

Quantitative marker of myocardial injury, Troponin I

# Vcheck Canine Tnl

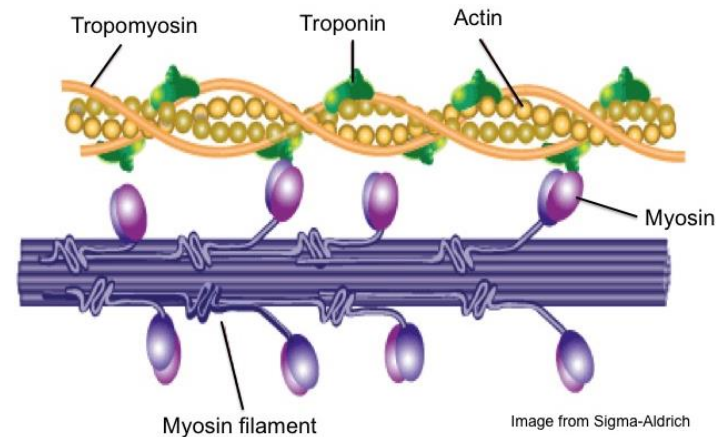
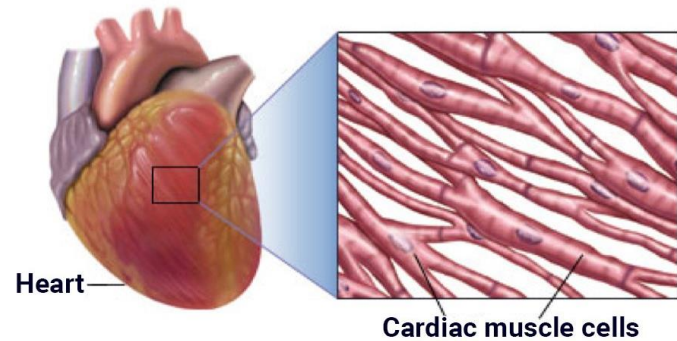
BIONOTE Marketing team

Aug 2021



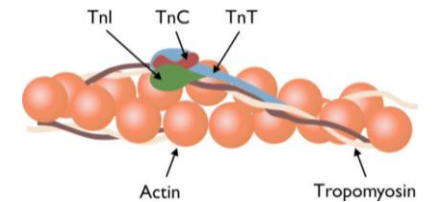
# New Cardiac Biomarker, TnI

## What is TnI?



In the Heart Muscle,  
The contractile apparatus is composed of ...

- **Actin**
- **Myosin**
- **Tropomyosin**
- **Troponin complex** (Regulatory proteins)
  - 1) Troponin T: binds to Tropomyosin
  - 2) Troponin C: binds to  $\text{Ca}^{2+}$
  - 3) **Troponin I**: inhibits the interaction



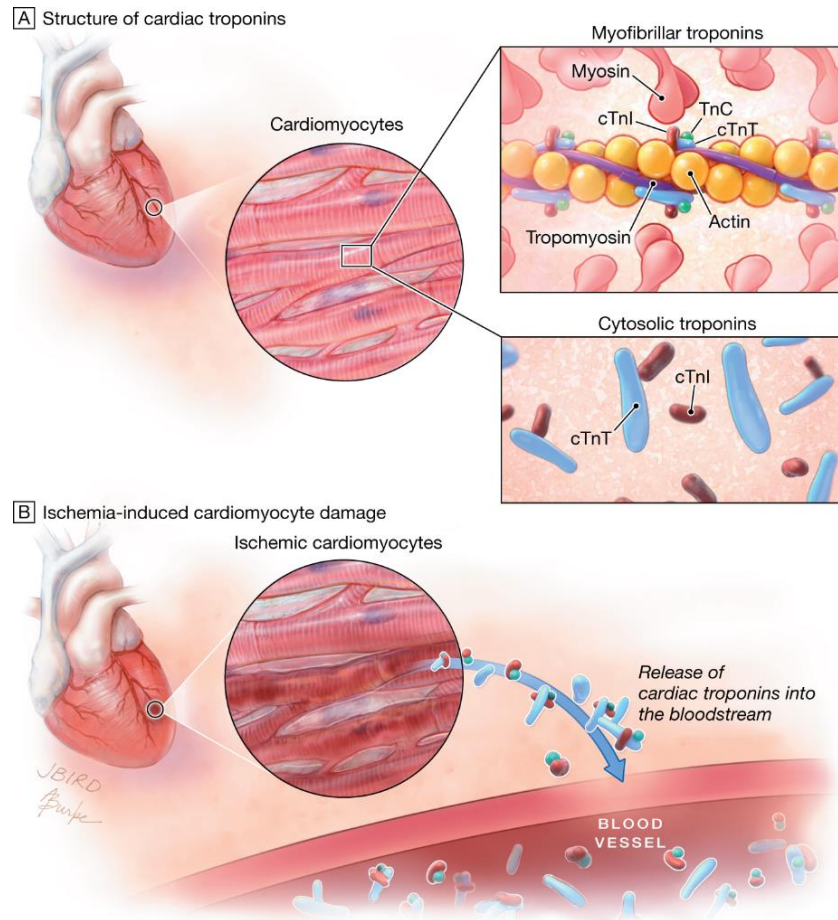
- ✓ Troponin I (TnI): cardiac, skeletal isoforms  
→ Only measures cardiac TnI
- ✓ **Cardiac TnI is a more sensitive marker of myocardial injury**  
when compared to cardiac TnT.

Click!



# New Cardiac Biomarker, TnI

## What is TnI?



- ✓ After cardiac insult, a rise of **Troponins** can be seen within 2-3 hours, and peak concentration is frequently reached in 18-24 hours.
  - A biomarker for the diagnosis of acute myocardial infarction (AMI) in humans, but AMI occurs very rarely in dogs and cats
- ✓ Still, dogs with cardiac diseases have chronically increased **Troponin** concentrations > signifying ongoing myocardial injury

# When TnI levels increase

In occasions which lead to cardiac injury,



## Cardiac Trauma

- hit-by-car trauma
- high-rise syndrome
- thoracic bite injuries

TnI for detecting or ruling out significant blunt cardiac injury



## Heart Disease

- mitral valve disease (MMVD)
- cardiomyopathy (DCM)
- congenital heart disease

TnI for signifying ongoing myocardial injury (worsening of cardiac function)



## Non-Cardiac Disease

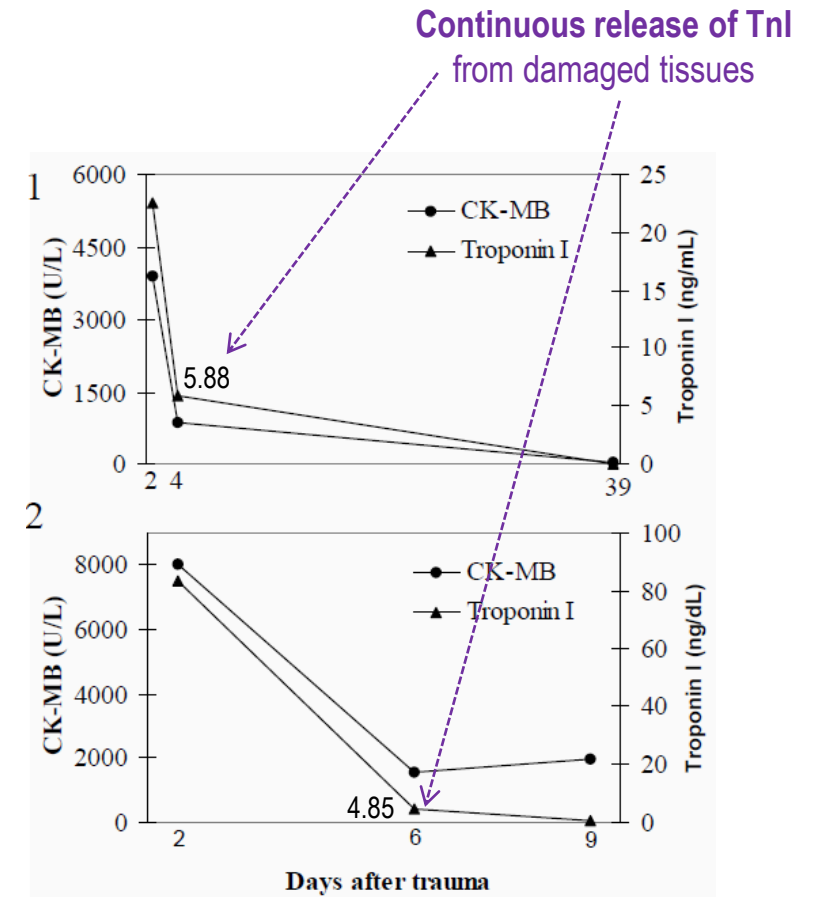
- inflammatory diseases
- neoplasia
- shock

TnI for discovering myocardial injury in critically ill individuals

# Clinical Utility of TnI in Dogs

## 1) After Cardiac Trauma

- **Direct cardiac trauma** occurs frequently in extreme conditions (hit-by-car trauma, high-rise syndrome, thoracic bite injuries)
  - The diagnosis of traumatic injury to the heart is important as it can lead to cardiogenic shock, acute heart failure, life-threatening arrhythmias, or structural damage.
- TnI levels accurately indicate **myocardial injury secondary to trauma.**



\*Normal range of TnI: < 0.03 ng/ml

\*Half-life of TnI: 1.85 h



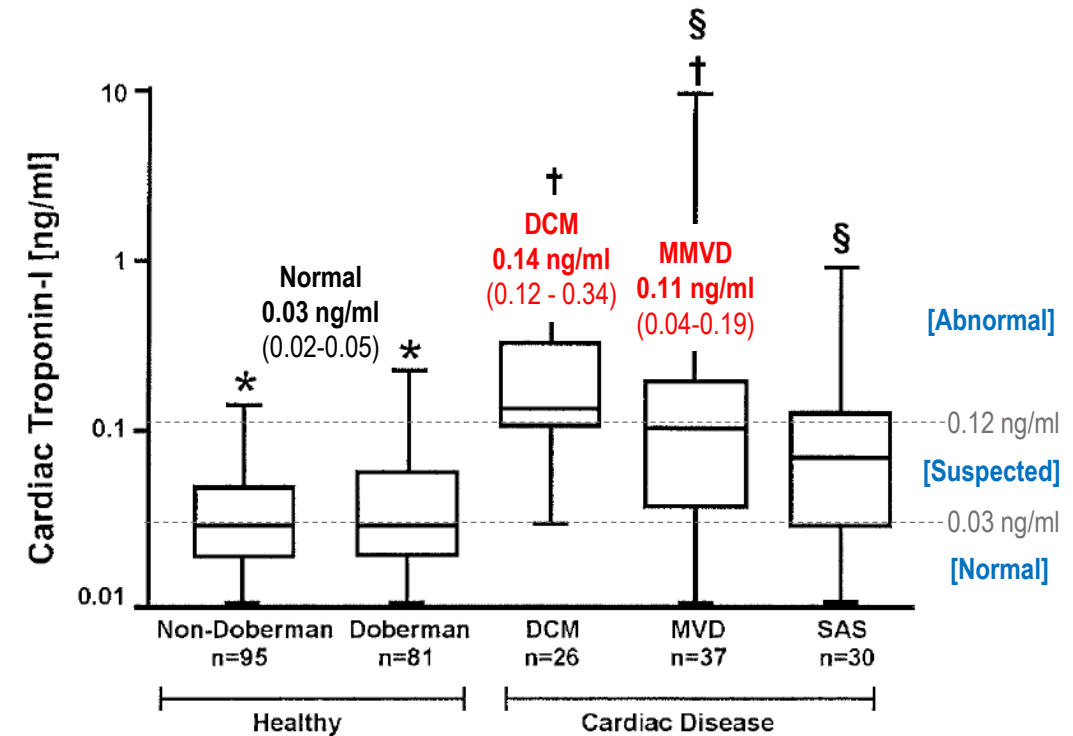
# Clinical Utility of TnI in Dogs

## 2) For Dogs with Primary Heart Diseases

- Increased TnI level means ongoing myocardial injury (damage).
  - 1) TnI levels are increased in dogs with heart disease.
    - **DCM**: median 0.14 ng/ml (0.12-0.34)
    - **MMVD**: median 0.11 ng/ml (0.04-0.19)
  - 2) TnI levels are significantly correlated with the left heart size. (correlation with La:Ao, LVDD, LVDs)



**TnI** for quantifying the extent of myocardial injury  
+ **Conventional examination** (X-ray, echocardiography)  
→ **Comprehensive evaluation for cardiac function**



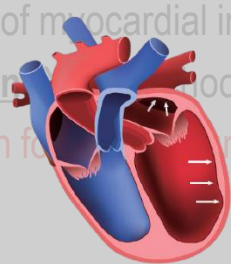
# Clinical Utility of Tnl in Dogs

## 2) For Dogs with Primary Heart Diseases

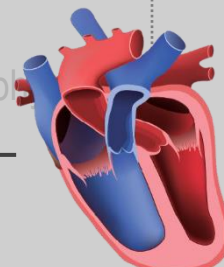
### DCM (Dilated Cardiomyopathy)

: Most common heart disease in large breeds  
(10% of all heart disease)

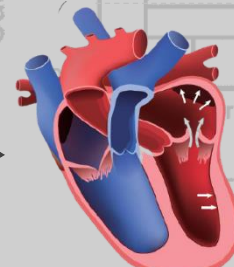
- **Age:** 3–7 years old
- **Breeds at elevated risk:** Large breeds (Boxer Dogs, Doberman Pinschers, Great Danes, Irish Wolfhounds, Saint Bernards)
- **Conditions:** Ventricular wall thinning  
→ Poor pumping ability



DCM



Normal

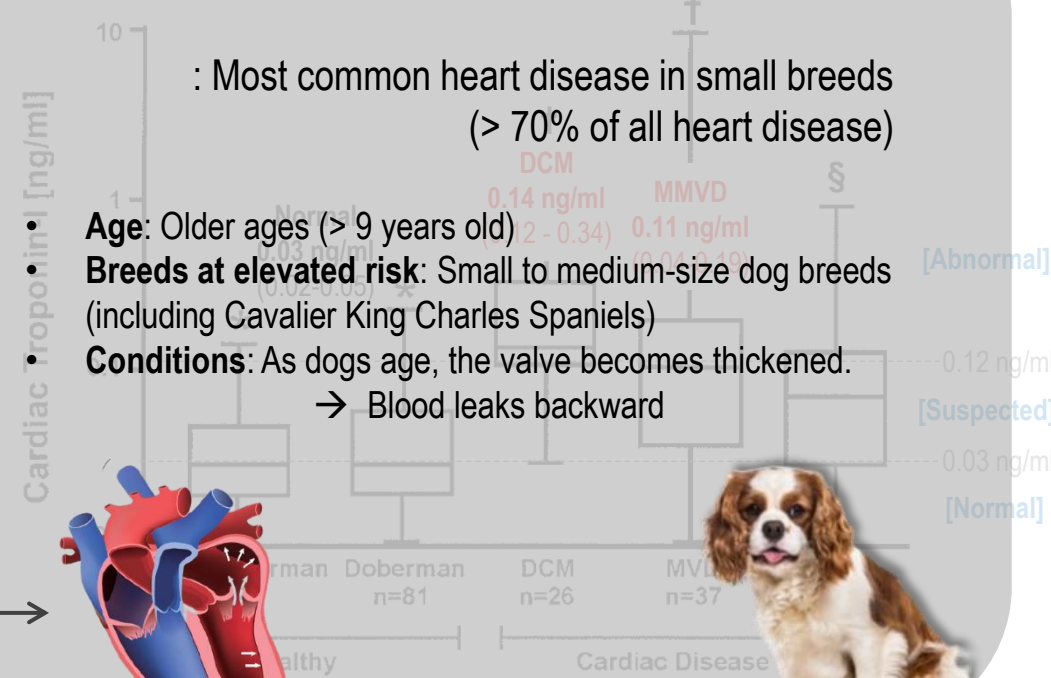


MMVD

### MMVD (Mitral valve disease)

: Most common heart disease in small breeds  
(> 70% of all heart disease)

- **Age:** Older ages (> 9 years old)
- **Breeds at elevated risk:** Small to medium-size dog breeds (including Cavalier King Charles Spaniels)
- **Conditions:** As dogs age, the valve becomes thickened.  
→ Blood leaks backward

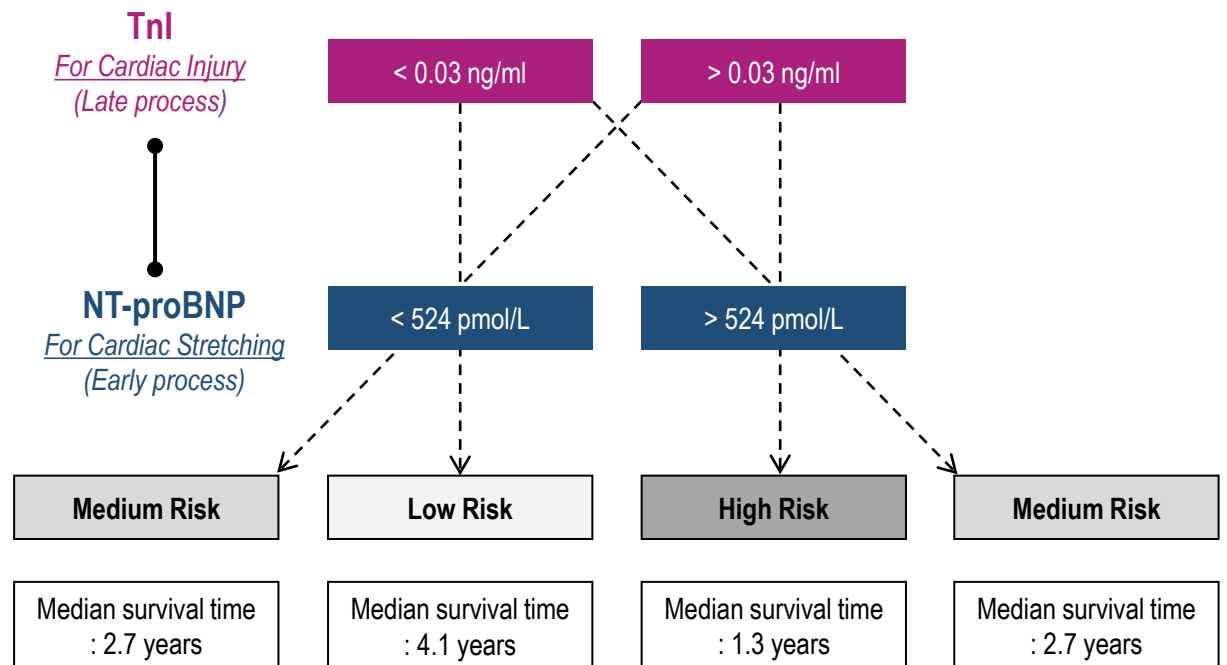


# Clinical Utility of Tnl in Dogs

## 2) For Dogs with Primary Heart Diseases

- Tnl and NT-proBNP should be measured in combination as a comprehensive evaluation.
  - **Tnl** for cardiac muscle injury
  - **NT-proBNP** for cardiac muscle stretching
- Combined measurement of Tnl and NT-proBNP is prognostically superior to measuring each alone.
- Monitor the rates of the changes of these markers **every 6 months** for further information

[Prognostic Algorithm] For dogs with MMVD of varying severity

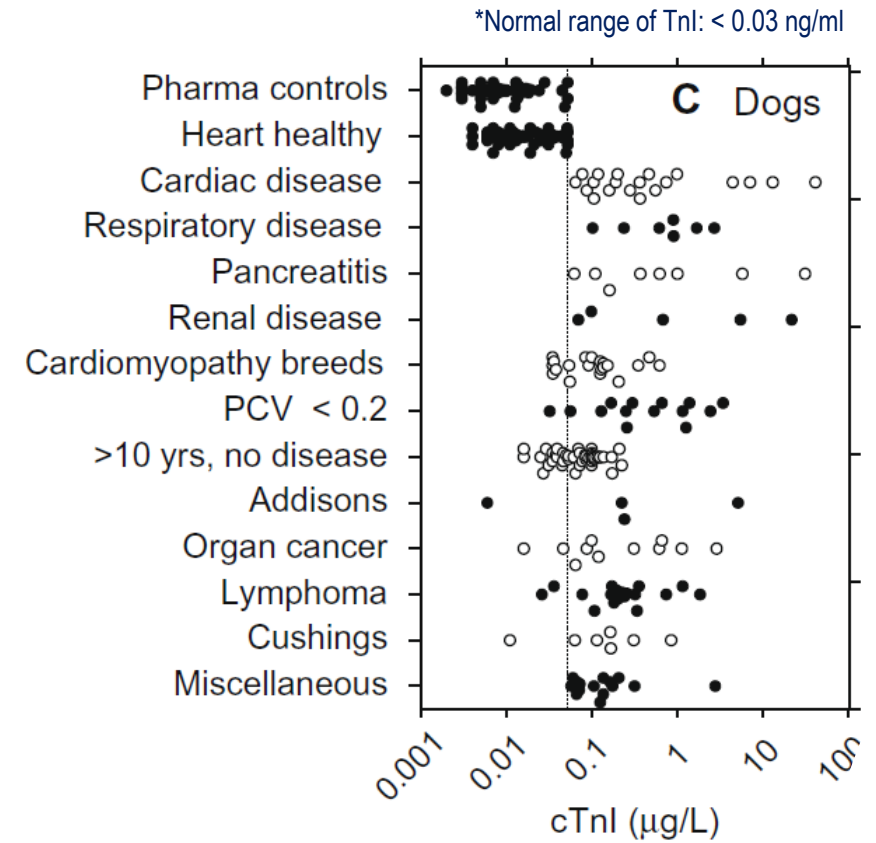




# Clinical Utility of Tnl in Dogs

## 3) In Dogs with Non-Cardiac Diseases

- Noncardiac critical disease can affect the heart, **causing myocardial injury**.
  - > **Need for close follow-up after hospital discharge**
- Interestingly, critically ill patients with noncardiac disease often have higher Tnl concentrations than the patients with severe primary cardiac disease.
- **Various diseases increasing Tnl levels**
  - Systemic inflammation
  - Parvoviral enteritis (parvovirus infection)
  - Pancreatitis
  - Cancer, lymphoma
  - Uncontrolled hyper(hypo)adrenocorticism
  - Respiratory disease
  - Anemia (moderate-marked)
  - Gastric dilatation-volvulus (GDV)
  - Infectious diseases (leptospirosis, leishmaniasis, babesiosis, ehrlichiosis)



# Clinical Utility of Tnl in Dogs

\*Normal range of Tnl: < 0.03 ng/ml

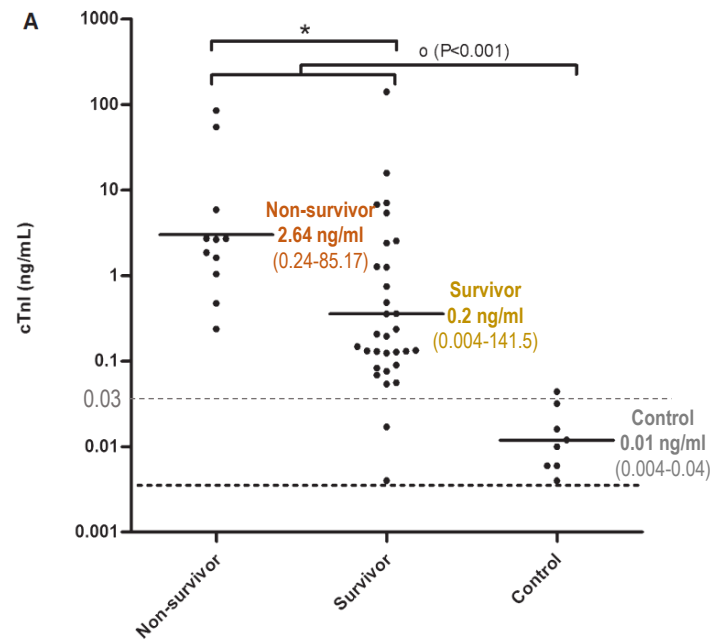
## 3) In Dogs with Non-cardiac Diseases

*For dogs with systemic inflammation*

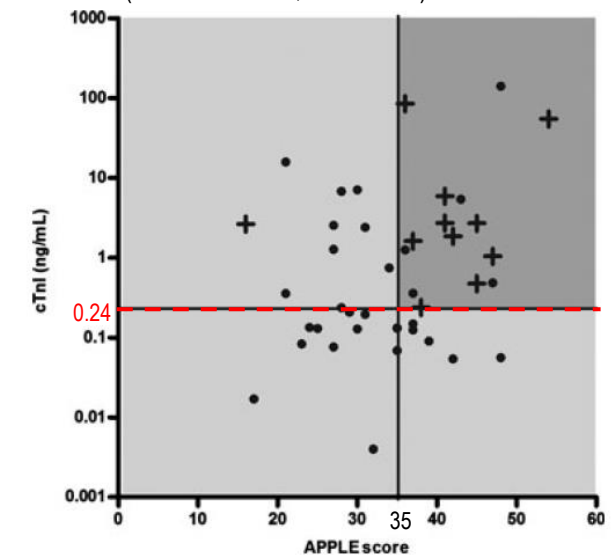
- Measurement of Tnl is necessary to discover the involvement of **myocardial injury in a patient's clinical status**.
- Presence of myocardial injury (high Tnl) is predictive of short-term survival (28-day case fatality).
- Cardiac Tnl is a marker of myocardial injury contributed independently to the APPLE score.
  - 1) APPLE score > 35
  - 2) Tnl > 0.24 ng/ml

→ Provides additional prognostic information

Graph 1. Tnl concentrations in dogs with systemic inflammation



Graph 2. The prognostic contribution of Tnl to APPLE score (+: non-survivors, •: survivors)



※APPLE: Acute Patient Physiologic and Laboratory Evaluation (Diagnosis-independent severity scores)



Product Introduction

# Vcheck Canine Tnl

# Product Introduction

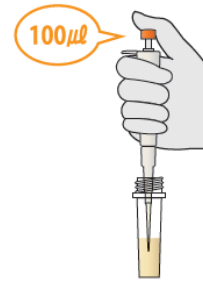
## Vcheck Canine Tnl

Quantitative marker of myocardial injury



- **Species:** Dog
- **Sample:** Serum 100  $\mu$ l
- **Testing Time:** 10 minutes
- **Measurement :** Quantitative
- **Measurement Range:** 0.01 – 20 ng/ml
- **Storage Condition:** 1 - 30 °C

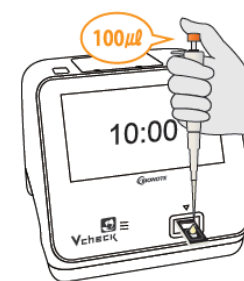
1 Add 100  $\mu$ l of the sample to the assay diluent tube



2 Mix well 5-6 times by using a 100  $\mu$ l pipette



3 Add 100  $\mu$ l of the mixed sample into the test device



※Samples should be tested immediately after collection.

(If not, freeze the samples at -20 °C or below for storage. Do not freeze and thaw repeatedly.)

| < 0.03 ng/ml | 0.03 – 0.12 ng/ml                             | > 0.12 ng/ml                                      |
|--------------|---|---|
| Normal       | Suspected<br>Possibility of myocardial injury | Abnormal<br>High possibility of myocardial injury |

※Tnl concentrations should not be used to either confirm or exclude primary cardiac disease without the simultaneous use of echocardiography.

# Product Introduction

## Vcheck Canine Tnl

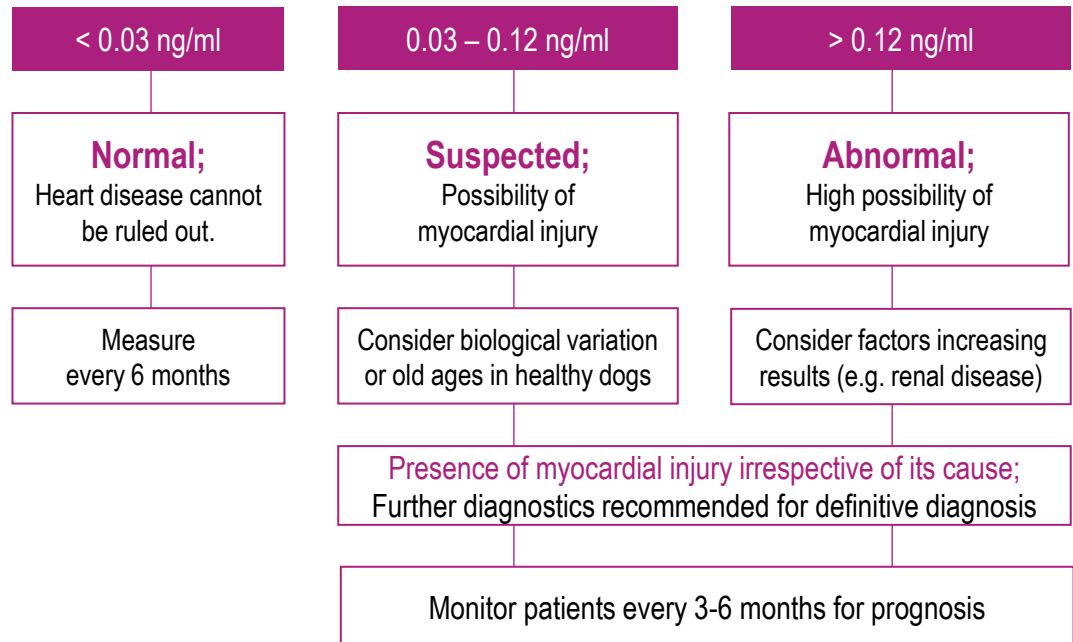
Quantitative marker of myocardial injury



- **Species:** Dog
- **Sample:** Serum 100 µl
- **Testing Time:** 10 minutes
- **Measurement :** Quantitative
- **Measurement Range:** 0.01 – 20 ng/ml
- **Storage Condition:** 1 - 30 °C

### • Diagnostic Algorithm

- Tnl levels reflect heart muscle injury from cardiac or non-cardiac diseases
- Include measurement of Tnl among routine biochemical testing (Renal, hepatic +Cardiac)



#### ※Caution

- Extreme exercise can cause transient myocardial injury in dogs, increasing Tnl levels.
- In older dogs, Tnl levels can be elevated because of myocardial changes leading to Tnl leakage.

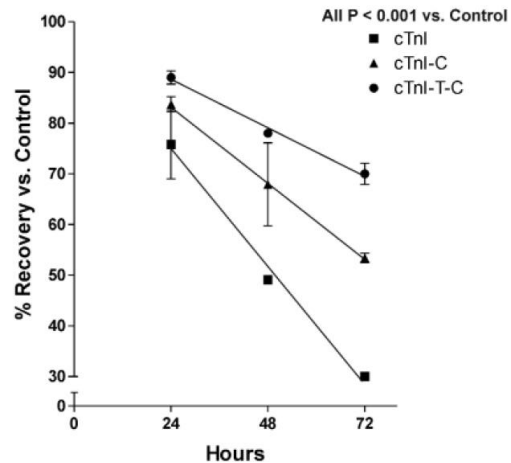
# Factors which may affect results

- Storage Conditions**

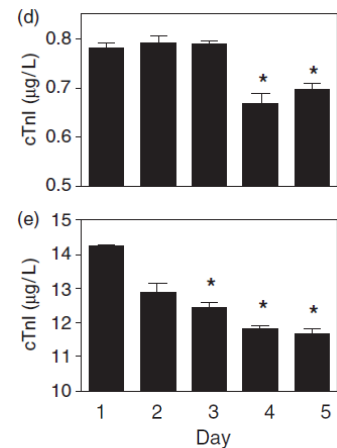
- Degradation by serum **proteases** reduces the stability of Tnl  
(**Not stable at room or refrigeration temperature**)
- The freeze-thaw cycles can affect Tnl concentrations.

→ **Samples should be tested immediately after collection.** (If not, freeze the samples at -20 °C or below for storage. Do not freeze and thaw repeatedly.)

**Graph 1.**  
At room temperature



**Graph 2.**  
At refrigerator temperature (2–8°C)  
→ 15-18% decrease



- Interference**

- **Vcheck Canine Tnl**

No interference was observed for each substance up to the concentration presented in the following table.

| Interfering substances | Concentration |
|------------------------|---------------|
| Hemoglobin             | < 150 mg/dl   |
| Intralipid             | < 2,500 mg/dl |
| Cholesterol            | < 250 mg/dl   |
| Bilirubin (total)      | < 20 mg/dl    |
| Vitamin C              | < 1 mg/ml     |

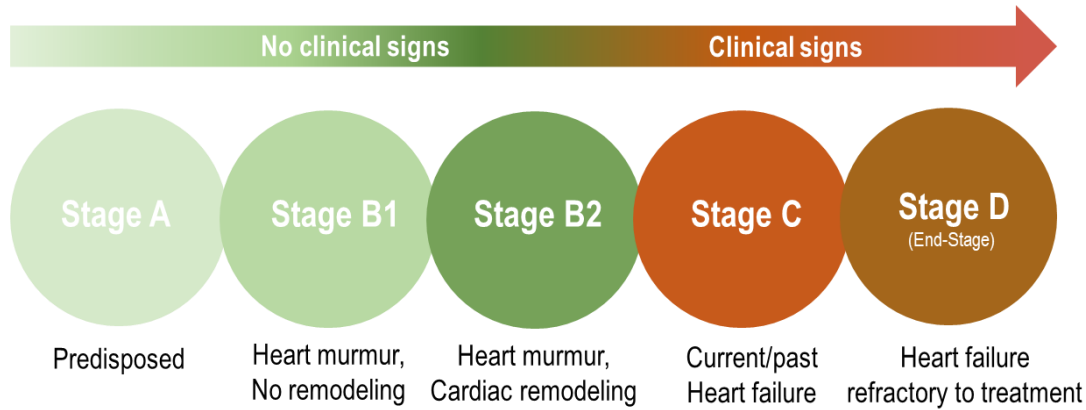
→ **Moderate or Severe hemolysis** can falsely increase Tnl levels.

- Other Tnl Increasing Factors**

- Renal disease
- Old ages (> 10 years)
- Extreme exercise

# Clinical evaluation

- Tnl concentrations in MMVD dogs**



**Table1.** Concentrations of Tnl according to ACVIM stages in dogs

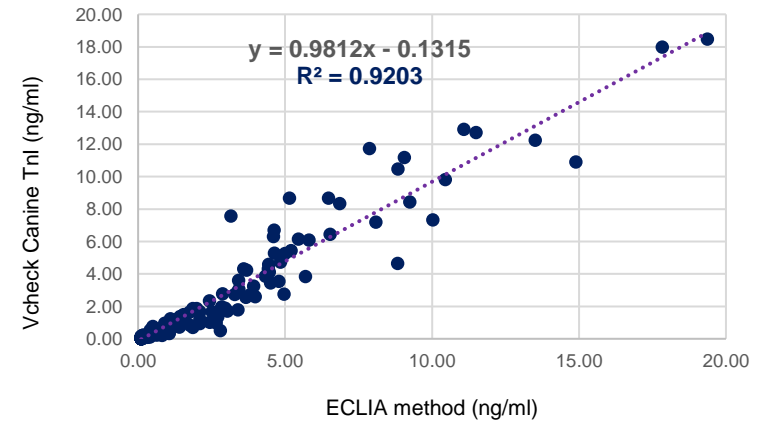
| AVG±SD (ng/ml) | Normal (N=50)          | Stage B1 (N=3)         | Stage B2 (N=9)          | Stage Cc (N=25)        | Stage D (N=2)          | P value |
|----------------|------------------------|------------------------|-------------------------|------------------------|------------------------|---------|
| <b>Vcheck</b>  | 0.06±0.06 <sup>a</sup> | 0.07±0.06 <sup>a</sup> | 0.11±0.07 <sup>a</sup>  | 0.24±0.45 <sup>b</sup> | 0.46±0.06 <sup>a</sup> | <0.001  |
| <b>Roche</b>   | 0.10±0.13 <sup>a</sup> | 0.20±0.18 <sup>b</sup> | 0.18±0.15 <sup>b</sup>  | 0.28±0.50 <sup>b</sup> | 0.46±0.51 <sup>b</sup> | <0.001  |
| <b>ANI***</b>  | 0.01±0.01 <sup>a</sup> | 0.04±0.03 <sup>b</sup> | 0.038±0.04 <sup>a</sup> | 0.25±0.71 <sup>b</sup> | 1.48±2.06 <sup>b</sup> | <0.001  |

\*Different alphabets mean significant differences between groups (p < 0.05).

- High correlation with a Reference method**

Vcheck Canine Tnl has a high correlation ( $Y=0.98X-0.13$ ,  $R^2=0.92$ ) with Roche Elecsys Troponin I.

**Graph1.** Correlation of Vcheck and Roche Troponin I (N=156)



| Comparative analysis of Tnl | Roche (ng/ml) |           | Total      |
|-----------------------------|---------------|-----------|------------|
|                             | > 0.12        | ≤ 0.12    |            |
| Vcheck (ng/ml) > 0.12       | 93            | 1         | 94         |
| Vcheck (ng/ml) ≤ 0.12       | 5             | 57        | 62         |
| <b>Total</b>                | <b>98</b>     | <b>58</b> | <b>156</b> |

- **Sensitivity 94.9%** (93/98), **Specificity 98.3%** (57/58)
- **Accuracy 96.2%** (150/156)

# Q&A Session

BIONOTE Marketing team

Aug 2021

