clinical Complex, Kamdhenu University, Navsari, South Gujarat. Despite the global availability of extensive literature on canine cardiology, regional data from India particularly Gujarat remain limited. The findings of this study underline the importance of early recognition and comprehensive diagnostic evaluation of cardiac disorders in canines. Hematological assessment revealed mild anemia and leukogram alterations consistent with stress and chronic cardiac compromise. Biochemical analysis indicated evidence of prerenal azotemia and hypoalbuminemia, suggesting early cardiorenal involvement and altered hepatic or nutritional status in affected dogs. Among the biomarkers evaluated, cardiac troponin-I and NT-proBNP proved highly valuable, with significant elevations indicating myocardial damage and increased cardiac stress. The marked rise in NT-proBNP reinforces its diagnostic utility, particularly in distinguishing cardiac from respiratory causes of clinical signs and in assessing disease severity and prognosis. Overall, the study demonstrates that integrating routine hematological and biochemical testing with advanced cardiac biomarkers substantially enhances diagnostic accuracy in canine cardiac disease. The results emphasize the need for increased awareness among dog owners and improved access to diagnostic facilities in the region. Continued research in diverse geographic and clinical settings across India is essential to better understand breed predispositions, disease patterns, and effective management strategies for canine cardiac disorders.

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