

Getting to the Heart of Cardiac Biomarkers

presented by dvm360



with Courtney Smith, DVM, MS, DACVIM (Cardiology)

Summary

In October of 2020 Courtney Smith, DVM, MS, DACVIM (Cardiology) along with dvm360's Adam Christman, DVM, MBA discussed the importance of cardiac biomarkers and the various diseases relating to the heart. Real life case studies were also presented. Below is the outline of the webinar presentation.

Outline

- What is a biomarker?
- Cardiac specific biomarkers
- NT-proBNP
- Testing options
- Brief review of canine heart disease
- Literature review
- Real world applications

What is a Biomarker¹?

- Combination of two words: biological & marker
- NIH Biomarkers Definitions Working Group definition:
 - “a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention”

Simple Summary

- Objective, quantifiable characteristic of a biological process

Ideal Biomarker

- Highly specific for organ/disease process
- Sensitive to detect small changes
- Detectable close to time of change/injury
- Easily quantified
- Affordable

Cardiac Specific Biomarkers³

- Myocardial necrosis/cell injury
 - Troponins⁴
 - cTnI and cTnT
- Hemodynamic stress
 - Natriuretic peptides & fragments
 - ANP, BNP, NT-proBNP, CNP
- Homeostasis of serum lipoproteins
 - High-density & low-density lipoproteins
- Cardiovascular inflammation
 - C-reactive protein

Natriuretic Peptide Family

- Structurally similar hormone family
 - ANP (atrial), BNP, and CNP (C-type)
- Key players in regulation of intravascular volume & syst. pressures
- Natriuresis - excretion of sodium in urine

B-type Natriuretic Peptide^{2,5,6}

- Stored as precursor hormone
 - In multiple regions of heart
- Myocardial stretch triggers release:
- BNP Fragment
 - Bioactive
 - Half-life ~90 sec
- N-terminal proBNP
 - Inactive
 - Half-life ~120 min
 - Increased stability

BNP

- Acts on the kidneys & peripheral circulation to unload the heart by:
 - Increasing excretion of sodium and water
 - Inhibiting release of renin and aldosterone
 - Inducing vasorelaxation

BNP Breakdown Via:

- Neutral endopeptidase enzymes within the circulation, liver, lungs and kidneys
- A specific natriuretic-peptide receptor binding degradation without activity

**Less is known about NT-proBNP breakdown - renal excretion is suspected main pathway **

Testing Options

- Utilize ELISA assay testing method
 - Reference lab testing
 - Accurate, quantifiable results
 - Not rapid
 - Point of care testing
 - Utilized mainly for cats
 - Most tests don't provide quantifiable results, with exception of the Vcheck test by Bionote

Vcheck Canine NT-proBNP

- Species: Dog
- Sample: Serum 100µl
- Testing Time: 15 minutes
- Measurement: Quantitative
- Measurement Range: 500-1,00 pmol/L
- Storage Condition: 2-8°C

Reference Range

- <900 pmol/L Normal
- 900-1,800 pmol/L Suspected*
- >1,800 pmol/L Abnormal*

*'Abnormal' or 'Suspected' NT-proBNP results should always be interpreted in combination and other diagnostic findings

** Concentration over 735 pmol/L in Doberman Pinschers indicated an increased risk for occult dilated cardiomyopathy

Vcheck Feline NT-proBNP

Important Note

- NT-proBNP levels above upper cutoff should never be used as sole parameter to diagnose heart disease²

Heart Basics

Myxomatous Mitral Valve Disease (MMVD)^{7,8}

- Slow progressive degeneration of the MV apparatus
- Subsequent left atrial and ventricular dilation

MMVD

- Cellular & intracellular matrix changes within leaflets
- Progressive deformation of leaflets impairs closure = valve leakage
- Progressive regurgitation = increased cardiac work & chamber remodeling
- Left sided enlargement = more regurgitation
- Est. ~10% of dogs have heart dz
 - ~75% of these cases = MMVD
- Inc. prevalence in dogs <20kg
- ~1.5 times more common in males
- Not all dogs with MMVD develop heart failure

MMVD Categories

- ACVIM Stage A
 - Dogs at higher risk without apparent disease
- ACVIM Stage B
 - Asymptomatic MMVD
 - B1: MR with minimal cardiac remodeling
 - B2: MR severe enough to result in enlargement sufficient enough to recommend medical management
- ACVIM Stage C
 - Severe enough disease to cause current or past clinical signs of CHF
- ACVIM Stage D
 - Clinical signs refractory to standard CHF therapy

Ways to Diagnose MMVD

- Physical Exam - left apical systolic murmur with appropriate signalment
- Gold standard - echo
- Thoracic radiographs

**Diagnostic challenge - often not to establish if MMVD present, BUT determination of whether MMVD is responsible for clinical signs

CHF Risk Factors

- Increased age
- Progressive heart enlargement
- Increased trans-mitral E wave flow velocity (echo)
- Increased LA and LV dimensions
- Increased NT-proBNP concentrations
- Increased resting heart rate and resting respiratory rate

Dilated Cardiomyopathy (DCM)⁹

- Final result of various myocardial insults
 - Genetic, nutritional, toxic, viral, tachycardia
- Signalment - large and medium dog breeds
 - Doberman Pinscher, Irish wolfhound, Great Dane, Cocker spaniel
- 2 Stages:
 - Asymptomatic (occult)
 - Symptomatic (overt)

DCM

- Enlargement and impaired contractility of one or both ventricles

Diagnosing Cardiac Disease

- What question are we trying to answer?
 - What's causing the coughing?
 - Are these signs of CHF?
 - What's the risk for CHF?
 - Is heart disease present?
- Heart disease ≠ heart failure

Clinical Signs of CHF

- Coughing
 - Mainstem bronchial compression by LA enlargement
- Exercise intolerance
- Tachypnea and dyspnea
 - Increased lung sounds on auscultation
 - Increased respiratory rate
- Night time restlessness/changes in sleeping position
- Decreased appetite
- Syncope

Differential Diagnoses

- Lower airway disease
- Tracheal instability
- Pulmonary fibrosis
- Neoplasia
- Heartworm disease
- Pneumonia

Diagnosing CHF

- Gold standard - thoracic radiographs
 - LA enlargement
 - LV enlargement
 - Pulmonary venous distension
 - Cardiogenic pulmonary congestion/edema
- Echocardiogram - can help provide evidence of disease & elevated LA pressures
- Physical examination
- History/presenting complaint
- NT-proBNP

** Can't diagnose CHF with just one criterion

NT-proBNP & MMVD¹⁰

- Can typically discriminate between normal dogs and dogs with CHF¹¹⁻¹⁷
- May have some ability to distinguish severity of MMVD¹⁵
 - LA size via echo or thoracic radiographs more specific¹⁰

- Can provide information on risk for development of CHF in dogs with MMVD^{18,19}

Group		Mean Plasma (NT-proBNP)
1	Asymptomatic, no HD	906.8 ± 1,173.4
2	Asymptomatic, compensated HD	1,593.8 ± 1,292.5
3	Respiratory distress, non-cardiogenic	2,064.5 ± 2,064.0
4	Respiratory distress, CHF	6,143.3 ± 4,517.3

Low NT-proBNP result with dyspnea makes L-CHF unlikely^{2,7,8,10,20}

Factors assoc. with inc. CHF risk^{18,19}

- VHS > 12.0 or...
 - rate of increase ≥ 0.07 per month since last visit
 - NT-proBNP >1500pmol/L or...
 - rate of increase > 750pmol/L
 - 60% since last visit
- = inc. risk for CHF development in 3-6 months

Take Home Message

- Owner communication of inc. risk & monitoring
- More frequent rechecks recommended
- Importance of in-home monitoring of SRR21

NT-proBNP & DCM

- NT-proBNP can help with diagnosis of occult DCM in Dobermans
 - Along with Holter monitor documenting >50 VPCs/24 hours²²
 - Can assist with prognosis/survival²²⁻²⁴

Testing Considerations...

- In a normal population there's a greater chance for false positives than in a population of dogs with heart disease^{10,18}

False Positive Causes

- Breed-related variation^{2,10}
 - Labradors and greyhounds
- Inc. levels with kidney disease²⁵
- Normal daily variability²⁶
- Heterophilic antibody interference²⁷
 - Animal's own antibodies bind to the test antigens

Cardiac Evaluation

- History
- Physical Exam
 - Concern for heart disease
 - Thoracic radiographs and/or echocardiogram
 - NT-proBNP?
 - Determining cause of dyspnea
 - Thoracic radiographs and/or echocardiogram
 - +/- NT-proBNP
 - Still equivocal, furosemide trial with monitoring SRR/RRR

Case Examples

Doberman Pinscher

- 6-year-old NM Doberman
- Asymptomatic, annual wellness exam
- Family aware of risk for cardiac disease
- PE: NSF

Maltese

- 10-year-old FS Maltese-poodle mix
- Coughing more, exercise intolerance
- PE: IV/VI left apical systolic heart murmur, regular rhythm, mildly tachycardic (HR: 160bpm), panting comfortably with mildly increased BV sounds, bad dental disease

Cavalier King Charles Spaniel

- 10-year-old MN CKCS
- Mildly inc. coughing, annual wellness exam
- Prior history: Diagnosed with MMVD ACVIM B2 ~12 months prior with echocardiogram and started pimobendan
- PE: Grade V/VI L apical systolic heart murmur, HR: 120bpm with regular rhythm, panting comfortably, clear lung sounds

Chihuahua

- 12-year-old NM Chihuahua
- Coughing at night for "awhile" with a severe inc. in cough for 24 hours
- PE: Grade III/VI L apical systolic heart murmur, HR: 120bpm, RR: 32/min, fine pulmonary crackles bilaterally with inc. BV sounds, coughing with stress/excitement

Others...

- 3-year-old asymptomatic Labrador retriever
- 16-year-old mixed breed with a new low intensity HM and CKD
- 5-year-old Labradoodle fed grain-free diet their whole life

Final Thoughts

- What can NT-proBNP tell us about the heart?
- Do you think you will use NT-proBNP testing more frequently?

Literature

1. Libby P, Gerszten RE, and Ridker PM: Biomarkers, Proteomics, Metabolomics, and Personalized Medicine. DL Mann DP Zipes P Libby et al. Braunwald's heart disease: a textbook of cardiovascular medicine. ed 10 2015 Saunders Philadelphia 84-92
2. Viera de Lima G and de Silveira Ferreira F: N-terminal-pro brain natriuretic peptides in dogs and cats: A technical and clinical review. *Vet World*. 10: 1072-1082 2017.
3. Jacob R and Khan M: Cardiac Biomarkers What Is and What Can Be. *Indian J Cardiovasc Dis Women WINCARS*. 3:240-244 2018
4. Langhorn R and Willesen JL: Cardiac Troponins in Dogs and Cats. *J Vet Intern Med*. 30: 36-50 2016
5. Scollan KF and Sisson DD: Pathophysiology of Heart Failure. Ettinger SJ Feldman EC Cote E. *Textbook of Veterinary Internal Medicine*. ed 8 2017 Elsevier St. Louis 1153-1162
6. Kim YS, Karisa N, Jeon WY, et al: High-level production of N-terminal pro-brain natriuretic peptide, as a calibrant of heart failure diagnosis, in *Escherichia coli*. *Appl Microbiol Biotechnol*. 103: 4779-4788 2019
7. Ljungvall I and Haggstrom J: Adult-Onset of Valvular Heart Disease. Ettinger SJ Feldman EC Cote E. *Textbook of Veterinary Internal Medicine*. ed 8 2017 Elsevier St. Louis 1250-1269
8. Keene BW, Atkins CE, Bonagura JD, et al: ACVIM consensus guideline for the diagnosis and treatment of myxomatous mitral valve disease in dogs. *J Vet Intern Med*. 33: 1127-1140 2019
9. Stern JA and Meurs KM: Myocardial Disease. Ettinger SJ Feldman EC Cote E. *Textbook of Veterinary Internal Medicine*. ed 8 2017 Elsevier St. Louis 1269-1277
10. Kittleson MD and Rishniw M: NT-proBNP Assay. NT-proBNP Assay- Medical FAQs. *VIN*. 2 July 2020. www.vin.com/doc/?id=3859730&pid=11200
11. Boswood A, Dukes-McEwan J, Loureiro J, et al: The diagnostic accuracy of different natriuretic peptides in the investigation of canine cardiac disease. *J Small Anim Pract*. 49: 26-32 2008
12. Tarnow I, Olsen LH, Kvarn C, et al: Predictive value of natriuretic peptides in dogs with mitral valve disease. *Vet J*. 180:195-201 2009
13. Hezzell MJ, Boswood A, Chang YM, et al: The combined prognostic potential of serum high-sensitivity cardiac troponin I and N-terminal pro-B-type natriuretic peptide concentrations in dogs with degenerative mitral valve disease. *J Vet Intern Med*. 26: 302-11 2012
14. Moonarmart W, Boswood A, Luis Fuentes V, et al: N-terminal pro B-type natriuretic peptide and left ventricular diameter independently predict mortality in dogs with mitral valve disease. *J Small Anim Pract*. 51: 84-96 2010
15. Fox PR, Oyama MA, Hezzell MJ, et al: Relationship of Plasma N terminal Pro brain Natriuretic Peptide Concentrations to Heart Failure Classification and Cause of Respiratory Distress in Dogs Using a 2nd Generation ELISA Assay. *J Vet Intern Med*. 29: 171-179 2015
16. Chetboul V, Serres F, Tissier R, et al: Association of plasma N-terminal pro-B-type natriuretic peptide concentration with mitral regurgitation severity and outcome in dogs with asymptomatic degenerative mitral valve disease. *J Vet Intern Med*. 23: 984-994 2009
17. Prosek R, Sisson DD, Oyama MA, et al: Distinguishing cardiac and non-cardiac dyspnea in 48 dogs using plasma atrial natriuretic factor, B-Type Natriuretic Factor, endothelin and cardiac troponin-I. *J Vet Intern Med*. 21:238-242 2007
18. Gordon SG: NT-proBNP Testing in the Dog. Circulations conversations with a cardiologist. CardiacEducationGroup.org. October 2014 www.cardiaceducationgroup.org/wp-content/uploads/2015/10/CEG_Circulations_Canine-Biomarkers_FINAL.pdf
19. Reynolds CA, Brown DC, Rush JE, et al: Prediction of first onset of congestive heart failure in dogs with degenerative mitral valve disease: the PREDICT cohort study. *J Vet Cardiol*. 14: 193-202 2012
20. Fine DM, DeClue AE, Reinero CR: Evaluation of circulating amino terminal-pro-B-type natriuretic peptide concentration in dogs with respiratory distress attributable to congestive heart failure or primary pulmonary disease. *J Am Vet Med Assoc*. 232: 1674-1679 2008
21. DG Ohad, M Rishniw, I Ljungvall, et al: Sleeping and resting respiratory rates in dogs with subclinical heart disease. *J Am Vet Med Assoc*. 243: 839-843 2013
22. Singletary GE, Morris NA, Lynne O'Sullivan M, et al: Prospective evaluation of NTproBNP assay to detect occult dilated cardiomyopathy and predict survival in Doberman Pinschers. *J Vet Intern Med*. 26: 1330-1336 2012
23. Wess G, Butz V, Mahling M, et al: Evaluation of N-terminal pro-B-type natriuretic peptide as a diagnostic marker of various stages of cardiomyopathy in Doberman Pinschers. *Am J Vet Res*. 72: 642-649 2011
24. Klüser L, Holler P, Simak J, et al: Predictors of Sudden Cardiac Death in Doberman Pinschers with Dilated Cardiomyopathy. *J Vet Intern Med*. 30: 722-732 2016
25. Raffan E, Loureiro J, Dukes-McEwan J, et al: The Cardiac Biomarker NTproBNP Is Increased in Dogs with Azotemia. *J Vet Intern Med*. 23: 1184-1189 2009
26. Ruaux C, Scollan K, Suchodolski JS, et al: Biologic variability in NT-proBNP and cardiac troponin-I in healthy dogs and dogs with mitral valve degeneration. *Vet Clin Pathol*. 44: 420-430 2015
27. Solter PF, Oyama MA, Sisson DD: Canine heterophilic antibodies as a source of False-positive B-type natriuretic peptide sandwich ELISA results. *Vet Clin Pathol*. 37: 86-95 2008