

## SOP FOR ULTRASOUND IMAGING

### Overview/Purpose

The objective of this SOP is to describe the procedure for conducting ultrasound imaging using Vevo2100 high frequency ultrasound imaging system (VisualSonic, Boston, MA) located in room 0020A BRT.

**Owner of SOP and Responsibilities:** **Kimerly Powell** is responsible for ensuring that the SOP is followed, updated as needed (including personnel changes) and that personnel conducting the procedures are appropriately trained.

**Species** – Mice, rats and rabbits

### Agent Administration

Agent Name	Vehicle **	Dose	Route	Volume	Frequency
Isoflurane	O <sub>2</sub> /air/ (95% O <sub>2</sub> + 5% CO <sub>2</sub> ) or medical air	0.5-5%	Inhalation	As needed	1x/imaging
Ketamine & Xylazine	Sterile saline or water	50-70mg/kg & 5-10mg/kg (rabbit)	Subcutaneous	<2mL/kg	1x/imaging
Ketamine	Sterile saline or water	25-35 mg/kg (rabbit)	Subcutaneous	< 2 ml/kg	1-2x /imaging for redosing if needed
Acepromazine	Sterile Saline or water	0.5-2 mg/kg (rabbit)	Subcutaneous	<2ml/kg	1x/imaging
Heparin	Sterile Saline or water	10-100 U/ml	Intravenous	As needed	1x/imaging
Depilatory Cream	None	As needed	Topical	As needed	1x/imaging
Microbubbles (**)	Sterile saline or water	50 microL- 6 ml	Intravenous	as needed, appropriate to animal size	1-2x\imaging
ECG electrode gel	None	As needed	Topical	As needed	1x/imaging
Ultrasound gel	None	As needed	Topical	As needed	As needed
Developmental contrast agent *	As needed	As needed	As needed	as needed, appropriate to animal size	1-2x\ imaging

(\*) Details will be provided in PI protocol

(\*\*) example of the microbubble are: untargeted or targeted MicroMarker™ Contrast (Bracco Research S.P.A)

### Procedure

- Animals will be anesthetized prior to imaging and then positioned as needed. Depth of anesthesia will be monitored by lack of response to noxious stimuli, such as toe pinch, cornea reflex (rabbit) and loss of spontaneous movement, and adjusted accordingly.

Additional information:

- Rabbits may be sedated prior to anesthesia if not contraindicated by the study.
  - Rodents will be anesthetized using isoflurane and precise vaporizer.
2. Fur may be removed, prior to imaging, over appropriate part of animal body  
Details of hair removal:
    - Animals will be kept under isoflurane anesthesia for this procedure. Hair will be removed using an electrical animal shaver and/or depilatory cream (eg. Nair®). After 30 to 90 seconds, the depilatory cream will be wiped off and any remaining agent will be thoroughly removed using wet tissue to prevent burns.
  3. The animal will be placed on an animal bed and secured with tape. The ECG electrode gel will be used for contact of the probes with animal body. Pre-warmed ultrasound gel will be applied on an animal body to ensure contact of the ultrasound transducer and animal body. The animal will remain under anesthesia for 5-60 minutes (depends of the study requirements).
  4. Physiological parameters will be monitored during ultrasound imaging as follows:
    - *Respiration* will be monitored during all imaging sessions (contact with bed)
    - *Temperature*: A rectal thermometer or temperature probe with body-contact will be used (depends on study requirements) for imaging session longer than 15 min. Body temperature will be maintained at species appropriate ranges once placed on ultrasound heated platform.
    - *Heart rate* and ECG signal of the animals will be monitored if required using the electrodes build into the ultrasound platform. ECG electrode gel is placed on the stripes, then animal paws are placed on the top to ensure that there is a contact between paws and ECG leads. For very small animals, the cupper tape can be used as an extension.
  5. If necessary, a microbubble will be used as a contrast agent and injected during imaging session. Agents which are not unavailable in a pharmaceutical grade will be used in accordance with the IACUC policy "USE OF PHARMACEUTICAL AND NON-PHARMACEUTICAL GRADE COMPOUNDS IN ANIMALS"
  6. Placement of I.V. catheter  
An I.V. catheter will be placed if needed for agent administration during the ultrasound procedure. All animals will be under anesthesia (isoflurane when possible) during catheter placement procedure.
    - Tail will be cleaned with 70% alcohol prep pad and slightly warmed up using the warm pad prior to catheter placement.
    - Sterile tubing and needle of required size (e.g. PE10 for mice) will be used for tail vein catheterization and secured in place. Tubing will be sterilized prior to use according to ULAR veterinary recommendation ( i.e. autoclave or gas sterilization)
    - Heparinized saline (0.1-1 ml heparin [1,000 USP units/ml] in 10 ml of sterile saline) will be used as an anticoagulant agent.
  7. Following imaging, animals will be monitored to confirm recovery from anesthesia (walking, active, etc.) prior to return to routine husbandry/housing.

**Personnel conducting procedure must be up to date on all IACUC study team requirements.** Personnel not associated with an active protocol must create a profile that is up to date on IACUC study team requirements including a training narrative providing detail on experience for the techniques described in the SOP.

1. Kimerly Powell
2. Anna Bratasz
3. Michelle Williams
4. Katie Gallagher
5. Personnel listed on protocols referencing this SOP may perform this activity

#### **Potential adverse effects**

None anticipated related to activities covered in this SOP.

Novel contrast agents or alternative dosing schedules must be covered by a research protocol including potential adverse effects.

#### **Early Removal Criteria**

In case of unexpected issue with animals at any time the PI/study team will be notified by the core staff and will be responsible for further animal monitoring, treatment or euthanasia as per the IACUC protocol.

Any unusual/unexpected symptoms or anesthetic deaths will be reported to the ULAR veterinary staff for consultation.

#### **History of Revisions**

**404-00** - new SOP approved by IACUC on 10/19/18