

132nd International Workshop on high- resolution respirometry

2018 August 24-25
Budapest, Hungary

Venue:

Department of Medical Biochemistry
Semmelweis University
Budapest, Hungary
37-47 Tuzolto Str, HU-1088

Host:

Prof. László Tretter
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The **132nd O2k-Workshop on high-resolution respirometry (HRR)** is held in cooperation with our O2k-Network Lab in Hungary. This O2k-Workshop presents a basic introduction to the **Oroboros O2k** with integrated real-time analysis by **DatLab**. We introduce the new **DatLab 7** software with innovative **DatLab-Protocols** and the concept of a quality control system including the MitoFit interlaboratory Proficiency Test.

HRR provides information on cell respiration with basic coupling control protocols. State-of-the-art OXPHOS analysis is extended using mt-preparations (permeabilized cells, permeabilized muscle fibers, tissue homogenate, isolated mitochondria), to evaluate coupling efficiencies and OXPHOS capacities with electron transfer into the Q-junction converging from NADH, FADH₂, succinate and α -glycerophosphate (N,F,S,Gp), to diagnose defects in respiratory electron transfer system pathways and the phosphorylation system. Novel developments are presented on **substrate-uncoupler-inhibitor titration (SUIT) protocols** in HRR using the **O2k-FluoRespirometer** for simultaneous measurement of respiration and hydrogen peroxide production (Amplex UltraRed®). Discussions are extended on comparison of measurement of mt-membrane potential using Safranin (fluorometric) versus TMRM (fluorometric) and on perspectives of HRR in mitochondrial physiology.

Lecturers and tutors

Gnaiger Erich	CEO, Oroboros Instruments (AT)
Doerrier-Velasco Carolina	CSO, Oroboros Instruments (AT)
Komlodi Timea	Research assistant, Oroboros Instruments (AT)
Meszaros Andras	CRO, Oroboros Instruments (AT)

Programme

1 Friday, August 24

*printed in workshop materials

Workshop 1		Weblink
08:30	Registration, welcome <i>Venue:</i> Department of Medical Biochemistry, Semmelweis University, Budapest, Hungary 37-47 Tuzolto Str, HU-1088	IOC132
09:00-09:30	Get-together: Introduction of participants and their research interests	
09:30-10:00	Application of the O2k Series H	
10:00-11:00	Real-time experiment: Oxygen calibration (instrumental quality control 1) and DatLab 7 DL-Protocol for air calibration	Gnaiger 2008 POS SOP: O2-calibration
11:00-11:30	<i>Coffee / Tea</i>	
11:30-13:00	Introduction and real-time experiment: high-resolution respirometry (mt-respiration and H₂O₂ production) with isolated mitochondria: studying the effect of oxygen concentration on H ₂ O ₂ production	Amplex UltraRed
13:00-13:30	DatLab analysis	Amplex UltraRed
13:30-14:30	<i>Lunch</i>	
14:30-15:30	Comprehensive OXPHOS analysis: substrate-uncoupler-inhibitor titration (SUIT) protocols for respiratory control by coupling and mitochondrial pathways, SUIT reference assay.	The Blue Book* SUIT reference protocol
15:30-16:30	DatLab data analysis and normalization of results Flux per volume, flux per mass, flow per cell, flux control ratio, flux control factor	MitoPedia: Respiratory control ratios MitoPedia: SUIT
16:30-17:00	<i>Coffee / Tea</i>	
17:00-18:00	Introduction to analysis of mitochondrial oxygen kinetics and O2kinetics software	
18:00	Questions and answers	

2 Saturday, August 25

Workshop 2		Weblink
08:30-10:30	Determination of mitochondrial (mt) membrane potential via Safranin and TMRM with isolated mitochondria: Introduction and Real-time experiment	Safranin,TMRM
10:30-11:00	<i>Coffee / Tea</i>	
11:00-12:00	Data analysis and interpretation: TMRM/safranin	
12:00-13:00	Final discussion	

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 FluoRespirometry
 - 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**

OROBOROS INSTRUMENTS

O2k

Mitochondria and cell research

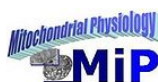


Oroboros: O2k in numbers



- **25 years** - since 1992
- **>950** instruments world-wide
- **>576** O2k-Network Labs in 49 countries
- **>2,600** O2k-Publications: www.orooboros.at
- **Oroboros-Team: 20**
- **131** O2k-Workshops

2018 Aug




MiP2018/MitoEAGLE
Jurmala LV

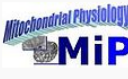
MiPschool
Tromso-
Bergen
2018



MiPschool Tromso-Bergen 2018



Tromso-Bergen, NO. 2018 Oct 20-24, 11th
MitoEAGLE Training School - MiPschool 2018.



More detail?

Gnaiger E (2014) Mitochondrial pathways and respiratory control. An introduction to OXPHOS analysis. 4th ed. Mitochondr Physiol Network 19.12. Oroboros MiPNet Publications, Innsbruck: 80 pp. » [Full text in Bioblast](#)

Doerrier C, Garcia-Souza LF, Krumschnabel G, Wohlfarter Y, Mészáros AT, Gnaiger E (2018) High-Resolution FluoRespirometry and OXPHOS protocols for human cells, permeabilized fibers from small biopsies of muscle, and isolated mitochondria. Methods Mol Biol 1782:31-70. » [Full text in Bioblast](#)

Komlodi T, Sobotka O, Krumschnabel G, Bezuidenhout N, Hiller E, Doerrier C, Gnaiger E (2018) Comparison of mitochondrial incubation media for measurement of respiration and hydrogen peroxide production. Methods Mol Biol 1782:137-55. » [Full text in Bioblast](#)

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

O2k-Protocols – <http://wiki.oroboros.at/index.php/O2k-Protocols>

>2,200 O2k-Publications – <http://wiki.oroboros.at/index.php/O2k-Publications: Topics>

COST Action CA15203 MitoEAGLE



MitoEAGLE preprint publication

[Mitochondrial respiratory states and rates: Building blocks of mitochondrial physiology](#)

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Mitochondria and cell research

O2k-Workshops are listed as [MitoGlobal Events](#)

