WG4: Blood Cells and Cell lines

MITOEAGLE data repositories (from WG2-4)

1. Deposit post-hoc datasets related to articles already published on topics of WG 2-4 by Consortium members on locations accessible for the Consortium.
2. Connect published articles to deposited datasets, using generally accessible tools such as PubMed Commons (http://www.ncbi.nlm.nih.gov/pubmedcommons/).
3. Along with development of SOPs, implement advance public deposition of protocols (Begley, Ioannidis 2015).
4. Deposit datasets with submitted manuscripts and finally connect these using tools such as PubMed Commons.
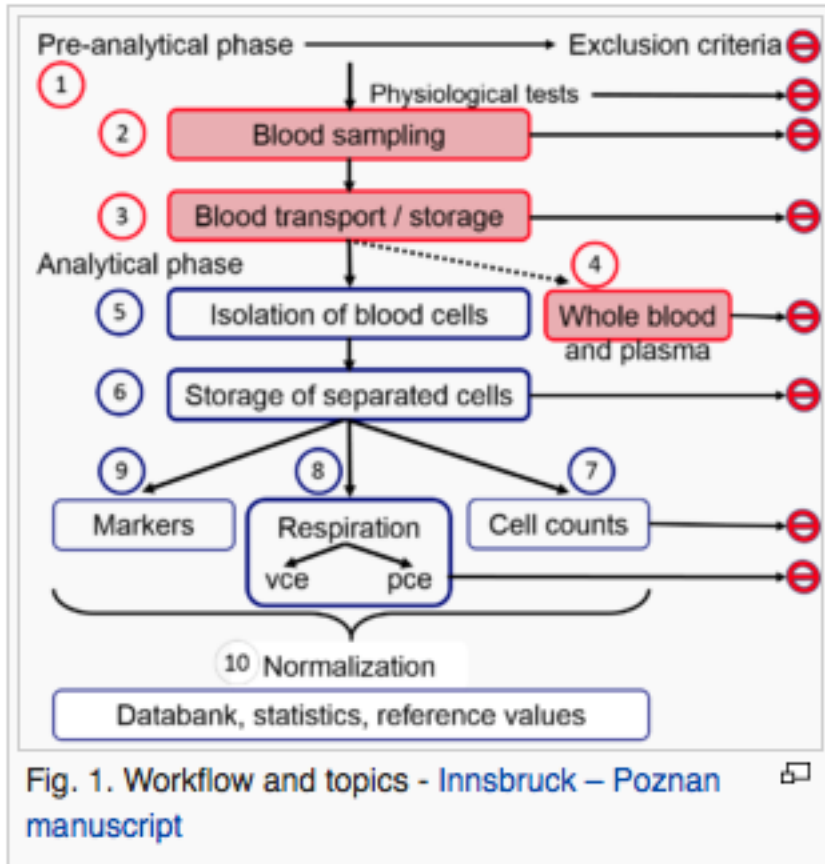
Milestones

1. Consensus on protocols, reporting schemes and work assignments.
2. Completed SOPs for cell preparation & laboratory protocols.
3. Application study finished and data transmitted to MITOEAGLE data repository.
4. Publication finished.

Deliverables

1. SOPs for blood cell separation and respirometric characterisation open for the research community.

2. MITOEAGLE data repository for comparative data evaluation, planning of future studies, data mining.

3. Publication with a set of reference data.

1) Consensus protocols for blood cells
 (Verona Innsbruck Poznan Lund)

Fig. 1. Workflow and topics - Innsbruck – Poznan manuscript

MITOEAGLE meetings in WG4 Blood cells
Poznan and Lund 2018
Maitrei 2019

1) Consensus protocols for blood cells
(Verona Innsbruck Poznan Lund)

SOP/Protocols for PMBC and Platelets (isolation, cryopreservation)

Templates for PBMC and PLT data base

» MitoEAGLE template PLT final.xlsx

» MitoEAGLE template PBMC final.xlsx

2) Data repository for comparative data evaluation, planning of future studies, data mining – cell lines

WG4:

Task 1
Please provide details of experience, current work/interest, data that can be made available to the group (published/unpublished)

Name	cell type experience preparation protocols/ mitochondrial function	current interest	Own published data & protocols (ref)	un-published data & protocols that can be made available(attach as pdf)
Nicoleta Moisoi	Models of Parkinson's Human fibroblasts, drosophila, mouse (respiration, ROS, ATP, Mito potential)	Blood cells from fibroblasts PD patients and controls and dopaminergic neurons derived from stem cells; genetic and pharmacologic cellular models of PD.	yes	yes
Maria Monsalve	Cell Type experience: Cell lines: C2C12, NIH/3T3, COS7, 293T, 293A, FAO Primary cells: HUVEC,	Humans: Mitochondrial biomarkers for personalized medicine. T2D, cancer (thyroid). Preclinical: Role of PGC-1a in tumor development. Alterations in oxidative	yes	yes

Task

Cell lines (control and disease condition):
- Human fibroblasts, Mouse fibroblasts
- SH-SY5Y
- HeLa, HEK293

Quantitative data
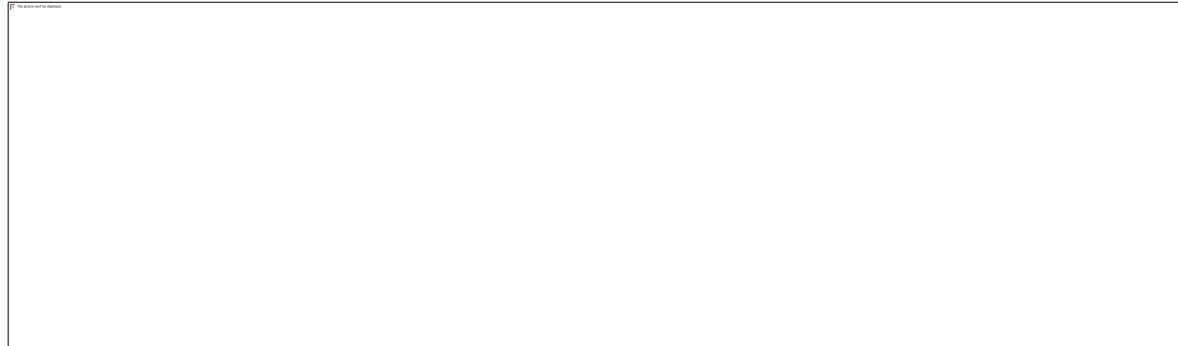
Respiration	ATP	ROS	MitoPotential
Higher than control (1), Lower than control (-1), No Change (0)			

Data collection

Parkinson's cellular models

- Human dermal fibroblasts: control, PINK1, LRKK2, VPS35
- SH-SY5Y (overexpression and downregulation)
- Cybrids (human platelets and SH-SY5Y or NT2 Rho zero)
- Mouse fibroblasts (control and transgenics)
- N2A

Parkinson's murine models

- Transgenic mice (PINK1KO, HtrA2 KO, Parkin KO)
- Toxin models (6OHDA, MPTP)

Alzheimer's cellular models

- SH-SY5Y (overexpression APPmut)
- HEK293
- CHO
- PC12

Alzheimer's murine models

- Transgenic mice: 3xTg, Thy1-APP

Quantitative data

				Information Quantitative					
Parameter 1	Parameter 2	Parameter 3	Parameter 4	Parameter 5	Parameter 6	Parameter 7	Parameter 8	Parameter 9	Parameter 10
background	Complex I	Complex II	Complex I+II	ETS (E) - uncoupled LEAK		OXPHOS	ROX		
no substrates	*driven*	*driven*	*driven*						

	MITOCHONDRIAL FUNCTION - OTHER PARAMETERS				
ROS-DCF(cytoplasm)	ROS - Mitosox Mitochondria	Mito potential	ATP production	Morphology	

Problem:
different measurements units, different parameters
and lack of original data
to be able to get the data into the same format

Qualitative data: scoring system

Parameter 1 Background	Respiration	ATP	ROS	MitoPotential
	Higher than control (1) Lower than control (-1) Not changes (0)			

Qualitative data
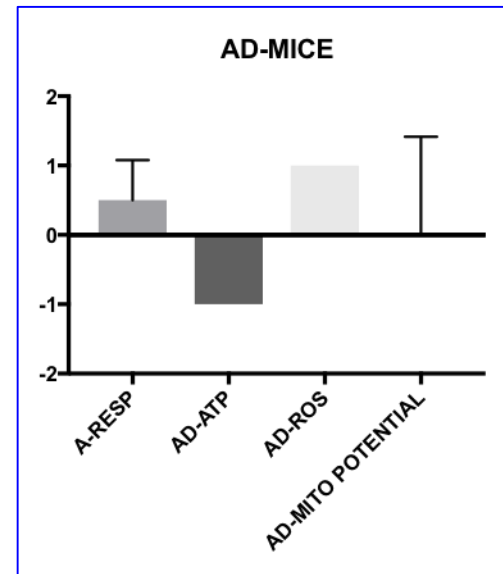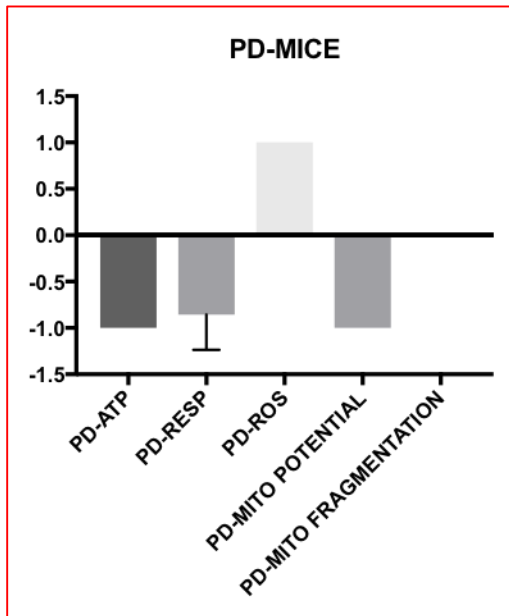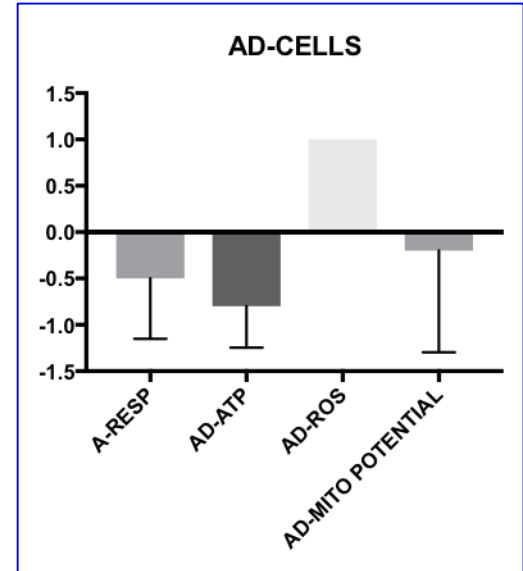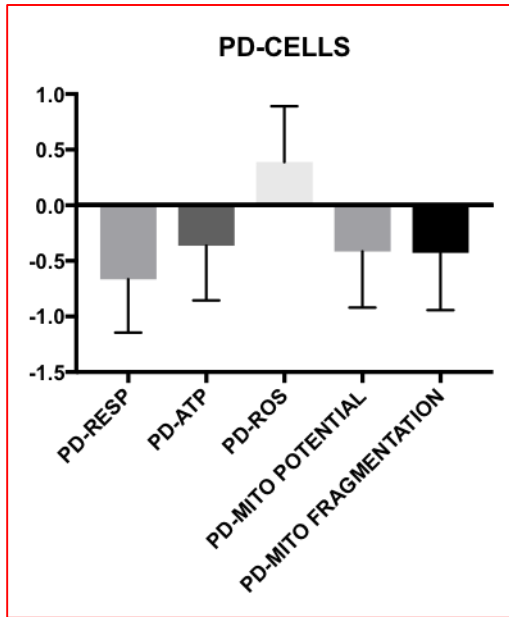
Future

•Blood cells: finish and publish the work

•Cell lines: Still to collect and compile data in cell lines