Appendix VIII

Cardiac Screening in Lynx

Authors: Laurent Locquet, Laurens Van Mulders
Reviewers: Linking Lynx Health Working Group

Introduction

In recent years there has been increasing recognition of the importance of cardiac health in Lynx populations as specific cardiovascular diseases have been observed in both wild individuals and in animals under human care. Wild lynx populations can be particularly vulnerable to introduction of (genetic) cardiovascular disorders due to their low population density. To improve our understanding of cardiac health in Lynx populations, the development of a specific cardiac screening protocol is imperative.

In this context, we present a focused cardiac ultrasound screening protocol designed for non-invasive assessment of the cardiovascular function in individual animals. By facilitatating specific disease detection, valuable data can be provided on the prevalence and distribution of cardiovascular disease within Lynx populations. This might prevent the introduction or spread of potential hereditary cardiac anomalies in wild Lynx populations.

Cardiac Screening Protocol

Instrumentation:

Echocardiographic examinations are optimally conducted using phased-array sector (or curvilinear) scanning transducers (Figure 1, right), incorporating a colour doppler modality coupled to a portable ultrasound device (Figure 1, left). To achieve superior temporal resolution, it is essential to reduce the field depth and minimize the sector angle (sector width). Recommended transducer frequencies for Lynx encompass the range of 4–8 MHz.

Figure 1. *(Left) Example of a portable ultrasound device with colour doppler modality. (Right) Example of a suitable phased-array sector transducer, that encompasses the range of 4–8 MHz.*

Preparation:

The animals should be positioned in right lateral recumbency with the ultrasound device placed on the cranial end of the animal, creating the preferred examination set-up (Figure 2, left). The transducer is applied from a ventral direction, often utilizing a specially designed table with cutouts for this purpose (Figure 2, right). The apex beat of the heart can be palpated or auscultated first to ensure selection of a correct transducer application site; this will usually be around the 4-5th intercostal space. Acoustic transmission can be enhanced by either trimming the hair over the transducer application site and/or by thoroughly saturating the fur with ultrasound coupling gel.

Figure 2. *(Left) Preferred examination set-up with the animal in right sternal recumbency and the ultrasound device placed on the cranial end of the animal. (Right) A specially designed table with cutouts allows application of the transducer from a ventral direction.*

Transducer Positions and Cardiac Views:

Transthoracic echocardiography provides access through relatively limited windows. On the right side of the ventral thorax, adjacent to the sternum **the right parasternal (RPS) window** is situated. Typically, there are two or more rib spaces available for obtaining (RPS) views, with a cranial location typically corresponding to the 4th intercostal space and a more caudal location at the 5th. The central axis of the left ventricle can be conceptualized as an imaginary line extending from the cardiac apex to the base at the center of the left ventricular lumen (Figure 3, top left).

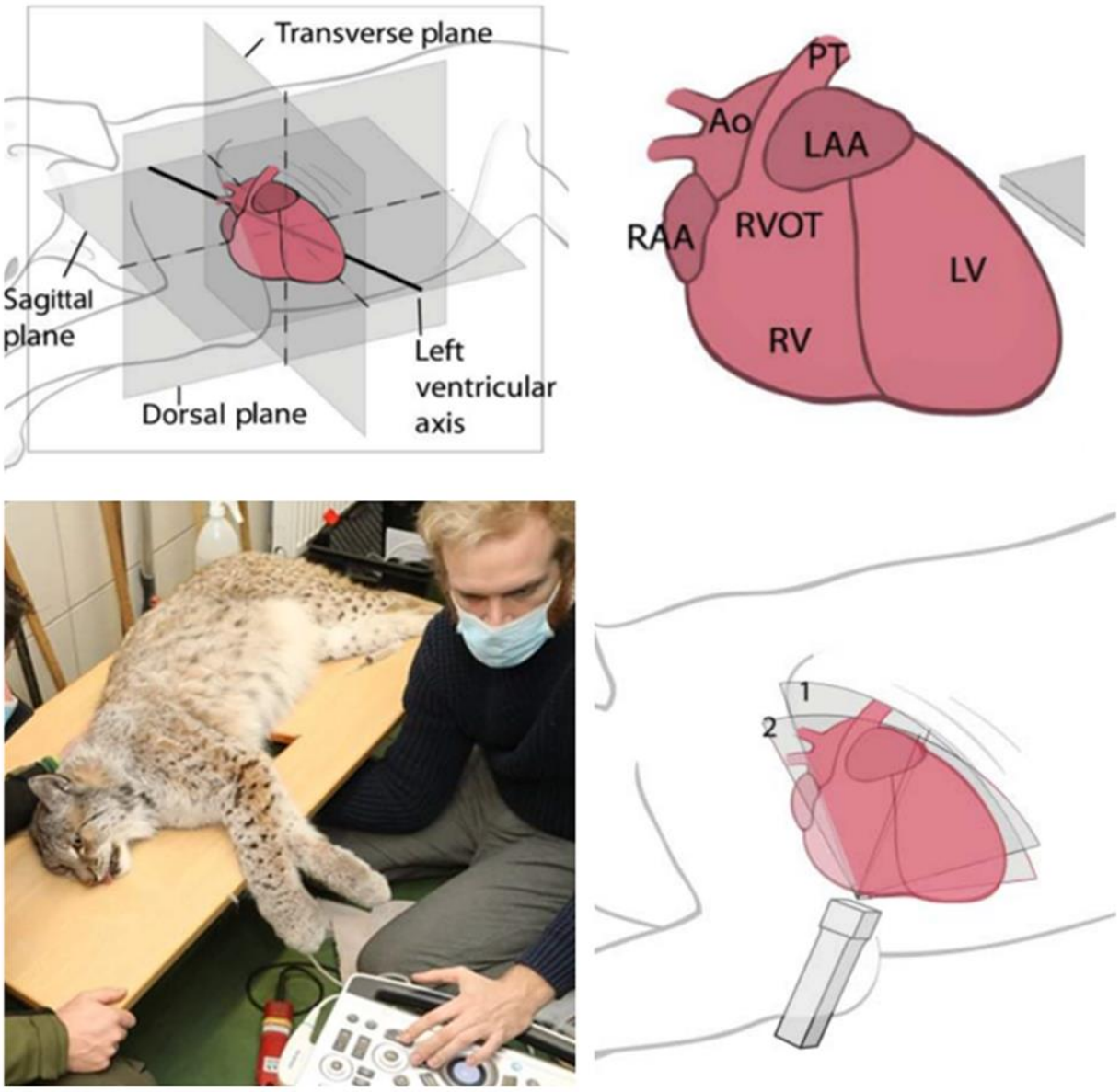


Figure 3. *(Top) The left ventricular axis can be conceptualized as an imaginary line extending from the cardiac apex to the base of the left ventricular lumen (LV; left ventricle, RV; right ventricle, Ao; aorta, PT; pulmonary truncus, RVOT; right ventricular outflow tract.) (Bottom) (1) Orienting the transducer to parallel this axis with the scanning plane gives* the *right parasternal four-chamber view, note that the central transducer beam is perpendicular to the left ventricular long axis. (2) Slight cranial angulation and counterclockwise twisting of the transducer, gives the right parasternal five-chamber view*

Right Parasternal Long-Axis Views

When orienting the transducer to include or parallel this axis within the scanning plane, a long-axis image is generated (Figure 3, bottom left and right). To capture suitable images, the transducer should be positioned within the chosen rib space in such a way that the central transducer beam is perpendicular to the left ventricular long axis (Figure 3, bottom right (1)), resulting in the **right parasternal four-chamber view** (Figure 4, top, **Image 1**), which enables visualization of the four heart chambers, cardiac walls and atrioventricular valves (Figure 4, top).

Subsequent slight cranial angulation and counterclockwise twisting of the transducer results in **the right parasternal five-chamber view** (Figure 3, bottom right, **Image 2**), which enables visualization of the aortic valve and root from this perspective (Figure 4, bottom).

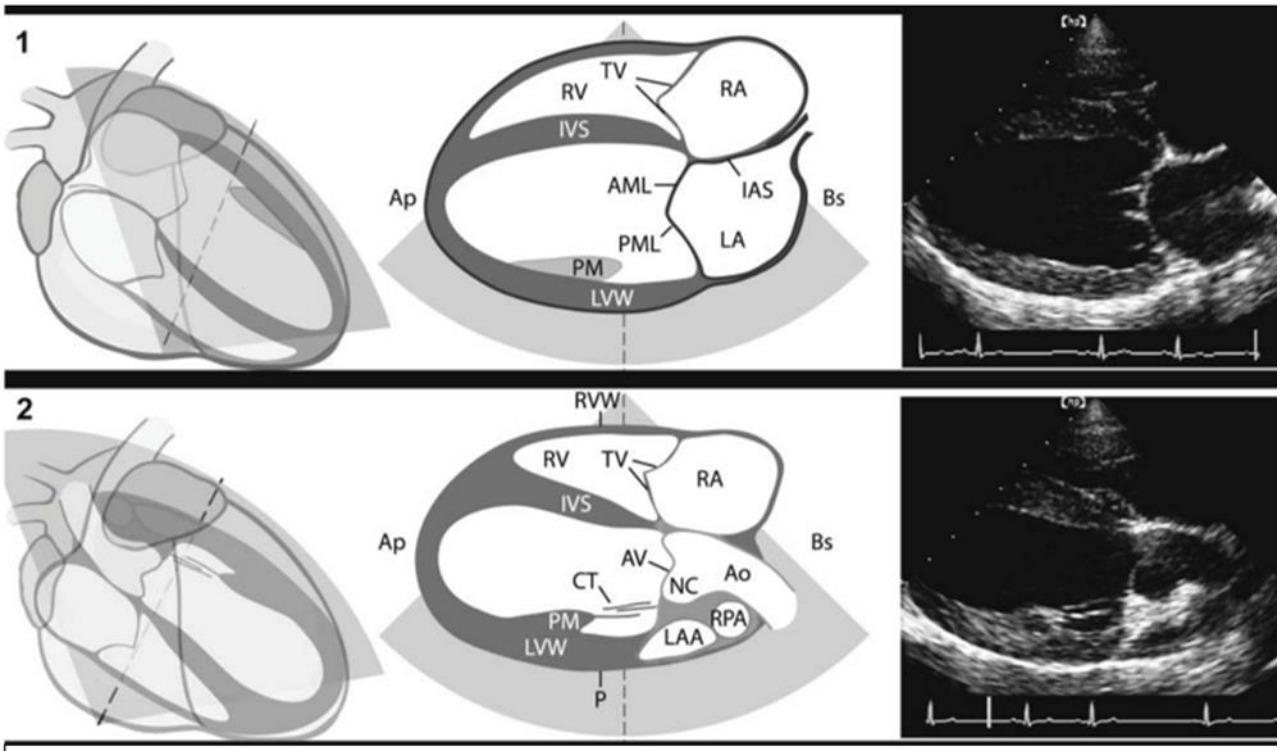


Figure 4. *(Top,* **Image 1***)* *Right parasternal four-chamber view: Visualisation of the left ventricular lumen and free wall (LVW), the mitral valve with bot leaflets (AML and PML), the left atrium, the interventricular (IVS) and interatrial septum (IAS), the right ventricular lumen (RV) and free wall (RVW), tricuspid valve (TV) and right atrium (RA). (Bottom,* **Image 2***) Right parasternal five-chamber view: Visualisation of the left ventricular lumen and free wall (LVW), the aortic valve (AV), the aortic root (Ao), the interventricular septum (IVS), the right ventricular lumen (RV) and free wall (RVW), tricuspid valve (TV) and right atrium (RA).*

Right Parasternal Short Axis Views

Starting from the right-parasternal four-chamber view, a counterclockwise rotation of the transducer towards a 90-degree position compared to the left ventricular axis and upward angulation of the transducer away from the animal, results in the **right parasternal short axis view at the level of the aortic root** (Figure 5, top left, **Image 3**), which enables additonal visualization of the right heart with the pulmonary valve and trunk, the left atrium and aortic valve/root from this perspective (Figure 5, bottom).

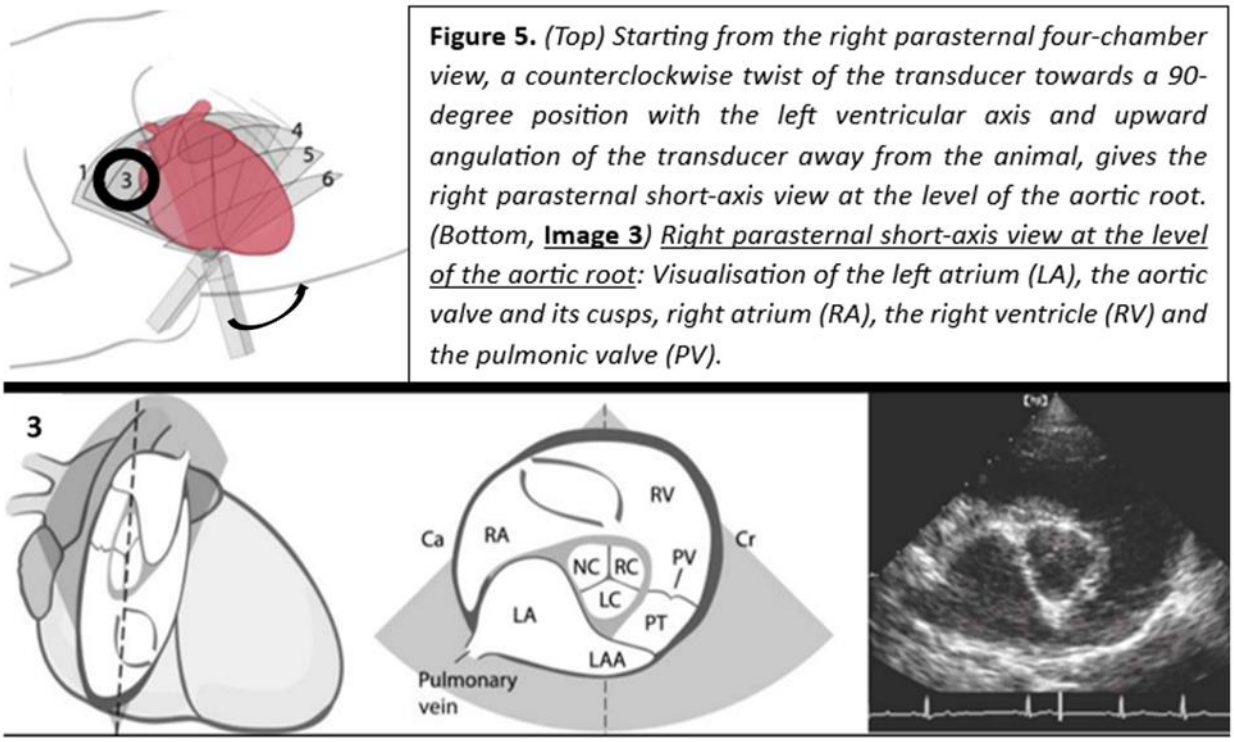


Figure 5. *(Top) Starting from the right parasternal four-chamber view, a counterclockwise twist of the transducer towards a 90-degree position with the left ventricular axis and upward angulation of the transducer away from the animal, gives the right parasternal short-axis view at the level of the aortic root. (Bottom,* **Image 3***) Right parasternal short-axis view at the level of the aortic root: Visualisation of the left atrium (LA), the aortic valve and its cusps, right atrium (RA), the right ventricle (RV) and the pulmonic valve (PV).*

The aortic root occupies the center of the image with its three cusps (hence the name of this view: short axis transaortic view). Only one or two of the cusps are usually visible. The right ventricular outflow tract is wrapped around the aortic root, allowing visualisation of the pulmonic valve and the main pulmonary artery. The left atrium and its appendage appear beneath the aorta, bordered by the right atrium on the left of the screen. The tricuspid valve forms a circular structure.

Additional images allong the right parasternal short-axis can be obtained by angulating the probe towards and away from the animal disecting the heart in a transverse plane at different levels (Figure 5, top left), an overview of these levels is presented below and can be found in Figure 6:

Right parasternal short-axis view at the:

- (Apex level)

On this view (referred to as a transapical view) only the extremity of the left ventricular cavity is visible. It is small in size compared to the predominant myocardium.

- **Papillary muscle level**

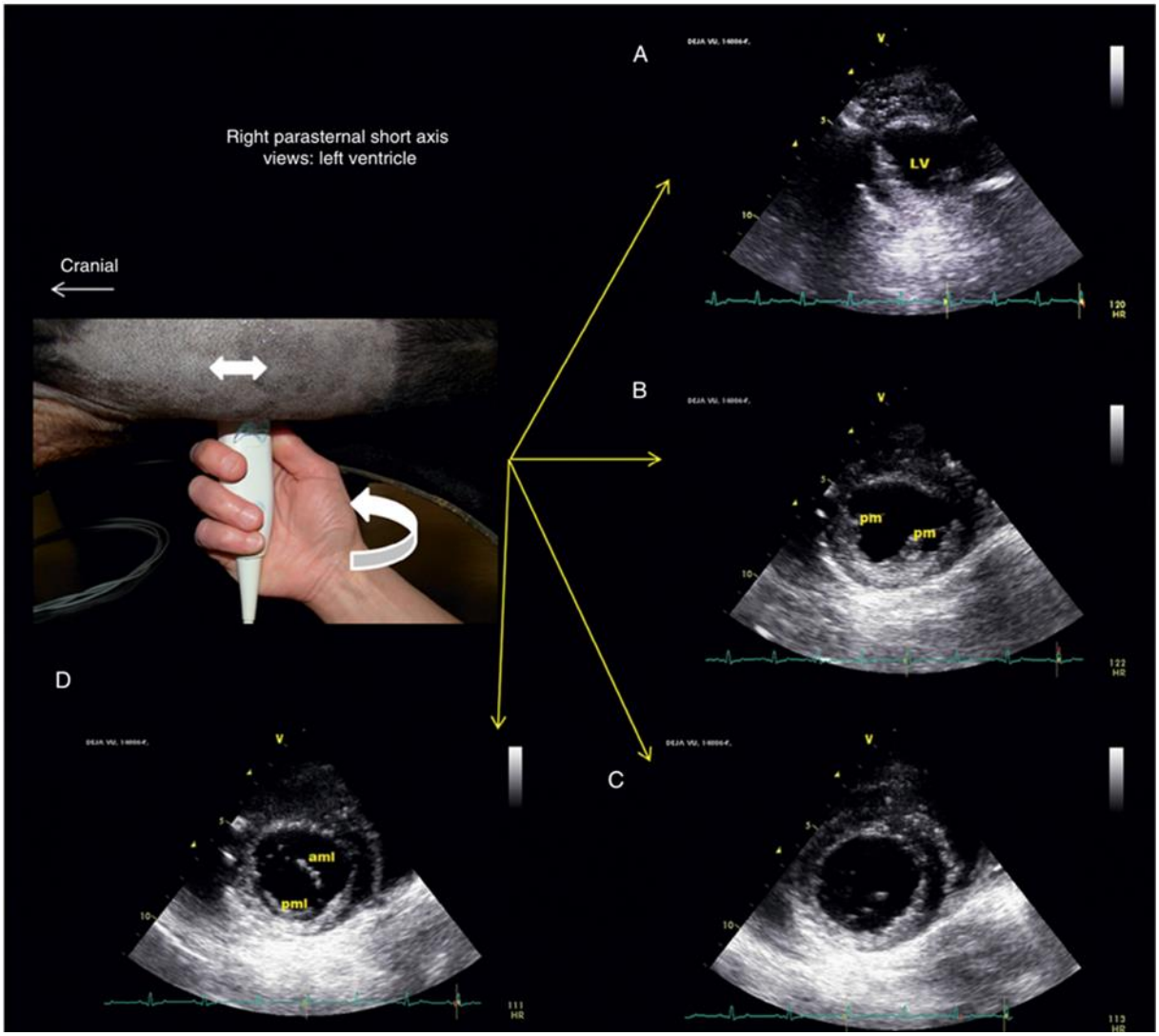
The right ventricular cavity becomes visible along with the cranial and caudal papillary muscles (hence the name of this view: short axis transpapillary view).

- (Chordae tendinae level)

At this level the left ventricular cavity is larger than on the preceding view (hence the name of this view: short axis transventricular view).

- (Mitral valve level)

At this level the mitral valve appears as a “fish mouth,” opening in diastole and closing in systole (hence the name of this view: transmitral short axis). The left ventricular cavity is now perfectly circular. The right ventricular outflow tract is visible.



Assesment and Documentation of Cardiac Images:

1) Right parasternal four-chamber view

- The left atrium should subjectively not be larger than ½ of the left ventricle
- The interatrial septum should be complete (artefact at the fossa ovalis might sometimes give the false impression of a atrial septal defect)
- The left ventricle should subjectively have a bullet shape with the tip of the bullet facing the apex.
- The tips of the leaflets of the mitral valve should not look nodular and irregular
- There should be no or only trivial mitral valve regurgitation with the colour doppler.

2) Right parasternal five-chamber view

- There should be no or only trivial aortic valve regurgitation with the colour doppler.

3) <u>Right parasternal short axis views:</u>

<u>At the level of the aortic root:</u>

- The ratio of the left atrium to the aortic root should be less than 1.6.
- The (tips of the) leaflets of the aortic, pulmonic and tricuspid valve should not look nodular and irregular.
- There should be no or only trivial tricuspid valve regurgitation with the colour doppler.
- There should be no or only trivial pulmonic valve regurgitation with the colour doppler.
- The interatrial septum should be complete

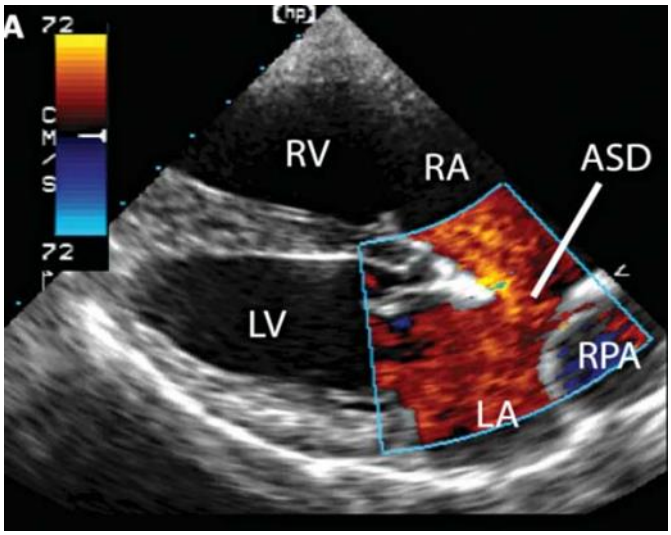
<u>At the level of the papillary muscles:</u>

- You should see 'a mushroom shape' in the left ventricle. The roof of the mushroom should not be flattened, but should be bulging upwards.
- There should be a good contraction visible. Poor contraction can be secondary due to e.g. the anesthesia (depends on the protocol), cardiac or systemic diseases.

<u>Examples of specific diseases:</u>

1) <u>Atrial septal defects</u>

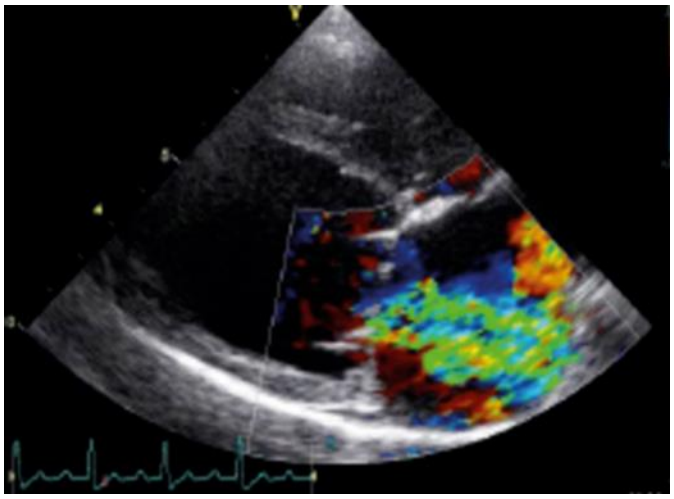
Atrial septal defects are congenital and potential hereditary cardiac disorders and have anecdotally been reported in free-roaming Lynx populations; therefore, specific attention should be paid to the interatrial septum during cardiac ultrasound screening.



Right parasternal four-chamber view and colour doppler imaging at the interatrial septum showing blood flow from the left to the right atrium compatible with an atrial septal defect.

2) <u>Chronic degenerative valve diseases</u>

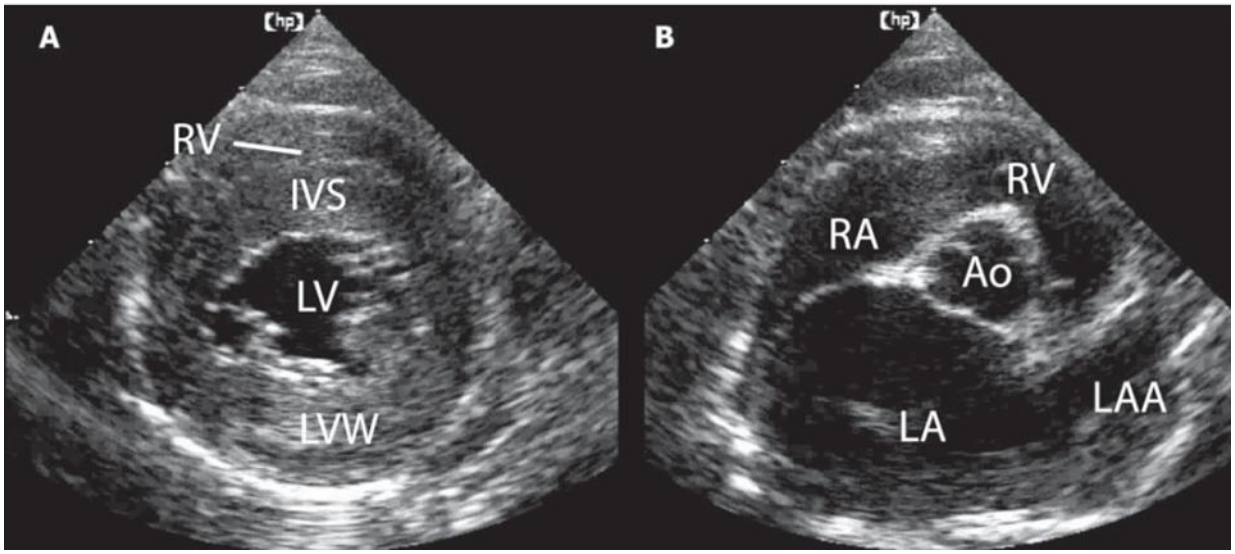
Chronic degenerative valve diseases are common acquired heart diseases and are potentially hereditary. Chronic degenerative mitral and tricuspid valve disease has been reported in free-roaming Lynxes and in animals under human care and have the potential to progress towards cardiomegaly and subsequent congestive cardiac failure; therefore, specific attention should be paid to the mitral (and tricuspid) valves during cardiac ultrasound screening.



Right parasternal four-chamber view and colour doppler imaging at the mitral valve showing turbulent blood flow from the left ventricle towards the left atrium, compatible with moderate to severe mitral valve regurgitation, often seen because of chronic degenerative mitral valve disease.

3) Hypertrophic cardiomyopathy

Hypertrophic cardiomyopathy (phenotype), defined as concentric thickening of the (most commonly) left ventricular wall, can be primary (including a possible hereditary component) or secondary to an underlying disease (e.g., chronic systemic hypertension). HCM is the most commonly acquired heart disease in small Felidae. A similar disease has been reported in free-roaming Lynx populations and in animals under human care, with the potential to progress towards cardiomegaly and subsequent congestive cardiac failure; therefore, specific attention should be paid to the ventricular walls, the interventricular septum and lumen of the left ventricle.



A) Right parasternal short axis view at the level of the papillary muscles showing thickening of the left ventricular free wall and the interventricular septum with a decrease in the left dimension of the ventricular lumen, compatible with a phenotype of hypertrophic cardiomyopathy. B) Right parasternal short axis view at the level of the aortic root showing atrial dilation compatible with left atrial enlargement.

Checklist:

Image		OK
1)	**Right parasternal four-chamber view**	
	General (video)	
	Colour dopler on mitral valve (video)	
	Colour dopler on interatrial septum (video)	
2)	**Right parasternal five-chamber view**	
	General (video)	
	Colour dopler on aortic valve (video)	
3)	**Right parasternal short axis views**	
	General (video) of all views – *at papillary muscle and aortic root*	
	Colour dopler on pulmonic valve (video) – *view at the aortic root level*	
	Colour dopler on the tricuspid valve (video) – *view at the aortic root level*	

Interpretation of images:

Images and videos can be send in DICOM-formats, together with additional documentation on the signalment and general health (including cardiac auscultation findings), to cardiorenalwildlife@gmail.com for further interpretation.

References:

Cardiac ultrasound images and protocols are directly adapted from:

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