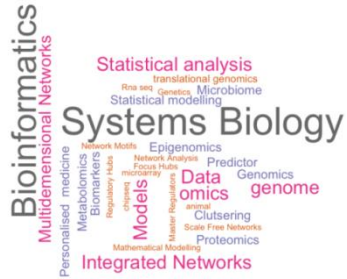


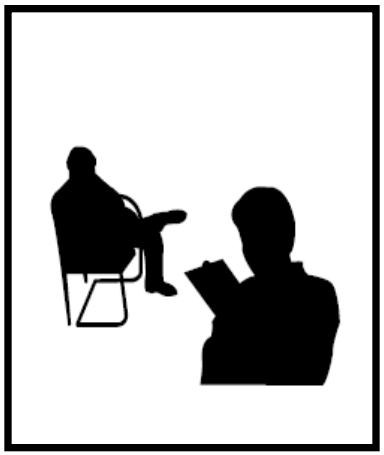
Mitochondria and brain disorders: A systems biology approach

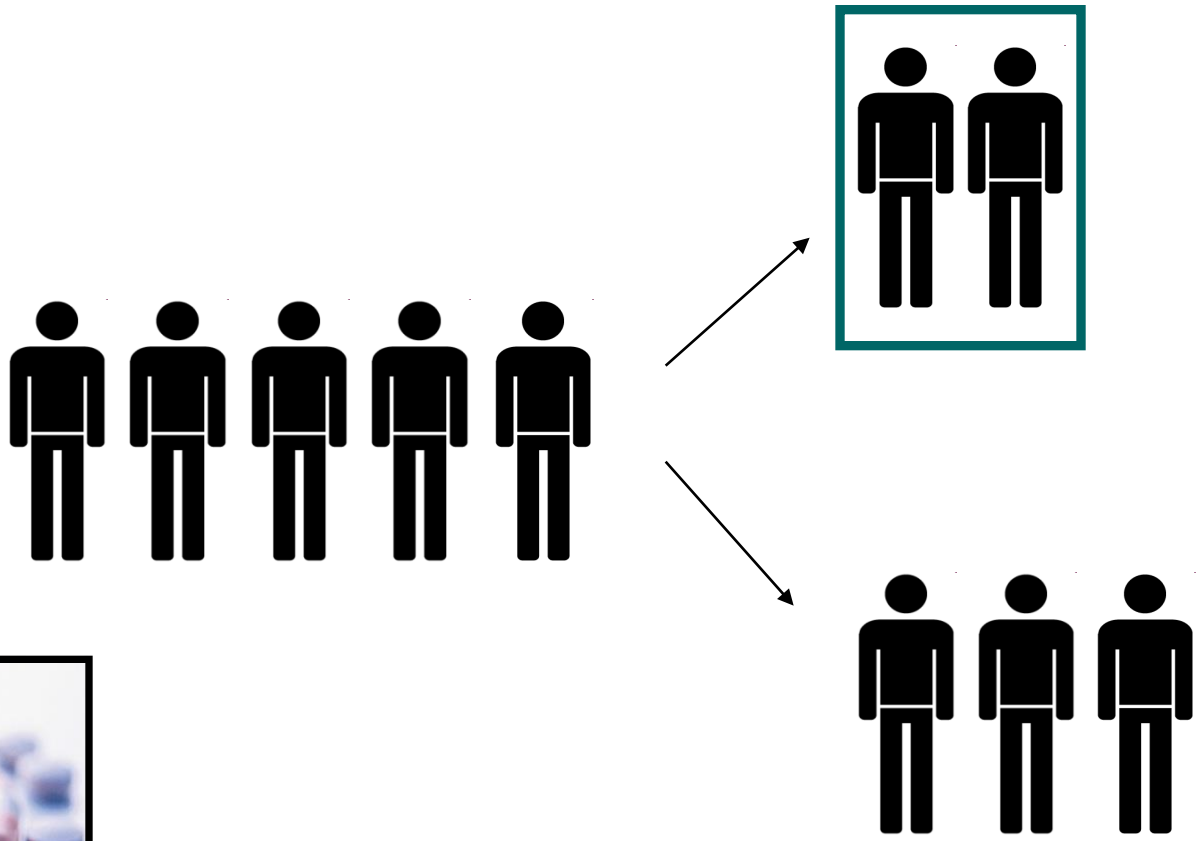
Michaela D. Filiou, PhD

Department of Stress Neurobiology and Neurogenetics

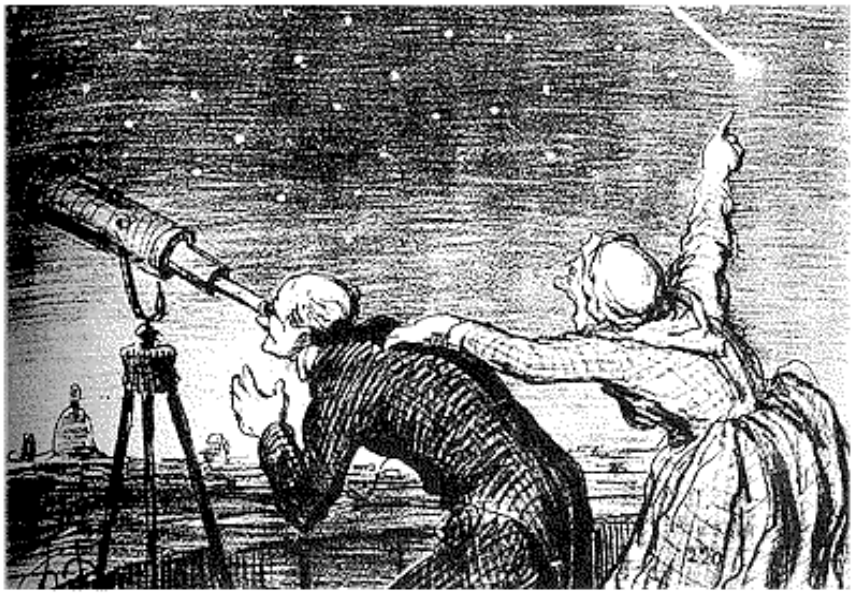
Max Planck Institute of Psychiatry, Munich

Problems, problems, problems





Why go data-driven?



Lithographie Honoré Daumier

New approaches to antidepressant drug discovery: beyond monoamines

Olivier Berton and Eric J. Nestler

Abstract | All available antidepressant medications are based on serendipitous discoveries of the clinical efficacy of two classes of antidepressants more than 50 years ago. These tricyclic and monoamine oxidase inhibitor antidepressants were subsequently found to promote serotonin or noradrenaline function in the brain. Newer agents are more specific but have the same core mechanisms of action in promoting these monoamine neurotransmitters. This is unfortunate, because only ~50% of individuals with depression show full remission in response to these mechanisms. This review summarizes the obstacles that have hindered the development of non-monoamine-based antidepressants, and provides a progress report on some of the most promising current strategies.

Filiou *Proteomics Clin Appl* 2015

Turck & Filiou *Mol Neuropsychia*try 2015

Outline

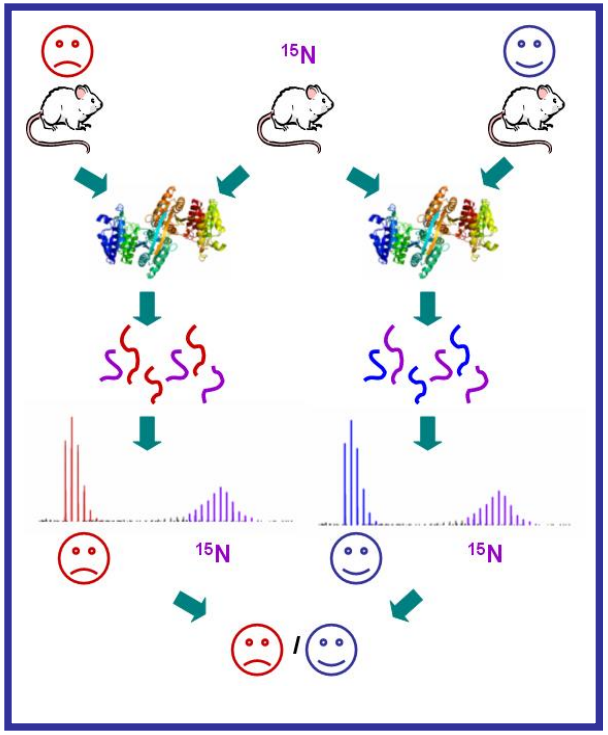
- A proteomics and metabolomics platform for biomarker discovery
- Implication of mitochondria in brain disorders
- Data-driven manipulation of mitochondrial function *in vivo*

A proteomics and metabolomics platform for biomarker discovery

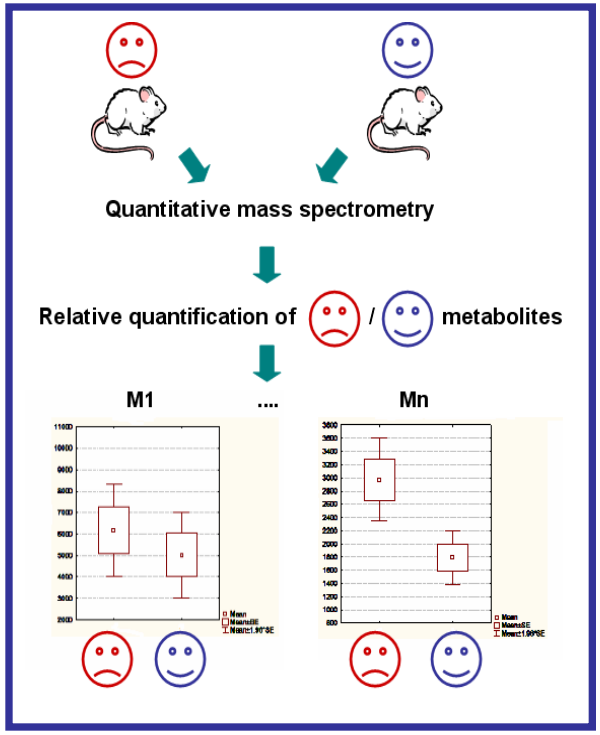
"Okay—who put my lunch through the mass spectrometer..?"

Mass spectrometry-based biomarker discovery

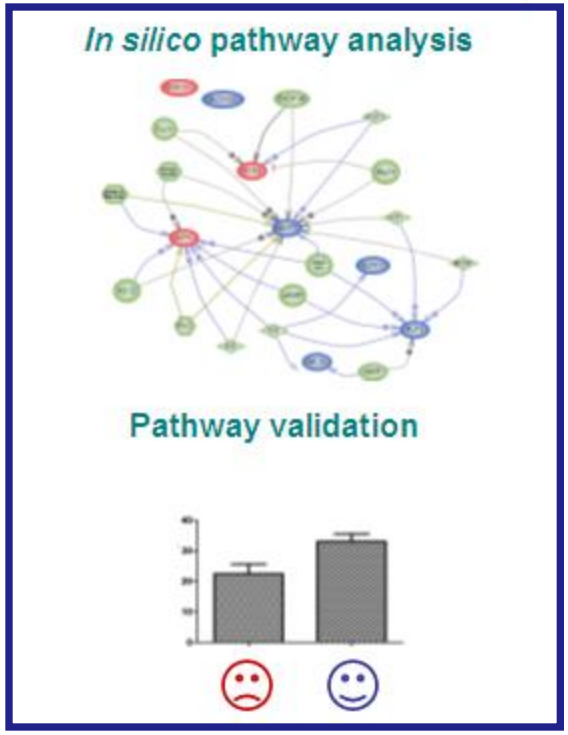
Proteomics



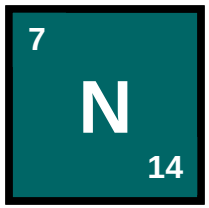
Metabolomics

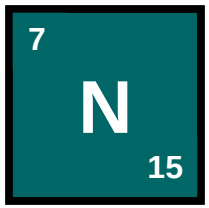


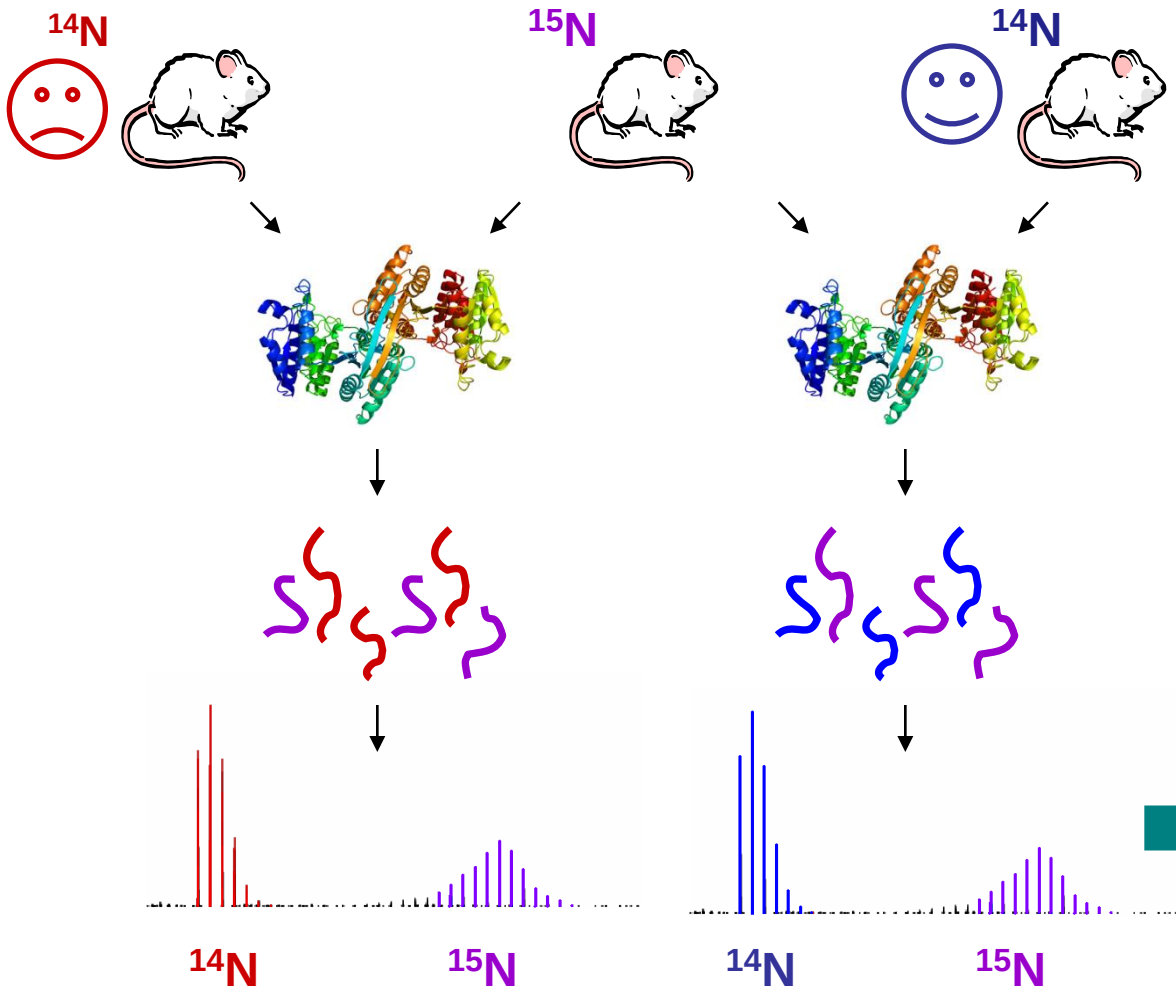
Bioinformatics



Quantitative proteomics





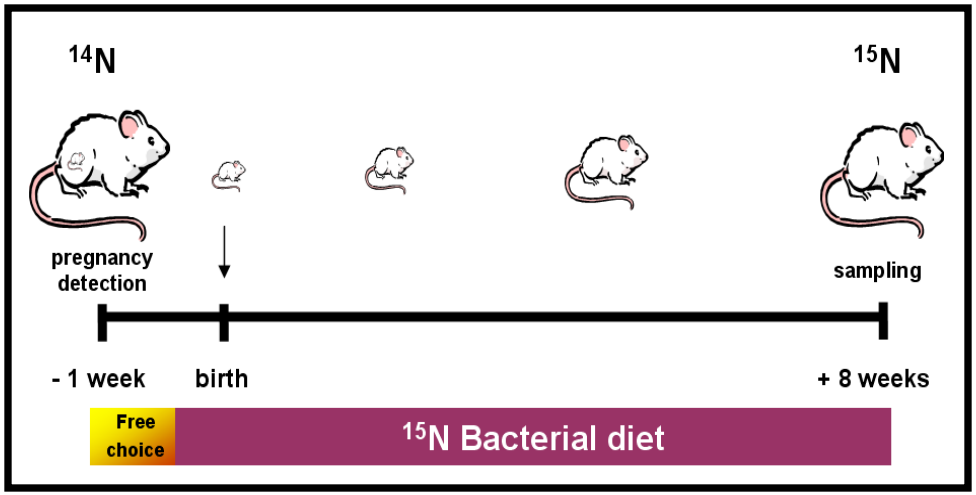


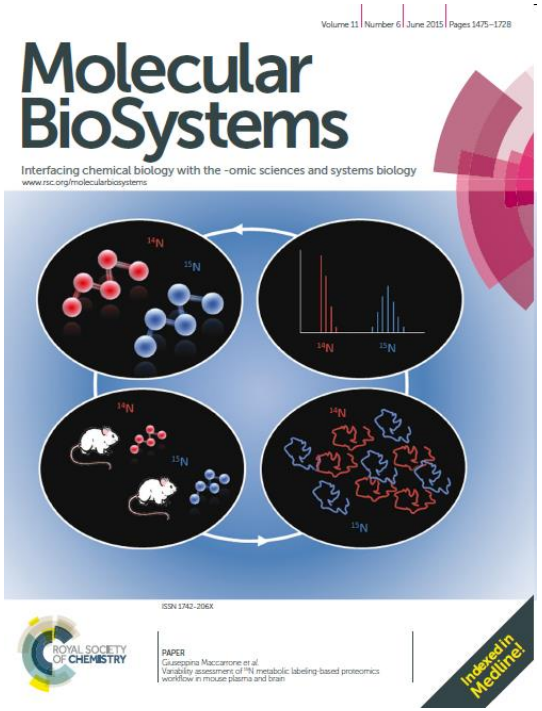
Filiou *Arch Clin Psychiatry* 2013

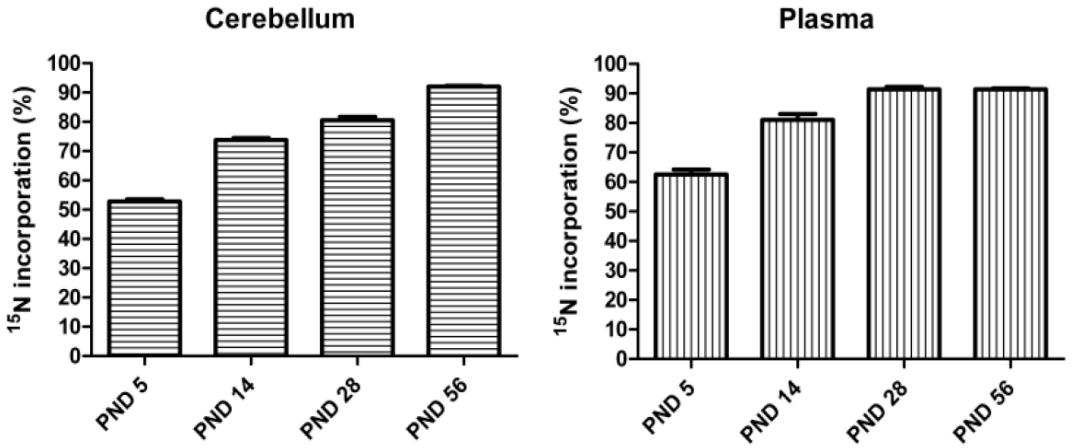
Filiou and Turck *Methods Mol Biol* 2012

In vivo ^{15}N metabolic labeling

^{15}N metabolic labeling protocol

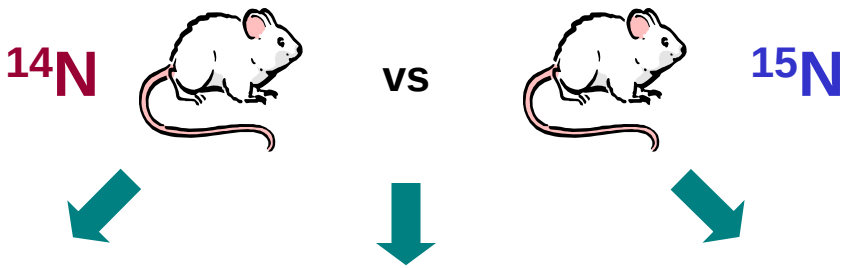




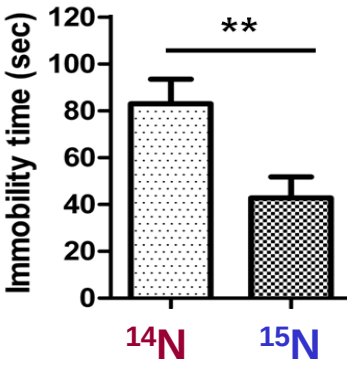


Filiou et al *Mol Biosyst* 2015
Zhang et al *Proteomics* 2009
Haegler et al *J Proteomics* 2009
Webhofer et al *J Proteomics* 2013
Filiou et al *Proteomics* 2012
Frank et al *PLoS ONE* 2009

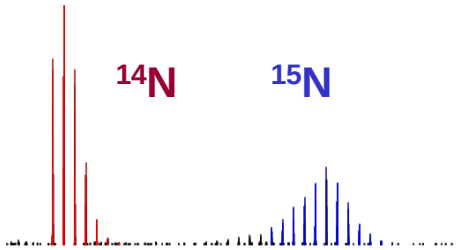
^{15}N isotope effect in mice



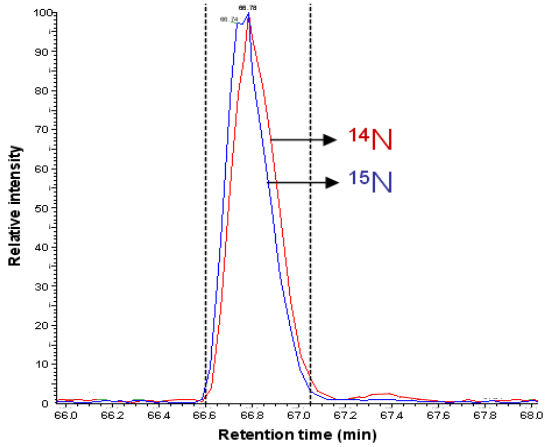
Behavioral phenotype changes



Altered protein expression in brain and plasma



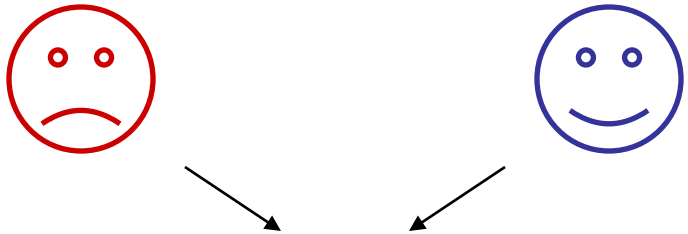
Retention time delay



Webhofer et al *J Proteomics* 2013

Filiou* Zhang* Gormanns* et al *Proteomics* 2012

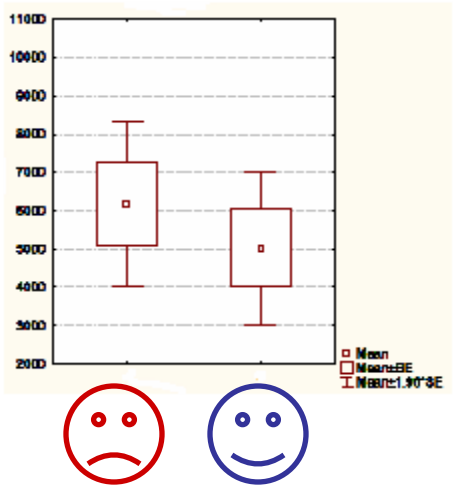
Targeted metabolomics

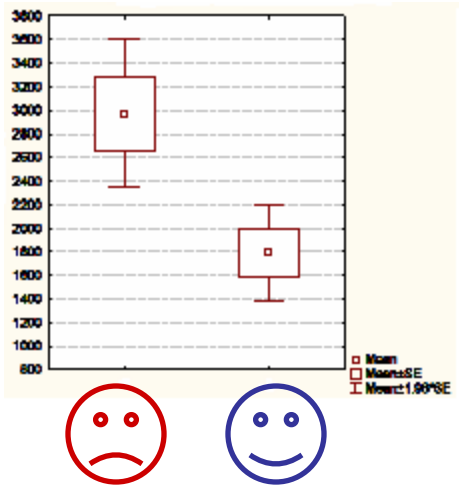


SRM-based

Relative quantification of 258 / metabolites

M1 M2 . . . M258



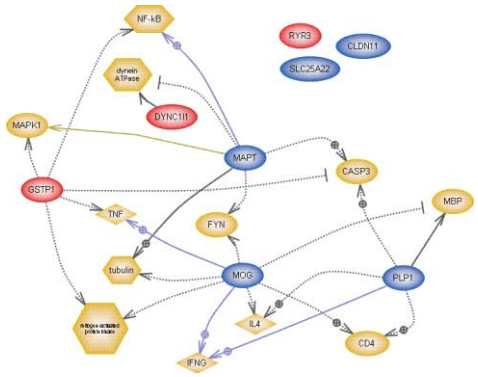


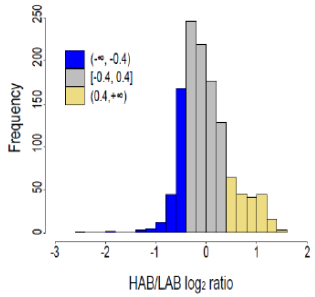
. . .

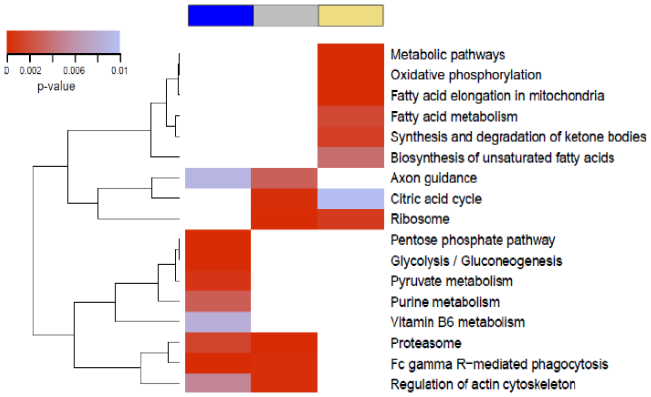
Yuan et al *Nat Protoc* 2012

Pathway analysis / Molecular and functional validation

In silico analysis

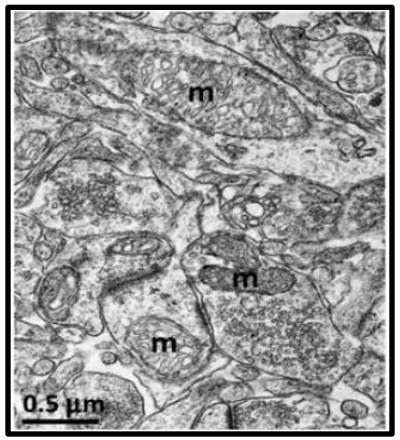


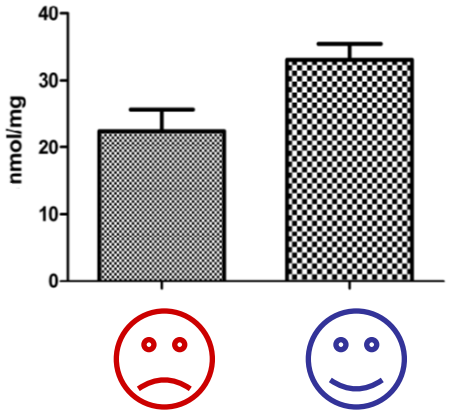




Metaboanalyst , Pathway Studio, DAVID, R

In vivo validation

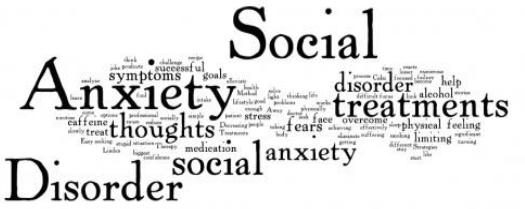


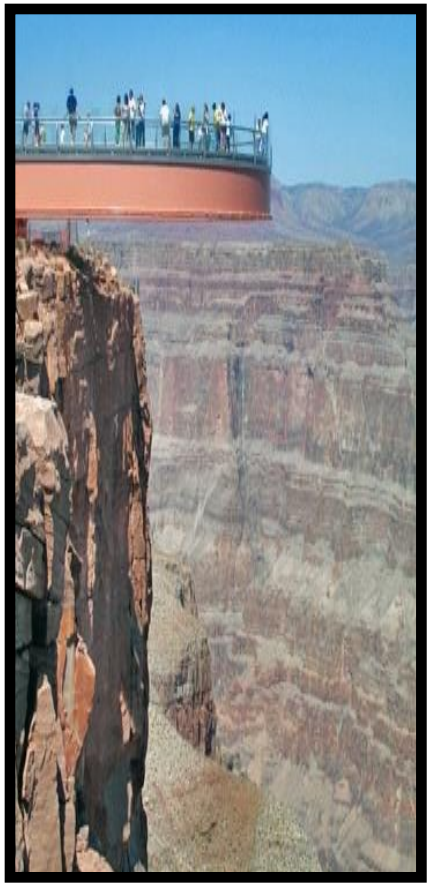


Iris et al *AJPN* 2014

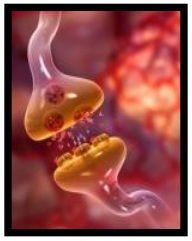
Implication of mitochondria in brain disorders

The HAB / LAB mouse model of trait anxiety

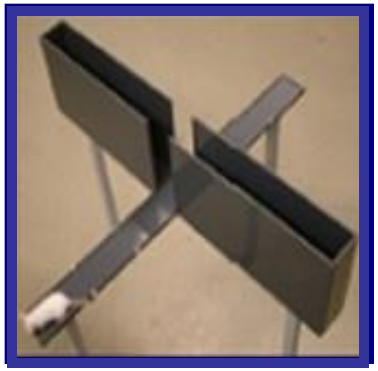




HAB



LAB

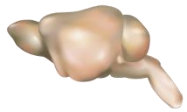


HAB: high anxiety-related behavior

LAB: low anxiety-related behavior

Quantification: 2678 proteins, 223 metabolites

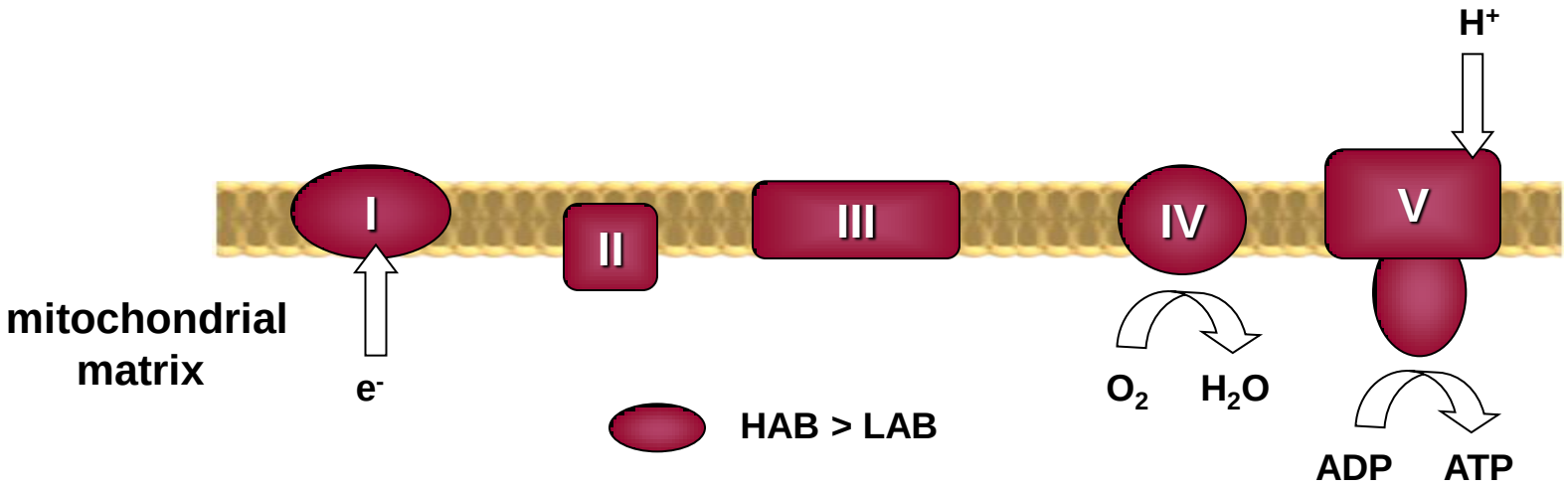
Altered levels: 273 proteins, 28 metabolites



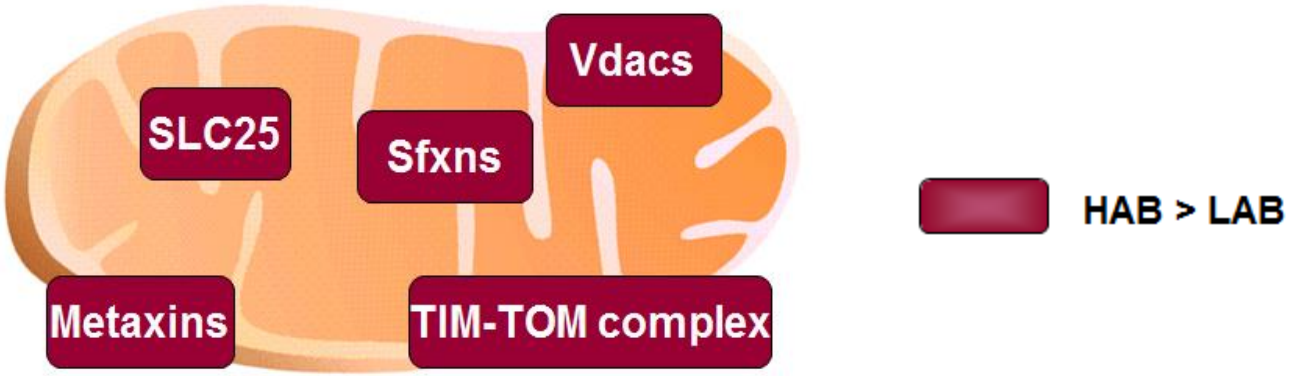
Krömer et al *J Neurosci* 2005

Protein changes in high anxiety

~ 60 oxidative phosphorylation proteins ↑ in high anxiety

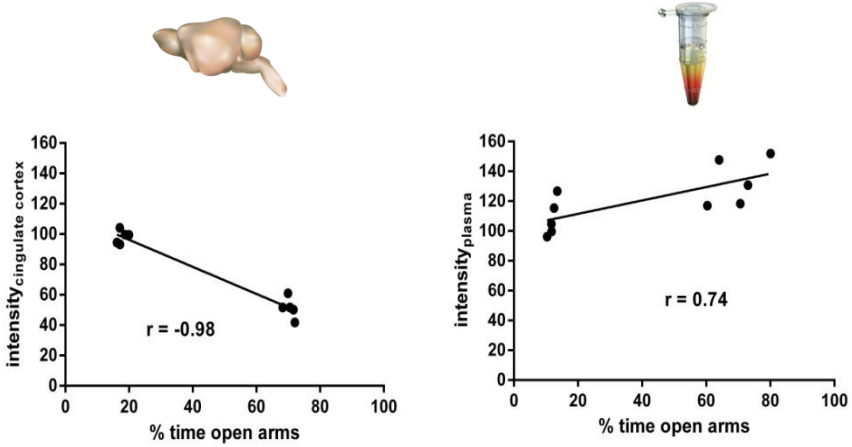


~30 mitochondrial import and transport proteins ↑ in high anxiety

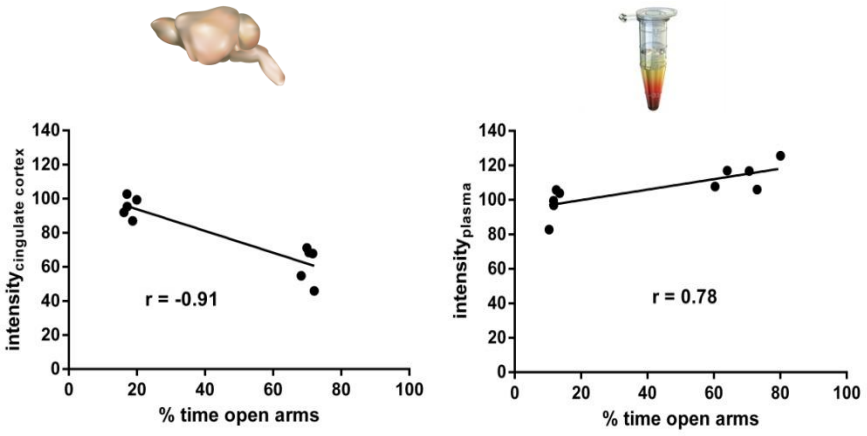


Filiou et al *Biol Psychiatry* 2011

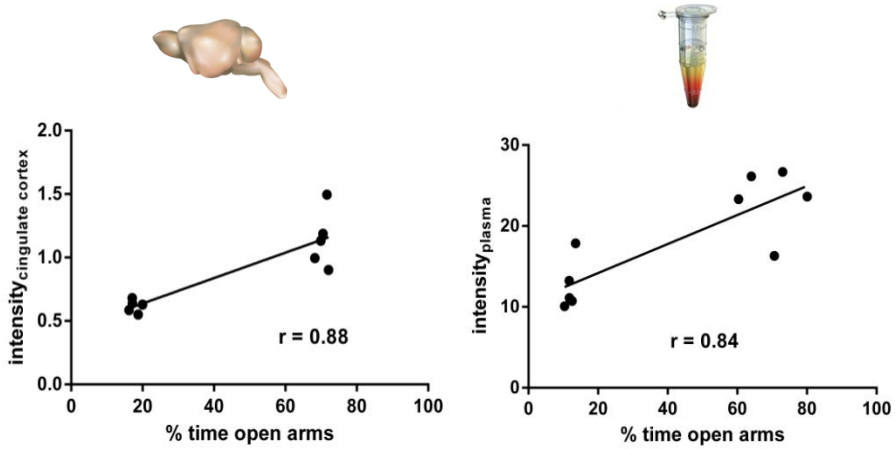
Correlation of metabolites with behavior



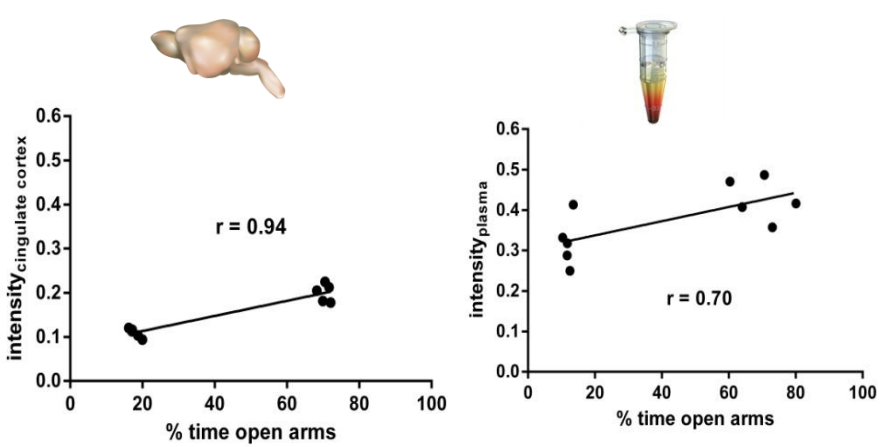
carnitine



acetylcarnitine



1-methyl-histidine



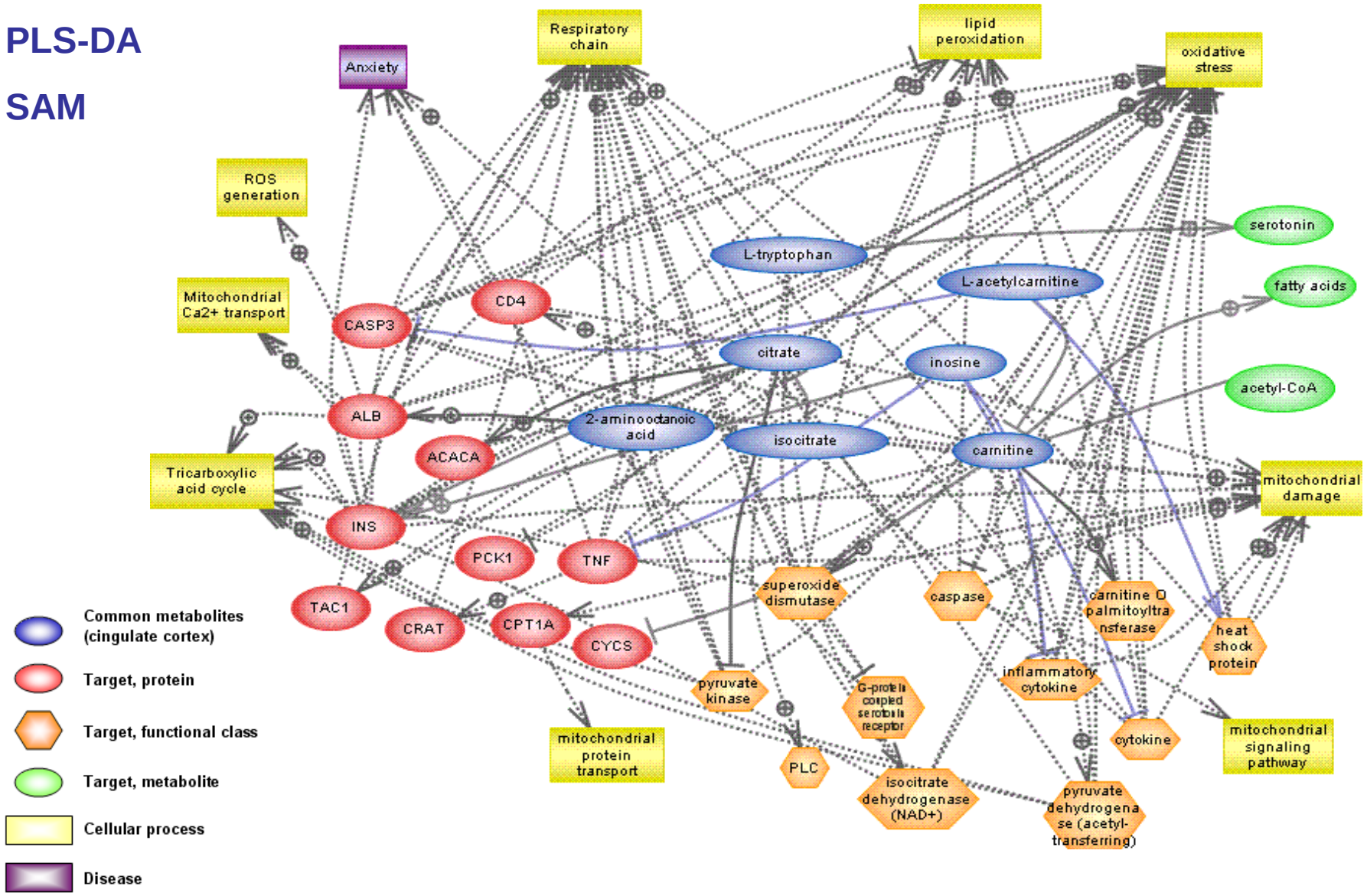
deoxyuridine

Filiou et al *J Psychiatr Res* 2014

What do *in silico* metabolomics analyses tell us?

PLS-DA

SAM

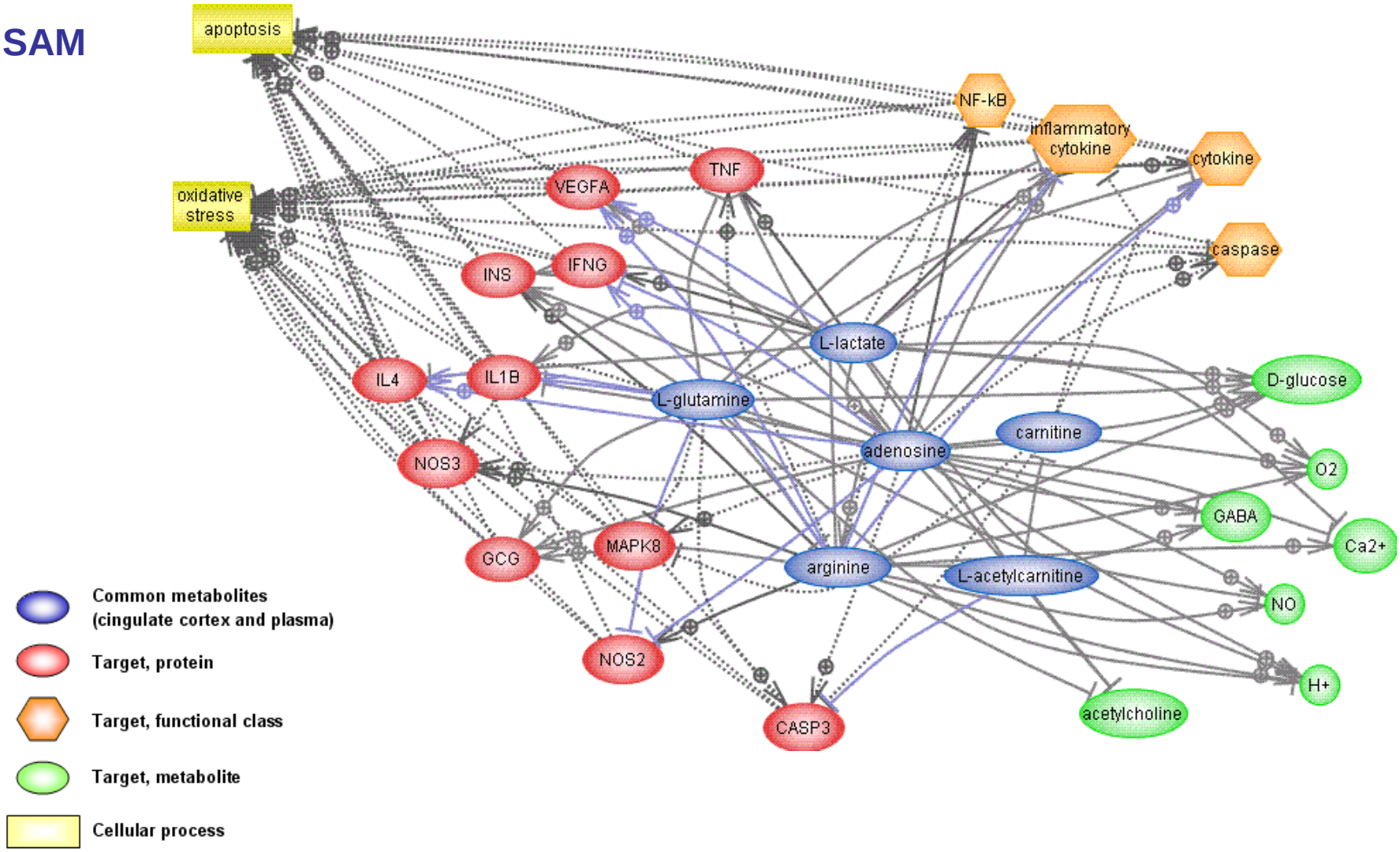


Filiou et al *J Psychiatr Res* 2014

What do *in silico* metabolomics analyses tell us?

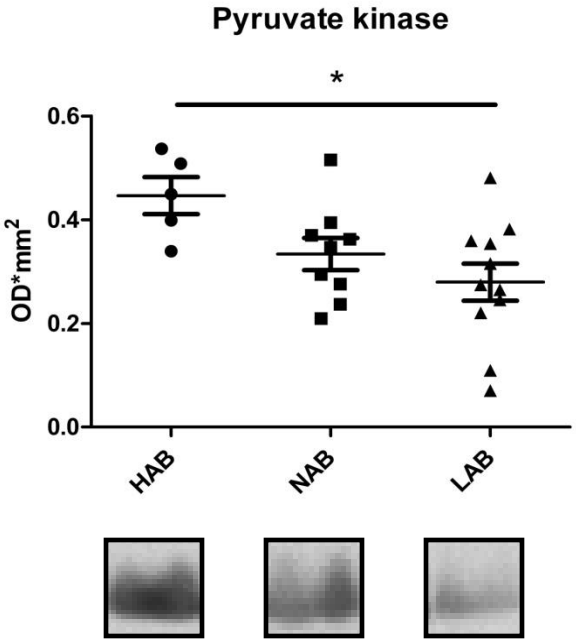
PLS-DA

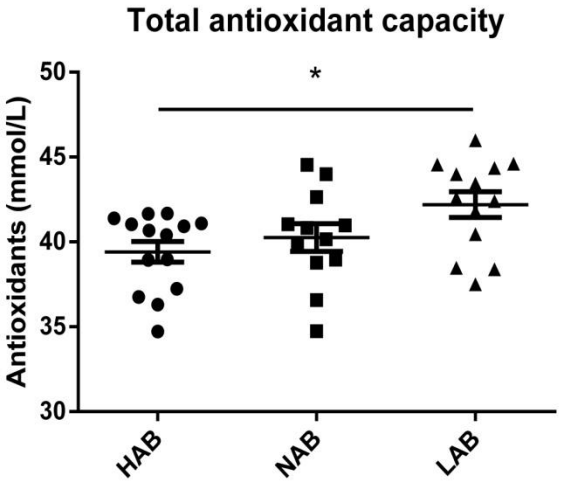
SAM

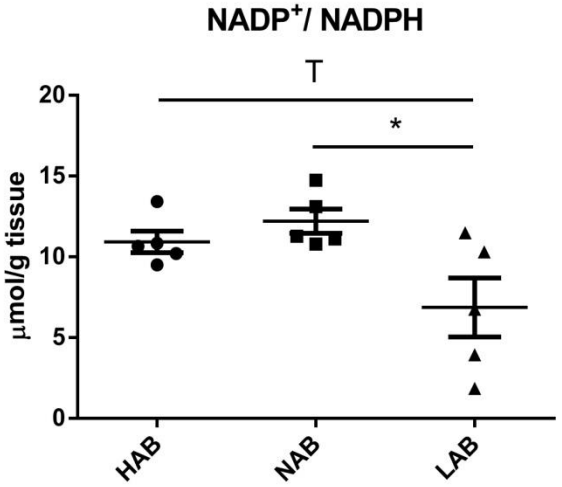


Filiou et al *J Psychiatr Res* 2014

Metabolomics-driven pathway validation

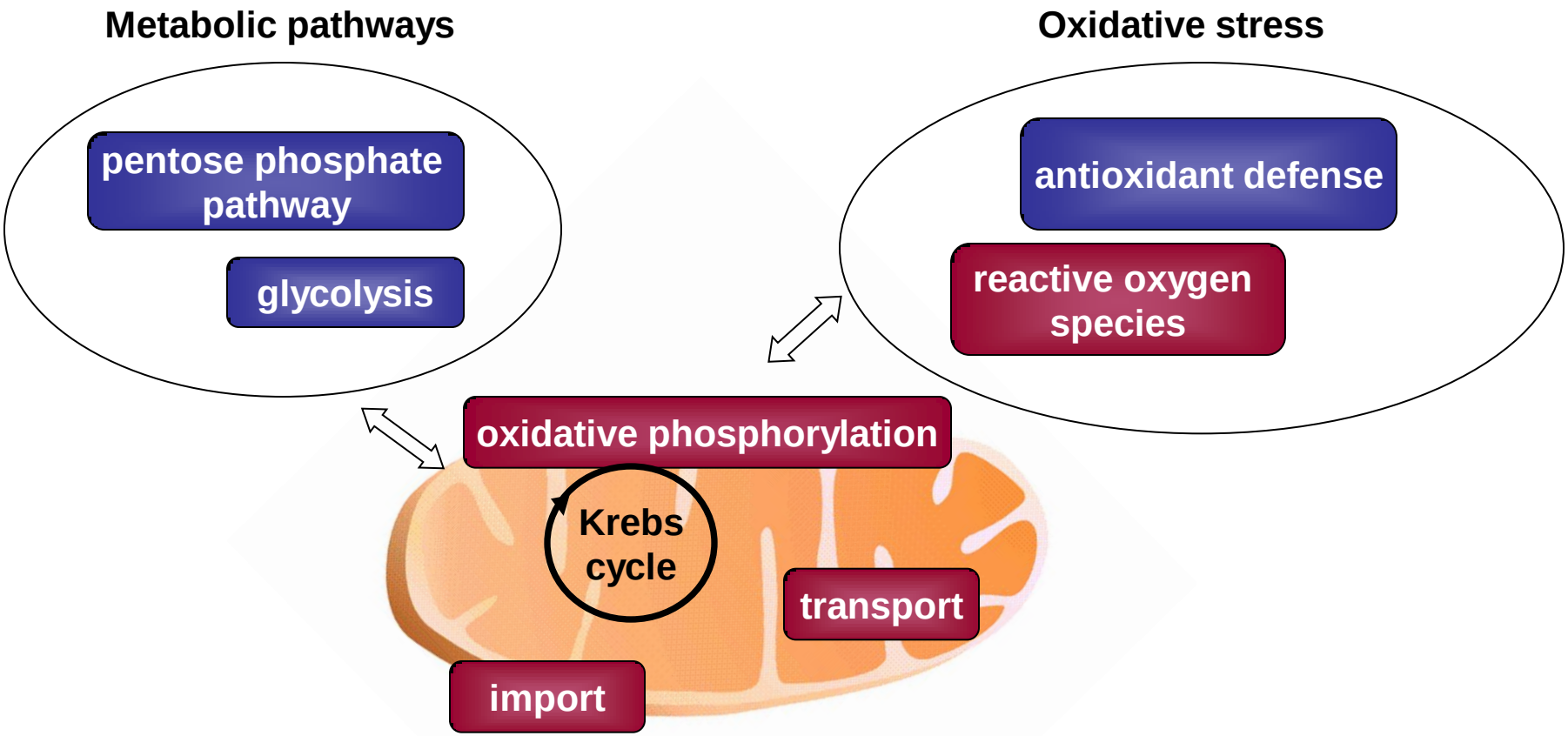






Filiou et al *J Psychiatr Res* 2014

Mitochondrial involvement in anxiety



HAB > LAB

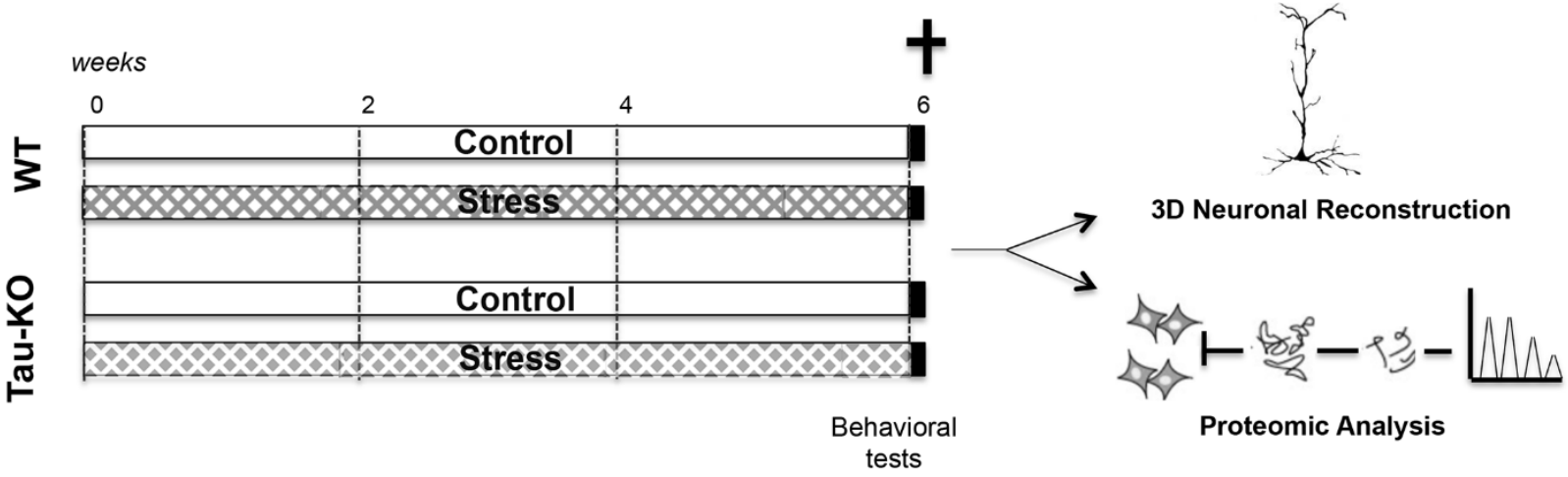
HAB < LAB

Filiou et al *Biol Psychiatry* 2011

Chronic stress and Tau

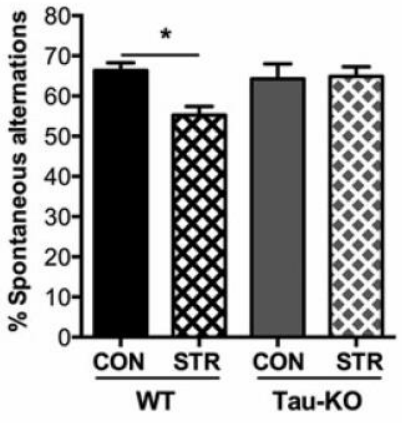
Stress

Tau-KO vs. WT



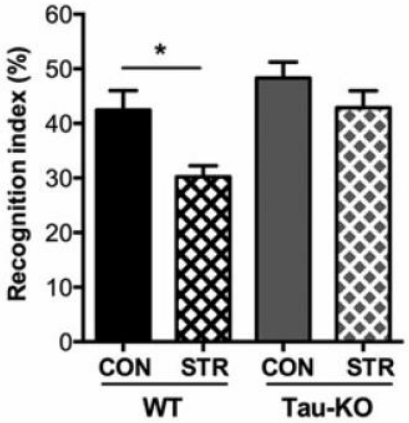
Tau deletion exerts neuroprotective effects

Tau deletion reverses stress-induced memory deficits



Y maze

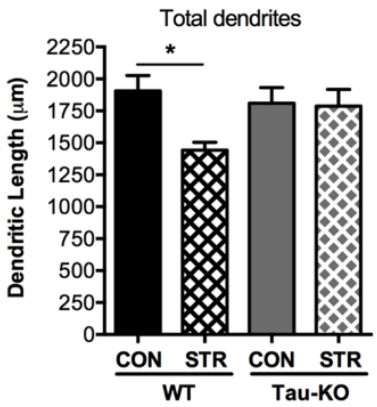
(working memory)

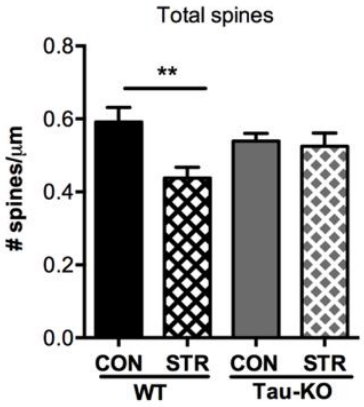


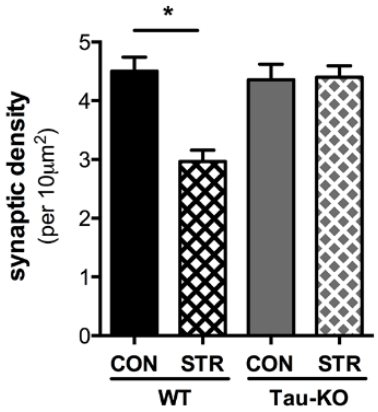
Novel place recognition

(recognition memory)

Tau deletion confers resilience to stress-induced neuronal atrophy and synaptic loss

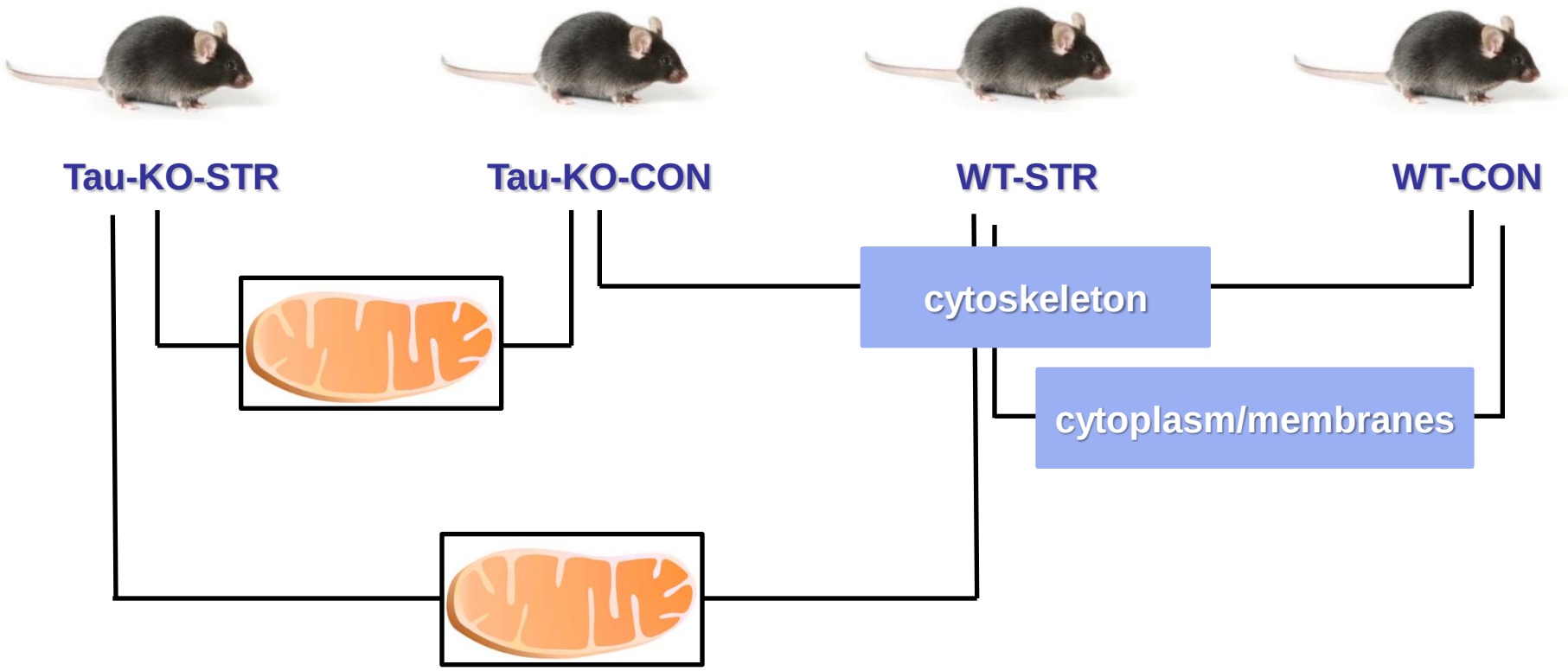






Lopes et al *Cereb Cortex* 2016

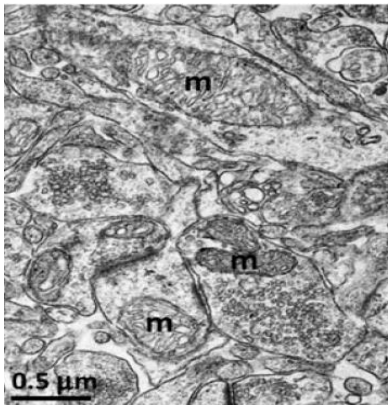
Mitochondria mediate stress effects in the absence of Tau

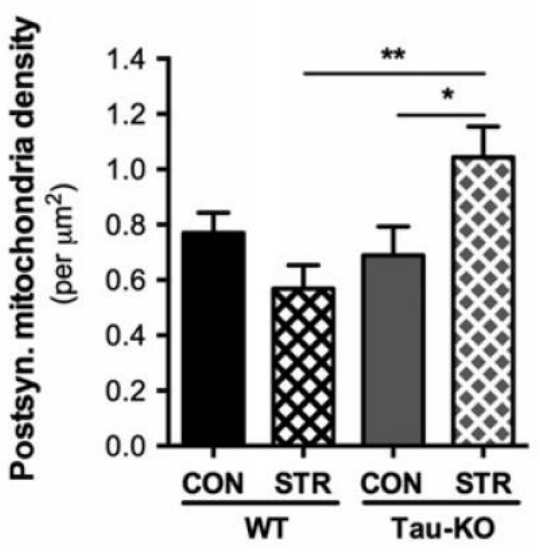


STR: stressed
CON: control

Lopes et al *Cereb Cortex* 2016

Altered mitochondrial density at the synapses





Cerebral Cortex, 2016, 1–12

doi: 10.1093/cercor/bhw057
Original Article

ORIGINAL ARTICLE

Tau Deletion Prevents Stress-Induced Dendritic Atrophy in Prefrontal Cortex: Role of Synaptic Mitochondria

Sofia Lopes[1,2], Larysa Teplytska[3], Joao Vaz-Silva[1,2], Chrysoula Dioli[1,2], Rita Trindade[1,2], Monica Morais[1,2], Christian Webhofer[3,4], Giuseppina Maccarrone[3], Osborne F.X. Almeida[3], Christoph W. Turck[3], Nuno Sousa[1,2], Ioannis Sotiropoulos[1,2] and Michaela D. Filiou[3]

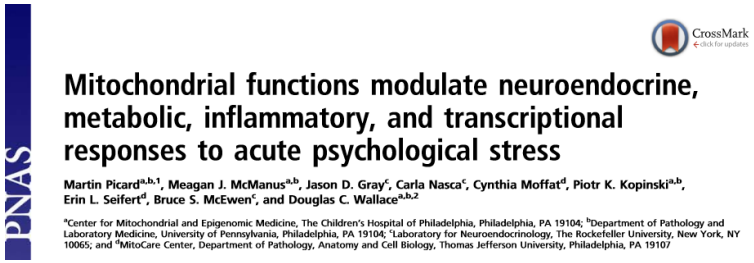
Lopes et al *Cereb Cortex* 2016

Take home message I

- Combination of proteomics and metabolomics can pinpoint affected/regulatory systems in an unbiased manner

- Follow up of data-driven approaches allows functional validation

- Mitochondria play a key role in the modulation of anxiety/stress-related pathologies



PNAS

Mitochondrial functions modulate neuroendocrine, metabolic, inflammatory, and transcriptional responses to acute psychological stress

Martin Picard[a,b,1], Meagan J. McManus[a,b], Jason D. Gray[c], Carla Nasca[c], Cynthia Moffat[d], Piotr K. Kopinski[a,b], Erin L. Seifert[d], Bruce S. McEwen[c], and Douglas C. Wallace[a,b,2]

[a]Center for Mitochondrial and Epigenomic Medicine, The Children's Hospital of Philadelphia, Philadelphia, PA 19104; [b]Department of Pathology and Laboratory Medicine, University of Pennsylvania, Philadelphia, PA 19104; [c]Laboratory for Neuroendocrinology, The Rockefeller University, New York, NY 10065; and [d]MitoCare Center, Department of Pathology, Anatomy and Cell Biology, Thomas Jefferson University, Philadelphia, PA 19107

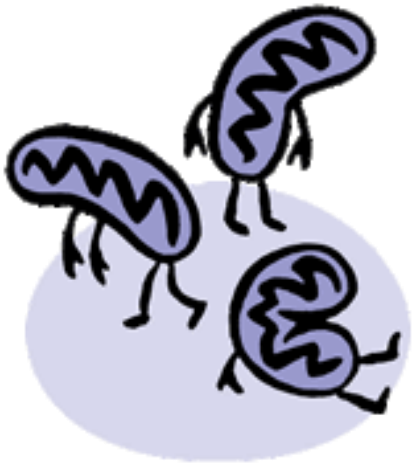
PNAS

Mitochondrial function in the brain links anxiety with social subordination

Fiona Hollis[a,1], Michael A. van der Kooij[a,1,2], Olivia Zanoletti[a], Laura Lozano[a], Carles Cantó[b], and Carmen Sandi[a,3]

[a]Brain Mind Institute, École Polytechnique Fédérale de Lausanne, CH-1015 Lausanne, Switzerland; and [b]Nestlé Institute of Health Sciences SA, CH-1015 Lausanne, Switzerland

Data-driven manipulation of mitochondrial function *in vivo*



Mitochondria as pharmacological targets

REVIEWS

British Journal of Pharmacology (2007) 151, 1154–1165

0007–1188/07 $30.00
www.brjpharmacol.org

Targeting mitochondria for cancer therapy

Simone Fulda, Lorenzo Galluzzi[‡§||] and Guido Kroemer[‡§||]*

REVIEW

Mitochondrial Medicine: Pharmacological targeting of mitochondria in disease

JS Armstrong

Department of Biochemistry, Faculty of Medicine, National University of Singapore, Singapore

ARTICLES

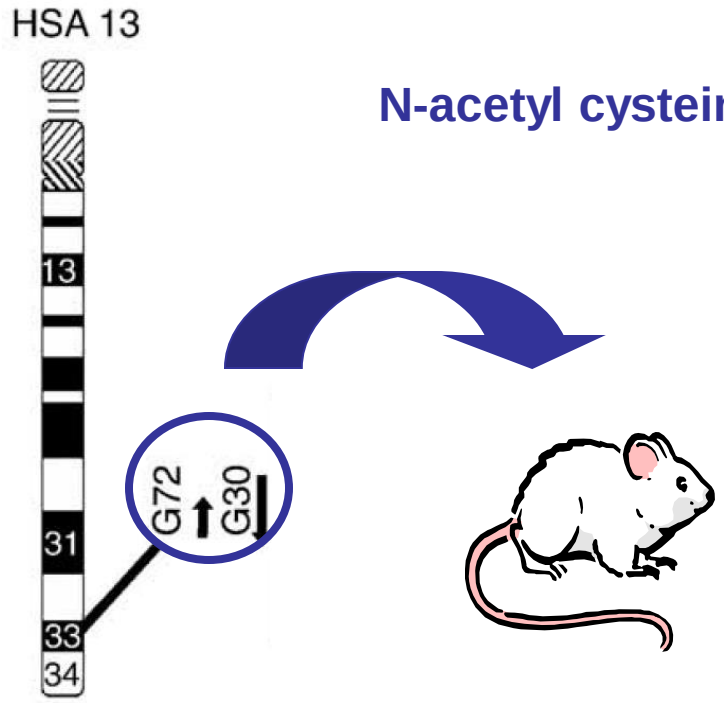
nature biotechnology

Nutrient-sensitized screening for drugs that shift energy metabolism from mitochondrial respiration to glycolysis

Vishal M Gohil[1–3,7], Sunil A Sheth[1–3,7], Roland Nilsson[1–3], Andrew P Wojtovich[4,5], Jeong Hyun Lee[6], Fabiana Perocchi[1–3], William Chen[1–3], Clary B Clish[2], Cenk Ayata[6], Paul S Brookes[4,5] & Vamsi K Mootha[1–3]

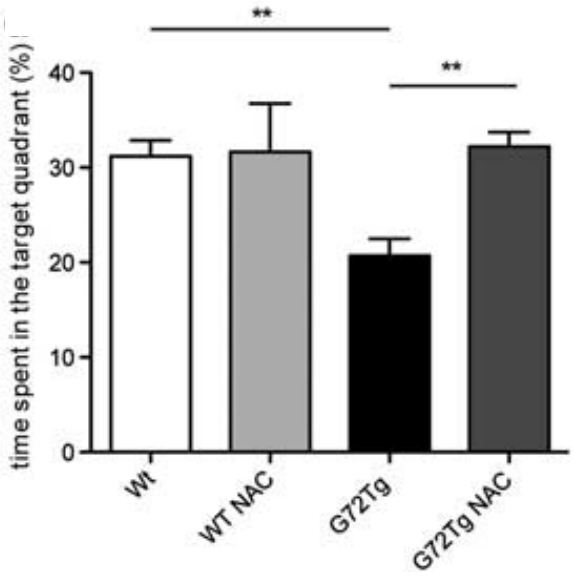
Oxidative stress as pharmacological target

imp
institut für molekulare psychiatrie



N-acetyl cysteine (NAC)

Morris water maze



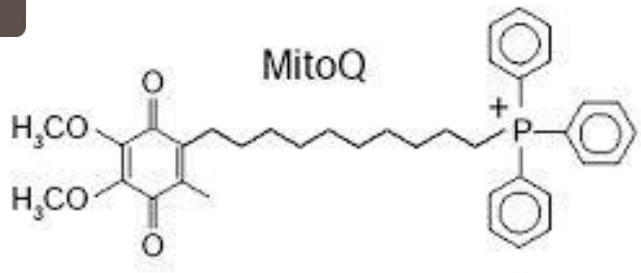
(cognitive function)

G72Tg: G72/G30 transgenic mice

Wood et al *Schizophrenia Res* 2014
Filiou et al *J Psychiatr Res* 2012
Otte et al *Neuropsychopharmacology* 2011
Otte et al *Eur Neuropsychopharmacol* 2009

Mitochondrial targeting in high anxiety

MRC Mitochondrial Biology Unit



glycolysis

reactive oxygen species

oxidative p[illegible]horylation

MitoQ: mitochondria-targeted antioxidant

quinone + triphenylphosphonium

HAB > LAB

HAB < LAB

Deutsche Forschungsgemeinschaft DFG

Experimental set up: *in vivo* MitoQ administration

HAB, 6 weeks (n=13 per group)

Habituation **10 week treatment** **Behavioral analysis**

DL OF

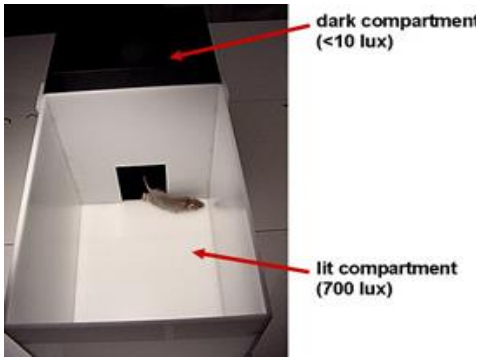
MitoQ **MitoQ in drinking water (400µM)**

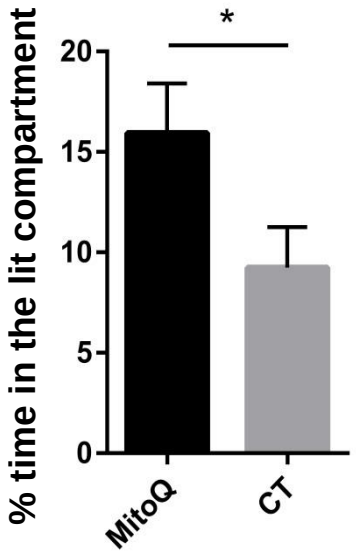
CT **drinking water**

DL	**Dark-light box**
OF	**Open field**

MitoQ administration exerts anxiolytic effects

Dark-light box





MitoQ **MitoQ treatment**
CT **Controls**

MW, p<0.05, n=13 per group

Neuropsychopharmacology (2015), 1–8

www.neuropsychopharmacology.org

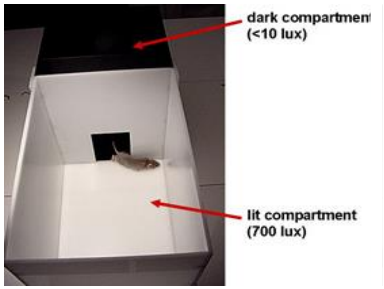
Selective Mitochondrial Targeting Exerts Anxiolytic Effects *In Vivo*

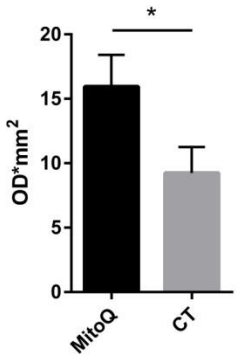
Markus Nussbaumer[1], John M Asara[2], Larysa Teplytska[1], Michael P Murphy[3], Angela Logan[3], Christoph W Turck[1] and Michaela D Filiou[*,1]

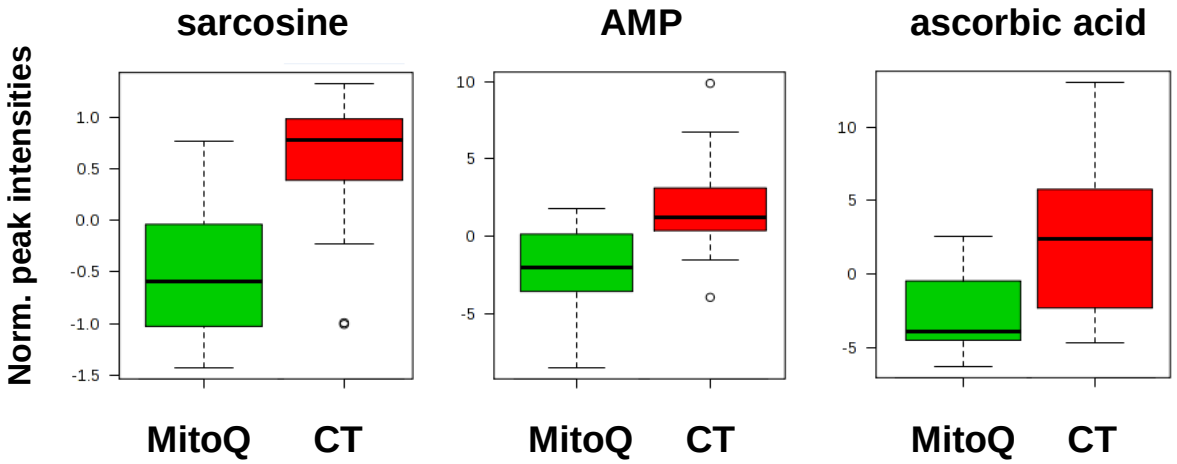
[1]Max Planck Institute of Psychiatry, Munich, Germany; [2]Division of Signal Transduction, Beth Israel Deaconess Medical Center, Department of Medicine, Harvard Medical School, Boston, MA, USA; [3]MRC-Mitochondrial Biology Unit, Cambridge, UK

Nussbaumer et al *Neuropsychopharmacology* 2015 Epub

MitoQ administration alters brain metabolite levels



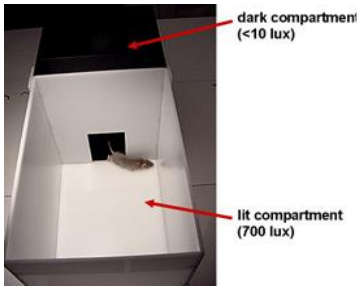


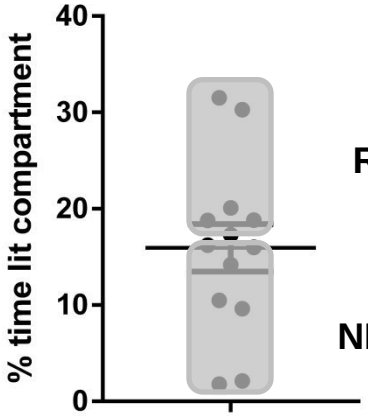


MitoQ	**MitoQ treatment**
CT	**Controls**

Nussbaumer et al *Neuropsychopharmacology* 2015 Epub

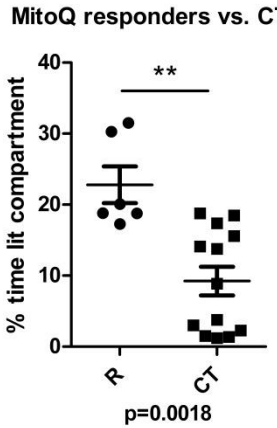
Response to MitoQ affects brain metabolite levels

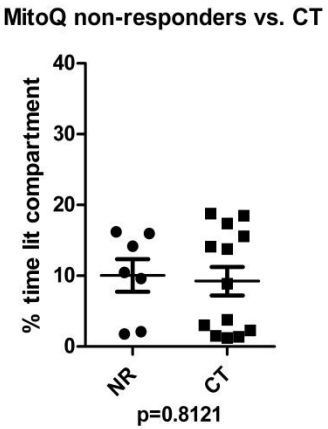


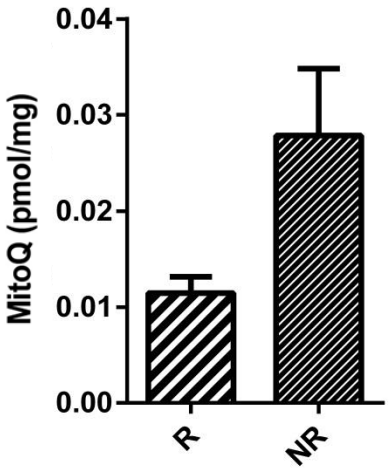


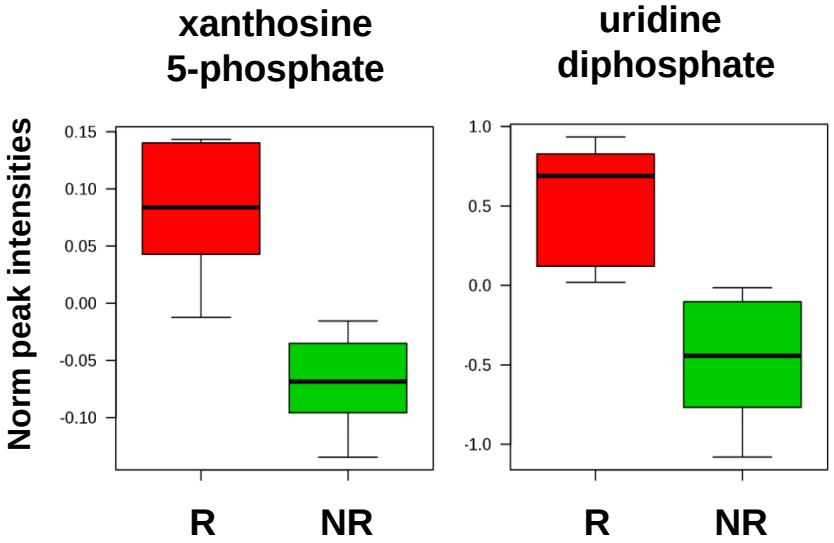
R : responders

NR: non responders









Is there a translational potential?

First *in vivo* application of selective mitochondrial targeting in psychiatric disorders

Movement Disorders
Vol. 25, No. 11, 2010, pp. 1670–1674

A Double-Blind, Placebo-Controlled Study to Assess the Mitochondria-Targeted Antioxidant MitoQ as a Disease-Modifying Therapy in Parkinson's Disease

Barry J. Snow, MD,[1*] Fiona L. Rolfe, BSc,[2] Michelle M. Lockhart, MM,[2] Christopher M. Frampton, PhD,[3] John D. O'Sullivan, MD,[4] Victor Fung, PhD, FRACP,[5] Robin A.J. Smith, PhD,[6] Michael P. Murphy, PhD,[7] Kenneth M. Taylor, PhD,[2] and On behalf of the Protect Study Group

Liver INTERNATIONAL
OFFICIAL JOURNAL OF THE INTERNATIONAL ASSOCIATION FOR THE STUDY OF THE LIVER

Liver International ISSN 1478-3223

CLINICAL STUDIES

The mitochondria-targeted anti-oxidant mitoquinone decreases liver damage in a phase II study of hepatitis C patients

Edward J. Gane[1], Frank Weilert[2], David W. Orr[1], Geraldine F. Keogh[3], Michael Gibson[3], Michelle M. Lockhart[3], Christopher M. Frampton[4], Kenneth M. Taylor[3], Robin A. J. Smith[5] and Michael P. Murphy[6]

Antipodean PHARMACEUTICALS

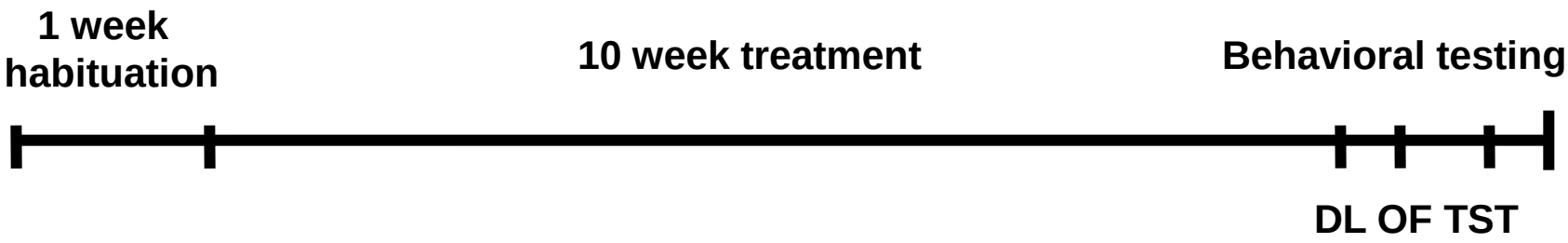
Perspectives

National and Kapodistrian
UNIVERSITY OF ATHENS

HT	Hydroxytyrosol
DL	Dark-light box
OF	Open field
Ole	Oleuropein
TST	Tail suspension test

Bl6, 3 weeks old, group housed

DBA, 3 weeks old, group housed



Bl6 Ole	Drinking water with Ole (150mg/L)	N = 15
Bl6 CT	Drinking water only	N = 15
DBA HT	Drinking water with HT (150mg/L)	N = 15
DBA CT	Drinking water only	N = 15

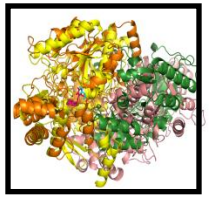
Take home message II

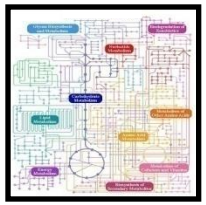
- Hypothesis-free discovery of altered pathways and their pharmacological manipulation have translational potential
- Pharmacological targeting of mitochondrial pathways *in vivo* exerts anxiolytic effects
- Potential of mitochondria-targeted compounds for therapeutic interventions

Overview

Identification of molecular pathways

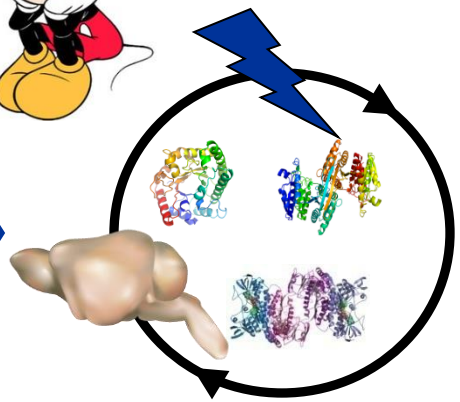
Animal models



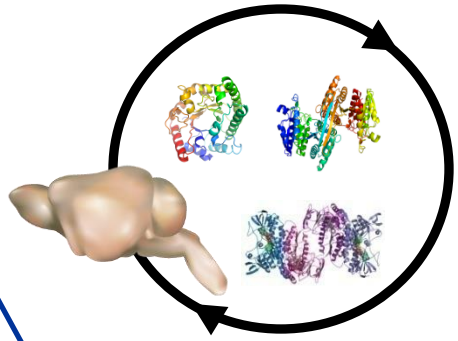


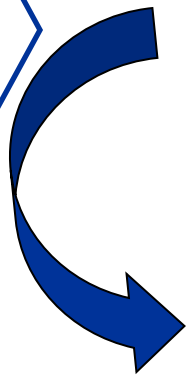
Systemic approaches

Manipualtion of molecular pathways



Reversal of molecular pathology



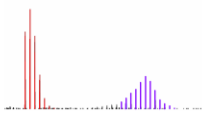


Thanks ☺ Questions?

Department Stress Neurobiology and Neurogenetics

Alon Chen
Carsten Wotjak
Markus Nussbaumer
Osborne Almeida
Mareen Engel
Evan Paul

Proteomics and Biomarkers



Chris Turck
Larysa Teplytska
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Proteomics Core Facility

Giuseppina Maccarrone
Christiane Rewerts

Harvard Medical School

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MRC Mitochondrial Biology Unit

Mike Murphy

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mfiliou@psych.mpg.de

Deutsche Forschungsgemeinschaft DFG

EMBO excellence in life sciences

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Call for papers

Journal of Chromatography B

Special Issue:
Advances in Mass Spectrometry-based Applications

Submission deadline: 31 May 2016

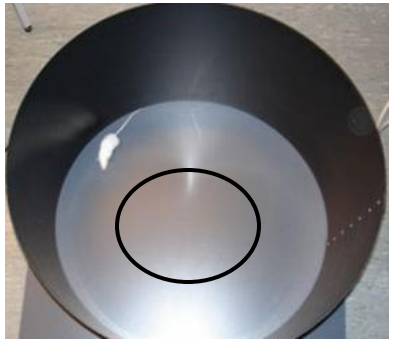
Guest editors:

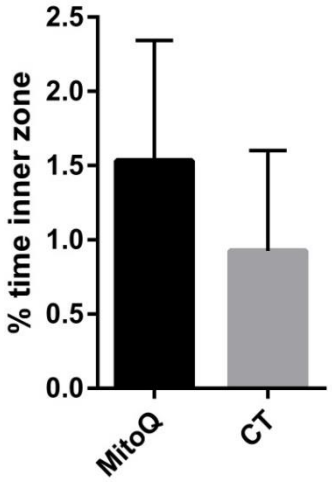
Evangelos Gikas vgikas@pharm.uoa.gr

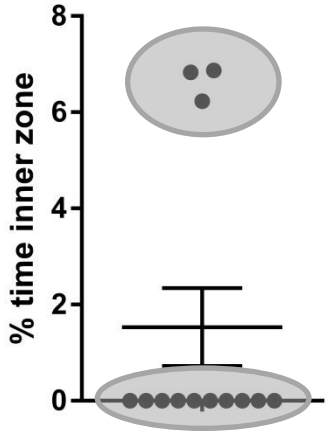
Michaela Filiou mfiliou@psych.mpg.de

MitoQ response has a distinct plasma metabolite signature

Open field



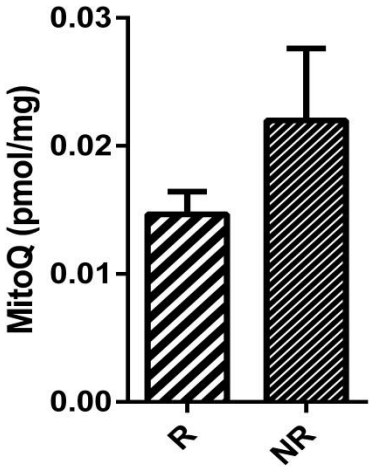




R : responders

NR : non-responders

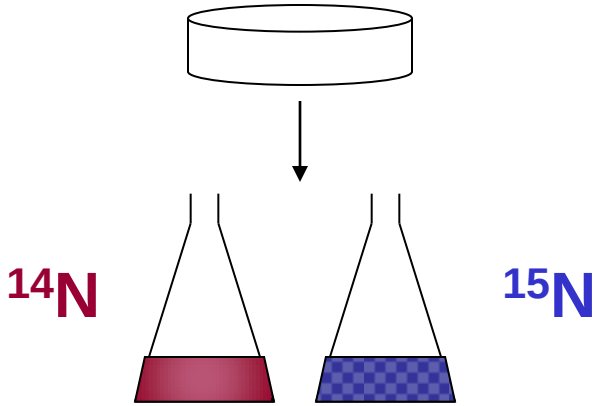
MitoQ levels



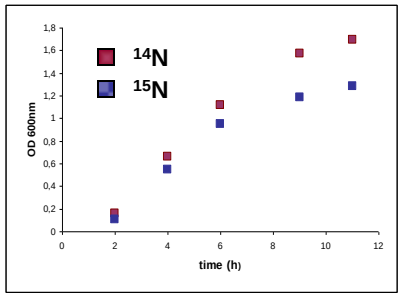
Metabolite	d.value	stdev	p-value	KEGG ID
Higher levels in MitoQ OF responders				
2-keto-isovalerate	3.51	1.19	0.005	C00141
alanine	3.59	2.65	0.005	C00041
dCMP	3.54	0.04	0.005	C00239
fumarate	4.09	1.12	0.003	C00122
maleic acid	3.57	1.20	0.005	C01384
myo-inositol	4.80	1.35	0.001	C00137
proline	3.61	3.45	0.005	C00148
Lower levels in MitoQ OF responders				
fructose-6-phosphate	-5.04	0.10	0.001	C00085
glutathione disulfide	-3.65	1.41	0.005	C00127
hexose-phosphate	-3.76	0.49	0.004	C05345
methylmalonic acid	-3.44	3.33	0.006	C02170

^{15}N isotope effect in bacteria

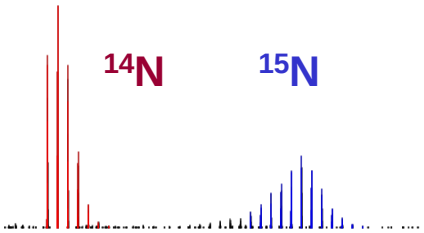
***E. coli* colonies**



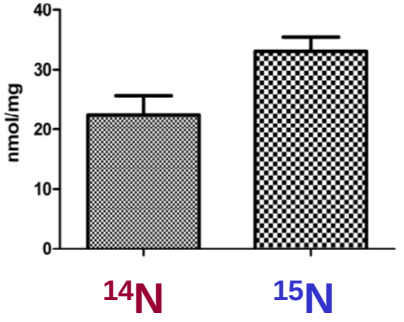
Altered growth rates



Altered protein expression

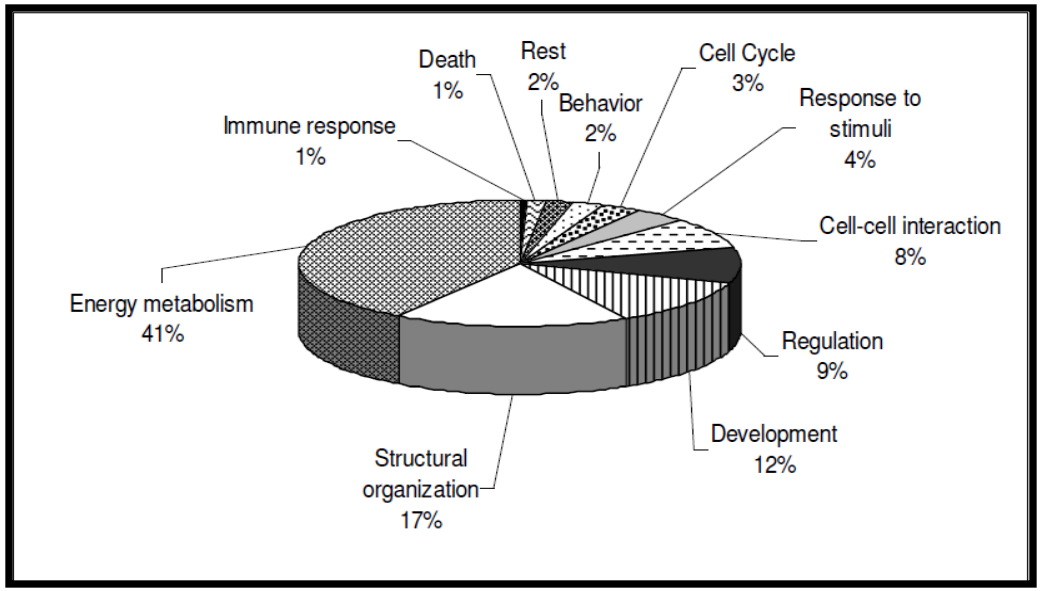


Altered metabolite levels



Filiou et al *Proteomics* 2012

Studying synapses: Synaptosomes



2980 proteins

only neuronal cells

4% vesicles

8% membranes

24% mitochondria

64% cytoplasm

Maccarrone & Filiou *Methods Mol Biol* 2015

Filiou et al *Electrophoresis* 2010