



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

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### **Faculty:**

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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>		
Oxidation	MPO, oxLDL, Protein carbonyls, ferritin	<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>
Inflammation	TNF $\alpha$ , IL-6, IL-8, V-CAM, Selectins, CRP	
Apoptosis	Cell death ELISA	
<b>Multiplex Immunoassays</b>		
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	<ul style="list-style-type: none"> <li>• Millipore MILLIPLEX® Analyzer 3.1 xPONENT System with MILLIPLEX® Analyst 5.1 software</li> <li>• BioTek Instruments ELx405™ HT Microplate Washer - for 96-well plates</li> </ul>
<b>Q-PCR</b>		
Inflammation	TNF $\alpha$ , IL-6, NF- $\kappa$ B	<ul style="list-style-type: none"> <li>• Applied Biosystems, Inc 7300 Real-Time PCR System</li> </ul>
Autophagy/Proteolysis	Lamp 2, Atg 5, 7, 12, Beclin, MurF 1, Atrogin 1, Foxo-3	
<b>Western blot</b>		
Oxidative stress	Nitrotyrosine, 4-HNE	<ul style="list-style-type: none"> <li>• Bio-Rad Criterion™ gel electrophoresis</li> <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>
Mitochondrial	Fusions & Fission (Opa1, Fis1, Mfn1, Drp1) PGC1 $\alpha$ , Tfam,	
Inflammation	TNF $\alpha$ Receptor I, NF- $\kappa$ B	
Apoptosis	Caspases 3 & 8, PARP, Endo G, & AIF	
Autophagy/Proteolysis	LC3B, MurF 1, Atrogin 1, Foxo-3	
<b>Activity assays</b>		
Oxidative	ROS production (H <sub>2</sub> O <sub>2</sub> ), Aconitase,	<ul style="list-style-type: none"> <li>• Bio-Tek Synergy HT multidetection microplate reader (fluorescence,</li> </ul>
Mitochondrial	Cox IV, citrate synthase	

Apoptosis	Caspase 3 & 9	absorbance and luminescence measurements)
<b>Novel techniques</b>		
Respirometry	Mitochondrial function/efficiency in permeabilized muscle fibers (State 3, State 4, uncoupling)	<ul style="list-style-type: none"> <li>• OROBOROS Oxygraph-2k system for high-resolution respirometry</li> </ul>
Imaging (Multiphoton and Confocal)	Mitochondrial function/Morphology/free iron levels	<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> </ul>
HPLC-ECD	DNA/RNA oxidation in blood and urine	<ul style="list-style-type: none"> <li>• ESA HPLC System with electrochemical detector and autosampler</li> </ul>

<b><u>Analysis technique</u></b>	<b><u>Measure</u></b>	<b><u>Specialized Equipment</u></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> </ul>
Object Location/Context/Recognition	Cue discrimination, spatial discrimination	
<b>MOTOR/SOMATOSENSORY</b>		
Locomotor Activity	Voluntary Activity	<ul style="list-style-type: none"> <li>• Activity Monitors (Med Associates). These devices are cubicles (43.5 X 43.5 X 23 cm). Arrayed along the sides are a series of infrared emitters and receivers which allow the automatic recording of an animal's position and movement in the chamber.</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>

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.	

