DAUGHERTY LAB
Saha Cardiovascular Research Center
University of Kentucky

Mouse Procedures\
Ultrasound for AAA Quantification

Ultrasonographic Measurements of Mouse Abdominal Aortic Lumen Using the Vevo 2100 System

PREPARATION

General Rules

1. If you have never used the Vevo 2100 machine, you will need to be trained by Jessica Moorleghen (jjmoorl@uky.edu)

2. Make sure you are on the SharePoints schedule for your particular date and time.

3. Bring your own memory stick to export data through USB port.

Starting the Machine and Software

1. Choose the proper transducer - MS550D (Blue) for the mouse abdominal imaging, MS400 (Red) for mouse cardiovascular, and MS250 (Yellow) for rat tumor and cardiovascular imaging. Secure the transducer in the platform holder perpendicular to the vessel to be imaged, with the notch facing left for proper orientation.

2. Turn on the heated physiological table via the touch pad (on/off). It is pre-set for 38 °C.

3. Push up the **Main Power** switch on the rear panel of the machine, then toggle the **Computer Standby** switch, located on the left side of the keyboard. Allow several minutes for the software to boot up.

4. Choose your application package and operator (NOTE: if you have been trained, your name should appear in the drop down menu)

5. Go to “new study” and type in your study name. In the “series” box, type your animal identification.

Preparing the Animal for Imaging

1. Rodents are anaesthetized with isoflurane at a concentration of 2-3%. Use the proper size induction chamber, depending on the number and species of animals (small = 1 mouse; large = 2-3 mice; large marked “RAT ONLY” = 1 rat).

2. Once the animal is anesthetized, lay in a supine position on the platform and insert the nose flush within the nose cone for constant isoflurane delivery and minimal isoflurane gas exposure to the user.

3. Apply a depilatory cream to remove the hair from most of the abdomen. Wait 1 minute then gently wipe with baby wipes or wet gauze to remove ALL traces of depilatory cream. Apply pre-warmed ultrasonic gel in this area.

   Optional - A small amount of gel can be placed on each of the 4 copper leads on the platforms and the paws taped to them (palms down). This will provide the ECG and respiratory physiology of the animal while anesthetized.

ULTRASONOGRAPHY

Imaging

1. Starting just below the sternum and moving the probe caudally, locate the right renal branch and use it as landmark. An acceptable suprarenal image can be taken ~ 0.5 to 2mm above the right renal branch, trying not to involve the superior mesenteric or celiac artery branch points.

2. Press the “frame store” button to collect one frame or “cine loop” for 100 frames (note* the 100 frames saved are those PRIOR to pushing the button). Each study animal should have an image of each renal branch point (used as internal control) and at least one suprarenal image.

3. When each animal’s scan is complete, remove the gel with warm sterile water, and place the animal back in its cage, on heating pad for recovery.

4. Press “study management” to return to the main screen and select “close series.” Then, select “new” and when prompted, select “new series” for each of the next study animal.

5. Once all animals are completed, shut off the isoflurane then the oxygen tank.

Data Export and Cleaning

1. To export data, insert your memory stick in one of the USB slots in the back of the machine. From the Study Management screen, → select your study → then “copy to”. This will give you a directory; choose the destination → click “OK”. These files will be in a “proprietary” .vsi format and can only be opened up within the Vevo 2100 software to be analyzed.

2. Once you have copied your study, turn the components off in the reverse order : Computer Standby switch (wait for screen to go blank) → Main Power switch in the back

3. Wipe any excess gel from probe and all other surfaces. Use antiseptic spray to wipe down the entire probe, physiological table, nose cone and any other

surfaces you or your animals came in contact with. Wash the induction chamber with soap and water and let air dry.

MEASUREMENT

Data Import

1. Using a computer that has the Vevo 2100 1.6v software, insert a 2100 dongle into the USB port. Open Vevo 2100 software.

2. To import your data for analysis, click “copy from” button on the upper left panel → select a user, choose the files, and click “OK” → once the data transfer is complete, click “OK”

3. Open an individual series to start the measurement by using the image with the largest luminal diameter.

Measurements of Maximal Luminal Diameters

1. Click the measurement icon ( ) on the lower left panel → Select the line icon ( ).

2. Draw lines vertically and horizontally and use the larger distance as your diameter (Figure 1).

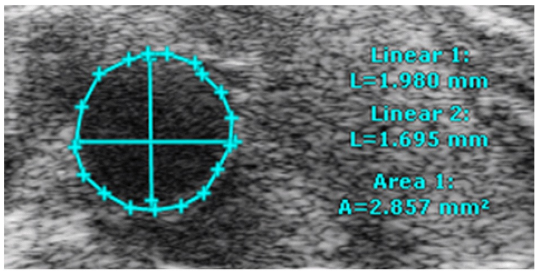

Figure 1. Ultrasound measurement of abdominal aortic lumen diameter and area.

3. Choose area icon ( ) and trace the lumen area. (Figure.1)

4. The image can be exported as a TIFF file at this time.

Exporting Data into Spreadsheet

1. Save the file by selecting your study → then click “copy to”. Save the file with measurement in a separate folder for a second observer to verify. Final quantification should be verified by a second observer who is blind to study groups.

Protocol Developed : Jing Liu on 2/24/14

Verified : Jessica Moorleghen on 2/27/14