VISUALSONICS
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Imaging Guide

Guide to Small Animal **Echocardiography** using the Vevo® Imaging Systems

System Compatibility: This guide contains instructions and suggestions for work on the Vevo2100, VevoLAZR, Vevo 3100 systems and transducers from the MS, MZ and MX series.

Objective

This document is intended as a guide for researchers through the steps of a complete echo exam in small animals with references to anatomy, imaging planes, imaging modes and related measurements and calculations.

Anatomical References

Murine bodies are parallel to the ground in the normal position and so the heart does not rest on the diaphragm like the human heart, thus having more room to move around within the pericardial cavity.

Generally, the murine heart has an ellipsoidal shape and given the position of the lungs, relative to the heart, and narrow sternum, specifically in mice, the optimum imaging windows are in the parasternal long axis (PLSA) on both the left and right side during transthoracic investigations.

The terms *superior (*cranial), *inferior (*caudal), *anterior* (ventral) and *posterior* (dorsal) are used to define the position of the transducer relative to the body of the animal.

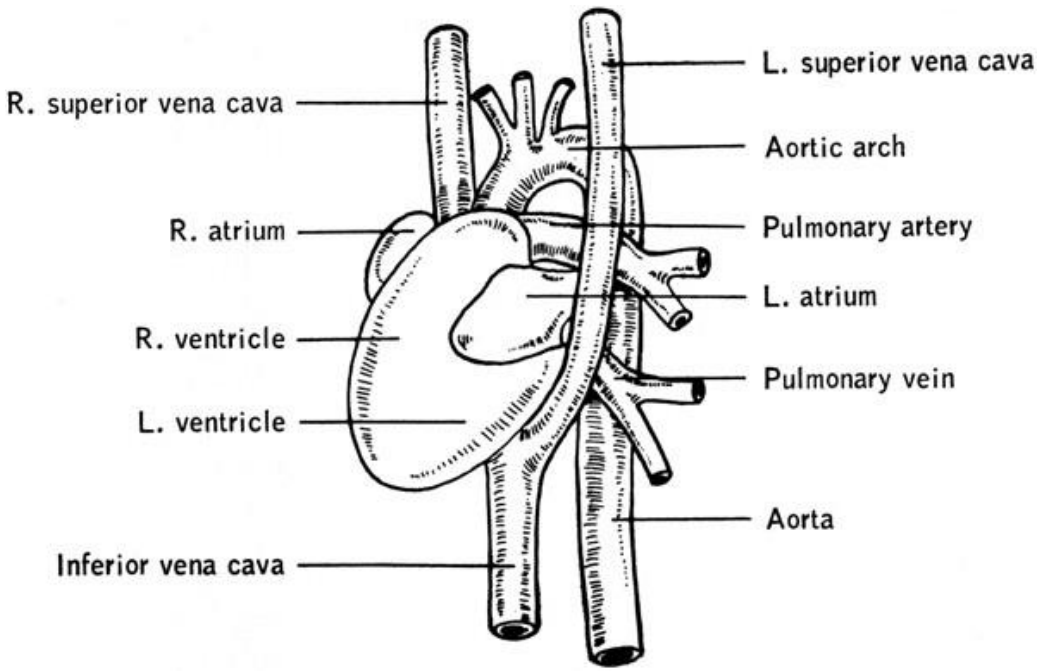

Figure 1 - Mouse cardiac anatomy

[http://www.informatics.jax.org/greenbook/figures/figure13-2.shtml]

Selecting the Appropriate Transducer

The criteria for selecting the appropriate transducer are: animal size and pathology.
For Adult Mice or Large size mice the ideal transducer would be the MS400 (30MHz). For *young mice and smaller sizes* excellent images can also be obtained using the MS550D (40MHz).
For Rat imaging, depending on the size of the animal, the best choices are the MS250 (25MHz) or the MS200 (20MHz) transducers.

Animal Preparation – Animal preparation should adhere to the research institutions SOP.

Left Ventricular Assessment

Parasternal Long Axis (PLAX) View

Visible anatomy in PLAX view:

- Left ventricle (LV)
- Right ventricle
- Left atrium
- LV Anterior wall
- LV Posterior wall
- Intraventricular septum (IVS)
- Aorta
- Pulmonary artery
- Aortic valve
- Mitral valve
- Pulmonary veins
- Papillary muscles

Transducer position:
Place the transducer in parallel to the longitudinal direction, with the notch pointing towards the animal's head. Then rotate the transducer approximately 35° counter-clockwise. The face of transducer will be positioned diagonally with the mouse imaging platform (aligning the corner of the board with the transducer is a way to find the correct angle).

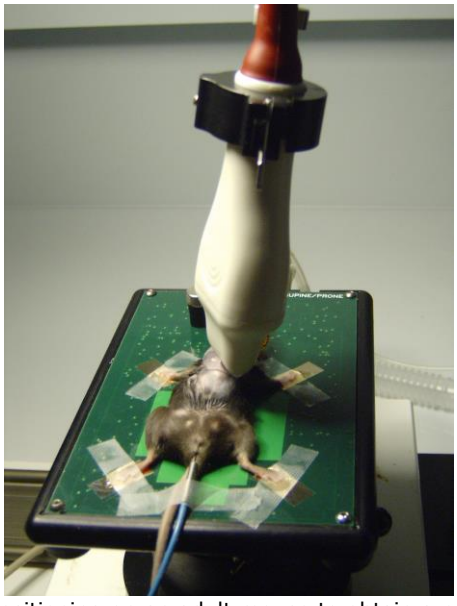

Figure 2 - Transducer positioning on an adult mouse to obtain a parasternal long axis view

Parasternal Long axis view in B-Mode

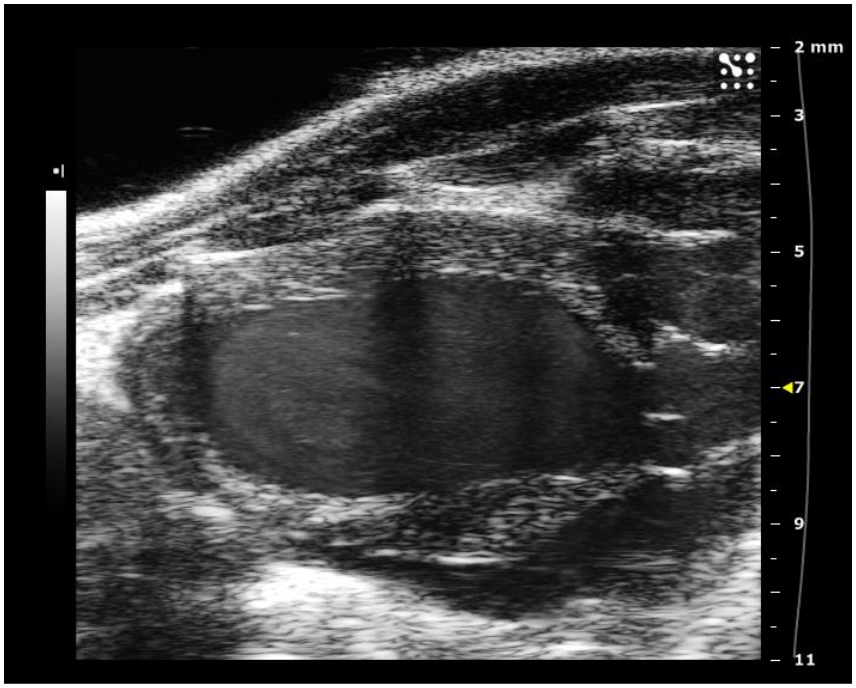

Figure 3 - B-Mode image of the left ventricle in the parasternal long axis view

Measurements and Calculations for PLAX in B-Mode

Measurements	Calculations
IVS:d,s	A:d,s
LVID:d,s	V:d,s
LVPW:d,s	SV
LA	FS
Ao Sinus	EF
LVEnL:d,s	CO
ENDOmajr:d,s	LV Mass corr
EPImajr:d,s	LV Mass
SimpLength:d,s	

Parasternal Long Axis view in M-Mode

To obtain a correct M-Mode image and data set, place the M-Mode sample volume the LV must be horizontal in orientation. Place the M-Mode axis immediately next to the papillary muscle.

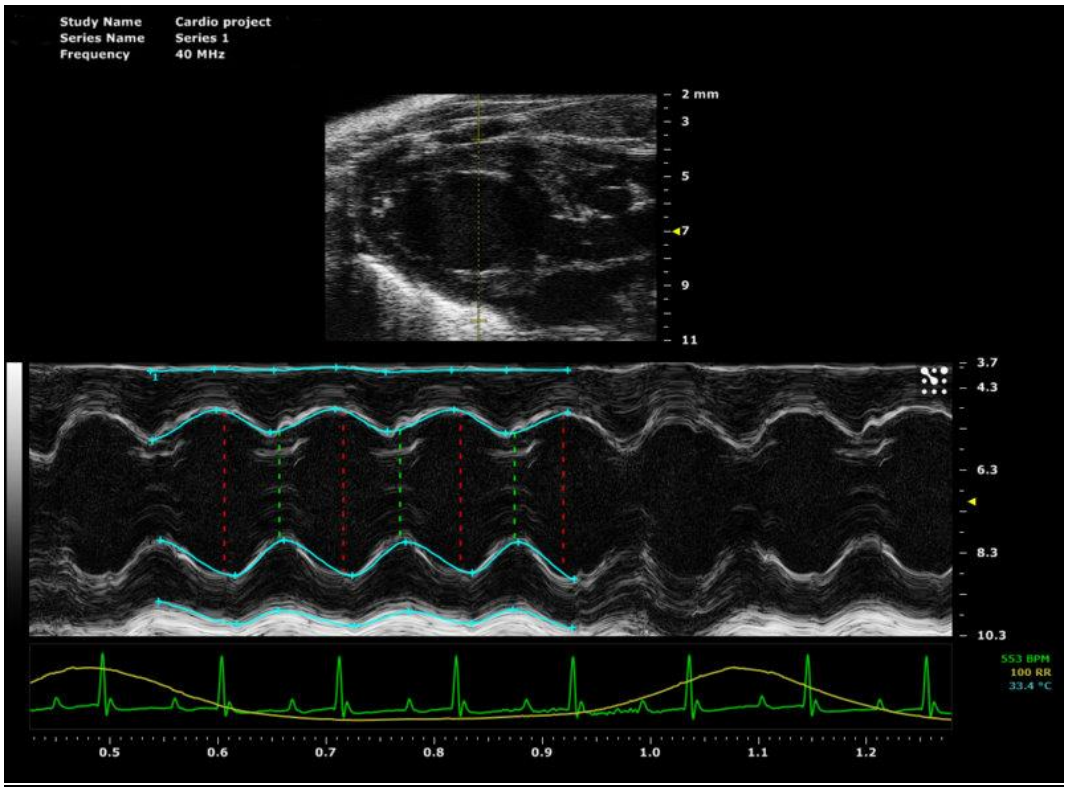

Figure 4 - M-Mode image of parasternal long axis view displaying motion of the anterior and posterior walls.

Measurements and Calculations for PLAX in M-Mode

Measurements	Calculations
RVID:d,s	A:d,s
IVS:d,s	V:d,s
LVID:d,s	SV
LVPW:d,s	FS
LA	EF
Ao Root	CO
LVET	LV Mass corr
LV Trace	LV Mass

Parasternal Long Axis in Color and PW Doppler Modes

Color and PW Doppler Modes are excellent tools for analysis of flow direction and velocity profiles. They are usually used in combination with B-Mode in that once the view has been found in B-Mode, through slight translation of the transducer a number of important vascular landmarks can be identified using Color Doppler Mode and then PW Doppler Mode.

Aortic Root
Pulmonary artery
Aortic valve
Mitral valve
Pulmonary veins

Aortic Root flow identification using Color Doppler Mode.

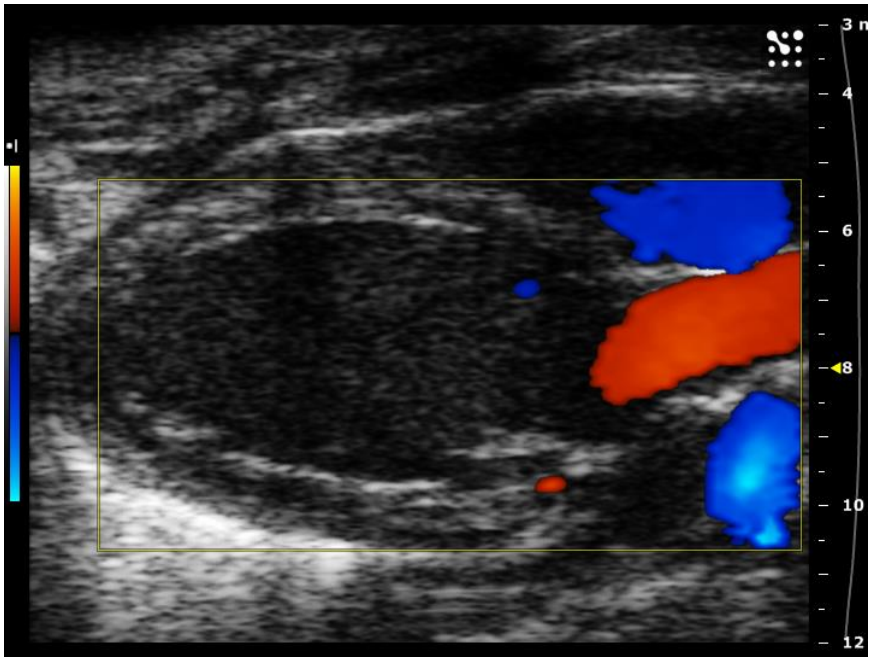

Figure 5 - Aortic Root in Color Doppler

Pulmonary Artery flow direction and velocity assessment using Color and PW Doppler Modes.

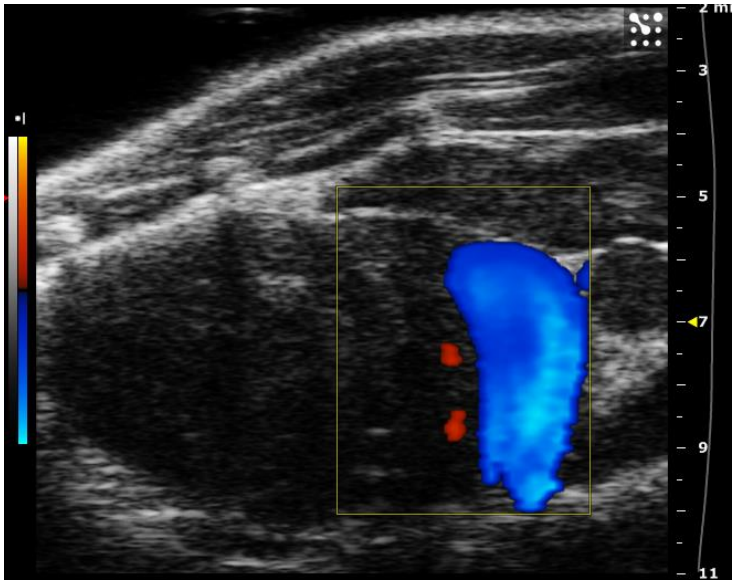

Figure 6 - Pulmonary Artery directional flow in Color Doppler Mode

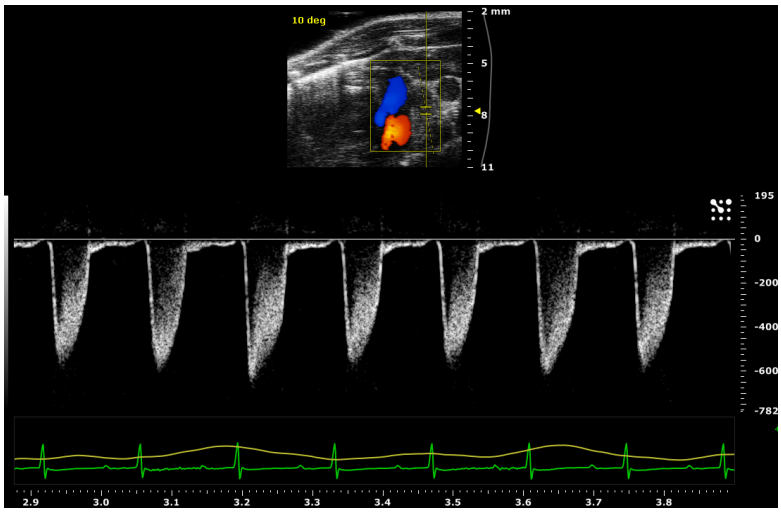

Figure 7 - Pulmonary Artery velocity display in PW Doppler Mode

Measurements and Calculations for PV in PW Doppler Mode

Measurements	Calculations
PV VTI	PV SV
PV Peak Velocity	PV CO
PAT	PVA
PET	PV Peak Pressure
	PAT/PET
	MPAP

Superior (Cranial) Vena Cava Flow Display in PW Doppler Mode

The right and left superior vena cava generally run parallel to the aorta and are located slightly to the right and left relative to the aorta. Their flow can be assessed by PW Doppler Mode.

Figure 8 - PW Doppler Mode flow distribution in the Superior Vena Cava.

Pulmonary Vein

Pulmonary veins run into the left atrium. They can be visualized and assessed in a standard parasternal long axis view.

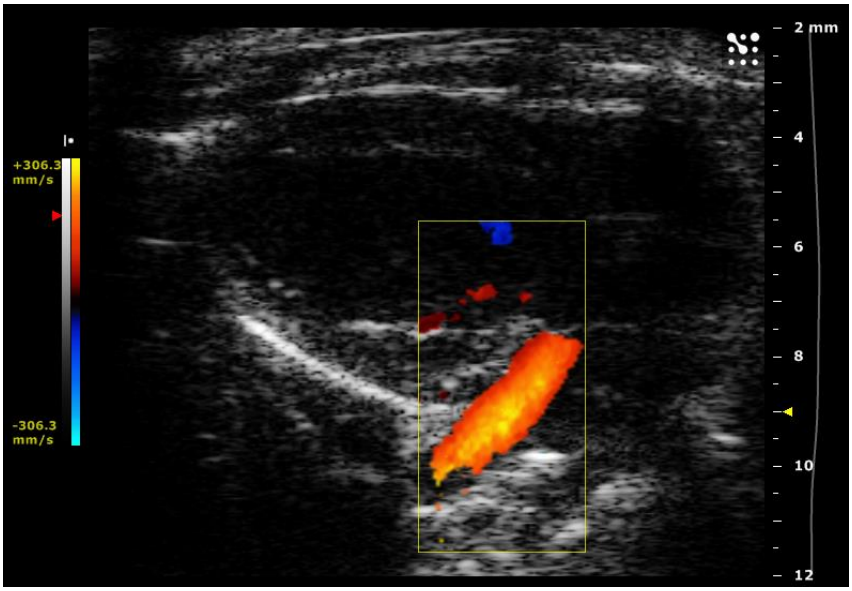

Figure 9 - Color Doppler Mode image of the Pulmonary Vein flow entering the Left Atrium.

Parasternal Short Axis View (PSAX)

Visible anatomy in PSAX view:

Left ventricle	Right ventricle
Anterior wall	Posterior wall
Intraventricular septum (IVS)	Papillary muscles

Transducer Positioning

Start with the transducer positioned for the PLAX view and rotate 90° clockwise. The heart will be displayed in the parasternal short axis view and the main landmarks are the papillary muscles. In order to have them in the view slight Y-axis translations by be necessary.

Figure 10 - Transducer positioning for the parasternal short axis view (PSAX)

Parasternal Short Axis View in B-Mode

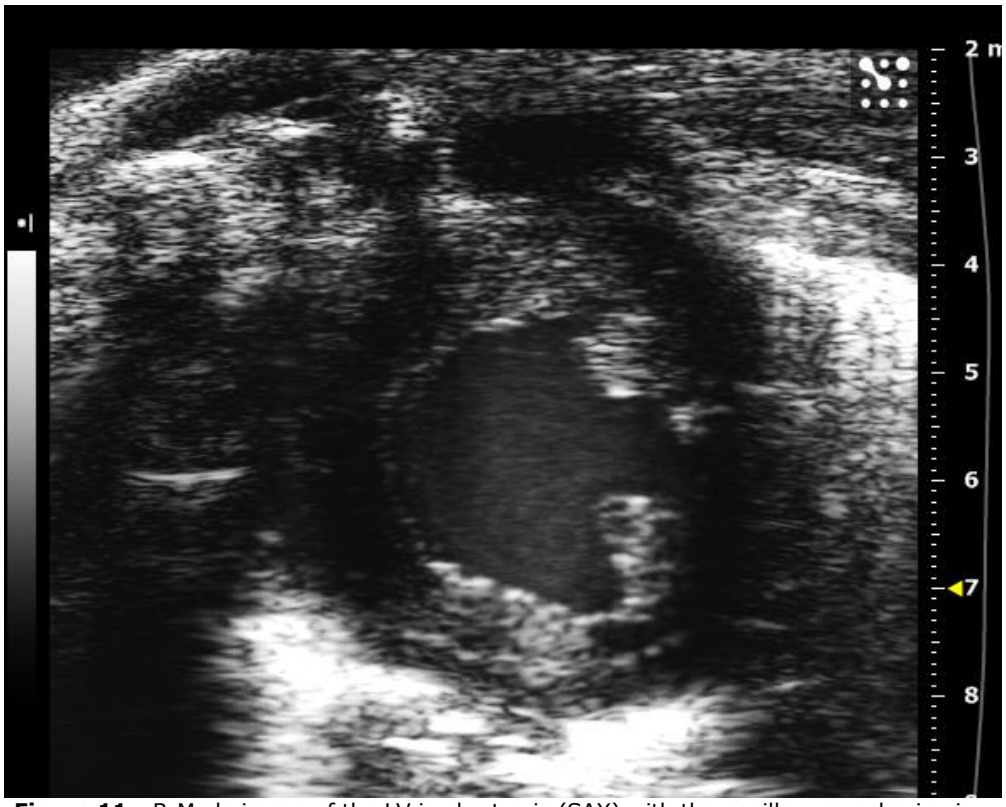

Figure 11 - B-Mode image of the LV in short axis (SAX) with the papillary muscles in view

Measurements and Calculations for SAX in B-Mode

Measurements	Calculations
IVS:d,s	LV V:d,s
LVID:d,s	LV SV
LVPW:d,s	LV FAC
LV Trace:d,s	LV CO
ENDO A:d,s	Endocardial SV
EPI A:d,s	Endocartial FAC
SimpArea Prox:d,s	Endocardial CO
SimpArea Mid:d,s	LV Mass
SimpArea Dist:d,s	LV Mass Corr

Parasternal Short Axis View in M-Mode

M-Mode axis should be placed at the mid-level of the LV, usually just medial of the papillary muscle. Measurements obtained here should correlate closely to measurements obtained in the long axis view.

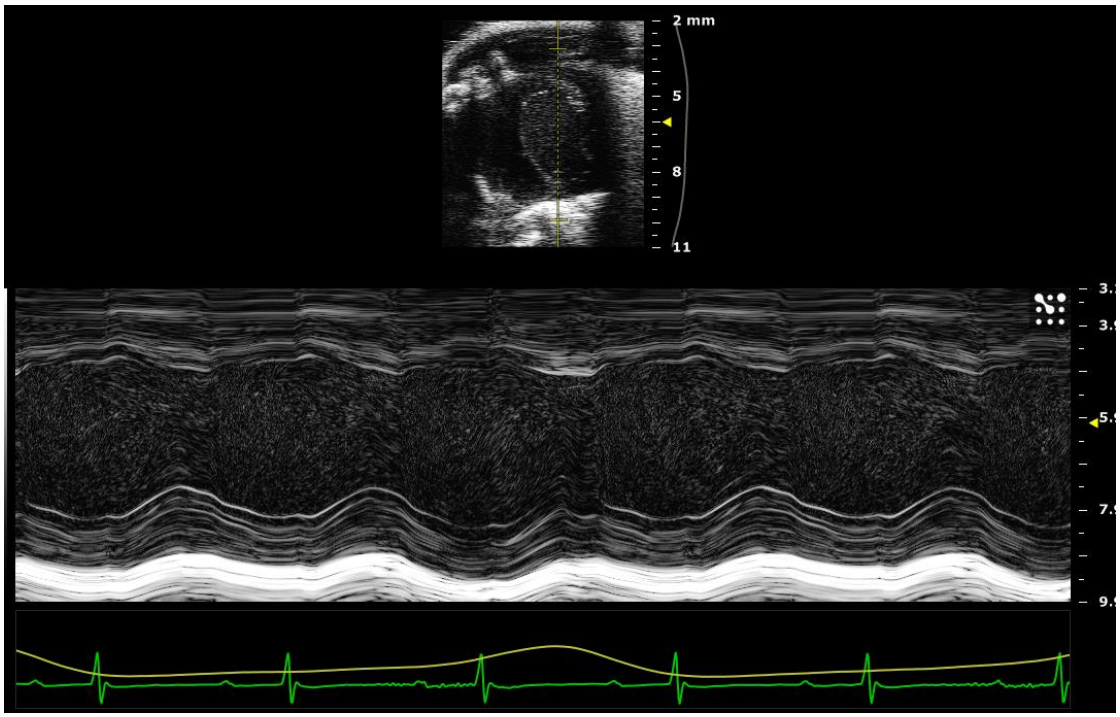

Figure 12 - M-Mode image of the LV in short axis (SAX) with the papillary muscles in view

Measurements and Calculations for SAX in M-Mode

Measurements	Calculations
IVS:d,s	LV V:d,s
LVID:d,s	LV SV
LVPW:d,s	LV FAC
LV Trace:d,s	LV CO
	LV Mass
	LV Mass Corr

Complete Cardiac Assessment

Apical Four Chamber View (Ap4)

Visible anatomy in the apical four chamber view:

- Left ventricle
- Right ventricle
- Mitral valve
- Left atria
- Right atria
- Tricuspid valve

Transducer positioning
Start with the transducer positioned for the short axis view. Lift the transducer arm using the height knob in order to allow space to tilt down the mouse imaging board with the upper left corner all the way down. Adjust the height of the transducer and angle it's base towards the user to achieve a coronal view of the heart looking from the apex.

Figure 13 - Transducer positioning for the Apical Four Chamber view.

Apical Four Chamber View in B-Mode

The apical four chamber view is obtained to visualize the right and left ventricles, with the atria at the bottom of the screen. This is the primary position for analysis of flow using PW Doppler Mode for the mitral and tricuspid valves.

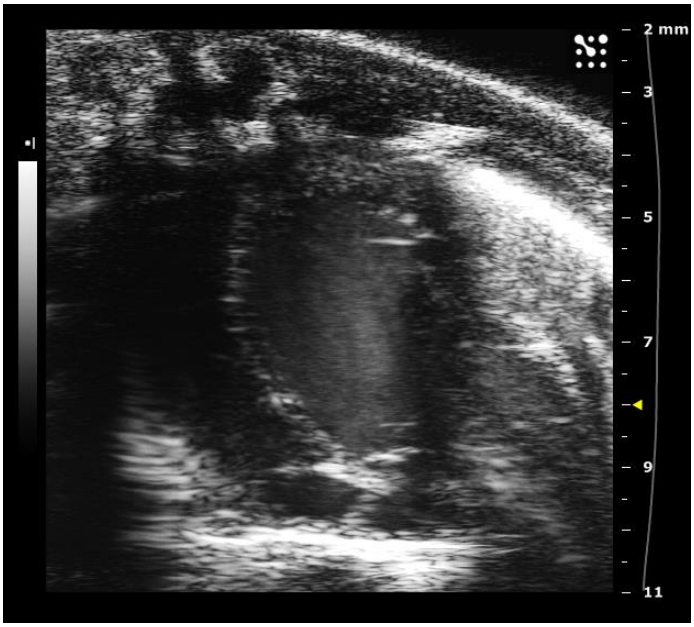

Figure 14 - B-Mode Apical 4 Chamber image displaying the apex at the top of the image, the left and partial right ventricles and mitral and tricuspid valves.

Apical Four Chamber View in Color Doppler Mode

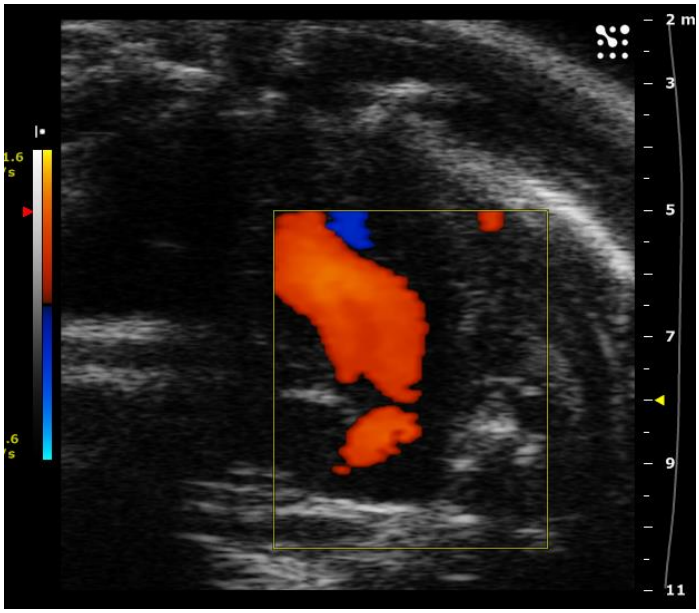

Figure 15 – Color Doppler Mode image displaying flow through the Mitral Valve.

Apical Four Chamber View – Mitral Flow display in PW Doppler Mode

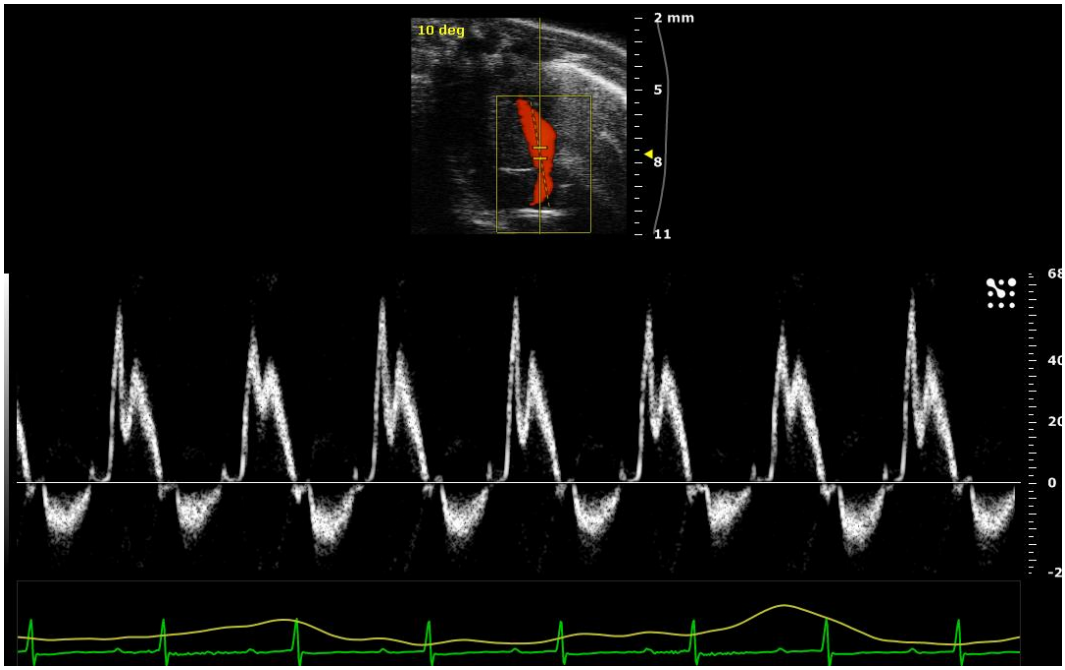

Figure 16 - PW Doppler Mode waveform of mitral valve flow in the apical four chamber view.

Apical Four Chamber - Tricuspid flow in PW Doppler Mode

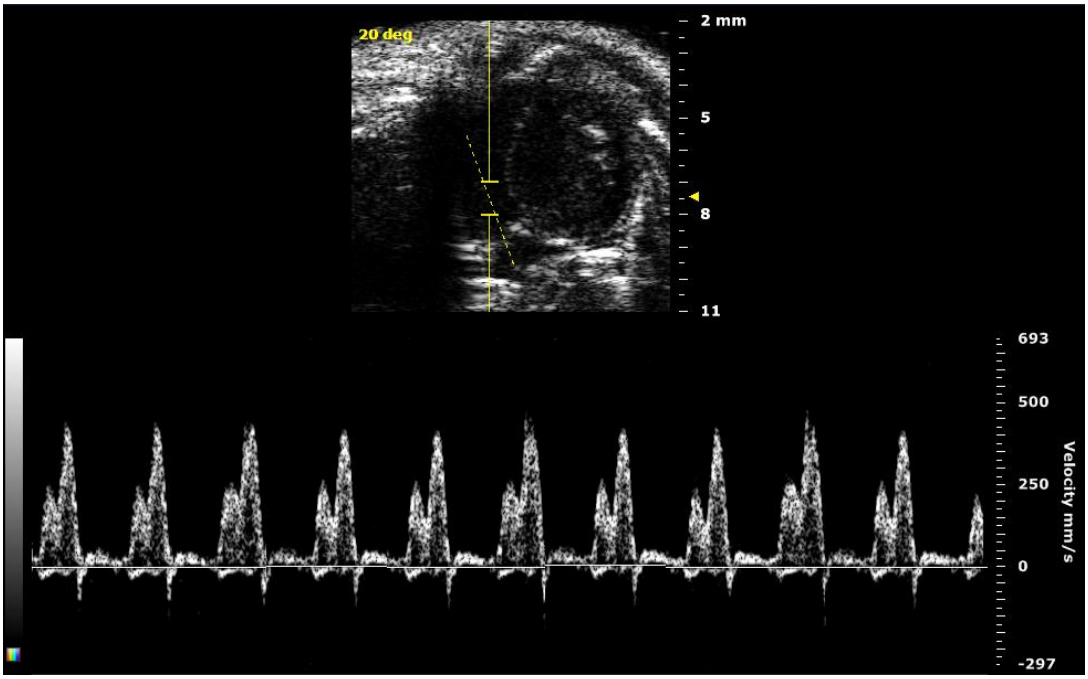

Figure 17 - PW Doppler Mode waveform of Tricuspid flow in the apical four chamber view.

Measurements and Calculations for MV and TV in PW Doppler

Measurements	Calculations
MV VTI	MV E/A
MV E	MV Area
MV A	LV MPI NFT
MV PHT	LV MPI IV
AET	MV PHT
MV Decel	MV Area
IVRT	TV Peak Pressure
IVCT	
MV ET	
TV VTI	
TV E	
TV A	
TR Peak Vel	

Apical Four Chamber – Mitral Valve Analysis in PW Tissue Doppler Mode

While the Apical 4 Chamber plane is optimal for MV flow assessment it also the best view for MV tissue analysis, at the annulus, using the Tissue Doppler Mode.

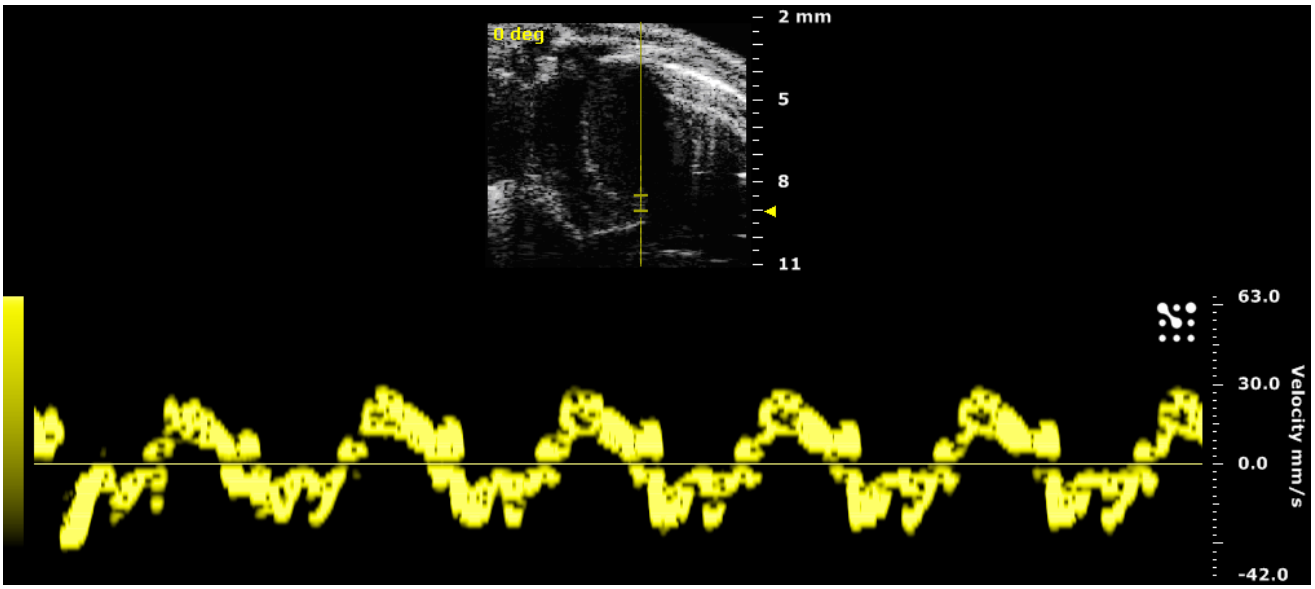

Figure 18 – PW Tissue Doppler display of tissue motion at the MV annulus.

Measurements and Calculations for MV and TV in PW Tissue Doppler

Measurements	Calculations
E′	E′/A′
A′	A′/E′
ET	MV E/E′
MV LW E′	MV LW E′/A′
MV LW A′	MV LW A′/E′
MV IVS E′	MV IVS E′/A′
MV IVS A′	MV IVS A′/E′
TV LW E′	TV E′/A′
TV LW A′	TV LW E′/A′
	TV LW A′/E′

Aortic Arch View

Visible anatomy in the aortic arch view:

- Ascending aorta
- Descending aorta
- Innominate artery
- Left common carotid
- Left subclavian artery

Transducer Positioning

The aortic arch view is obtained from a modified right parasternal view with the transducer positioned with the notch pointing towards the chin of the animal and slightly rotated clockwise.

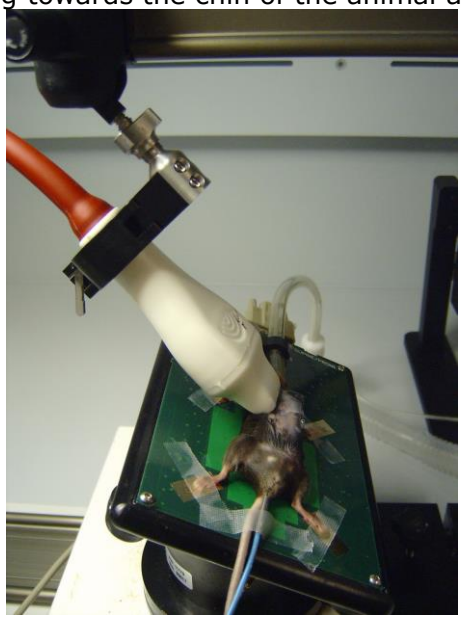

Figure 19 - Transducer positioning for the Aortic Arch view.

Aortic Arch View in B-Mode

Figure 20 – B-Mode image of the Aortic Arch with Ascending and Descending aorta, Innominate Artery, Subclavian Artery and Left Common Carotid artery in view

Measurements and Calculations for the Aortic Arch in B-Mode

Measurements	Calculations
Trans Arch	N/A
Asc Ao	N/A
Desc Ao	N/A

Aortic Arch View in Color Doppler Mode

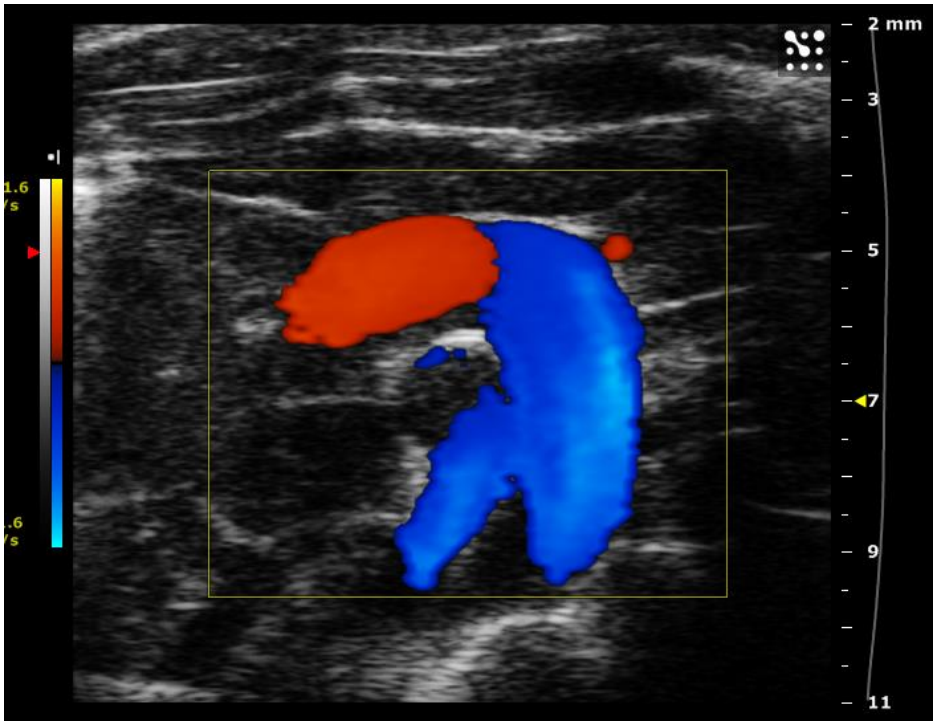

Figure 21 – Color Doppler image of the Aortic Arch with display of directional flow in the Ascending and Descending aorta

Aortic Arch in PW Doppler Mode

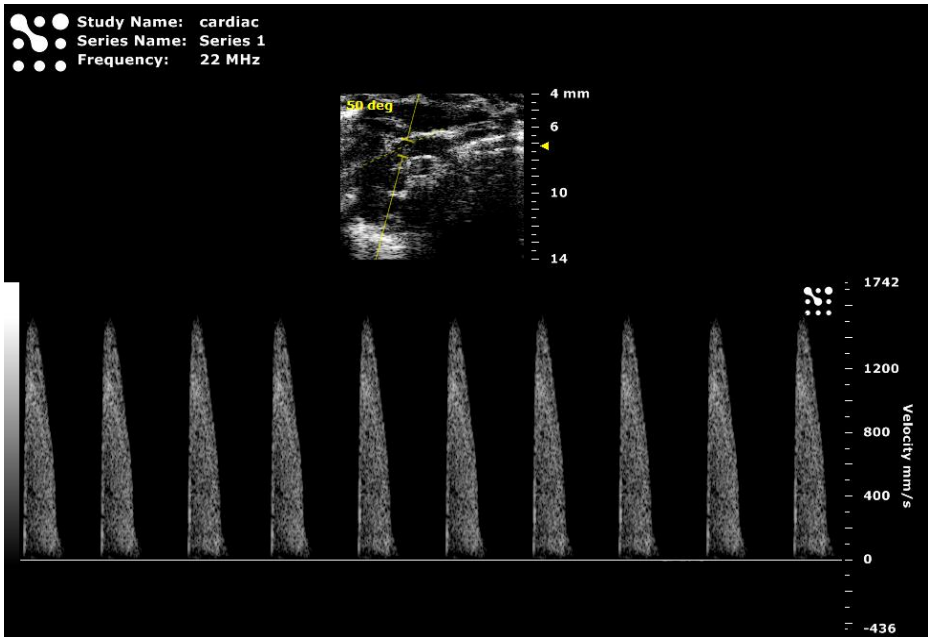

Figure 22 – Flow Distribution display in PW Doppler Mode on the Ascending Aorta

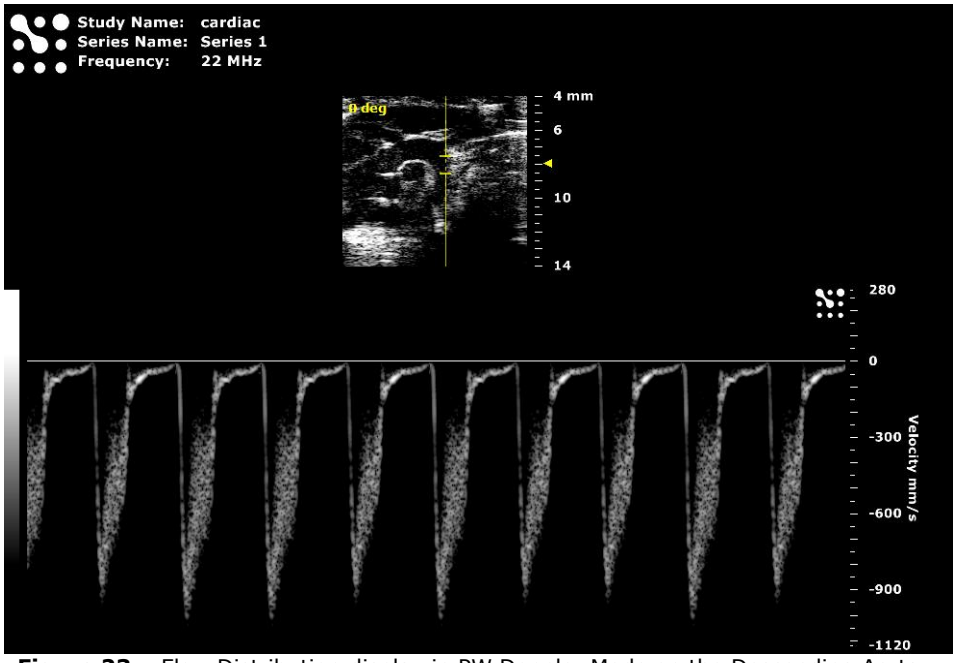

Figure 23 – Flow Distribution display in PW Doppler Mode on the Descending Aorta

Measurements and Calculations for the Aortic Arch in PW Doppler Mode

Measurements	Calculations
LVOT	AV Peak Pressure
AV Peak Vel	LVOT SV
LVOT VTI	LVOT CO
AoV VTI	AVA
Desc Ao Vel	AoV SV
AI PHT	AoV CO
AAT	AAT/AET
AET	
Desc Ao Vp	
Desc Ao Vd	

Right Parasternal Long Axis View

General anatomical aspects in murine anatomy limit visualization and analysis of the right side of the heart. That being said, imaging with the Visualsonics Integrated Imaging system presents the advantage of placing the animal platform such that users could benefit of analysis for the right ventricle.

Transducer Positioning
Start with the position for the parasternal long axis view, angle the base of the probe towards the animal's right by ~45°. Place het platform slightly to the animal's left to bring the right ventricle into the imaging plane.

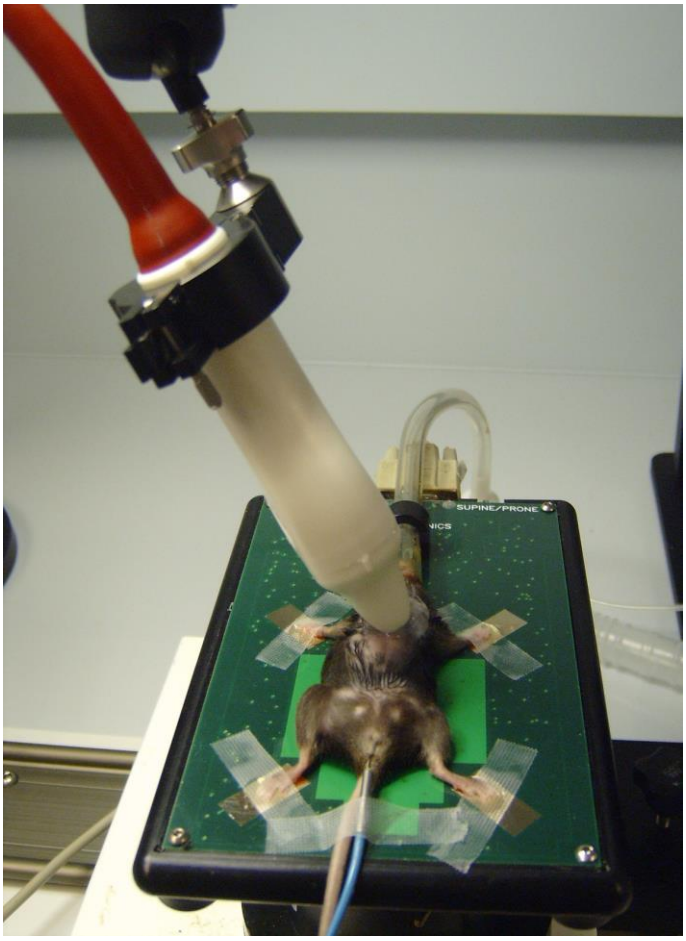

Figure 24 – Transducer positioning for the Right Parasternal Long axis view.

Right Parasternal Long Axis in B-Mode

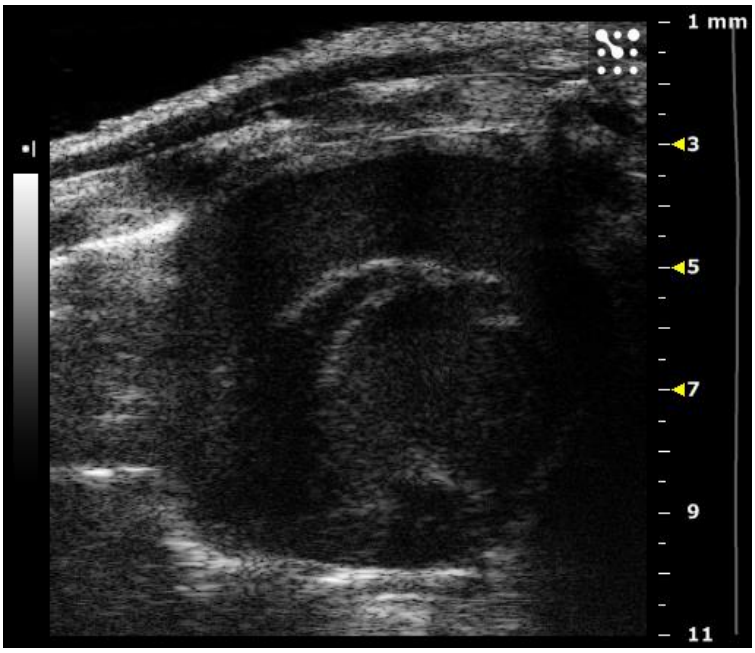

Figure 25 - B-Mode image of the right parasternal long axis view.

Measurements and Calculations for Right Ventricle

Measurements	Calculations
RVOT	
PV	

Right Parasternal Long Axis in Color and PW Doppler

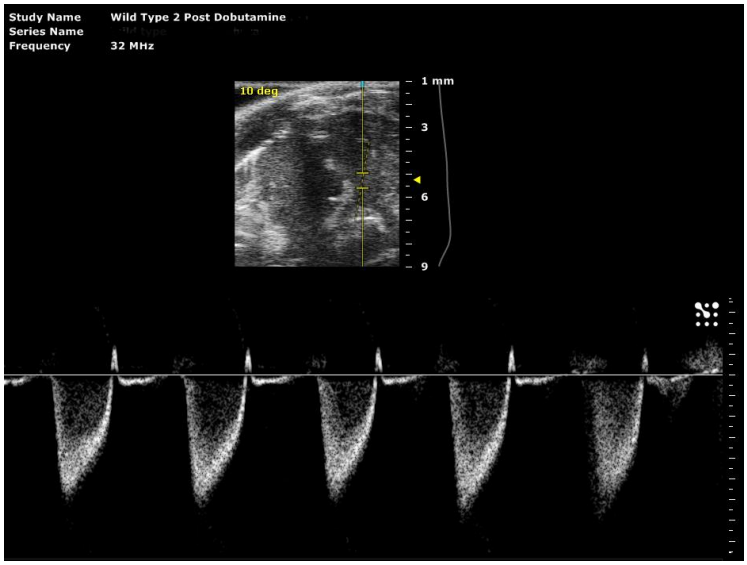

Figure 26 – PW Mode image of flow through the Pulmonary Valve

Measurements and Calculations for Right Ventricle

Measurements	Calculations
RV VTI	RV SV
IVRT;r	RV CO
IVCT;r	RV MPI NFT
NFT;r	RV MPI IV
PET	

Suprasternal View

Visible anatomy in suprasternal view:

- Aorta
- Left ventricle
- Aortic valves

The ascending aorta can be clearly seen in the parasternal long axis view. However, for accurate PW Doppler Mode flow distribution analysis the angle of interrogation in between the transducer beam and the direction of the vessel has to be less than 60°. Therefore, the suprasternal view is performed to generate this angle in order to obtain accurate measurements. With the advent of Doppler steering in the Vevo imaging systems, one can steer the vertical axis up to 25°, the original angle of the aorta can be as high as 85° (60° + 25°).

Transducer positioning
Place the transducer on the midline of the mouse at the level of the thoracic inlet. Then angle the base of the probe towards the animal's head by ~50°. This position is very similar to the like long axis view only the angle between the animal and transducer is very sharp. To obtain aortic PW Doppler Mode, place the Doppler sample volume just above the aortic leaflets.

Figure 27 – Transducer positioning for the Suprasternal View

Suprasternal View on B-Mode

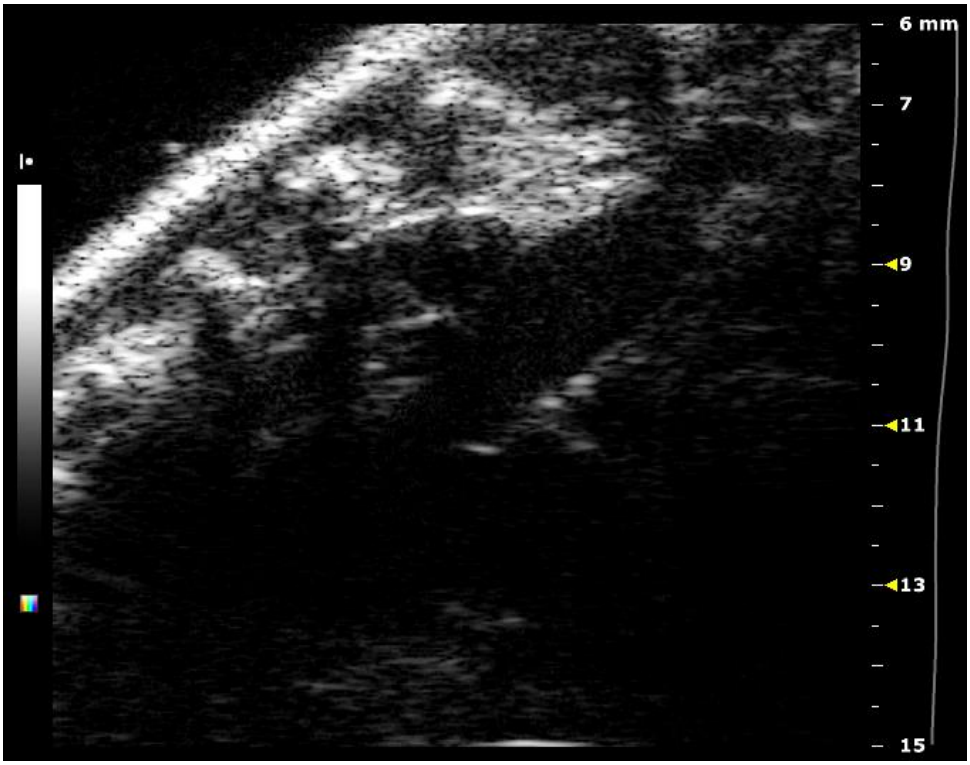

Figure 28 - B-Mode image of the Suprasternal view

Suprasternal View in PW Doppler Mode

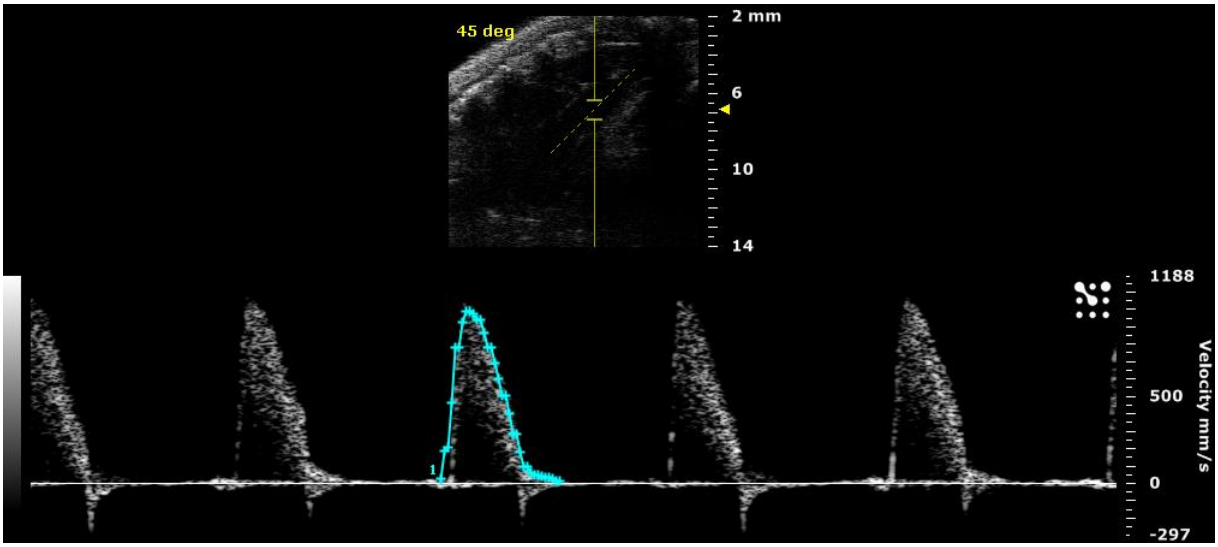

Figure 29 - PW Doppler Mode waveform of the aortic valve outflow from a suprasternal view, showing a VTI trace.

Measurements and Calculations for Aortic analysis

Measurements	Calculations
Ao VTI	AV Peak Press
AAT	AVA
Ao Peak Vel	AoV SV
AET	AoV CO
	AAT/AET

Note: The velocity range measured in PW Doppler Mode is 100-120 cm/sec, in a normal adult mouse model.

3D Cardiac Imaging

The Vevo imaging systems have capabilities that enable ECG-gated 3D cardiac imaging which could be particularly powerful with regards to precise volumetric measurements. Diastolic and systolic volumes can be measured in order to yield accurate stroke volume and cardiac output calculations.

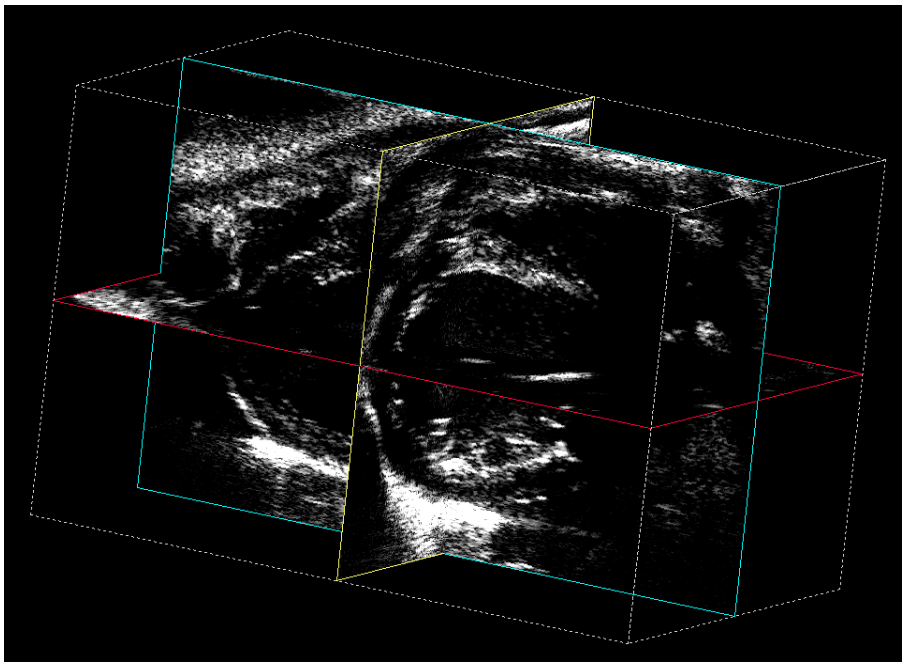

Figure 30 – 3D rendering of B-Mode image of the heart at Diastole.

Figure 31 - 3D volumetric mesh of Endo and Epicardial regions.

Transducer Positioning
3D imaging of the heart could be done starting from either the parasternal long axis position or the short axis position with the 3D motor travel set accordingly.

A. 3D Cardiac Imaging in Diastole
Enable respiration and T1 ECG gating. Adjust T1 time delay until the maximum diameter of the LV is observed and start imaging. Save and label the volume for further analysis.

B. 3D Cardiac Imaging in Systole
Enable respiration and T2 ECG gating. Adjust T2 time delay until the minimum diameter of the LV is observed and start imaging. Save and label the volume for further analysis.

C. 3D Measurements and Calculations
Use the 3D volume trace tool to measure endo- and epicardial LV volumes in diastole and systole.

- Stroke volume = diastolic – systolic endocardiac volume
- Cardiac output = (diastolic – systolic endocardiac volume) x Heart rate
- LV mass = (epicardial – endocardial volume) x 1.06 g/µL

Note: Specific gravity of muscle = 1.06 g/µL

Reference – Values measured in normal adult mice and rats

Note: These values are only guidelines as they greatly vary according to animal strain, size, sex, age and health condition.

Parameter	Mice	Rats
Ejection Fraction	55-85%	55-80%
Fractional Shortening	30-50%	30-50%
Left Ventricle Mass	65-90 mg	60-100 mg
Stroke Volume	40-70 µL	100-135 µL
Cardiac Output	20-35 mL/min	40-55 mL/min

FUJIFILM VisualSonics, Inc.
T.1.416.484.5000
Toll Free (North America) 1.866.416.4636
Toll Free (Europe) +800.0751.2020
E. info@visualsonics.com
www.visualsonics.com