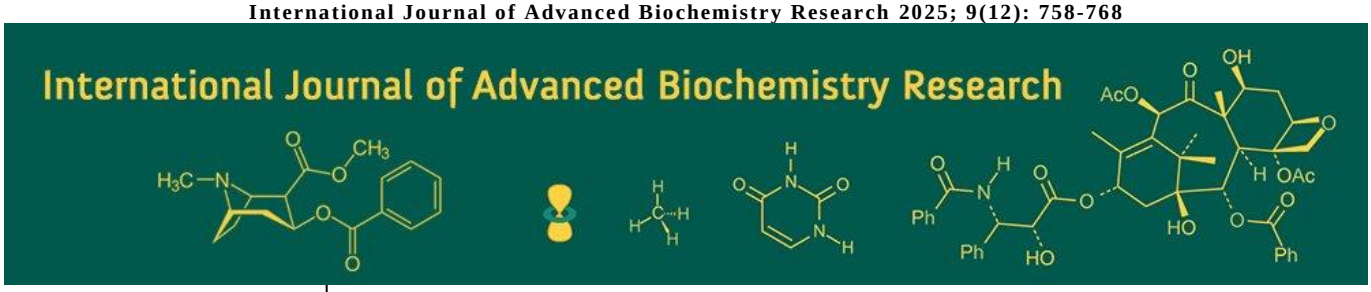


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# Cardiovascular anomalies in dogs: An anatomical perspective

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**Abstract**
Cardiovascular anomalies (CAs) in canines are significant developmental disorders that often arise from disruptions during embryogenesis. Normal heart development involves stages like heart tube formation, cardiac looping, and chamber septation (Anderson *et al.,* 2020). Disturbances in these processes can lead to structural malformations such as patent ductus arteriosus (PDA), persistent right aortic arch (PRAA), subaortic stenosis (SAS), and septal defects. These issues can affect hemodynamic efficiency, leading to symptoms like exercise intolerance and heart failure, although some may remain subclinical until acute events occur. Recent advancements in veterinary cardiology emphasize the role of blood-based biomarkers alongside imaging techniques. N-terminal pro–brain natriuretic peptide (NT-proBNP) and cardiac troponins serve as indicators of myocardial stress and dysfunction, with elevated NT-proBNP levels linked to various CAs (Hariu *et al.,* 2013). Combining biomarker data with echocardiography and radiography enhances diagnostic accuracy, especially in breeds prone to specific anomalies (Schober and Baade, 2000). Understanding the connection between embryology, anatomical deviations, and biomarkers is essential for tackling canine CAs. Establishing reference ranges for these biomarkers across breeds is vital for future research (Klein *et al.,* 2022). By integrating developmental anatomy with innovative diagnostic tools, veterinarians can improve detection, management, and breeding practices, thus enhancing canine cardiac health.

**Keywords:**

## 1. Introduction
The development of the heart is a complex process. During embryonic and fetal development, the heart grows and transforms from a valveless tubular structure into a four-chambered organ with valves. The heart is one of the earliest organs to develop and take on its vital functions during embryonic development. Throughout the intricate process of embryogenesis, the nascent heart undergoes significant morphological changes, skillfully reshaping a population of cardiac progenitor cells into a sophisticated four-chambered organ (Moorman and Christoffels, 2003) [41]. This remarkable transformation results in the formation of two atria, two ventricles, and four essential heart valves, each playing a critical role in the organ's functionality. Achieving this complex reorganization necessitates a series of intricate cellular processes, including migration, proliferation, apoptosis, and differentiation. Furthermore, these processes are tightly orchestrated through precise cellular signaling pathways, ensuring that each step occurs in a well-coordinated manner to establish a fully functional heart (Gittenberger *et al.*, 2005) [21].
Congenital heart disease (CHD) is defined as a morphologic defect of the heart or associated great vessels present at birth. The etiology of CHD is multifactorial, with several factors contributing to its risks (Dyer and Rugonyi 2021) [20]. Well-known and relatively well-studied factors include embryonic genetic and epigenetic anomalies, and maternal conditions such as undernutrition, diabetes, infectious diseases, exposures to teratogens, and abnormal blood flow can lead to a variety of CHDs (Midgett *et al.*, 2017) [39]
Congenital cardiovascular anomalies in dogs by examined through the dual lens of developmental anatomy and gross structural deviation. During this complex sequence, disruptions in progenitor cardiac cell migration, extracellular matrix interactions, or

hemodynamic forces can result in anomalies such as persistent right aortic arch results from abnormal vascular remodeling of the aortic arches, leading to esophageal compression, ventricular septal defects, and transposition of the great arteries, subaortic stenosis (SAS) is caused by an abnormal ridge of fibrous tissue beneath the aortic valve, obstructing blood flow from the left ventricle, patent ductus arteriosus (PDA) is caused by the persistence of the fetal ductus arteriosus postnatally, ventricular septal defects (VSD) is due to improper development of interventricular septum formation (Aanhaanen *et al.*, 2009) [1].

Clinically, these cardiovascular defects may present with nonspecific symptoms such as exercise intolerance, heart murmurs, or signs of congestive heart failure. However, in some cases, dogs may remain asymptomatic until a sudden cardiac event occurs, underscoring the need for thorough anatomical and developmental evaluation in at-risk breeds (Buchanan, 2001) [11].

Diagnostic modalities of canine cardiac illnesses also gained momentum, starting from heart catheterization and angiocardiography, radiography, electrocardiography and echocardiography to recently developed cardiac biomarkers and metabolomics. However, wide variability among different dog breeds may also mislead the interpretation of ECG, echocardiography, and thoracic radiograph (Schober and Baade, 2000) [51]. Thus, the identification and monitoring of blood-based cardiac biomarkers along with or in combination with other diagnostic tools could be a promising approach to diagnose cardiac diseases (Atkinson *et al.*, 2001) [7].

A comprehensive understanding of these anomalies, grounded in the principles of embryology, is vital for the precise identification and interpretation of developmental variations. This knowledge not only deepens a veterinary anatomist's expertise but also significantly enhances their capacity to collaborate effectively within multidisciplinary teams, particularly in the fields of diagnostic imaging and pathology. By bridging the gap between embryological theory and practical application, veterinarians can improve diagnostic accuracy and treatment outcomes.

## 2. Embryological Development of the Heart

### 2.1 Formation of the Primitive Heart Tube

Normal formation of the heart is crucial for understanding cardiac pathologies and congenital malformations. Early cardiogenesis begins with the organization of the cardiac area and continues through gastrulation, ending with the formation of two endocardial tubes that are externally covered by myocardial lineage cells. This stage occurs at weeks 4-8 of embryonic development and begins with the formation of the straight tube.

Progenitor cardiogenic mesodermal cells in the cranial primitive streak migrate cranio-laterally to form the first heart field around the cranial neural folds. Heart field develops medially and caudally to the pharyngeal mesoderm. The primitive heart begins to beat around day 21 and starts pumping blood by days 24–25. The heart tube forms through a complex process involving all three germ layers. As the embryo grows, folding in the cranio-caudal and lateral axes brings the paired endocardial tubes together at the midline, where they fuse into a single primitive heart tube—the earliest identifiable anatomical precursor to the mature heart (Kloesel *et al.*, 2007) [32].

The primitive heart tube consists of five regions, arranged cranio-caudally: the truncus arteriosus, bulbus cordis, primitive ventricle, primitive atrium, and sinus venosus. Each segment gives rise to distinct anatomical structures in the mature heart, forming the foundation for later differentiation and morphogenesis. The tube is composed of myocardium, formed by myocardial cells, and endocardium, formed by endothelial cells, separated by an extracellular matrix known as cardiac jelly. The epicardium develops later from migrating proepicardial precursor cells (Yamagishi *et al.*, 2009) [60].

Research has shown that the initial heart tube, formed by first heart field cells, contributes only a small portion to the final four-chambered heart. While these cells provide a scaffold, the majority of the heart develops from precursor cells in the second heart field, which also contribute to elongation of the heart tube. The rearrangement of the endocardial tube is essential for forming the heart chambers and connecting them to the vasculature, with cardiac looping driven by the migration of these precursor cells (Kelly, 2012) [28].

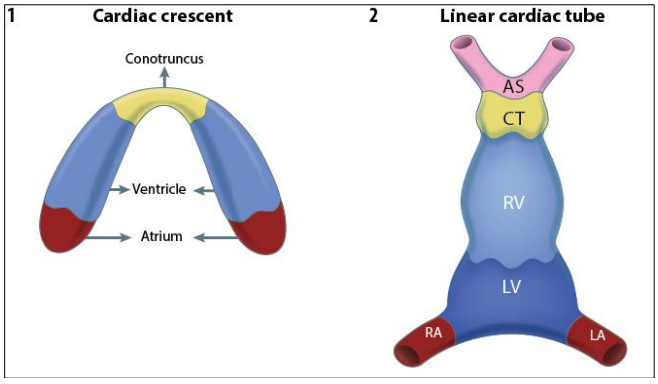


**Fig 01:** Formation of a single heart tube from two paired tubular structures. 1. Ventral view at approximately 21 days showing the beginning of fusion. 2. At 22 days, fusion is almost complete.

### 2.2 Cardiac Looping and Chamber Specification.

The linear heart tube undergoes rightward looping to establish the spatial orientation of the atria and ventricles. In dogs, this process is completed by day 22 of gestation and is essential for correct chamber alignment. The primitive ventricle shifts to the left, while the bulbus cordis and truncus arteriosus are displaced anteriorly and to the right (Anderson *et al.*, 2020) [4]. This morphological change produces the characteristic S-shaped configuration of the heart.

Between days 23 and 28 of embryonic development, the endocardial tube bends and folds to form the cardiac loop.

This pivotal transformation involves the shifting of the outflow tract and atrioventricular canal toward the midline phenomenon known as convergence. As development progresses, septation occurs, dividing the primitive ventricles and outflow tract into separate pathways for systemic circulation (aorta) and pulmonary circulation (pulmonary artery). This division is achieved through a combination of wedging and counterclockwise rotation of the outflow tract.

These coordinated movements position the aortic valve strategically behind the pulmonary trunk, laying the groundwork for the mature heart's intricate architecture.

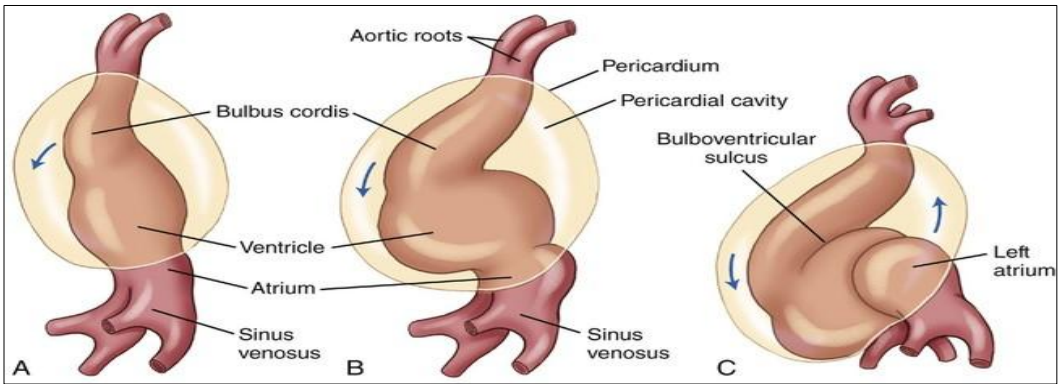


**Fig 2:** Formation of the atrio-ventriculobulbar loop at approximately 22 days A. at approximately 23 days, B. at approximately 24 days, C. As this loop is formed, a common atrium is formed and enters the pericardial cavity.

### 2.3 Septation of the Heart Chambers

The development of the interatrial and interventricular septa is a tightly coordinated process that begins with the proliferation of endocardial cushions in the atrioventricular canal. These mesenchymal structures contribute to the formation of both septa and valves.

- **Atrial Septation:** The interatrial septum develops from two components: the septum primum, which grows downward from the roof of the primitive atrium, and the septum secundum, which overlaps the septum primum, leaving the foramen ovale as a fetal communication (Kirby, 2002) [30]. During the fourth and fifth weeks of gestation, the septum primum grows from base to apex, dividing the left and right atria. Cell death in the cranial portion produces fenestrations, forming the ostium secundum. The septum primum later fuses with the endocardial cushions. Around day 33, the crescent-shaped septum secundum forms in the right atrium, creating the foramen ovale, which allows right-to-left blood flow during gestation. Postnatally, the septa fuse, closing the foramen (Kloesel *et al.*, 2017) [32].
- **Ventricular Septation:** Expansion of the primitive ventricles, along with migrating myocardial cells, results in the formation of the muscular interventricular septum, which grows from the apex toward the atrioventricular endocardial cushions. The interventricular foramen - a gap above this septum—is later closed by fusion of the superior and inferior atrioventricular cushions, with additional contribution from the outflow tract cushions, forming the membranous septum (Gittenberger *et al.*, 2005) [21].
- **Valve Development:** The atrioventricular valves develop from mesenchymal cells originating in the atrioventricular cushions during the crucial fifth and sixth weeks of gestation. As these cushions grow, the superior, inferior, and lateral components work together to subdivide the common atrioventricular canal into distinct left and right canals, effectively shaping the heart's structure. The final development of the mitral and tricuspid valves involves the intricate migration of epicardial cells onto the cushions and the parietal leaflets, completing the formation of these essential cardiac valves (Lin *et al*., 2012) [36].
- **Outflow Tract Septation:** The truncus arteriosus and bulbus cordis undergo a remarkable transformation as they are divided by the spiral aorticopulmonary septum. This vital structure, intricately shaped and guided by the contributions of cardiac neural crest cells, plays a crucial role in the separation of the heart's major vessels (Dyer *et al.*, 2021) [20]. As a result of this partitioning, the ascending aorta and the pulmonary trunk emerge, each serving as the primary pathway for oxygenated and deoxygenated blood, respectively, to flow from the heart to the body and lungs. This intricate process is essential for the proper formation of the circulatory system during development (Waldo *et al.*, 2005) [57].

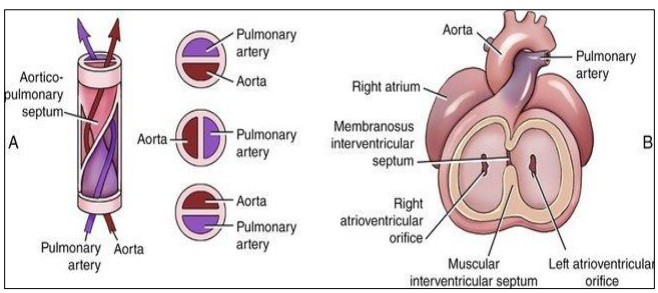


**Fig 3:** A. Formation of the aorticopulmonary septum, B. Division of the aorta and pulmonary artery twisting around each other.

**Sinu-atrial node and Atrio ventricular node:** Specialised myocardial cells responsible for the initiation and conduction of the electrical impulses which regulate the rate of cardiac contractions, develop in myocardial tissue. These cells form structures referred to as pacemakers. The first pacemaker is located in the caudal part of the left cardiac tube. Subsequently, a site in the right horn of the sinus venosus assumes this role. When the right horn of the sinus becomes incorporated into the definitive right atrium, the specialised tissue is referred to as the sino-atrial node (SA node) (McGeady *et al*., 2006) [38].

The Atrio Ventricular node develops from the atrioventricular canal (AVC), a region of the embryonic heart situated between the atria and ventricles. Studies indicate that the AVC contains precursors to the AV node, AV ring bundle, and the myocardium of the AV junction and these precursors are derived from heart field (Aanhaanen *et al.*, 2009) [1].

The AV node lies at the lower back section of the interatrial septum near the opening of the coronary sinus, which conducts the normal electrical impulse from the atria to the ventricles. It is located at the center of Koch's triangle—a triangle enclosed by the septal leaflet of the tricuspid valve, the coronary sinus, and the membranous part of the interatrial septum (Christoffels *et al.*, 2010) [14].

### 2.4 Aortic Arch Artery Remodeling

The vascular system originates from the heart field, where progenitor cells are induced to form cardiac myoblasts that give rise to paired dorsal aortae. Each pharyngeal arch is accompanied by a cranial nerve and an aortic arch artery. These aortic arches develop sequentially in a cranio-caudal order and connect the aortic sac (the distal part of the truncus arteriosus) to the paired dorsal aortae.

In mammals, five paired aortic arches are identified during development and are numbered I, II, III, IV, and VI (the fifth arch does not form). Their derivatives are as follows (Schorn *et al*., 2021) [52]

- **First aortic arch (I):** Forms the maxillary arteries.
- **Second aortic arch (II):** Gives rise to the hyoid and stapedial arteries.
- **Third aortic arch (III):** Contributes to the formation of the common carotid arteries and the first part of the internal carotid arteries.
- **Fourth aortic arch (IV):**

**a)** **Left side:** Forms part of the aortic arch.
**b)** **Right side:** Forms part of the right subclavian artery.

- **Sixth aortic arch (VI):**

**a)** **Left side:** Forms the left pulmonary artery and the ductus arteriosus.
**b)** **Right side:** Forms the right pulmonary artery.

This remodeling process is closely linked to the development of the pharyngeal arches and is influenced by neural crest cell migration and hemodynamic forces within the embryonic vasculature. Any disruption during this transformation can result in vascular anomalies, such as a persistent right aortic arch or double aortic arch, which may cause esophageal compression in affected animals.

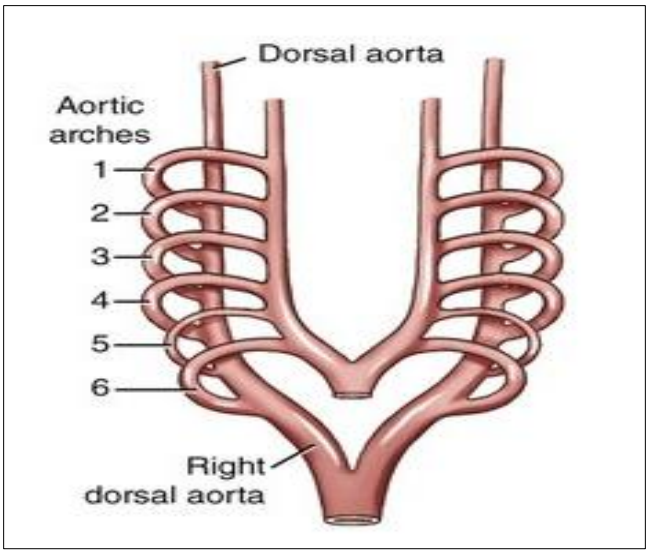


**Fig 4:** Aortic arches and dorsal aortas before transformation into the definitive vascular pattern.

## 3. Biomarkers in the Congenital Heart Anomalies

Biomarkers have emerged as indispensable tools in veterinary cardiology, offering a window into the physiological and pathological processes occurring within the body without the need for invasive procedures. A biomarker is defined as a measurable indicator of normal biological processes, pathological changes, or responses to a therapeutic intervention In the context of congenital heart anomalies in dogs, biomarkers can be used to detect abnormalities in cardiac structure or function even before birth (Strimbu and Tavel, 2010) [55]. During gestation, early identification of congenital heart defects (CHDs) is of immense clinical importance because timely detection can allow for better prognostication, perinatal management, and selective breeding strategies to reduce genetic predisposition in canine populations (Oyama and Sisson, 2004) [45]. These biomarkers may be present in:

- Maternal circulation – where fetal derived molecules cross the placental barrier.
- Amniotic fluid – which directly reflects fetal metabolic and physiological status.
- Fetal blood – obtained via specialized veterinary obstetric techniques in experimental or clinical research settings.

Common biochemical forms of biomarkers include proteins, enzymes, hormones, nucleic acids, and metabolites, each reflecting different aspects of cardiac health.
In prenatal veterinary cardiology, diagnostic and prognostic biomarkers hold the most value because they can help anticipate postnatal and prenatal health status and determine the viability of affected fetuses.

### 3.1 Natriuretic Peptides

Natriuretic peptides (NPs) are cardiac hormones secreted predominantly by atrial and ventricular myocytes in response to increased myocardial wall tension or volume overload. These hormones counteract excessive fluid retention by promoting natriuresis (sodium excretion), diuresis and vasodilation while inhibiting the renin–angiotensin–aldosterone system (RAAS) (De Lima and Ferreira, 2017) [17].
The most common form of B-type Natriuretic Peptide (BNP) is released from the ventricles that BNP is initially synthesized as pro BNP, which is enzymatically cleaved into the biologically active BNP and the inactive N-terminal pro BNP (NT-pro BNP) fragment.
In clinical practice, NT-pro BNP is more frequently measured because it is more stable in circulation and has a longer half-life.
In canine cardiology, NT-pro BNP levels are significantly elevated in diseases such as chronic mitral valve disease and congenital anomalies causing volume overload, including ventricular septal defects (VSDs) and patent ductus arteriosus (PDA) (Boswood *et al.*, 2008) [9].

#### Production and receptor interaction

The production, conversion, excretion, processing, and receptor interactions of natriuretic peptides involve several key steps. The NPPA and NPPB genes encode paralogous precursors known as preproANP and preproBNP, respectively. These precursors undergo cleavage and O-glycosylation in atrial cardiac myocytes. Additionally, endoproteolytic proteases such as corin and furin further process these precursors into the biologically active hormones ANP and BNP (Semenov *et al*., 2009) [53].
Under normal conditions, the atria, which are the upper chambers of the heart, mainly produce and release two important hormones called ANP and BNP. However, when the heart is under stress, the atria produce and release more of these hormones, and the lower chambers of the heart (the ventricles) also start to make and secrete ANP and BNP. Besides mechanical stretching of the heart, different hormones in the body can also increase the secretion of ANP and BNP (Kim *et al*., 2013) [29].
Natriuretic Peptides exert their physiological actions in multiple tissues through NP receptor-mediated signaling. Three types of NP receptors exist: NP receptor type A, NPR-A (encoded by NPR1), NPR-B (encoded by NPR2) and NPR-C (encoded by NPR3).
NP binding to the NPR-A induces the intracellular increase of cGMP, which in turn activates downstream signaling cascades, including cGMP-dependent protein kinases (PKGs), cGMP-gated ion channels and cGMP-regulated cyclic nucleotide phosphodiesterases (Goetze *et al.*, 2020) [22].
All three receptors are found in heart muscle cells and heart fibroblasts. However, NRP-B is usually inactive in the heart's lower chambers under normal conditions. All natriuretic peptides (NPs) can activate these three receptors, but they bind with different strengths. Atrial natriuretic peptide (ANP) and brain natriuretic peptide (BNP) mostly activate receptor A (NPR-A), while C-type natriuretic peptide (CNP) mainly binds to receptor B (NPR-B). Receptor C (NPR-C) acts as a clearance receptor, its function is to take in and break down the NPs. NPR-C has the weakest attraction to BNP, which helps BNP stay in the bloodstream longer (Keane *et al*., 2011) [25].

### 3.3 Cardiac Muscle and the Troponins

The striated muscle cell sarcomere consists of thin filaments (I bands) made of actin and tropomyosin, and thick filaments (A bands) composed of myosin chains and light chains. Its striated appearance comes from alternating A and I bands. The Z lines, which define the sarcomere's boundaries, are primarily made of α-actin and other proteins. The M-band at the center contains cross-linking elements of the cytoskeleton, with myosin filaments cross-linked by myomesin.

#### Biology of the Cardiac Troponins

Troponin are a group of three proteins: troponin C (TnC), troponin I (TnI), and troponin T (TnT), present in a set ratio of one of each. TnC binds to calcium, TnI inhibits muscle contraction, and TnT connects to tropomyosin, regulating heart muscle cell sensitivity to calcium and contraction.
Components of cardiac myocytes, cardiac troponins T (cTnT) and I(cTnT) are myofibrillar regulatory proteins which are highly specific to myocardial cells. They are expressed throughout ventricular and atrial tissue and conserved across species, serving as a “translational biomarker” in both human and veterinary medicine, indicating cardiac damage and congenital anomalies causing volume or pressure overload prior to treatment (Bottio *et al*., 2006).

#### Troponin C

It is 18 kDa calcium-binding protein has two domains with four EF-hands for calcium binding. The N-terminal binding sites control its regulatory function and are shared with slow skeletal muscle.

#### Troponin I

It is 24 kDa protein that inhibits actin-activated myosin ATPase activity by binding to actin when calcium is low. Increased intracellular calcium promotes conformational changes allowing cTnI to alternate binding between TnC and actin. Both cTnI and its skeletal isoform are present during fetal development, transitioning to cTnI in the heart postnatally.

#### Troponin T

The largest subunit at 37 kDa, binds to cTnI, cTnC, actin, and tropomyosin. cTnT1 and TnT2 are primarily fetal isoforms, while cTnT3 is predominant in healthy adults. The N-terminal region is hypervariable and contains several phosphorylation sites, which may affect calcium-dependent ATPase activity by altering the binding affinity of TnT for tropomyosin (Patil *et al.*, 2011) [46].

#### Increases in circulating cardiac troponins

Cardiac troponins (cTn) are vital proteins found in various forms, including cTnT and cTnI, along with the cTnI-TnC

complex and a composite of cTnT, cTnI, and TnC. These troponins play a critical role in the structure of heart cells, with approximately 3–8% of the total troponin present in a heart cell existing freely in the cytoplasm. This free troponin is believed to be responsible for the initial increase in troponin levels in the bloodstream following heart damage, while subsequent elevations are thought to arise from the breakdown of structural proteins. The extent to which troponins may leak from healthy cells remains uncertain.

Research indicates that TnT and TnI isoforms contribute to the modulation of calcium (Ca2+) sensitivity within the contractile system, particularly during cardiac development. Myofilament Ca2+ sensitivity can be evaluated using controlled Ca2+ concentration in various experimental setups, such as skinned fiber bundles, isolated myocytes, myofibrils, or reconstituted systems featuring purified contractile proteins (troponin-tropomyosin-actin-myosin). Data reveals that the relationship between force and Ca2+ concentration (force-Ca2+ relation) or between myofibrillar ATPase and Ca2+ concentration (ATPase-Ca2+ relation) demonstrates cooperative binding of Ca2+ by myofilaments (Puceat *et al.*, 1990) [47].

The study of myofibrils is complex, influenced by the variety of preparations used and the simultaneous isoform transitions of different myofibrillar proteins occurring during development. Moreover, the Ca2+ sensitivity within the contractile system may also be affected by posttranslational modifications, such as the phosphorylation of contractile proteins. Specifically, the phosphorylation of cardiac TnI by cAMP-dependent protein kinase is linked to reduced myofilament sensitivity in cardiac myofibrils (Noland and Kuo, 1991) [43].

Furthermore, when protein kinase C phosphorylates troponin T (TnT) and troponin I (TnI), there is a reduction in the activity of ATPase, the enzyme responsible for energy generation, particularly when calcium is present. This decreased effectiveness of ATPase coincides with the release of cardiac troponin into the bloodstream for a certain duration (Clement *et al.*, 1992) [15].

In veterinary studies, cTnI can be detected within 2–3 hours after myocardial damage in dogs, peaking within 24 hours and exhibiting a short half-life. Elevated levels of cTnI have been observed in various canine cardiac conditions, including myocarditis, endocardiosis, and congenital anomalies such as tetralogy of Fallot (Dunn *et al.*, 2011) [19].

A deeper understanding of the mechanisms and functionality surrounding cTnT phosphorylation and degradation could pave the way for innovative diagnostic and prognostic tools, as well as new therapeutic strategies.

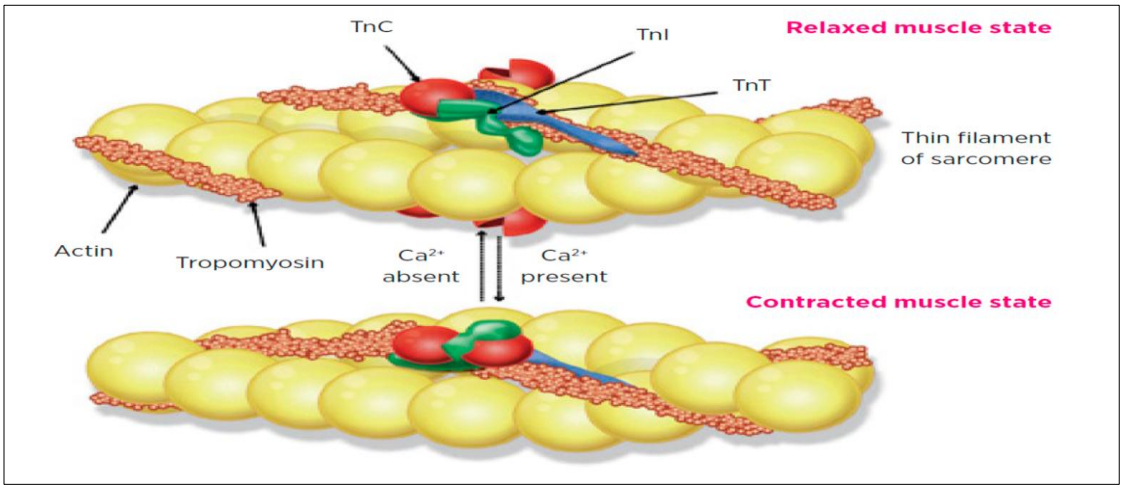


**Fig 5:** Schematic representation of the cardiac troponin complex's role in muscle contraction. Cardiac muscle showing location of cardiac troponin I (TnI), cardiac troponin T (TnT), and cardiac troponin C (TnC).

## 4. Anatomical overview of common cardiovascular anomalies in dogs

### 4.1 Patent ductus arteriosus (PDA)

Anatomically the ductus arteriosus originates from the left sixth aortic arch and plays a critical role in fetal circulation by diverting blood away from the non-ventilated lungs.

**Physiological closure process:** After birth, ductal closure occurs in two distinct phases:

1. **Functional closure:** Achieved by constriction of the vascular smooth muscle, typically within the first 72 hours of life.

**Mechanisms:**

- Drop in pulmonary vascular resistance.
- Decrease in vasodilatory prostaglandins ($PGE_2$) due to placental removal.
- Increased oxygen tension postnatally.

As oxygen tension increases, a complex constrictor mechanism is activated, involving cytochrome P450, which acts as a crucial oxygen sensor and endothelin-1, serving as the primary effector in this process. The heightened oxygen levels inhibit voltage-gated potassium channels, leading to a state of depolarization in the smooth muscle cells. This depolarization triggers the activation of voltage-dependent calcium channels, allowing calcium ions to flow into the cells. The resulting vasoconstriction effectively reduces blood flow through the ductus, demonstrating a finely-tuned response to changes in oxygen availability in the surrounding environment (Akaike *et al.*, 2014) [2].

2. **Anatomical (Permanent) closure:** Occurs over days to weeks and involves fibrous transformation of the ductus into the ligamentum arteriosum.

**Mechanisms (Coceani and Baragatti, 2012) [16]**

- Formation of intimal cushions.
- Turbulent blood flow causing mechanical stress on the narrowing lumen.
- Intramural hypoxia due to collapse of vasa vasorum.

- Platelet adhesion and aggregation at the ductal wall.

3. In Patent ductus arteriosus, the failure of the muscular wall of the ductus to constrict and undergo fibrosis after birth results in a persistent vascular connection between the aorta and the pulmonary artery. This leads to a left-to-right shunt due to the pressure gradient between the systemic and pulmonary circulations (Buchanan, 2001) [11].

**Hemodynamic patterns**

- **Left-to-Right Shunt (most common)**

Occurs when pulmonary vascular resistance is lower than systemic resistance.
Blood flows from the aorta to → pulmonary artery, increasing pulmonary blood flow.
Chronic shunting leads to volume overload of the left atrium and left ventricle, potentially causing left-sided heart failure.

- **Right-to-Left Shunt (reversed PDA – rare)**

Develops when pulmonary vascular resistance exceeds systemic resistance.
Blood flows from the pulmonary artery → aorta.
Often associated with Eisenmenger syndrome, where chronic pulmonary overcirculation causes severe pulmonary hypertension and shunt reversal.
Leads to systemic hypoxemia in the caudal body and differential cyanosis (McDonald, 2006) [37].

**Biomarker relevance:**

- N-terminal pro-B-type Natriuretic Peptide (NT-proBNP) levels are often elevated in PDA.
- This reflects myocardial strain and cardiac volume overload due to persistent left-to-right shunting (Hariu *et al*., 2013) [24].

### 4.2 Persistent Right Aortic Arch (PRAA)

Embryologically, the aorta develops out of an aortic sac, which is connected to the bilateral dorsal aortae by six paired aortic arches that develop bilaterally to the pharynx (Schorn *et al.,* 2021) [52].
When the right fourth aortic arch and the right dorsal aorta enlarge instead of the left, so persistent right aortic arch develops. Physiologically, the right ductus arteriosus degenerates, and the left ductus arteriosus persists, forming a connection between the left pulmonary artery and the abnormal right aortic arch. So these anomalies of the aortic arch are important to recognize because they may be associated with vascular ring formation (Hanneman *et al*., 2017) [23].
This congenital disorder of the aortic vasculature involves complete or partial encircling of the esophagus and trachea with secondary esophageal compression. Dogs with esophageal compression often regurgitate solid food after eating (Morgan and Bray 2019) [42]. Persistent right aortic arch is one of the most common ring anomalies in dogs, with a prevalence of about 7%.

### 4.3 Pulmonic stenosis

Pulmonic stenosis is a condition characterized by an obstruction to the outflow of blood from the right ventricle, primarily occurring at the level of the pulmonary valve, known as valvular stenosis. However, it can also manifest as subvalvular or supravalvular stenosis. From an anatomical perspective, valvular stenosis typically results from the fusion or malformation of the semilunar cusps of the pulmonary valve, which leads to a narrowing of the valve annulus. This narrowing may subsequently cause post-stenotic dilation of the main pulmonary artery (Snarr *et al.,* 2007) [54].
The right ventricle attempts to pump blood against this increased resistance, the free wall of the ventricle often undergoes hypertrophy, thickening in response to the pressure overload. In cases of chronic pulmonic stenosis, dissection of the affected areas reveals not only thickened pulmonary cusps but also the presence of a jet lesion on the wall of the pulmonary artery, a consequence of turbulent blood flow. These pathological changes are indicative of the ongoing stress and adaptation that the cardiovascular system endures due to the obstruction (Bussadori *et al*., 2000).
Cardiac troponin I and NT-pro BNP is often elevated due to right ventricular pressure overload. Severe cases may also exhibit increased cTnI, reflecting myocardial stress or damage (Saunders *et al.,* 2004) [47].

### 4.4 Subaortic stenosis

During the intricate process of embryogenesis, the heart begins its formation as a linear heart tube. This tube undergoes a series of complex morphological changes, including looping and septation, ultimately evolving into the intricate four-chambered structure that is essential for proper circulatory function. A critical component of this development is the conotruncal region, which arises from neural crest cells, this area plays a pivotal role in the formation of the aortic and pulmonary outflow tracts, ensuring the proper routing of blood flow to the systemic and pulmonary circulations (Lahaye *et al*., 2014) [34].
In canines, one of the most prevalent congenital cardiac anomalies is subaortic stenosis. This condition is characterized by the presence of a fibromuscular ridge that encircles or partially narrows the left ventricular outflow tract (LVOT), located just below the aortic valve. The presence of this ridge can significantly impede blood flow from the heart, leading to various complications and challenging the overall cardiovascular health of affected dogs (Kobayashi *et al*., 2014) [33].
Abnormal migration or proliferation of these cells can lead to subvalvular obstructions such as subaortic stenosis (Moorman and Christoffels, 2003) [41]. A failure in the resorption of the conal septum or improper alignment of the aortic outflow tract during foetal development may result in fibrous tissue formation beneath the aortic valve. This obstruction impedes blood flow from the left ventricle to the aorta, causing increased cardiac workload and potential left ventricular hypertrophy. This lesion is typically located just below the aortic valve, within the muscular septum. Over time, chronic pressure overload caused by the obstruction leads to left ventricular concentric hypertrophy, reducing ventricular compliance and increasing the risk of arrhythmias. This pathogenesis reflects a failure in embryological remodelling rather than a simple structural deformity (Tidholm and Jonsson, 1997) [56].
Subaortic stenosis (SAS) is a condition characterized by a narrowing below the aortic valve, which results in increased pressure within the left ventricle of the heart. This pressure overload can lead to significant cardiovascular stress, often manifesting as elevated levels of NT-pro BNP, a biomarker indicative of heart strain and dysfunction. In more severe instances, patients may also exhibit mild increases in cardiac

troponin I (cTnI), a protein released into the bloodstream when there is damage to the heart muscle. These biochemical changes highlight the impact of SAS on cardiac health and its potential progression if left unchecked.

### 4.5 Ventricular septal defect (VSD)

The interventricular septum separates the left and right ventricles. A Ventricular Septal Defect is an abnormal opening in the septum, allowing direct communication between the two chambers. VSDs may occur in the membranous portion (more common) near the origin of the aorta at the heart base, or in the muscular portion near the apex of the septum.

The membranous septum develops from the fusion of the endocardial cushions, conotruncal septum, and muscular septum. Failure of this fusion leads to a membranous VSD. Although VSDs may develop anywhere along the septum, they are most frequently located in the membranous portion. In dogs, VSD is an uncommon congenital anomaly (Saunders *et al*., 2004) [47].

**Classification – Minette and Shan (2006) describe four types**

1. **Membranous ventricular septal defect:** An opening in the membranous portion of the interventricular septum, located near the atrioventricular node, just below the aortic valve and adjacent to the tricuspid valve.
2. **Muscular ventricular septal defect:** Occurs within the muscular septum. When the defect lies just beneath the aortic valve, inadequate valve support may cause mild aortic dextroposition and aortic insufficiency.
3. **Inlet ventricualr septal defect:** Located near the inflow portion of the ventricles near the artioventricular valves. The degree of shunting depends on the defect size and the pulmonary/systemic vascular resistance ratio.

Ventricular septal defects often cause left-to-right shunting, leading to left-sided volume overload. NT-proBNP levels are typically elevated, reflecting myocardial strain. Mild increases in cTnI may occur with subclinical myocardial injury (De Madron *et al*., 2019) [18].

### 4.6 Atrial Septal Defect (ASD)

Atrial Septal Defects are rare in dogs. The atrial septum develops from four main embryonic structures:

- Superior and inferior atrioventricular cushions
- Primary atrial septum with its mesenchymal cap
- Dorsal mesenchymal protrusion

The atrioventricular cushions and the mesenchymal cap develop from the endocardium through a process known as epithelial-to-mesenchymal transformation. This transformation allows the epithelial cells to acquire migratory and invasive properties, contributing to the formation of these crucial structures. In contrast, the dorsal mesenchymal protrusion emerges from the second heart field, playing a vital role in the overall architecture of the heart during its embryonic development (Wessels *et al*., 2000) [59].

**Formation Process**

1. The superior and inferior atrioventricular cushions fuse at the atrioventricular canal.
2. The primary atrial septum grows from the atrial roof toward the cushions, partially dividing the common atrium.
3. The mesenchymal cap at the leading edge of the septum fuses with the cushions (anteriorly) and the dorsal mesenchymal protrusion (posteriorly) to close the ostium primum.
4. During this closure, dissolution of the upper part of the primary septum creates the ostium secundum.
5. The secondary (secundum) septum forms from the atrial roof on the right side of the primum and eventually closes the ostium secundum

**Defects:**

- Primum septum atrial septal defect - Ostium primum atrial septald (low in the atria, near the AV valves, often associated with endocardial cushion defects and ventricular septal defect).
- Secundum septum atrial septal defect - Ostium secundum atrial septal defect (at or above the foramen ovale, most common in dogs).

Blood is redirected from the left atrium to the right atrium, resulting in an increased volume load on the right side of the heart. This shift leads to notable dilation of both the right atrium and the right ventricle, prompting a rise in NT-proBNP levels as the heart struggles to accommodate the excess blood flow. In most cases, cardiac troponin I (cTnI) levels remain within normal ranges unless there is a development of secondary myocardial injury or the onset of pulmonary hypertension, which can compromise cardiac function (Schleich *et al*., 2016).

### 4.7 Tetralogy of Fallot

Tetralogy of Fallot is the most common form of cyanotic congenital heart disease. It is characterized by four defects:

1. Ventricular septal defect (VSD)
2. Overriding aorta
3. Right ventricular outflow tract obstruction
4. Right ventricular hypertrophy (Apitz and Webb, 2009)

It results from the abnormal development of the conotruncal septum, leading to varying degrees of pulmonary stenosis, pulmonary artery under development, misalignment of the infundibular septum, and a Ventricular Septal Defect (MacDonald, 2006).

The severity of pulmonary stenosis determines the direction of blood flow:

- Mild stenosis → Right ventricular pressure is only slightly increased → blood flows mostly left-to-right.
- Severe stenosis → Right ventricular pressure becomes very high → blood flows right-to-left through the VSD.

Right-to-left shunting reduces blood flow to the lungs, causing hypoxemia and cyanosis, which lead to weakness, fatigue, and shortness of breath. Severe obstruction also causes progressive polycythemia, exercise intolerance, and a shortened lifespan (Keith and Oyama, 2016) [26].

Biomarkers like NT-pro BNP frequently show elevated levels in response to pressure overload conditions, indicating the heart's struggle to cope with increased demand. Meanwhile, cardiac troponin I (cTnI) levels may rise, serving as a key indicator of myocardial stress and potential damage to cardiac tissue. Additionally, monitoring

oxygen saturation is crucial, as it provides important insights into the severity of respiratory or cardiovascular conditions and the overall adequacy of oxygen supply to tissues (Oyama and Singletary, 2010) [44].

### 4.8 Tricuspid Valve Dysplasia (TVD)

Tricuspid valve dysplasia is a congenital heart defect characterized by a spectrum of malformations that affect the tricuspid valve and its supporting structures, including the valve leaflets, chordae tendineae and papillary muscles. These abnormalities can present in various forms such as elongation or shortening of the valve leaflets, which may alter their normal shape and function. Additionally, there can be thickening of the tissue components, leading to a stiffer valve that struggles to open and close properly. In some cases, the leaflets may fuse together, resulting in restricted movement that impairs blood flow (Kellihan *et al*., 2015) [27].

Furthermore, abnormal attachments or misalignments of the chordae tendineae can disrupt the coordinated mechanisms that ensure the valve operates smoothly, ultimately compromising the delicate balance required for effective blood circulation. Each of these distinct variations can significantly impede the tricuspid valve's ability to manage the flow of blood between the right atrium and right ventricle. This dysfunction can consequently lead to a range of cardiovascular complications, manifesting as issues such as heart murmurs, enlargement of the heart chambers, or even heart failure over time (Snarr *et al.*, 2007) [54].

Normally, the tricuspid valve opens during diastole, allowing blood to flow from the right atrium to the right ventricle, and closes during systole to prevent backflow. In tricuspid valve dysplasia, the valve does not close properly, causing tricuspid regurgitation (Ware, 2013) [58]

**This leads to**

- Right atrial dilation
- Right ventricular eccentric hypertrophy (compensation for reduced stroke volume)
- Progressive right-sided heart enlargement
- Worsening regurgitation over time (Chan, 2017) [13]

Over time, the heart increasingly struggles to meet the body's demands, ultimately resulting in right-sided heart failure. This condition is marked by a concerning buildup of fluid in the body, known as edema, which can cause noticeable swelling in the extremities and abdomen. Additionally, as the heart's efficiency diminishes, blood flow to the lungs may become compromised, leading to a feeling of breathlessness. In more severe instances, patients may experience unsettling irregular heart rhythms, such as atrial fibrillation, which can further complicate their overall health status (Beijerink *et al*., 2017) [8].

NT-pro BNP levels often increase significantly when the heart experiences volume overload, serving as a critical indicator of the organ's difficulty in managing an excess of fluid. This biomarker reflects the strain on the cardiac system as it tries to maintain proper function under challenging conditions. In more advanced stages of heart disease, levels of cardiac troponin I (cTnI) may show slight elevations, signaling myocardial stress and potential injury. This rise in cTnI points to the heart working tirelessly and straining itself to cope with the demands imposed by the underlying condition, revealing the intricate relationship between heart function and the stressors it faces (Langhorn and Willesen, 2016) [35].

### 4.9 Mitral Valve Dysplasia (MVD)

Mitral valve development takes place during the latter part of the first trimester of fetal growth. The leaflets of the valve originate from the atrioventricular cushions, which are specialized structures that help guide the formation of the heart's internal architecture. As the heart continues to develop, these leaflets also incorporate a portion of the surrounding ventricular wall. To ensure proper function, they are anchored within the ventricle by a robust network of muscular tissue. This tissue eventually transforms into the chordae tendineae, thin yet strong strands that play a crucial role in connecting the leaflets to the papillary muscles. These muscles, located within the ventricles, work in concert to maintain the valve’s integrity and ensure proper blood flow throughout the heart (McGeady *et al*., 2006) [38].

In mitral valve dysplasia (MVD), various components of the valve complex—including the leaflets, chordae tendineae, and papillary muscles—may exhibit malformations. These irregularities can manifest as thickened leaflets that hinder proper closure, incomplete separation from the ventricular wall that compromises the valve's function, or abnormalities in the chordae tendineae that disrupt their supportive role. Such defects frequently result in valvular insufficiency and mitral regurgitation, which in turn leads to an overload of volume in the left side of the heart, creating challenges for its optimal performance (Andelfinger *et al.*, 2003) [3].

NT-pro BNP serves as a dependable biomarker for assessing cardiac strain, indicating how hard the heart is working to pump blood. In more severe cases, the presence of cardiac troponin I (cTnI) can provide insight into potential damage to heart muscle tissue. Furthermore, in the context of chronic disease, it becomes crucial to regularly monitor kidney function by evaluating levels of creatinine and urea, as these parameters can reflect the kidneys' ability to filter waste and manage fluid balance effectively (Klein *et al.*, 2022) [31]

## 5. Conclusion

Congenital heart anomalies in dogs present in a diverse array of forms, each affecting the heart's structure and function in unique ways. These abnormalities can severely influence a dog's overall quality of life, leading to symptoms that range from mild to life-threatening. A comprehensive understanding of these conditions and their contributing factors is crucial for veterinarians. This knowledge not only aids in the accurate diagnosis of these issues but also informs effective clinical management strategies and responsible breeding practices, ultimately ensuring better health outcomes for affected dogs and enhancing the well-being of future generations.

Many congenital cardiovascular defects—such as subaortic stenosis, patent ductus arteriosus, persistent right aortic arch, and ventricular septal defects—arise from interruptions that occur during crucial phases of embryonic heart development. These defects often stem from anomalies in the intricate process of forming and remodeling the cardiac tube, the septa that separate the heart chambers, and the outflow tracts that direct blood to the lungs and the rest of the body. Disruptions during these formative stages can lead to significant structural abnormalities, ultimately impacting the heart's function throughout life.

The integration of a diverse array of biomarkers, with a particular emphasis on cardiac proteins, presents a compelling opportunity to refine and enrich diagnostic practices in veterinary medicine. Veterinarians can achieve a deeper understanding of a dog's health by leveraging multiple indicators, which significantly enhances the accuracy of their diagnoses. To fortify this innovative approach, it is imperative that future research dedicates itself to studying large groups of dogs. This will not only aid in establishing robust reference ranges but also in determining diagnostic thresholds that can be seamlessly applied in clinical environments. Such advancements are vital for elevating the standard of care and enhancing treatment outcomes for canine patients, ultimately ensuring they receive the highest quality of medical attention tailored to their unique needs.

## References


1. Aanhaanen WTJ, Brons JF, Domínguez JN, Rana MS, Norden J, Airik R, *et al*. The Tbx2+ primary myocardium of the atrioventricular canal forms the atrioventricular node and the base of the left ventricle. Circ Res. 2009;104:1267-1274.
2. Akaike T, Minamisawa S. Role of ion channels in ductus arteriosus closure. Human Genetic Embryology. 2014;3:116-120.
3. Andelfinger G, Wright KN, Lee HS, Siemens LM, Benson DW. Canine tricuspid valve malformation, a model of human Ebstein anomaly, maps to dog chromosome. J Med Genet. 2003;40:320-324.
4. Anderson RH, Brown NA, Moorman AF. Development and structure of the heart: a key to understanding congenital heart disease. Heart. 2020;106:100-109.
5. Apitz C, Webb GD, Redington AN. Tetralogy of Fallot. Lancet. 2009;374:1462-1471.
6. Atkins CE, Gallo AM. Pulmonary hypertension and Eisenmenger's syndrome. In: Bonagura N, editor. Kirk's Current Veterinary Therapy XIII: Small Animal Practice. Philadelphia: W.B. Saunders; 2001. p. 756-761.
7. Atkinson AJ, Colburn WA, Degruttola VG, Demets DL, Downing GJ. Biomarkers and surrogate endpoints: preferred definitions and conceptual framework. Int J Clin Pharmacol Ther. 2001;69:89-95.
8. Beijerink NJ, Oyama MA, Bonagura JD. Congenital heart disease. In: Ettinger SJ, Feldman EC, Cote E, editors. Textbook of Veterinary Internal Medicine. 8th ed. Missouri: Elsevier Saunders; 2017. p. 1225-1227.
9. Boswood A, Dukes-McEwan J, Loureiro J, James RA, Martin M, Stafford-Johnson M, *et al*. The diagnostic accuracy of different natriuretic peptides in the investigation of canine cardiac disease. J Small Anim Pract. 2008;49:26-32.
10. Bottio T, Vida V, Padalino M, Gerosa G, Stellin G. Early and long-term prognostic value of Troponin-I after cardiac surgery in newborns and children. Eur J Cardiothorac Surg. 2006;30:250-255.
11. Buchanan JW. Patent ductus arteriosus: pathophysiology, clinical aspects, and therapy. J Vet Cardiol. 2001;3:7-16.
12. Bussadori C, Amberger C, Le Bobinnec G, Lombard CW, Domenech O. Pulmonic stenosis: pathophysiology and morphology in dogs. J Vet Cardiol. 2000;2:17-24.
13. Chan KMJ. Anatomy of the tricuspid valve and pathophysiology and functional tricuspid regurgitation. In: Chan KMJ, editor. Functional Mitral and Tricuspid Regurgitation. 1st ed. Springer International Publishing; 2017. p. 157-162.
14. Christoffels VM, Smits GJ, Kispert A, Moorman AFM. Development of the pacemaker tissues of the heart. Circ Res. 2010;106:240-254.
15. Clement OP, Puceat M, Walsh MP, Vassort G. Protein kinase C enhances myosin light chain kinase effects on force development and ATPase activity in rat single skinned cardiac cells. Biochem J. 1992;285:311-317.
16. Coceani F, Baragatti B. Mechanisms for ductus arteriosus closure. Semin Perinatol. 2012;36:92-97.
17. De Lima GV, Ferreira FS. N-terminal-pro brain natriuretic peptides in dogs and cat: a technical and clinical review. Veterinary World. 2017;10:1072-1082.
18. De Madron E, Chetboul V, Bussadori C. Clinical Echocardiography of the Dog and Cat. 2nd ed. Elsevier; 2019. p. 170-171.
19. Dunn ME, Coluccio D, Hirkaler G, Mikaelian I, Nicklaus R. The complete pharmacokinetic profile of serum cardiac troponin I in the rat and the dog. Toxicol Sci. 2011;123:368-373.
20. Dyer LA, Rugonyi S. Blood flow and genetic mutations in conotruncal congenital heart disease. J Cardiovasc Dev Disord. 2021;8:90-97.
21. Gittenberger-de Groot AC, Bartelings MM, Deruiter MC, Poelmann RE. Basics of cardiac development for the understanding of congenital heart malformations. Pediatr Res. 2005;57:169-176.
22. Goetze JP, Bruneau BG, Ramos HR, Ogawa T, De Bold MK, De Bold AJ. Cardiac natriuretic peptides. Nat Rev Cardiol. 2020;17:698-717.
23. Hanneman K, Newman B, Chan F. Congenital variants and anomalies of the aortic arch. Radiographics. 2017;37:32-51.
24. Hariu CD, Saunders AB, Gordon SG, Norby B, Miller MW. Utility of N-terminal pro-brain natriuretic peptide for assessing hemodynamic significance of patent ductus arteriosus in dogs undergoing ductal repair. J Vet Cardiol. 2013;15:197-204.
25. Keane FM, Nadvi NA, Yao TW, Gorrell MD. Neuropeptide Y, B-type natriuretic peptide, substance P and peptide YY are novel substrates of fibroblast activation protein-alpha. FEBS J. 2011;278:1316-1332.
26. Keith SN, Oyama M. Congenital heart disease. In: Smith FWK, Oyama MA, Tilley LP, Sleeper MM, editors. Manual of Canine and Feline Cardiology. 5th ed. St. Louis: Riverport Lane; 2016. p. 218-238.
27. Kellihan HB, Chun R, Henik RA, Stepien RL. Use of echocardiography and biomarkers in diagnosis of congenital heart diseases in dogs. J Vet Cardiol. 2015;17:239-246.
28. Kelly RG. The second heart field. Curr Top Dev Biol. 2012;100:33-65.
29. Kim M, Platt MJ, Shibasaki T, Quaggin SE, Backx PH, Seino S, *et al*. GLP-1 receptor activation and Epac2 link atrial natriuretic peptide secretion to control of blood pressure. Nat Med. 2013;19:567-575.
30. Kirby ML. Molecular embryogenesis of the heart. Pediatr Dev Pathol. 2002;5:516-543.
31. Klein S, Nolte I, Soler JLG, Leitz P, Sehn M, Raue JF, *et al*. Evaluation of new and old biomarkers in dogs

with degenerative mitral valve disease. Vet Res. 2022;18:256-260.
32. Kloesel B, DiNardo JA, Body SC. Cardiac embryology and molecular mechanisms of congenital heart disease - a primer for anesthesiologists. Anesth Analg. 2016;123:551-569.
33. Kobayashi K, Hori Y, Chimura S. Plasma N-terminal pro B-type natriuretic peptide concentrations in dogs with pulmonic stenosis. J Vet Med Sci. 2014;76:827-831.
34. Lahaye S, Lincoln J, Garg V. Genetics of valvular heart disease. Curr Cardiol Rep. 2014;16:1-9.
35. Langhorn R, Willesen JL. Cardiac biomarkers in dogs: a review. J Vet Intern Med. 2016;215:36-50.
36. Lin CJ, Lin CY, Chen CH, Zhou B, Chang CP. Partitioning the heart: mechanisms of cardiac septation and valve development. Development. 2012;139:3277-3299.
37. McDonald KA. Congenital heart diseases of puppies and kittens. Vet Clin North Am Small Anim Pract. 2006;36:503-531.
38. McGeady TA, Quin PJ, Fitzpatrick ES, Ryan MT. Cardiovascular system. In: Veterinary Embryology. 1st ed. Wiley-Blackwell; 2006. p. 117.
39. Midgett M, Thornburg K, Rugonyi S. Blood flow patterns underlie developmental heart defects. Am J Physiol Heart Circ Physiol. 2017;312:H632-H642.
40. Minette MS, Sahn DJ. Ventricular septal defects. Circulation. 2006;114:190-197.
41. Moorman AFM, Christoffels VM. Cardiac chamber formation: development, genes, and evolution. Physiol Rev. 2003;83:1223-1267.
42. Morgan KRS, Bray JP. Current diagnostic tests, surgical treatments and prognostic indicators for vascular ring anomalies in dogs. J Am Vet Med Assoc. 2019;254:728-733.
43. Noland TA, Kuo JF. Protein kinase C phosphorylation of cardiac troponin I or phosphorylation of cardiac troponin I or myosin MgATPase activity. J Biol Chem. 1991;266:4974-4978.
44. Oyama MA, Singletary GE. The use of NT-proBNP assay in the diagnosis of heart failure in dogs. Vet Clin North Am Small Anim Pract. 2010;40:545-558.
45. Oyama MA, Sisson DD. Cardiac troponin-I concentration in dogs with cardiac disease. J Vet Intern Med. 2004;18:831-839.
46. Patil H, Vaidya O, Bogart D. A review of causes and systemic approach to cardiac troponin elevation. Clin Cardiol. 2011;34:723-728.
47. Puceat M, Clement O, Lechene P, Pelosin JM, Ventura-Clapier R, Vassort G. Neurohormonal control of calcium sensitivity of myofilaments in rat single heart cells. Circ Res. 1990;67:517-524.
48. Ro WB, Kang MH, Park HM. Serial evaluation of cardiac biomarker NT-proBNP with speckle tracking echocardiography in a 6-year-old Golden Retriever dog with subaortic stenosis and dilated cardiomyopathy. Vet Q. 2020;40:77-82.
49. Saunders AB, Miller MW, Gordon SG. Pulmonary embolization of vascular occlusion coils in dogs with patent ductus arteriosus. J Vet Intern Med. 2004;18:663-666.
50. Schleich JM, Abdulla T, Summers R, Houyel L. An overview of cardiac morphogenesis. Arch Cardiovasc Dis. 2016;106:612-623.
51. Schober KE, Baade H. Comparability of left ventricular M-mode echocardiography in dogs performed in long-axis and short-axis. Vet Radiol Ultrasound. 2000;41:543-549.
52. Schorn C, Hildebrandt N, Schneider M, Schaub S. Anomalies of the aortic arch in dogs: evaluation with the use of multidetector computed tomography angiography. BMC Vet Res. 2021;17:387.
53. Semenov AG, Postnikov AB, Tamm NN, Seferian KR, Karpova NS, Bloshchitsyna MN, *et al*. Processing of pro–brain natriuretic peptide is suppressed by O-glycosylation in the region close to the cleavage site. Clin Chem. 2009;55:489-498.
54. Snarr BS, Wirrig EE, Phelps AL, Trusk TC, Wessels A. Spatiotemporal evaluation of the contribution of the dorsal mesenchymal protrusion to cardiac development. Dev Dyn. 2007;236:1287-1294.
55. Strimbu K, Tavel JA. What are biomarkers? Curr Opin HIV AIDS. 2010;5:463-466.
56. Tidholm A, Jonsson L. Subaortic stenosis in the dog: a retrospective study of 51 cases. J Small Anim Pract. 1997;38:311-316.
57. Waldo KL, Hutson MR, Ward CC, Zdanowicz M, Kirby ML. Cardiac neural crest is necessary for normal addition of the myocardium to the arterial pole from the secondary heart field. Dev Biol. 2005;281:66-77.
58. Ware WA. Congenital cardiovascular diseases. In: Ware WA, editor. Cardiovascular Disease in Small Animal Medicine. 4th ed. London: Boehringer Ingelheim; 2013. p. 246-247.
59. Wessels A, Anderson RH, Markwald RR, Webb S, Brown NA, Viragh S, *et al*. Atrial development in the human heart: an immunohistochemical study with emphasis on the role of mesenchymal tissues. Anat Rec. 2000;259:288-300.
60. Yamagishi H, Maeda J, Uchida K, Tsuchihashi T, Nakazawa M, Aramaki M, *et al*. Molecular embryology for an understanding of congenital heart diseases. Anat Sci Int. 2009;84:88-94.