NT-proBNP Application Overview

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Cardiac Biomarkers

- Cardiac Troponins – Markers reflecting heart tissue damage
- Natriuretic Peptides (NP's) – Indicators of heart function
- C-Reactive Protein – Markers of inflammatory diseases (not specific)
- Homeostasis of serum lipoproteins (high-density lipoproteins and low-density lipoproteins)

Natriuretic Peptides (NPs)

- Natriuretic Peptides (NPs) – composed of a group of peptides with similar structure, secreted through endocrine, paracrine and/or autocrine pathways, with natriuretic effect

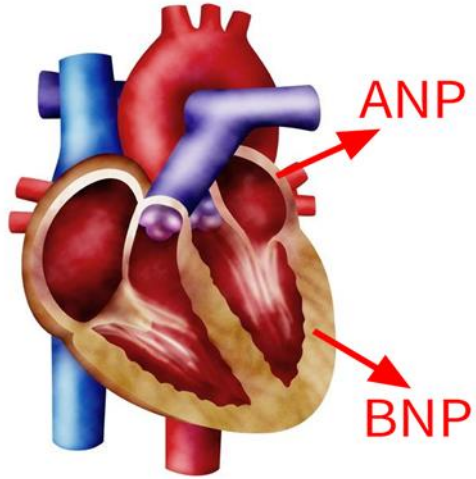
- Mainly Includes:
  - Atrial Natriuretic Peptide (ANP)
  - Brain Natriuretic Peptide (BNP)
  - C-Type Natriuretic Peptide (CNP)
  - Renal Natriuretic Peptide (RNP)

- NPs are similar in structure but have different physiological effects

Natriuretic Peptides (NPs)

The heart not only has the function of pumping blood, but it is also an endocrine gland.

So far, it has been confirmed that two polypeptides are secreted by the heart and stretched into the blood circulation through myocardial cells. They are atrial natriuretic peptide (ANP) and B-type natriuretic peptide (BNP)

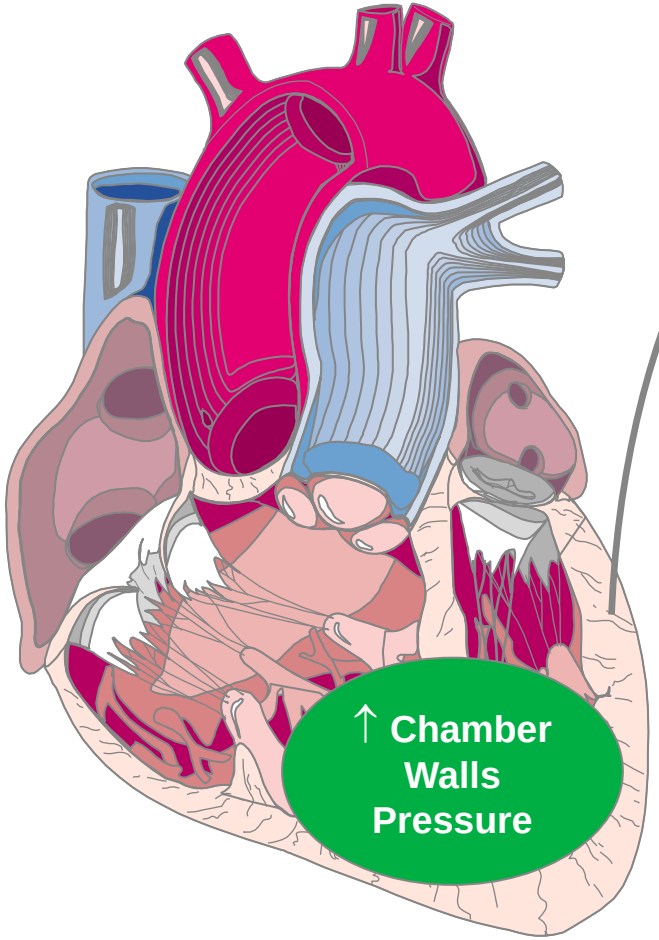


ANP is mainly secreted by the atria while BNP is secreted by the ventricles, mainly ventricular myocytes. It has been discovered that cardiac fibroblasts can also produce BNP.

The main factor that stimulates the release of ANP and BNP is the increase in myocardial tension. The regulation of secretion and release of ANP is mainly at the level of atrial storage, with few newly synthesised ones. The regulation of synthesis and secretion and release of BNP is mainly at the level of gene expression.

The physiological and biochemical functions of the two natriuretic peptides are interdependent. They are the main regulators that balance the body's water and sodium metabolism, and maintain blood pressure.

NT-proBNP/BNP Physiology



Pre-Pro-BNP$_{1-134}$

26-aa signal sequence

Pro-BNP$_{1-108}$

Cardiomyocytes

Blood

N-terminal Pro-BNP$_{1-76}$

$t_{1/2}$ = 60-120 min

BNP$_{77-108}$

$t_{1/2}$ = 18 min

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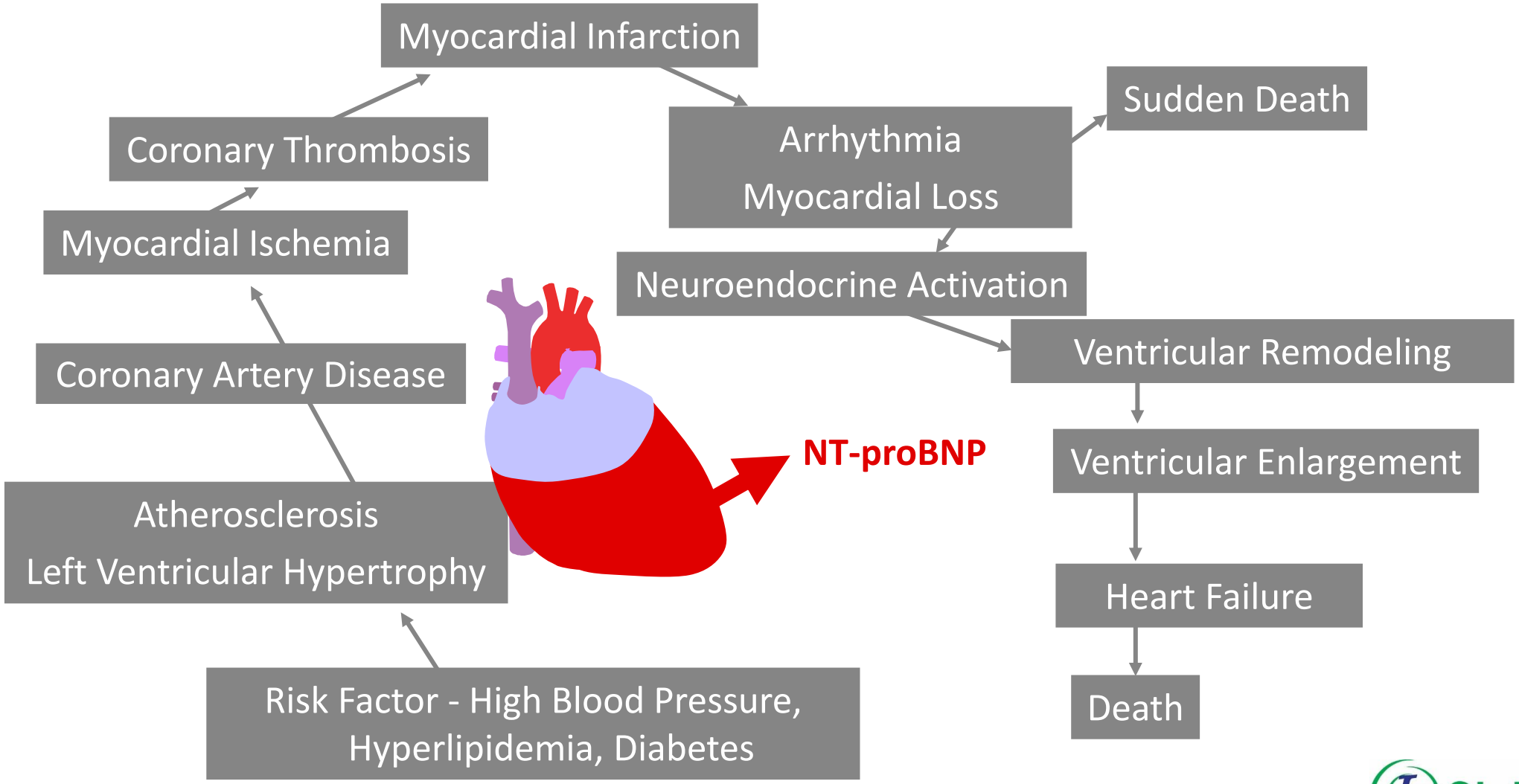
Heart Failure – End Stage of Cardiovascular Disease



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NT-proBNP Biological Characteristics

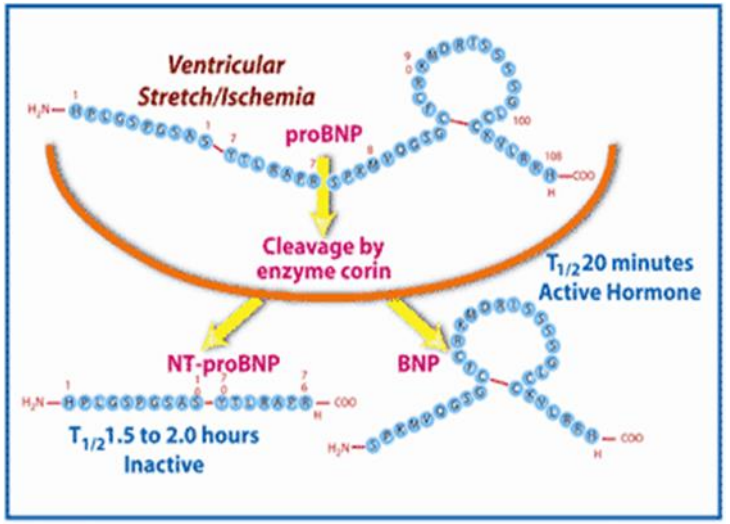
NT-proBNP is a type of hormone fragment secreted by the heart.

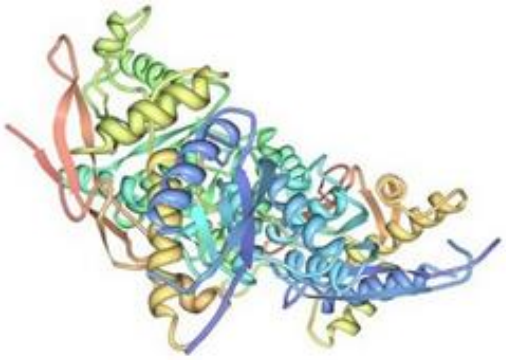
The heart not only has the function of blood transport, but it is also an endocrine gland. So far, it has been confirmed that two peptides can be secreted by the heart and stretched into the blood circulation through cardiomyocytes.

They are Atrial Natriuretic Peptide (ANP) and Brain Natriuretic Peptide (BNP), both belong to the family of natriuretic peptides. ANP is mainly secreted by the atria while BNP is mainly secreted by the ventricles.

The physiological and biochemical functions of the two hormones depend on each other. They are the main regulators that balance the body's water and sodium metabolism and maintain blood pressure.

NT-proBNP is stable and has a long half life, making it a more desirable biomarker.





Natriuretic Peptide
Protein Structure

BNP is synthesised as a prohormone and is called proBNP (BNP precursor). After being stimulated by cardiomyocytes (for example, cardiomyocytes are stretched), proBNP is cleaved into inactive NT-proBNP and the biologically active hormone BNP under the action of proteases. Both peptides are released into the blood circulation.

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BNP and NT-proBNP

BNP exists in the ventricular septal granules and its secretion depends on the volume expansion and pressure load increase of the ventricle.

When the cardiomyocytes receive traction stimulation, they first secrete precursor pro-B-type natriuretic peptide (pre-proBNP) and then form pro-B-type natriuretic peptide (pre-proBNP peptide, proBNP). proBNP is cleaved under the action of endonuclease into BNP with beneficial sodium, diuretic, vasodilator and other biological activities and non-biologically active N-terminal pro-B-type natriuretic (N-terminal pro-B-type natriuretic) peptide, NT-proBNP.

BNP is mainly degraded in large blood vessels and other places while NT-proBNP is mainly excreted through the kidneys. Therefore, measuring the level of NT-proBNP in plasma can diagnose and evaluate heart failure.

NT-proBNP Biological Characteristics

NT-proBNP

- Secreted by the ventricle and stretched into the blood circulation through the cardiomyocytes. It is a peptide containing 76 amino acids
- Linear structure, no biological activity, cleared by the kidney
- Half-life is 90 – 120 mins, stable in vitro
- High blood concentration (15 – 20 times of BNP), blood concentration is positively correlated with the degree of cardiac dysfunction
- NT-proBNP level is not affected by factors such as receptor position and daily activities, nor does it have intra-day or inter-day fluctuations, and has good repeatability

Physiological Function

- Acts on tissues involved in sodium regulation and maintaining blood pressure homeostasis
- Promote urinary sodium excretion and diuretic effect
- Dilate blood vessels
- Antagonise the renin-angiotensin-aldosterone system (RAAS)
- Increased blood NT-ProBNP levels in patients with congestive/chronic heart failure
- NT-proBNP has no physiological activity. When BNP is secreted, it enters the blood at the same time in a ratio of 1:1

NT-proBNP Secretion Regulation

Normal Heart Function:

- Low circulation level

During Myocardial Infection:

- Gene level, explosive synthesis
- Ventricular cells undergo phenotypic modification and secondary expression of certain fetal genes, making the ventricle an important source of NT-proBNP which in turn leads to a significant increase in blood NT-proBNP levels
- The change of ventricular volume and the increase of ventricular wall tension are the key to affecting the secretion of NT-proBNP
- Myocardial ischemia and hypoxia can also stimulate the production of NT-proBNP. Other factors include increased heart rate, vasoconstriction, antidiuretic effect, myocardial hypertrophy and cell proliferation etc.

NT-proBNP Superiority as a Clinical Diagnostic Index for Heart Failure

Early/mild heart failure often has no changes in ventricular structure and symptoms are not obvious.

- ❑ Asymptomatic heart failure accounts for a large proportion of clinical cases
- ❑ Echocardiography often fails to detect changes in the ventricles

Traditional clinical diagnosis is difficult, and there is a high misdiagnosis rate.

NT-proBNP is the only objective diagnostic indicator of heart failure.

Medical History	**Check for risk factors, heart disease and triggers**
Clinical Symptoms and Signs	**Non-specific**
Electrocardiogram	**Can only be used for lesion location**
Chest Radiograph	**Subjective interpretation, lack of standardisation**
Echocardiography	**Heart structure and function, subjective & objective**
NT-proBNP	**Objective, quantitative and accurate**

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BNP and NT-proBNP

NT-proBNP is mainly eliminated in the kidneys while BNP is mainly eliminated in the peripheral circulation and partly in the kidneys. Renal insufficiency will affect these two indicators to varying degrees.

In clinical, heart failure and renal failure often coexist and are mutually cause and effect. The interpretation of BNP/NT-proBNP requires more caution.

Pathological factors such as chronic glomerulonephritis, diabetes and hypertension have an impact on renal function which will then affect NT-proBNP.

In the case of advanced renal failure, it is not suitable to use NT-proBNP or even BNP to diagnose heart failure.

NT-proBNP Data Interpretation

Canine NT-proBNP	pg/mL	pmol/L
Low Risk	<9490	<900
Suspicion of MVD or DCM or Cardiomegaly	>9490	>900
High Risk Heart Failure	>18981	>1800

Feline NT-proBNP	pmol/L
Low Risk	<100
Suspicion of MVD or DCM or Cardiomegaly	100-270
High Risk Heart Failure	≥270

The reference range of this product is a suggestion. A veterinarian must determine the final result.

Under no circumstances can it be diagnosed with one diagnosis method. Please make a comprehensive judgment based on clinical symptoms, diagnosis records and results

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NT-proBNP

Guidelines for Using NT-proBNP Assay in Dogs and Cats:

- ❑ Normal or low levels of NT-proBNP are more common in non-heart diseases while high levels are a typical feature of heart diseases
- ❑ In asymptomatic heart disease animals, high levels of NT-proBNP may be misdiagnosed as having non-heart disease causes, such as dyspnea, etc.
- ❑ NT-proBNP level has a high predictive value. When there is no echocardiogram, NT-proBNP may be a suitable substitute parameter
- ❑ NT-proBNP level can be used to predict the increase in cardiac capacity and indirectly assess survival rate based on the damage caused

NT-proBNP Main Application

Clinical Diagnosis and Differential Diagnosis:

- ❑ Such as the differential diagnosis of dyspnea (cardiogenic or pulmonary)
- ❑ Diagnosis of congestive heart failure
- ❑ Diagnosis of hypertensive myocardial hypertrophy

Evaluation of Heart Function:

- ❑ NT-proBNP concentration is related to the degree of heart failure and is an objective indicator for judging heart failure and its severity

Prognosis Estimation and Risk Classification of Cardiovascular Diseases:

- ❑ Such as prognostic evaluation of heart failure, prediction of re-morbidity and mortality
- ❑ Prognostic evaluation of acute myocardial injury, prediction of mortality, estimation of injury range of myocardial ischemia

Monitoring of Treatment Effect:

- ❑ The concentration change of NT-proBNP is related to the curative effect and the drug dose can be adjusted according to the change to estimate the curative effect

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Canine NT-proBNP Rapid Quantitative Test

Woodley have developed a rapid, accurate and reliable, highly sensitive detection method for Canine NT-proBNP.

The InSight V-IA Canine NT-proBNP Rapid Quantitative Test is a fluorescence immunoassay used with the InSight V-IA Veterinary Immunoassay Analyser for quantitative determination of NT-proBNP concentration in canine serum or plasma.

The test is used to evaluate myocardial function.

It can be stored at room temperature.

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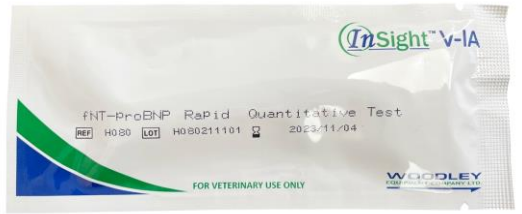
Feline NT-proBNP Rapid Quantitative Test

Woodley have developed a rapid, accurate and reliable, highly sensitive detection method for Feline NT-proBNP.

The InSight V-IA Feline NT-proBNP Rapid Quantitative Test is a fluorescence immunoassay used with the In Sight V-IA Veterinary Immunoassay Analyser for quantitative determination of NT-proBNP concentration in feline serum or plasma.

The test is used to evaluate myocardial function.

It can be stored at room temperature.



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Thank You

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