

## **Ultrasound Imaging**

### **I. Objective**

Ultrasonic imaging in live adult rodents. This procedure is broadly applicable to various soft tissue ultrasound imaging procedures, including echocardiography.

### **II. Comments and Recommendations**

- This procedure has been written with specific reference to the Integrated Physiology Facility (IPF), the Vevo3100 ultrasound may only be used at this facility by IPF approved experienced operators
- Use of the Vevo3100 ultrasound and any variation to this SOP must be described in a relevant animal ethics application
- Users should read and understand the associated Risk Assessment prior to operation: 20230 Rodent Anaesthesia using Isoflurane, 7720 UQBR Use of Inhalation Anaesthetics (Isoflurane), 3657 UQBR Handling and restraint of laboratory animals; 3940 Handling rats and mice (available on the [UQSafe](#) website)
- Equipment/software failures and animal escapes need to be reported to the animal facility manager immediately
- All incidents/injuries should be reported via [UQSafe](#) online
- Spills must be cleaned up immediately
- This SOP should be read in conjunction with [LAB\\_060 Rodent anaesthesia – isoflurane, 2.70.16 Working safely with isoflurane](#)
- Use of the Vevo3100 ultrasound involves mouse handling and appropriate care should be taken, refer to [LAB\\_006 Handling and restraint in mice and neonates](#), and [LAB\\_039 Handling and Restraint in Rats and Neonates](#)
- Wild type and genetically modified animals must be transported to equipment as per OGTR guidelines and [LAB\\_003 Transportation of Laboratory Rodents](#)
- The IPF is a shared space with unknown commensal microbial status. Once transported to a shared space it is often not possible, for biosecurity reasons, to return rodents to their original animal facility. Arrangements for transportation and ongoing care of experimental animals must be made with relevant animal facility managers when planning projects that aim to use a shared facility

### **III. Equipment**

- Minimum Personal Protective Equipment (PPE) consists of gloves, gown, closed in shoes, eye protection and face mask. Additional PPE may be required based on added risk e.g., working with infectious animals (P2 fitted mask and viral gown)
- Anaesthetic machine and vaporiser calibrated for isoflurane use, induction box, rodent anaesthetic circuit and scavenger system
- Vevo 3100 ultrasound transducer/s
- Imaging stage containing non-invasive respiration/ECG monitor, heating pad and temperature probe

- Heat lamp
- Eye lubricant (e.g. Viscotears®, GenTeal®)
- Depilatory cream (e.g. Nair™ for sensitive skin)
- Gentle adhesive tape (e.g. Micropore™, silicone tape)
- Ultrasound gel, stored within a gel warmer (maintains gel at 37°C)
- Disposable gauze cotton swabs and/or cotton tips
- Disinfectants (e.g. 1-2% Virkon, 70% Ethanol)
- Clinical waste bin

#### **IV. Preparation**

- Removing hair is necessary for successful ultrasound imaging, therefore thermoregulation while under general anaesthesia must be considered
- Check booking date/s and time/s on PPMS
- All animal arrivals/departures and euthanasia's must be recorded on the Mosaic movement sheet available in the animal facility

#### **V. Procedure**

1. Turn on the Vevo 3100 ultrasound, imaging stage, heating pad and gel warmer approximately 15 minutes prior to performing procedure

- Select and attach the correct transducer for your application
- Login as Administrator, password: 'vevo'
- Select Study Browser to label your study or series: In Study Browser select New and then either New Study or New Series, label appropriately and add any other relevant information, select Done. Choose your application from the transducer panel. The B mode imaging window appears, and the equipment is ready to collect B mode data.

2. Ensure all equipment and worksurfaces are clean using 70% ethanol

3. Check animal identification and enter details into Vevo®3100 operating software

4. Place induction box on top of heating pad

5. Turn on oxygen flow (0.8L/min is suitable for the Vevo®3100 anaesthetic machine) and place mouse in the induction chamber

6. Introduce 4% isoflurane by turning on the vaporiser to induce anaesthesia and monitor breathing pattern

7. Once adequately anaesthetised, place animal onto the preheated imaging stage (typically in the supine position, dependant on organ/tissue of interest). Maintain anaesthesia (~1.5-2% isoflurane) via use of a nose cone and apply a small amount of corneal lubricant onto each eye

8. Apply a small amount of ultrasound gel to each metal ECG surface electrode, and carefully tape all four paws in position using the gentle adhesive tape

*Contact must be made with the electrode for effective ECG recording*

9. Lubricate the rectal temperature probe with ultrasound gel and carefully insert into the rectum. Tape probe and tail to stage to prevent movement. Monitor temperature throughout and use additional heat lamp if required (body temperature should be maintained between 35-37°C)

*The rectal temperature probe must be cleaned and disinfected between animals*

## 10. Remove hair as required

*Hair should be removed only from the required area. Removing excessive amounts of fur will impact the animal's ability to thermoregulate effectively*

- Depilatory cream should be applied using a cotton swab or cotton tip. Once the required duration has passed ensure all depilatory cream has been removed with a damp gauze (water)

11. Place warm gel on area of interest (try to minimize bubbles) and start imaging using the appropriate transducer

12. Monitor the animal's temperature and respiration rate throughout the procedure. The animal should not be under anaesthesia for longer than 3 hours (ultrasound typically takes no longer than 15 minutes)

13. Once images are acquired remove all traces of ultrasound gel from the animal's skin and wipe with a damp gauze (water). Carefully remove the temperature probe and all tape securing the paws.

14. Turn off the vaporiser and place the animal in a heated cage. Monitor until conscious and freely moving, then return to home cage

*In the event your mouse does not recover or seems unwell refer to LAB\_022 UQBR Veterinary Care Program. Report any Unexpected Adverse Events to the AEC*

15. Ensure vaporiser and oxygen supply have been turned off. Turn off all additional equipment and ultrasound machine (once fan has stopped)

16. Clean image station, transducer, induction box and work surfaces with 70% ethanol (or another relevant hard surface disinfectant)

## VI. Parameters

### B Mode Images

Select Mode Pre-sets and select application

Adjust the image area, depth, and gain (contrast) using the controls on the touch screen

- To freeze image, select freeze (these frames can be viewed and saved)
- To resume, slide to scan
- To save a clip select save clip

### M Mode images

Locate the area of interest using B Mode.

Select M mode (on the touch screen menu)

Place the vertical yellow bar over the exact location of interest. Adjust the horizontal bars to bracket your area of interest

Select start

### Colour Doppler Images

Locate the area of interest using B Mode

Select Color (on the touch screen menu)

Adjust the yellow rectangle to outline the area of interest

### Pulse Wave Doppler Images

Locate the area of interest using B Mode

Select PW Doppler (on the touch screen menu)

Adjust the location, size and angle of the yellow bars to area of interest.

### **Tissue Doppler Images**

Locate the area of interest using B Mode

Select Tissue Doppler (on the touch screen menu)

Adjust the location, size and angle of the yellow bars to area of interest.

### **Viewing Images**

To view most recent clip/s for a current series, select current series (top of screen) and the thumbnail of the clip you want to view

OR

Select study browser (top left), select the study and series

Images will appear as thumb nails, select a thumbnail to view image.

Use play speed button to control the speed an image is replayed at.

### **Measurements**

Find image/s as above

Select measurements (right side of screen)

Select appropriate measurement and move cursor/callipers

Measurements will be stored with the image

Return to either Current Series or Study Browser and select More and Report to view the report

### **Exporting**

Insert storage device into USB port on the right front corner of the control panel

Find series/images in Study Browser and select Export

Either export to VEVO LAB for analysis or to other file types (e.g., tiff, MP4) to use outside the Vevo imaging system