Ascending Aortic Doppler Blood Velocity

Equipments**:** **20 MHz transcutaneous pulsed Doppler mainframe** (Indus Instruments, Houston, Texas) http://www.indusinstruments.com/

20 MHz hand-held probe (Matec Instrument Companies Incorporated, Northborough, M.A.) http://www.matec.com/

THM 100 Temperature and Heart Rate Monitoring System for Mice (Indus Instruments, Houston, Texas) which includes a heated mouse pad with ECG electrodes, a body temperature and heart rate control module and a RET-3 mouse rectal temperature probe (manufactured by Physitemp Instruments, Inc., Clifton, N.J.) http://www.indusinstruments.com/

Supplies: ultrasound gel
conducting gel
isoflurane inhalation anesthetic
heat lamp
masking tape
gauze or Kleenex

General Information:

We use a 20 MHz hand-held ultrasound probe to obtain *in vivo*, non-invasive, transcutaneous ascending aortic blood velocity waveforms from anesthetized mice. The probe is connected to a Doppler module, which generates ultrasound transmitter signals and processes the received signal. The received signal is then used to generate the in-phase and the quadrature (time domain) signals, which contain the information about the ascending aortic blood velocity. The Indus Instruments software is used to analyze the waveforms to obtain several indices of cardiovascular function (see below). We can also accurately measure *in vivo* aortic diameters using our ultrasound biomicroscope (VisualSonics VS40, Toronto, Canada) which, when combined with the stroke distance or the mean blood velocity, can be used to calculate cardiac output. Aortic diameter measurements are not routinely performed as a primary screen.

Procedure:

The mouse is anesthetized with 1-2% isoflurane in 700 mL O_2/minute (induced in a chamber at 5% isoflurane and maintained by face mask). To help maintain a consistent body temperature of 37-38 °C, the work area where the Doppler measurements are carried out is heated with a heat lamp and the anesthetized mouse is laid on a heated mouse pad. The mouse's body temperature is measured with a mouse rectal temperature probe and is monitored throughout the entire procedure. After applying conducting gel to the ECG electrode pads, the paws are taped to the pads and the animal's heart rate and ECG is continuously displayed.

Ultrasound gel is placed on the mouse's chest. The Doppler probe is placed under the suprasternal notch (the segment of bone that covers the thoracic inlet) and is directed towards the mouse's heart. The probe is lifted slightly (0-10° above the horizontal plane) and the angle of the probe is adjusted until a good ascending aortic waveform is observed. For adult mice, the Doppler range is set at 4 mm for the measurement of ascending aortic blood velocity. The ascending aortic waveform is recognized by the high peak velocity (80-130 cm/second), steep ascending slope, convex descending slope, and end-systolic flow reversal associated with valve closure. Approximately 15-20 ascending aortic waveforms (total duration of 2 seconds) are acquired for each mouse. The mouse's heart rate and body temperature at the time of collection of the waveforms are recorded. When the desired number of waveforms have been collected, the anesthetic is turned off, the rectal temperature probe is removed and the ultrasound and conducting gels are cleaned off of the mouse. The mouse is allowed time to wake up while breathing 100% oxygen and is then returned to its cage. Each test takes ~15 min including anesthetic induction and recovery time.

Using the Doppler Signal Processing Workstation, the 15-20 acquired waveforms are viewed offline on a computer screen. The waveform with the greatest amplitude is selected for a detailed analysis. The waveform is marked

using 4 analysis markers. These include FVS (flow velocity start), LAE (linear acceleration end), PV (peak velocity), and FVE (flow velocity end). The following parameters are then automatically calculated;

- Pre-ejection time (ms)
- Ejection time (ms)
- Peak velocity (cm/s)
- Mean velocity (cm/s)
- Peak acceleration (cm/s^2)
- Stroke distance (cm)

In addition, heart rate (beats/min) is determined from the accompanying ECG signal.

Reported Results :

For each mouse analyzed, a summary report is generated. The report includes the mouse's heart rate, and the pre-ejection time, ejection time, peak velocity, mean velocity, peak acceleration, and stroke distance for the mouse's ascending aortic blood velocity waveform.

Acknowledgements:

The CMHD requests that the users of our screening service acknowledge the technical assistance of our facility in any presentations or publications that report results generated by our services. A suitable acknowledgement for publications is as follows: "The authors would like to acknowledge the Samuel Lunenfeld Research Institute's CMHD Mouse Physiology Facility for their technical screening services (www.cmhd.ca)."

Additionally, please send reprints or information on such publications or presentations when they are submitted or available. Such acknowledgements will help promote the use of our service and assist us in obtaining continued financial support to help defray service fees.