UC Davis MMPC-Live Protocol

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Summary:

Body composition NMR exploits differences in the time domain (T1 and T2) relaxation time (rather that the specrum itself) to quantify the amount of protein (lean mass), fat and water in the entire mouse. This methodology has the critical adavantages of being very fast (typical scans are under 3 minutes) and not requiring anesthesia, allowing for repeated, longitudinal assessment of body composition changes.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Body Composition Analyzer	EchoMRI	100V
NMR mouse tube	EchoMRI	
Plastic mouse restrainer plunger	EchoMRI	
Plastic wrap		
70% EtOH		
Extra paper towels		
USB flash drive		
Household (10%) ammonia		

Protocol:

Setup
1. Cover keyboard with plastic wrap and wipe down all metal surfaces with 70% EtOH (sprayed on a paper towel, NOT directly on the MRI machine).

Computer
1. Select Folder →Browse/New → Select C:\EchoScans\[PI] → OK.
   Note: If you need a new folder select C:\EchoScans → type a backslash (\) followed by a new folder name into the upper window of the dialog -> OK
   Note: Software will refuse to scan in a folder which is a sub-folder of any folder in which scans have been previously collected.
2. Perform System Test (Alt +Y):
   a. When not in use, fat standard should remain in NMR machine to maintain constant temp and accurate reading (19.23g canola oil).

      b.   Select **Yes** for system test (~1.5 min).
3. If NMR was moved or System Test fails, calibrate the signal amplitude: Review → Calibration (Cont B) → Calibrate Canola. Calibration includes a system test and calibrates as needed.
4. After system test/calibration, gently return fat standard back into drawer and lock.

Assessing Body Composition
1. Place mouse into tube and secure with plunger. Place tube with mouse and plunger into NMR machine. Adjust the height of the plunger so that the mouse in hunched over, but can still breath easily.
   Note: It may be necessary to lean plastic card box on plunger to prevent mouse from jumping/moving. ***DO NOT USE ANY METAL TO WEIGHT PLUNGER DOWN***
2. Start Scan (F5) → Label: enter the animal's unique identifier. → Select **OK**.
   Note: Do not touch machine while scanning. *Your electromagnetic field will alter results.*
   Note: If forgot to label or mislabeled before scan, can edit label post-scan: Review -> Post-Scan Comments
3. When finished with all mice, return mice to their home room (covered with filter-top lids) and return to original cages if necessary.

Extracting Data
4. Select all scans that you wish to save/export. Only the scans that are selected will be exported. Shift + mouse click OR control + mouse click allows you to select scans to save → Review (F8) → Extract Table → Choose default directory for your file(s): C:\EchoScans\[PI]\ExtractedTables and name the file(s)
5. Go to the export directory and open the file in Excel/Open Office Calc to verify that the desired scans were exported.
6. Drag the exported file onto the USB flash drive.   The EcoMRI computer is not connected to the network, so this is the method for getting your data off of that Eco.
7. Exit EchoMRI program (click red X in top right corner). Ready to quit = yes

   Note: Do NOT shut down or power off unless moving NMR. Powering down NMR the system requires significant time (> 24 hours) for the magnet to return to operating temperature before the system can be used again.

MRI Equipment Clean-Up
8. Discard plastic wrap from computer keyboard and computer mouse.
9. Spray paper towel with 70% EtOH and wipe down all metal surfaces.
10. Wash out NMR tubes and plunger in warm soapy water to remove visible contamination.
11. Soak NMR tubes and plunger in 10% ammonia overnight in the chemical hood. After soaking, dispose of ammonia (pour down the drain) and leave NMR tubes in chemical hood until traces of ammonia have evaporated.

Reagent Preparation:

- None