



## **Metabolism and Translational Science Core (RC2)**

### **Leader:**

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### **Faculty:**

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### **Staff:**

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The University of Florida Institute on Aging's Claude D. Pepper Older Americans Independence Center Metabolism and Translational Science Core is capable of performing all of the analyses listed below as well as many others that fall within each category (e.g., most ELISAs and multiplex immunoassay panels, additional physical/cognitive performance measures).

In addition, the Core

- 1) provides guidance on the selection of analyses best suited for an investigator's project
- 2) provides consultation on the development of standard operating procedures for the collection, processing and storage of biological samples as well as on IACUC protocols.
- 3) And, the Core can provide budget estimates for both currently funded projects as well as grant proposals.

<b><u>Analysis technique</u></b>	<b><u>Assay</u></b>	<b><u>Specialized Equipment</u></b>
<b>ELISA's</b>	<b>Sample type: plasma, serum, tissue</b>	<ul style="list-style-type: none"> <li>• Bio-Tek Synergy HT multi-detection microplate reader (fluorescence, absorbance and luminescence measurements) with Gen5 Data Analysis Software</li> <li>• BioTek Instruments ELx405™ HT Microplate Washer - for 96-well plates</li> </ul>
Oxidation	MPO, oxLDL, Protein carbonyls, ferritin	
Inflammation	TNFα, IL-6, IL-8, V-CAM, Selectins, CRP	
Apoptosis	Cell death ELISA	
<b>Multiplex Immunoassays</b>	<b>Sample type: plasma, serum, tissue</b>	<ul style="list-style-type: none"> <li>• Millipore MILLIPLEX® Analyzer 3.1 xPONENT System with MILLIPLEX® Analyst 5.1 software (Luminex 200)</li> <li>• BioTek Instruments ELx405™ HT Microplate Washer - for 96-well plates</li> </ul>
Cardiovascular Disease, Metabolic Disorders, Inflammation	Adiponectin, MPO, Soluble E-Selectin, Soluble ICAM-1, Soluble VCAM-1, Total PAI-1, IFN-gamma, IL-10, IL-12, IL-12, IL-13, IL-1beta, IL-5, IL-6, IL-8, TNF-alpha	
<b>Q-PCR</b>	<b>Sample type: tissue, cells, whole worms</b>	<ul style="list-style-type: none"> <li>• Bio-Rad Laboratories CFX96 Touch™ Real-Time PCR Detection System</li> </ul>
Inflammation	TNFα, IL-6, NF-κB	
Autophagy/Proteolysis	Lamp 2, Atg 5, 7, 12, Beclin, MurF 1, Atrogin 1, Foxo-3	
Aging-related transcription factors	FOXO/DAF-16, FOXA/PHA-4, NRF2/SKN-1, nuclear hormone receptors, etc	
<b>Western blot</b>	<b>Sample type: tissue, cells</b>	<ul style="list-style-type: none"> <li>• Protein Simple Jess System for capillary Western immunoassays (chemi, IR, NIR detection for multiplexing)</li> <li>• Bio-Rad Criterion™ gel electrophoresis</li> </ul>
Oxidative stress	Nitrotyrosine, 4-HNE	
Mitochondrial	Fusions & Fission (Opa1, Fis1, Mfn1, Drp1), PGC1α, Tfam,	
Inflammation	TNFα Receptor I, NF-κB	
Apoptosis	Caspases 3 & 8, PARP, Endo G, & AIF	

Autophagy/Proteolysis	LC3B, MurF 1, Atrogin 1, Foxo-3	<ul style="list-style-type: none"> <li>• Bio-Rad Trans-Blot® SD semi-dry transfer cell</li> <li>• Bio-Rad Gel Doc XR high-resolution gel/blot imaging system</li> </ul>
<b>Activity assays</b>	<b>Sample type: cells, tissue</b>	<ul style="list-style-type: none"> <li>• Bio-Tek Synergy HT multidetection microplate reader (fluorescence, absorbance and luminescence measurements)</li> </ul>
Oxidative	ROS production (H <sub>2</sub> O <sub>2</sub> ), Aconitase,	
Mitochondrial	Cox IV, citrate synthase	
Apoptosis	Caspase 3 & 9	
<b>Novel techniques</b>	<b>Sample type: cells, tissue</b>	
Mitochondrial Function	Mitochondrial function of permeabilized muscle fibers and cells (respiration, reactive oxygen species emission, sensitivity to permeability transition)	<ul style="list-style-type: none"> <li>• OROBOROS Oxygraph-2k fluorometer system for high-resolution mitochondrial function assessment</li> <li>• Agilent/Seahorse XFe96 Flux Analyzer <ul style="list-style-type: none"> <li>○ BioTek Cytation 1 cell imaging multi-mode reader</li> </ul> </li> </ul>
Imaging (Multiphoton and Confocal)	Mitochondrial function/morphology/free iron levels  Neuromuscular junction morphology	<ul style="list-style-type: none"> <li>• Zeiss 510 NLO multiphoton laser scanning confocal microscope equipped with a META spectral detector and a Coherent Chameleon XR tunable femtosecond pulsed Ti-Sapphire laser</li> <li>• Leica SP8 inverted confocal microscope with super-resolution Hyvolution (120 nm in x-y)</li> </ul>

<b>Analysis technique</b>	<b>Measure</b>	<b>Specialized Equipment</b>
<b>COGNITION/ATTENTION/EXECUTIVE FUNCTION</b>		
Water Maze	Morris Swim Task	<ul style="list-style-type: none"> <li>• Noldus Ethovision automated tracking system</li> <li>• Pools (mice/rats)</li> <li>• Stereoscopes (C. elegans)</li> <li>• Chemotaxis plates (C. elegans)</li> <li>• Incubators (C. elegans)</li> </ul>
Object Location/Context/Recognition	Cue discrimination, spatial discrimination	
Chemotaxis	Olfactory sensation and olfactory learning	
<b>MOTOR/SOMATOSENSORY</b>		
Locomotor Activity	Voluntary Activity	<ul style="list-style-type: none"> <li>• Noldus Ethovision automated tracking system</li> <li>• Customized Wormlab C. elegans locomotion tracker (MBF Bioscience) (also applicable to larval Drosophila)</li> </ul>
Running Wheel	Voluntary Exercise Activity	<ul style="list-style-type: none"> <li>• Nalgene Activity Wheels (rat). Each wheel is equipped with a magnetic switch and a counter with liquid crystal display (LCD) that records the number of wheel revolutions.</li> </ul>
Treadmill	Exercise/exercise tolerance testing.	<ul style="list-style-type: none"> <li>• Five-lane Rodent Treadmill (Panlab/Harvard Apparatus)</li> </ul>
Grip Strength	Forelimb grip strength (mean force in grams will be divided by body weight). Average measurements from three successful trials will be taken as the final outcome.	<ul style="list-style-type: none"> <li>• Automated Grip Strength Meter (Columbus Instruments, Columbus Ohio): computerized electronic pull strain gauge that is fitted directly to the grasping ring.</li> </ul>
<b>Body Composition</b>		
Nuclear Magnetic Resonance	NMR analyzer delivers body composition measurements of fat, lean, free water, and total water masses in live animals weighing up to 700 grams.	<ul style="list-style-type: none"> <li>• EchoMRI™-700</li> </ul>

Auditory		
ABR	Auditory Brainstem Response (ABR) is an electrical signal generated from the brainstem in response to sound: measures the brainstem's response to sound.	<ul style="list-style-type: none"> <li>• Intelligent Hearing System with Smart-EP v10 software</li> <li>• Tucker Davis Technologies Evoked Potentials and EEG system</li> <li>• Sound Isolation Booth (GretchKen Industries, Inc.)</li> </ul>
DPOAE	DPOAE (distortion product otoacoustic emission) are generated in the cochlea in response to two simultaneous tones of a given frequency and sound pressure level presented in the ear canal: indicative of outer hair cells function, providing information as to the source of hearing loss and is a general method to test hearing levels in animals.	