



Course on High-Resolution Respirometry

IOC75. *Mitochondrial Physiology Network* 18.03: 1-8 (2013)

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75th International Workshop on HRR

2013 April 09-14
Schröcken, Vorarlberg, Austria



The **75th Workshop on High-Resolution Respirometry (HRR)** is the **29th** International Oxygraph Course held in Schroecken since 1988. The workshop includes experiments with biological samples, providing a practical overview of the **Oxygraph-2k**, with integrated on-line analysis by **DatLab**, applications of the **TIP2k**, the new **O2k-Fluorescence LED2-Module**, and perspectives of HRR in mitochondrial physiology. Hands-on training will be done in six parallel groups.



An international team of experienced tutors guides small working groups step-by-step through the approach of HRR. Six O2k (12 chambers) are available for do-it-yourself applications of both hardware and software. Combined with an introduction and demo experiments, it is best to put the O2k into action yourself.



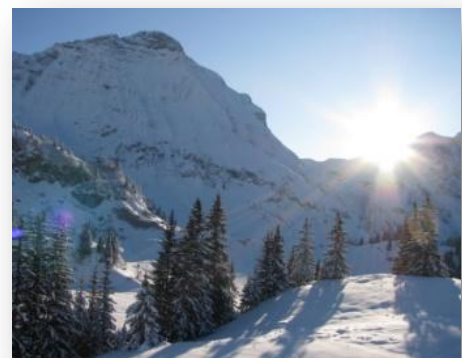
Lunch breaks provide an opportunity for skiing, relaxing walks and talks, enjoying the refreshing scenery of the secluded alpine environment, or using the spare time for specific tutorials. With DatLab 5 (new), we accomplish data analysis on-line, providing final results and their graphical presentation by the end of an experimental run. Thus, we gain sufficient time to see the Titration-Injection microPump TIP2k with feedback-control in action and practice its simple and automatic operation.

Lecturers and Tutors

[Gnaiger Erich](#)
[Fontana-Ayoub Mona](#)
[Laner Verena](#)

Guest Tutors

[Kiss Gergely](#) (Semmelweis University, HU)
[Konrad Csaba](#) (Semmelweis University, HU)
[Sumbalova Zuzana](#), PhD (Comenius University, SK)



Programme IOC75



1 Tuesday, April 09	
Arrival	
15:00	Arrival in Bregenz: Meeting point Bregenz train station at 3:00 pm; approx. 1 hour bus drive to Schröcken and Hochtannberg (Salober). Transfer/walk to Hotel Körbersee
18:30	<i>Welcome reception at Hotel Körbersee</i>
19:00	<i>Dinner</i>
20:30-21:00	Get-together: Introduction of participants and their research interests

2 Wednesday, April 10		
	Workshop 1	Weblink
07:30-08:30	<i>Breakfast</i>	
08:00	<i>Organize loan of skiing equipment</i>	
O2k-Core	Principles of high-resolution respirometry - from switching on the Oxygraph-2k to the experimental result	Gnaiger 2008 POS
08:30-09:00	Erich Gnaiger: Get O2k-Connected with your OROBOROS-USB: a guided tour to the O2k	get O2k-Connected
09:00-10:00	Instrumental quality control 1: The oxygen sensor OroboPOS - calibration, stability testing, and evaluation of sensitivity to measure oxygen flux.	O2k-Calibration
10:00	<i>Organize special lunch or bad weather task groups (fibre and homogenate prep, fluorometry and spectrophotometry, NO, pH and Ca, Bioblast wiki login) and loan of skiing equipment</i>	
11:00	<i>Practice - skiing, walk & talk, or individual O2k-tasks / Lunch</i>	
14:00	<i>Coffee / Tea</i>	
14:30-16:00	O2k-Demo experiment 1: Respiration of intact cells and on-line DatLab Analysis: Simultaneous measurement of oxygen consumption (O2k-Core) and H₂O₂ production (add-on O2k-Fluorescence LED2-Module)	O2k-Fluorometry Workshop
16:00	<i>Coffee / Tea</i>	
16:30-18:00	DatLab Guide through the menus: DL-Demo files and DL-Excel templates	DatLab Guide
18:30	<i>Dinner</i>	

20:00-20:30	Hot MiP-Topics: 10+5 min presentations of abstracts by participants	IOC75 Abstracts MiPNet18.03
20:30-21:00	Erich Gnaiger & Verena Laner: Taking mitochondria to the extremes: high-resolution, high-altitude, low-temperature	

3 Thursday, April 11		
Workshop 2		Weblink
07:30	Breakfast	
O2k-Basic	O2k instrumental setup	OroboPOS service
08:30-09:15	Groups A and B	Groups C and D
09:15-10:00	Groups C and D	Groups A and B
10:00	Practice - skiing, walk & talk, or individual O2k-tasks / Lunch	
14:00	Coffee / Tea	
14:30-16:00	Instrumental quality control 2: O2k-Background test and on-line analysis of oxygen flux.	O2k-Background
16:00	Coffee / Tea	
16:30-18:30	Hands-on (6 groups): O2k-Background from air saturation to zero oxygen concentration; or for permeabilized muscle fibres in the high-oxygen range of 500 - 200 µM. O2k-Background with automatic TIP2k or manual titrations.	O2k-Background
18:30	Dinner	
20:00-21:00	MiPNet-Lectures by guest tutors Konrad Csaba (HU): Fluorimetric applications of the O2k Oxygraph using the LED2 module on isolated mitochondria Kiss Gergely (HU): Functional importance of mitochondrial diaphorases in maintaining phosphorylation potential during inhibition of electron transport chain	

4 Friday, April 12		
Workshop 3		Weblink
07:30	Breakfast	
08:30-09:30	Experimental design 1: Coupling control of mitochondrial respiration: LEAK, OXPHOS, ETS, ROX	DatLab Flux Analysis
09:30-10:00	DatLab O ₂ flux analysis: Flux per volume, flux per mass, flow per cell, flux control ratio	Glossary: Respiratory states
10:00	Practice - skiing, walk & talk, or individual O2k-tasks / Lunch	
14:00	Coffee / Tea	
14:30-16:00	O2k-Demo experiment 2: SUIT protocol with mouse permeabilized fibres and on-line DatLab analysis	Pesta 2012 Methods Mol Biol

16:00	<i>Coffee / Tea</i>	
16:30-17:30	Hands-on: SUIT experiment continued with DatLab Analysis and guide through Excel templates	DatLab Flux Analysis
17:30-18:00	Experimental design 2: Coupling Control Protocol with intact cells: ROUTINE, LEAK, ETS, ROX	Cells: PCP
18:00-18:30	Experimental design 3: Substrate and coupling control of mitochondrial respiration.	CI+II
18:30	<i>Dinner</i>	
20:00-21:00	Hot MiP-Topics: 10+5 min presentations of abstracts by participants	IOC75 Abstracts MiPNet18.03

5 Saturday, April 13		
Workshop 4	Weblink	
07:30	<i>Breakfast</i>	
8:30-09:15	Working groups: Elaborate answers to the 'Questions for the O2k-Workshop'	IOC-Questions
9:15-09:45	IOC-Questions - discussion of 'Answers'	
09:45-10:15	The O2k-Workshop continues with the Bioblast wiki	www.bioblast.at
10:15	<i>Coffee / Tea</i>	
10:45-12:00	O2k-Demo experiment 3: Respiration and steady-state feedback control of oxygen levels with the TIP2k.	TIP2k User Manual
12:00	<i>Lunch</i>	
13:00-13:45	Mona Fontana-Ayoub: Introduction into trouble shooting	
14:00	<i>Walk (snowshoe rental) to the Alpmuseum: Guided tour and reception: 15 €</i>	www.alpmuseum.at
17:30	<i>Coffee / Tea</i>	
18:00-19:00	Erich Gnaiger: MitoPathways through Complexes I+II - perspectives of comparative mitochondrial physiology	Gnaiger 2009 Int J Biochem Cell Biol
19:00	<i>Dinner</i>	
20:30-21:00	Panel Discussion - Feedback IOC75	O2k-Feedback - Bioblast
Farewell party		

6 Sunday, April 14	
Departure	
<i>Breakfast</i>	
Early morning: Departure	

IOC75 Participants

Name	Lab
Asghar Mohammad	US_TX Houston_Asgar M: Department of Pharmacological & Pharmaceutical Sciences, College of Pharmacy - University of Houston. - Aging, GPCRs, Oxidative and inflammatory stress, sodium transporters
Ateba Dorine Louise	NO_Oslo_Eide L: Oslo University Hospital HF. - Diagnostics, mitochondrial respiratory chain diseases
Calloway Jones Jessica	US_MA Cambridge_Vernochet C: Cardiovascular and Metabolic Diseases Research Unit, Pfizer Research Technology Center (RTC). - Energy wasting, Diabetes
Camacho-Pereira Juliana	US_MN Rochester_Chini EN: Mayo Clinic, Rochester. - CD38, NAD, Deacetylases
Chung Weiliang	AU_Melbourne_Stepto NK: Institute of Sport, Exercise and Active Living, Victoria University. - Exercise, sport, adaptation
Dohlmann Tine	DK_Copenhagen_Dela F: Faculty of Health Sciences, University of Copenhagen. - Exercise, mitochondria
Fessel Joshua Patrick	US_TN Nashville_Fessel JP: Vanderbilt University Medical Center, Pulmonary and Critical Care Medicine. - Pulmonary hypertension, vascular biology, free radical biology/biochemistry
Fontana-Ayoub Mona (team tutor)	AT_Innsbruck_OROBOROS INSTRUMENTS
Frost Simon	DK_Copenhagen_Boushel RC: Copenhagen University Hospital at Rigshospitalet/Bispebjerg. - Hyperoxic sepsis, COLD sepsis
Gehrig Saskia	CH_Zurich_Lundby C: Center for Integrative Human Physiology (ZIHP), University of Zurich, Institute of Physiology. - High-intensity interval training, mitochondrial myopathy, muscle mitochondrial respiratory capacity, muscle morphology, respiratory chain enzyme activity
Gnaiger Erich (Organizer, Tutor)	AT_Innsbruck_OROBOROS INSTRUMENTS: DSL, Dept Visceral, Transpl Thoracic Surgery, Medical Univ Innsbruck.
Gomez Rodriguez Alexia	ES_Madrid_Barja G: Department of Animal Physiology-II, Faculty of Biological Sciences, Complutense University of Madrid (UCM). - Mitochondria, Aging, pO ₂
Goncalves Vera	PT_Oeiras_Teixeira M: Universidade Nova de Lisboa, Instituto de Tecnologia Química e Biológica. - Flavo/Iron proteins: a novel type of oxygen/nitric oxide reductases
Hägler Patrizia	CH_Basel_Kraehenbuehl S: Clinical Pharmacology & Toxicology University Hospital Basel. - Mitochondria, toxicity, liver, vitamin B12 deficiency
Hara Naomi	JP_Tokyo_Uchino H: Department of Anesthesiology, Tokyo Medical University. - Sepsis, mitochondrial dysfunction
Kallijärvi Jukka	FI_Helsinki_Mervaala E: Folkhälsan Research Center, Biomedicum, University of Helsinki. - GRACILE, BCS1L, Rieske, complex III, hepatopathy
Kilbaugh Todd	US_PA Philadelphia_Margulies S: Children's Hospital of Philadelphia. - Traumatic Brain Injury and other CNS disease processes
Kiss Gergely (guest tutor)	HU_Budapest_Chinopoulos C: Semmelweis University, Budapest
Khalifa Abdel Rahman Medhat	ET_Giza_Ali SS: Zewail City of Science & Technology, Giza. - Mitochondria, Synaptosomes, Cardiomyocytes, Oxidative stress, Aging
Konrad Csaba (guest tutor)	HU_Budapest_Chinopoulos C: Semmelweis University, Budapest

Kotb Islam Abo El Yazeed	ET_Giza_Ali SS: Zewail City of Science & Technology, Giza. - Mitochondria, Synaptosomes, Cardiomyocytes, Oxidative stress, Aging
Laner Verena (team tutor)	AT_Innsbruck_OROBOROS INSTRUMENTS
Mercer John	UK_Cambridge_Mercer J: Division of Cardiovascular Medicine, University of Cambridge, Addenbrookes Hospital. - Mitochondrial dysfunction in Atherosclerosis
Oliveira Pinto Matheus	BR_Rio de Janeiro_Oliveira MF: Institute of Medical Biochemistry, Federal University of Rio de Janeiro. - Schistosoma mansoni, metabolism, energy
Pfeiffer Annika	DE_Mainz_Methner A: Klinik und Poliklinik für Neurologie, Johannes Gutenberg-Universität Mainz, Forschungszentrum Translationale Neurowissenschaften (TN). - Mitochondrial function, mitochondrial respiration, mitochondrial biogenesis, disease, exercise training
Piel Sarah	SE_Lund_Elmer E: NeuroVive, Pharmaceuticals AB, Mitochondrial pathophysiology. - Pharmacological modification of mitochondria. Drug Toxicity
Porter Craig	US_TX Galveston_Finnerty CC: University of Texas Medical Branch, Shriners Hospital for Children, Department of Surgery, Metabolism Unit. - Skeletal muscle, adipose tissue, aging, burn injury
Prouteau-Angebault Claire	FR_Montpellier_Prouteau-Angebault C: INSERM U1051 Institut des Neurosciences de Montpellier, Hôpital St Eloi. - Cardiolipin, cytochrome c oxidase, OPA 1
Puskarich Michael	US_MS Jackson_Hosler J: Department of Biochemistry, University of Mississippi Medical Center. - Platelet, sepsis, post cardiac arrest
Ruud Lima Lene	NO_As_Egelandsdal B: Department of Chemistry, Biotechnology and Food Science, UMB, Norwegian University of Life Sciences. - SUIT protocols
Soerensen Liv Karin	NO_Tromsøe_Ingebretsen OC: Laboratoriemedisin, University Hospital of Northern Norway. - Diagnosis of human mitochondrial disorders
Schoettl Theresa	DE_Freising_Fromme T: Molecular Nutritional Medicine, Technische Universität München, Else Kröner Fresenius Center. - Mature white adipocytes; isolated white adipocyte mitochondria; Insulin resistance
Schlotte Volkhard	DE_Frankfurt_Leeuw T: Sanofi Aventis GmbH. - Respiration and aging
Scott Mark	NO_Oslo_Eide L: Oslo University Hospital HF. - Fish, mitochondria, anoxia, oxygen consumption
Sumbalová Zuzana (guest tutor)	SK_Bratislava_Sumbalova Z: Comenius University, Bratislava
Treberg Jason	CA_Winnipeg_Treberg JR: Department of Biological Sciences, University of Manitoba. - Fish, ROS, antioxidants, temperature, diet
Valenzuela Limiñana Rita	ES_Santiago de Compostela_Mendez-Alvarez E: Laboratory of Neuroanatomy and Experimental Neurology, Department of Morphological Sciences, University of Santiago de Compostela. - Parkinson disease, rat brain and cell culture isolated mitochondria, ROS production, Neuroprotection
Van Hove Johan	US_CO Denver_Van Hove J: University of Colorado, Department Pediatrics, Section of Clinical Genetics and Metabolism. - Diagnosis of inherited mitochondrial diseases - identification of new genetic mitochondrial diseases
Van Schaardenburgh Michel	NO_Trondeim_Rognmo O: Cardiac Exercise Research Group, Department of Circulation and Medical Imaging, Faculty of Medicine, Norwegian University of Science and Technology (NTNU). - Mitochondrial function in patients with peripheral arterial disease

MiPNet Abstracts IOC75: 10+5 min

Hot topics in Mitochondrial Physiology

Online: www.bioblast.at/index.php/MiPNet18.03_IOC75_Abstracts

Continue the discussion:

www.bioblast.at/index.php/Talk:MiPNet18.03_IOC75_Abstracts

Camacho-Pereira J, Matalonga J, Escandes C, Chini EN (2013) **CD38 as a regulator of mitochondrial function.**

Dohlmann T, Hindsoe M, Larsen S, Dela F, Helge JW (2013) **Effects of high-intensity interval training on mitochondrial respiratory capacity.**

Gomez Rodriguez A, Sanchez-Roman I, Naudi A, Lopez-Torres M, Pamplona R, Barja G (2013) **β -adrenergic receptor signaling interruption with two β -blockers (atenolol or nebivolol) can modify different oxidative stress related parameters linked to longevity in mouse liver.**

Goncalves V, Pinto L, Testa F, Giuffre A, Sarti P, Vicente JB, Teixeira M (2013) **FLAVODIIRON PROTEINS: A Novel Type of Oxygen/Nitric Oxide Reductases.**

Kallijarvi J (2013) **Respiratory chain dysfunction in the complex III assembly disorder GRACILE.**

Oliveira Pinto M, Oliveira MF (2013) **Studies on the energy and redox metabolism of the blood fluke *Schistosoma mansoni*.**

Porter C, Herndon DN, Radhakrishnan RS, Boersheim E, Sidossis LS (2013) **Severe thermal trauma alters adipose tissue mitochondrial function in children.**

Prouteau-Angebault C, Sarzi E, Seveno M, Gueguen N, Hamel CP, Reynier P, Lenaers G (2013) **Are cardiolipins involved in the mitochondrial respiratory chain defect in OPA1 mouse model?**

Seok-Hyung K, Scott SA, Bennett MJ, Carson RP, Fessel JP, Brown HA, Ess KC (2013) **Structural, Functional, and Signaling Abnormalities in Mitochondria and Multiple Organ Systems in a Novel Zebrafish Model of Multiple Acyl-CoA Dehydrogenase Deficiency (MADD).**

Van Schaardenburgh M (2013) **Five Plus Study.**

Accommodation and Location

Hotel Körbersee www.koerbersee.at

T +43 5519 265; hotel@koerbersee.at



Further information

O2k-Manual – www.oroboros.at/?O2k-Manual

O2k-Protocols – www.oroboros.at/?O2k-Protocols

O2k-Publications – www.oroboros.at/?O2k-Publications

Recommended Reading

- Pesta D, Gnaiger E (2012) High-resolution respirometry. OXPHOS protocols for human cells and permeabilized fibres from small biopsies of human muscle. *Methods Mol Biol* 810: 25-58.
- Lemieux H, Semsroth S, Antretter H, Hofer D, Gnaiger E (2011) Mitochondrial respiratory control and early defects of oxidative phosphorylation in the failing human heart. *Int J Biochem Cell Biol* 43: 1729-1738.
- Pesta D, Hoppel F, Macek C, Messner H, Faulhaber M, Kobel C, Parson W, Burtscher M, Schocke M, Gnaiger E (2011) Similar qualitative and quantitative changes of mitochondrial respiration following strength and endurance training in normoxia and hypoxia in sedentary humans. *Am J Physiol Regul Integr Comp Physiol* 301: R1078-R1087.
- Gnaiger E (2009) Capacity of oxidative phosphorylation in human skeletal muscle. New perspectives of mitochondrial physiology. *Int J Biochem Cell Biol* 41: 1837-1845.
- Gnaiger E (2008) Polarographic oxygen sensors, the oxygraph and high-resolution respirometry to assess mitochondrial function. In: *Mitochondrial Dysfunction in Drug-Induced Toxicity* (Dykens JA, Will Y, eds) John Wiley: 327-352. – *A methodological introduction into high-resolution respirometry.*
- Hütter E, Renner K, Pfister G, Stöckl P, Jansen-Dürr P, Gnaiger E (2004) Senescence-associated changes in respiration and oxidative phosphorylation in primary human fibroblasts. *Biochem J* 380: 919-928.
- Gnaiger E (2001) Bioenergetics at low oxygen: dependence of respiration and phosphorylation on oxygen and adenosine diphosphate supply. *Respir Physiol* 128: 277-297. – *A detailed introduction into high-resolution respirometry with particular emphasis on kinetics and measurements at low oxygen.*
- Gnaiger E, Kuznetsov AV, Schneeberger S, Seiler R, Brandacher G, Steurer W, Margreiter R (2000) Mitochondria in the cold. In: *Life in the Cold* (Heldmaier G, Klingenspor M, eds) Springer, Heidelberg, Berlin, New York: 431-442. – *Isolated mitochondria and permeabilized muscle fibres, MiRO5.*

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www.orooboros.at/?MitoCom-Tyrol



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