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141st O2k-Workshop on high-resolution respirometry

2019 Sep 23rd-28th
Schroecken, Vorarlberg, Austria

The **141st O2k-Workshop on high-resolution respirometry (HRR)** is the **42nd** International Oxygraph Course held in Schroecken since 1988. We will provide an overview of the **O2k-FluoRespirometer**, including real-time analysis by **DatLab 7.4 (*new*)** and applications of the **Titration-Injection microPump TIP2k**. O2k-Demo experiments will be used to demonstrate the unique advantages and limitations of simultaneous monitoring of oxygen concentration, respiration, and hydrogen peroxide production. During the hands-on sessions, HEK 293T cells are used as a biological reference sample, which are easily stored and shipped on dry-ice introducing the MitoFit Proficiency Test.

The course provides a full introduction to learn how to do the instrumental setup and service of the polarographic oxygen sensor (**OroboPOS**), followed by hands-on practice in 10 teams. A wide range of mitochondrial topics will be covered, including abstracts and experimental experiences presented by the participants.

On previous workshops, IOC participants invariably asked for a detailed discussion about protocol design. The **Blue Book** (5th edition in prep.) and **Mitochondrial respiratory states and rates** provide a basic introduction to mitochondrial physiology, complemented by overview presentations with examples, including **DatLab Analysis** of demo files. During the course, the students will be introduced to our **instrumental quality control system**, which is a fundamental component of HRR and will be put to the practical test in teams using seven O2k (14 chambers). Finally, the **O2k-FluoRespirometer** and its **O2k-MultiSensor** applications, particularly fluorescence measurements, will be introduced with hands-on sessions about ROS production measurement with Amplex® UltraRed.

The 141st workshop is a unique opportunity to learn about the new developments for HRR. We will introduce our new tool for selecting the best SUIT protocol for your research question: **Oroboros SUITbrowser**. Also, a parallel session with the NextGen-O2k and **Q-redox** measurements will be run (Q-Module). Lunch breaks provide an excellent opportunity for relaxing *Walks&Talks*, enjoying the refreshing scenery of the secluded alpine environment or using the spare time for individual practice with the O2k.

Lecturers and tutors

Cardoso Luiza	Postdoc, Oroboros Instruments (AT)
Di Marcello Marco	Research Assistant, Oroboros Instruments (AT)
Garcia-Souza Luiz Felipe	PhD student, Oroboros Instruments (AT)
Gnaiger Erich	CEO, Oroboros Instruments (AT)
Iglesias-Gonzalez Javier	Principal Investigator, Oroboros Instruments (AT)
Komlodi Timea	Postdoc, Oroboros Instruments (AT)

NextGen O2k

Oroboros - as a driving force in mitochondrial physiology - extends the analytical and diagnostic power of high-resolution respirometry by integration of NADH- and Q-redox monitoring in the **NextGen-O2k**. We aim at establishing the Oroboros quality control management for dissemination to our worldwide O2k-Network laboratories. This will become an effective contribution to address the acute *reproducibility crisis* of scientific investigation. In the spirit of Open Science and global networking, we will enable data sharing across projects and institutions in an Open Access database on mitochondrial physiology and pathology, to resolve the *inflation crisis* and ultimately the *value-impact crisis* of present academic publication. This will support key developments in mitochondrial medicine. In addition, we expand our business to algal biotechnology and ecology with the photobiology module of the NextGen-O2k, widening our focus from medicine to environment and climate.

Programme

1 Monday, Sep 23rd

*printed in workshop materials

	Arrival	Weblink
15:00	**Arrival in Bregenz:** Meeting point Bregenz train station at 3:00 pm; approx. 1 h bus drive to Schroecken and Hochtannberg (Salober); walk to Hotel Körbersee (approx. 40 min)	IOC-travel
18:30-19:30	*Welcome reception at Hotel Körbersee & **get-together:*** Introduction of participants and their research interests - a welcome by Oroboros Instruments	Schroecken
19:30	*Dinner*	

2 Tuesday, Sep 24th

	Workshop 1			Weblink
07:30-08:30	*Breakfast*			
08:30-10:30	**O2k-Series H and DatLab 7** O2k instrumental setup – overview with video clips			O2k-FluoRespirometer MitoPedia: DatLab DL-Protocols O2k-Videosupport O2k-Start
10:30-12:30	**Hands-on (11 teams; HRR & Q)**			
	DatLab 7	**OroboPOS service**	**Q service**	
10:30-11:15	Teams 1-5	Teams 6-10	Q Team	POS Service
11:15	*Coffee / Tea*			
	DatLab 7	**OroboPOS service**	**DatLab 7 & DatLabPro**	POS Service
11:45-12:30	Teams 6-10	Teams 1-5	Q Team	O2k-Start
12:30	*Lunch packages/ Walk & Talk* *Alternative:* individual O2k-practice			
14:30-15:30	**Hands-on (7 teams + Q team)**			Gnaiger 2008 POS SOP: O2-calibration
	Oxygen calibration (instrumental quality control 1) DL-Protocol: O2k-cleaning BeforeUse DL-Protocol: O2 calibration air		**Q team: Cyclic voltammetry (instrumental quality control 1)** DL-Protocol: O2k-cleaning BeforeUse DatLab Pro: Cyclic voltammetry	
15:30	Coffee / Tea			
16:00-18:00	**Hands-on (7 teams): cell respiration** Respiration and simultaneous measurement of H_2O_2 production in permeabilized cryopreserved HEK cells DL-Protocol (O2&AmR): SUIT-026 AmR mt D064 DL-Protocol: O2k-cleaning AfterUse		**Hands-on (Q-Team)** Oxygen calibration and Q measurement (imt) DL-Protocol: O2k-cleaning BeforeUse DL-Protocol: O2 calibration air DL-Protocol: Q-measurement DL-Protocol: O2k-cleaning AfterUse	MitoPedia: SUIT SUIT-026_AmR_mt_D064
18:30	*Dinner*			
20:00-21:00	**DatLab analysis:** Quality control and reproducibility of technical repeats			DatLab-Analysis

3 Wednesday, Sep 25th

	Workshop 2		Weblink
	HRR Team	Q Team	
07:30-08:30	*Breakfast*		
08:30-09:45	**Experimental design:** Pathway and coupling control of mitochondrial respiration		MitoPedia: Respiratory states
09:45-10:00	**Oroboros SUITbrowser**		Oroboros SUITbrowser
10:00	*Coffee / Tea*		
10:30-11:15	**Substrate-uncoupler-inhibitor titration (SUIT) protocols** – fundamental principles	**Q team-**Technical development and discussion about the new Q-Module	MitoPedia: SUIT
11:15-12:30	**Hands-on (7 teams) - getting started with an O2k experiment:** washing, stirrer test, air calibration DL-Protocol: O2k-cleaning BeforeUse DL-Protocol: O2 calibration air	**Q team-Round table**	SOP: O2k-cleaning and ISS SOP: POS-calibration
12:30	*Lunch packages / Walk & Talk alternative:* individual O2k-tasks		*The Blue Book p 56**
14:00-16:30	**Hands-on (7 teams) - O2k-experiment** Respiration of permeabilized cells: Measurement of oxygen consumption with RP1 (SUIT-001) and RP2 (SUIT-002) with 7 Power-O2k DL-Protocol (O2): SUIT-001 O2 ce-pce D003 and SUIT-002 O2 ce-pce D007 DL-Protocol: O2k-cleaning AfterUse	**Hands-on (Q-Team) - getting started with the Q-Module** Oxygen calibration and Q measurement (pce) DL-Protocol: O2k-cleaning BeforeUse DL-Protocol: O2 calibration air DL-Protocol: Q-measurement DL-Protocol: O2k-cleaning AfterUse	SUIT reference protocol SUIT-001_O2_ce-pce_D003 SUIT-002_O2_ce-pce_D007
16:00	*Coffee / Tea – Take turns in your team to continue the experiment*		
16:30-17:45	**DatLab analysis and SUIT protocols** Flux per volume, flux per mass, flow per cell, flux control ratio, flux control factor		MitoPedia: Respiratory control ratios MitoPedia: SUIT
17:45-18:45	**DatLab analysis: hands-on in teams** Analysis of the hands-on experiment with permeabilized cells		O_2-Flux Analysis MitoPedia: DatLab
19:00	*Dinner + registration for the walk to the Alpmuseum*		
20:30-21:30	**Tony Moore:** tba.		

4 Thursday, Sep 26th

	Workshop 3		Weblink
07:30-08:30	*Breakfast*		
08:30-09:00	**Introduction to H_2O_2 measurements**	**Q team: Cyclic voltammetry (instrumental quality control 1)** DL-Protocol: O2k-cleaning BeforeUse DatLab Pro: Cyclic voltammetry	Amplex UltraRed H_2O_2
09:00-10:30	**Hands-on (7 teams): H_2O_2 production produced by reverse electron transfer in permeabilized cells using 7 O2ks** DL-Protocol (O2&AmR): SUIT-018_AmR_mt_D041 DL-Protocol: O2k-cleaning AfterUse		SUIT-018_AmR_mt_D041
10:00	*Coffee / Tea – Take turns in your team to continue the experiment*		
10:30-11:00	**Hands-on (7 teams): continuation** DL-Protocol (O2&AmR): SUIT-018_AmR_mt_D041 DL-Protocol: O2k-cleaning AfterUse	**Hands-on (Q-Team) - getting started with the Q-Module** Oxygen calibration and Q measurement (pce) DL-Protocol: O2k-cleaning BeforeUse DL-Protocol: O2 calibration air DL-Protocol: Q-measurement DL-Protocol: O2k-cleaning AfterUse	SUIT-018_AmR_mt_D041
11:00-12:30	**H_2O_2 data analysis: introduction**	**Q data analysis: introduction**	
12:30	*Lunch packages / walk & talk alternative:* individual O2k-tasks		
14:30-15:50	**DatLab analysis: hands-on in teams and summary discussion**	**Q DatLab analysis: hands-on and summary discussion**	O_2-Flux Analysis
15:50-16:30	**From isolated mitochondria to tissue fibers and tissue homogenate preparation:** The PBI-Shredder (overview with video clips)		MiPNet17.03 Shredder vs Fibres O2k-Videosupport
16:30	*Coffee / Tea*		
17:00-18:00	**Blue Book: chapter 8**		MitoPedia: SUIT Oroboros SUITbrowser
18:00-19:00	**Quiz: data interpretation using SUIT protocols** **OXPHOS analysis: diagnosis of respiratory defects**		MitoPathways and respiratory control Oroboros SUITbrowser
19:00	*Dinner*		
20:30-22:00	**O2k perspectives:** 10+5 min presentations of abstracts 1-6		

5 Friday, Sep 27

	Workshop 4	Weblink
07:30-08:30	*Breakfast*	
08:30-09:00	**Introduction to instrumental O_2 background** (Demo-Experiment), using the TIP2k DL-Protocol: Instrumental O2 background TIP2k	SOP: O2 background TIP2k manual
09:00-11:00	**Hands-on (7 teams + Q team): Instrumental O_2 background (instrumental quality control 2)** O2 background test with the TIP2k; analysis of oxygen flux; O2 background from air saturation to zero oxygen concentration; or for permeabilized muscle fibres in the high-oxygen range of 500 – 200 µM Q background and stoppers DL-Protocol: Instrumental O2 background TIP2k	SOP: O2 background
10:30	*Coffee / Tea – Take turns in your team to continue the experiment*	MiPNet18.10 O2kvsMultiwell*
11:00-12:00	**Data analysis**	The Blue Book* pp 43-57
12:00	*Lunch packages*	
12:30-15:30	*Walk to the Alpmuseum - guided tour and reception: € 15.-*	Alpmuseum*
15:30	*Coffee / Tea*	
16:00-17:00	**NextGen-O2k: The revolutionary all-in-one instrument to conquer mitochondrial disease**	NextGen-O2k
17:00-17:30	**Introduction to mitochondrial membrane potential**	Mitochondrial membrane potential
17:30-18:00	**MitoFit Preprint Archives**	MitoFit Preprint Archives O2k-Publications
18:30	*Dinner*	
20:00	**Tutorial on the Bioblast wiki** www.bioblast.at/ *Feedback discussion: Next steps in the individual projects*	O2k-Network www.bioblast.at

6 Saturday, Sep 28th

	Departure
06:30-7:30	*Breakfast*
	Early morning: departure from Hotel Körbersee at 08:15 am, bus departure 9.00 am at Salober.

O2k-Workshop: OUR COMMON AIMS

- **Mitochondrial physiology:**
  Study mitochondrial function in the **context** of cell physiology and pathology
- **Instrumental performance – the O2k:**
  - Learn **high**-resolution respirometry
  - Gain **hands-on** experience
  - Extend to O2k-**Multi**Sensor applications
- **Excellence in research:**
  - Instrumental **quality** control
  - Experimental design for **innovation**
  - Data analysis meeting superior **standards**

List of participants

Participant	Institution
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*Asterisks indicate the number of O2k instruments in the participant's lab.

MiPNet24.02 Abstracts IOC141: 10+5 min

O2k perspectives

1. N Tremblay, L Leiva, J Haafke, CL. Meunier, M Boersma. (2019) Effects of low-frequency noise and temperature on copepod performance. Mitochondr Physiol Network 24.02.

Offshore wind farms (OWF) are often touted for their "green energy" etiquette and their artificial reef-like structures that promote secondary production by benthic invertebrates. As the number of OWF are bound to increase as a mitigation strategy to reduce the emission of greenhouse gases, it is crucial to address all of their potential impacts on key ecosystem components in detail. Especially, the chronic effect of noise created during OWF turbine operations (duration 20-25 years) must be understood. In cephalopods exposed to low-frequency exposures, complete mitochondria in cells adjacent to sensory organs were degenerated (André et al. 2011, Front Ecol Environ, doi: 10.1890/100124). This is one of the reasons why the mitochondria are in the spotlight. Life-history theory predicts adaptive shifts in response to stress, namely earlier reproduction, smaller age/size at maturity, and higher relative investment into reproduction. Such shifts should bring about reduced life expectancy. The overall project aims to assess the trade-off for key crustacean species between the effect of a long-term exposure to OWF operational noise on performance (physiological and ecological) and the protection offered by those artificial refuges. Many anti-ageing mechanisms have been identified in crustaceans, but the effect of environmental stressors on premature senescence remains unclear in those organisms (Vogt. 2012, Zool Anz, doi: 10.1016/j.jcz.2011.05.003).

Here, we carried out experiments with one model key organisms, the pelagic copepod Acartia tonsa. Because of copepod's feeding modes, which relies on setal receptors that sense the vibration and velocity of the particles they feed on, the effect of low-frequency noise generated by OWF turbines could potentially altered their capacity in gathering the energy required to fulfil all their biological functions. The copepod species is commonly used as a proxy for a range of fundamental processes that relate to marine planktonic crustaceans. Given that higher temperatures increase metabolic demands, the experiment was conducted at three different temperature levels (18, 21, 24°C) combined with silent and noise treatments. So far, we assessed the combined effects on energetic balance and oxidative stress indicators. First results from our work indicate no changes in feeding and respiration rates when copepods were exposed to low-frequency noise coupled with higher temperatures. However, an important decrease in the antioxidant system defenses was

observed, except the activity of the glutathione S-transferase. The latter enzyme is involved in detoxification processes, which means that animals were not healthy. By measuring mitochondrial capacity and the production of hydrogen peroxide under noise exposure, it will be possible to assess if noises are potentially disruptors of the general mitochondrial dynamic and functioning, which would ensue suboptimal biological processes, and jeopardize the population sustainability.

2. Dora Ravasz, David Bui, Alex Kitayev, Bennett Greenwood, Collin Hill, Timea Komlodi, Carolina Doerrier, Oliver Ozohanics1, Anthony L Moore, Erich Gnaiger, Michael Kiebish, Krasimir Kolev, Thomas N Seyfried, Wayne T Willis, Niven Narain, Vera Adam-Vizi, Christos Chinopoulos (2019) Endogenous quinones sustain a moderate NADH oxidation by complex I during anoxia. Mitochondr Physiol Network 24.01.

Anoxia leads to over-reduction of mitochondrial quinone pools hampering complex I from oxidizing NADH, leading to a profound decrease in the matrix NAD+/NADH ratio. As a consequence of this, the function of matrix dehydrogenases is impaired. Yet, under certain anoxic conditions catabolism of metabolites converging through the ketoglutarate dehydrogenase complex (KGDHC) is known to occur yielding succinyl-CoA, in turn supporting substrate-level phosphorylation substantiated by succinate-CoA ligase. Here, by measuring simultaneously oxygen partial pressure and NADH autofluorescence or quinone redox state we show that in isolated mitochondria complex I utilizes endogenous quinones oxidizing NADH during anoxia. Untargeted metabolomic analysis of matrix metabolites of anoxic mitochondria and in the presence of ETC inhibitors inferred that NAD+ arising from complex I is utilized by KGDHC yielding succinyl-CoA for succinate-CoA ligase, thus maintaining substrate-level phosphorylation during anoxia. The amount of endogenous quinones was estimated to be in the millimolar range and was unaffected by dietary intake of vitamin K3 (menadione). The quinone pools could be reduced by complexes I and II and the electron transfer flavoprotein-ubiquinone oxidoreductase (ETF-QO) system during anoxia, exhibiting a descending order of affinity and reciprocally, increasing order of capacity. Our results highlight the importance of quinone availability in conjunction to complex I-mediated NADH oxidation in maintaining substrate-level phosphorylation during anoxia.

3. E Rodríguez, FM Camus, Nick Lane (2019) Mitochondria as "flux capacitors": the effect of mitonuclear incompatibilities on mitochondrial physiology, metabolomics profile and gene expression in D. *melanogaster*. Mitochondr Physiol Network 24.01.

Recent findings place mitochondria as more than simple "powerhouses" of eukaryotic cell, because of their fundamental role in regulating cellular function. Mitochondria integrate metabolic flux and stress, signal the physiological status of the cell to the nucleus and coordinate nuclear gene expression accordingly: they can thus be seen as "flux capacitors". This idea fits with their important implication in a wide range of diseases and the aging process, although the molecular mechanisms behind this remain poorly understood. An important feature of mitochondrial function is the fact that the respiratory proteins of the electron transport system are encoded by 2 genomes: nuclear and mitochondrial, which differ in their mode of inheritance and mutation rates. Incompatibilities or even subtle mismatches arising from mutations in either genomes can profoundly affect protein and cellular function, gene expression, to the point of disrupting normal health, fertility and lifespan. The consequences of these mismatches can be difficult to predict because of the variable penetrance of mtDNA mutations and their tissue specificity (among others), meaning that mitochondrial function can be affected by diet, temperature and stress in different ways. Linking alterations in mitochondrial function to changes in downstream metabolic flux and differences in gene expression is therefore needed to understand the underlying signalling processes. In order to do so, Drosophila melanogaster can prove a useful model whereby manipulations of the mitochondrial and nuclear genomes can create

genotypes that vary in the degree of match, and yield individuals on which fine-scale biochemical and genetic analysis can be performed. We aim to test the effects of three different treatments (2 drugs, 1 diet) in 1 coevolved and 2 mismatched fly lines. One of these mismatched lines harbors 30 SNPs differences in their mtDNA (compared to wild-type), mostly in complex I of the electron transport system, while the other has a single critical SNP difference at the level of complex IV. The drugs N-acetyl cysteine (NAC, an antioxidant modulating oxidative stress via glutathione metabolism), and Nicotinamide Riboside (NR, a precursor of NAD synthesis), will be tested; while the dietary treatment will consist in a high protein diet, which can alter TCA cycle flux via changes in carbohydrate and amino acid metabolism. Using the O2k-FluoRespirometer in different respiratory states, we will measure mitochondrial respiration, ATP synthesis, H2O2 flux, and membrane potential in adult thorax and reproductive tissues, of male and female flies at three time points during their lifetimes. Following these mitochondrial physiology measurements, we will look at the effects of the treatments on metabolomic profiles, gene expression and phenotype (fertility and longevity) to ultimately construct a predictive metabolic flux model.

4. Axelrod CL, Davuluri G, Zunica ERM, Noland RC, Hoppel CL, Kirwan JP (2019) Administration of BAM15 to Obese C57BL/6J Mice Increases Skeletal Muscle Fatty Acid Oxidation Independent of OXPHOS or ETC Capacity. Mitochondr Physiol Network 24.01.

Current pharmacologic strategies for the treatment of obesity remain ineffective at achieving long-term weight control. This is due, in part, to difficulties in identifying tolerable and efficacious small molecules and biologics capable of regulating systemic nutrient homeostasis. Mitochondria present a unique opportunity for drug targeting by regulating systemic nutrient flux across tissues and cell types. However, unfavorable pharmacokinetic properties, off-target effects, and poor tolerability have limited clinical application. Herein, we evaluated the effects of BAM15 on body weight regulation and skeletal muscle mitochondrial function. 16 (n=8 per group) diet induced obese (DIO) male C57BL/6J mice were randomized to 3 weeks of high fat diet (HFD) or BAM15 (HFD + 0.01% w/w BAM15). After 3 weeks, mixed gastrocnemius muscle was harvested after euthanasia and assessed for oxidative phosphorylation (OXPHOS) and electron transport (ETC) capacity (1), as well as [1-14C] palmitate oxidation (2), as described previously. Mice treated chronically with BAM15 were resistance to dietary weight gain, attributable to reductions in fat accrual. BAM15 treated animals displayed increased skeletal muscle fatty acid oxidation. However, OXPHOS and ETC capacity with glucose or fatty acid substrates remained unchanged between control and BAM15 treated animals. We conclude that BAM15 is tolerable and efficacious small molecule for the treatment of obesity. Importantly, chronic administration of BAM15 does not result in mitochondrial fatigue or dysfunction, warranting further investigation into pre-clinical efficacy and tolerability.

References

1. Ye F, Hoppel CL. Measuring oxidative phosphorylation in human skin fibroblasts. Anal Biochem. 2013;437(1):52-8. Epub 2013/03/07. doi: 10.1016/j.ab.2013.02.010. PubMed PMID: 23462540.
2. Koves TR, Ussher JR, Noland RC, Slentz D, Mosedale M, Ilkayeva O, Bain J, Stevens R, Dyck JR, Newgard CB, Lopaschuk GD, Muoio DM. Mitochondrial overload and incomplete fatty acid oxidation contribute to skeletal muscle insulin resistance. Cell Metab. 2008;7(1):45-56. Epub 2008/01/08. doi: 10.1016/j.cmet.2007.10.013. PubMed PMID: 18177724.

5. Scandalis L, Kramer P, Nelson B, Dozier S, Stone J, Kitzman D, Molina AJA (2019) Oxygen consumption of Skeletal Muscle in Heart Failure with Preserved Ejection Fraction. Mitochondr Physiol Network 24.01.

Heart failure with preserved left ventricular ejection fraction (HFpEF) is the most prevalent form of heart failure (HF) and is nearly unique to older adults, particularly older women. This disorder manifests as severe exercise intolerance as evidenced by significantly reduced peak exercise oxygen uptake (peak VO2). Previous studies indicate that, in addition to underlying cardiac dysfunction, 'non-cardiac' factors contribute to this reduction in exercise tolerance.
Several lines of evidence indicate that older adults with HFpEF have altered skeletal muscle metabolism. We have previously reported that older HFpEF patients have abnormal skeletal muscle

oxygen utilization, mitochondrial content, and oxidative capacity compared to healthy controls. The relationships of these mitochondrial parameters with measures of exercise capacity indicate that these deficits may contribute to impaired skeletal muscle aerobic metabolism and severely reduced exercise tolerance in patients with HFpEF. In the current study, we examine skeletal muscle mitochondrial bioenergetics in patients with HFpEF. We obtained Vastus lateralis biopsies from 33 patients with HFpEF and 44 healthy controls. High resolution respirometry of permeabilized skeletal muscle fiber bundles revealed significantly lower oxygen consumption rates across states, including respiration driven by complex 1, complex 2, and MAX ETS.

6. Kramer P, Distefano G, Monte J, Mills A, Edmunds L, Jurczak M, Cummings SR, Kritchevsky S, Newman AB, Hepple RT, Goodpaster B, Coen PM, Molina A (2019) The Study of Muscle, Mobility, and Aging (SOMMA): A Multi-Institutional Respirometry Study. Mitochondr Physiol Network 24.01.

The decline in muscle mass and function with age can result in significant losses to mobility, independence, quality of life, and pose a significant financial burden to older adults. The cause of age-related loss of muscle mass and function are not adequately defined but may be due to a decreased mitochondrial efficiency and capacity for ATP production. The Study of Muscle, Mobility, and Aging (SOMMA) is a prospective, observational multi-institutional study that began enrollment in May 2019. The goal of the study is to recruit 875 older adults (>70 YOA) who are at risk of mobility disability at the University of Pittsburgh, and Wake Forest University. We will then track loss of mobility over a 3 year follow up period. Muscle biopsies are being collected at baseline to measure mitochondrial function. We hypothesize that mitochondrial function will predict slowing and incidence of mobility disability. As of August 2019, >90 respirometry assays have been performed, and internal and external quality control measures have been implemented between the two institutions. Quality control methods include muscle wet weight normalization, cytochrome c injection for outer membrane integrity, and assay timing data to account for variations in processing, permeabilization, and assay start time. The primary respirometry protocol consists of serial injections of pyruvate, malate, sub-saturating ADP (x2), saturating ADP, cytochrome c, glutamate, succinate, FCCP (x4), Antimycin A, and ascorbate/TMPD. The samples are run in duplicate on the Oroboros High-Resolution Respirometer. Assessing the mitochondrial function in muscle from older adults will provide valuable insight into the loss of muscle mass and mobility with age, and quality control measures will ensure reliable respirometry profiles between clinical sites.

MiP2019/MitoEAGLE Belgrade RS

Belgrade RS, 13-16 Oct 2019. 14th Conference on Mitochondrial Physiology: Mitochondrial function: changes during life cycle and in noncommunicable diseases - COST MitoEAGLE perspectives and MitoEAGLE WG and MC Meeting.

Accommodation and location

Hotel Körbersee www.koerbersee.at
T +43 5519 265 hotel@koerbersee.at

More detail?

Gnaiger E (2019) Mitochondrial pathways and respiratory control. An introduction to OXPHOS analysis. 5th ed. Mitochondr Physiol Network 24.05. Oroboros MiPNet Publications, Innsbruck:96 pp. » Full text in Bioblast

O2k-Manual – http://wiki.oroboros.at/index.php/O2k-Manual

O2k-Procedures – http://wiki.oroboros.at/index.php/O2k-Procedures

>3,300 O2k-Publications – http://wiki.oroboros.at/index.php/O2k-Publications:_Topics

COST Action CA15203 MitoEAGLE

MitoEAGLE Mitochondrial respiratory states and rates. MitoFit Preprint Arch doi:10.26124/mitofit:190001.v6
Mitochondrial respiratory states and rates

MitoFit Preprint Archive

The Open Access preprint server for mitochondrial physiology and bioenergetics

Articles can be submitted by Email as a pdf file with embedded figures accompanied by the abstract as a word (text) document.
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» https://www.mitofit.org/index.php/MitoFit_Preprint_Archives

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Mitochondria and cell research

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