Cincinnati Children's
Heart Institute

Impaired fatty-acid metabolism in tafazzin-deficient mice

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Overview

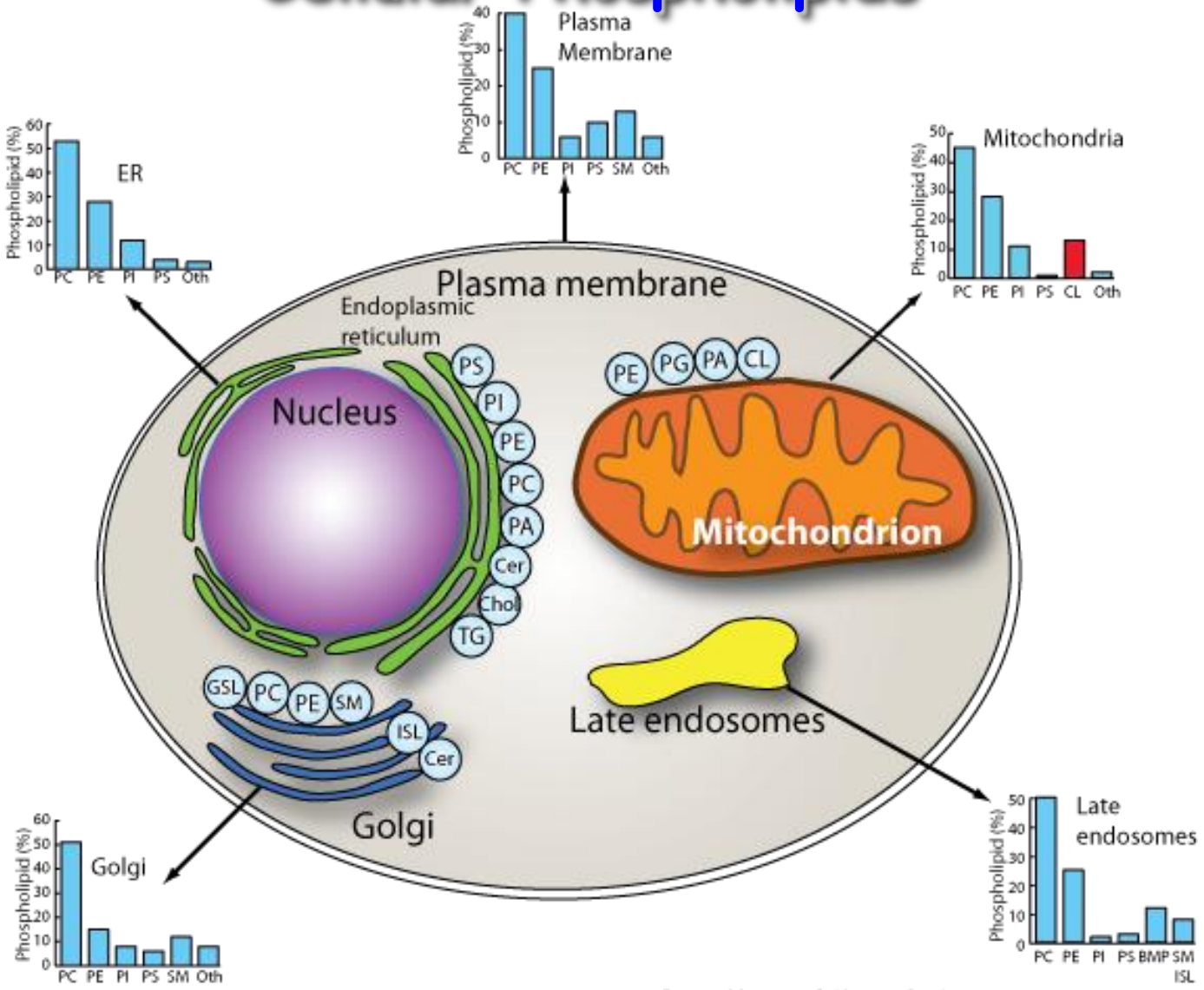
- Background: phospholipids, cardiolipin, Barth syndrome, mouse model, mitochondrial defects in sarcomeric tissue.

- Indirect calorimetry: Energy expenditure, Oxygen consumption, Respiratory Exchange Ratio (RER) in basal and stressed conditions. Cold exposure and forced exercise on treadmill.

- Mitochondrial respiration in taz-deficient neonatal cardiomyocytes. Mitochondrial proteomics.

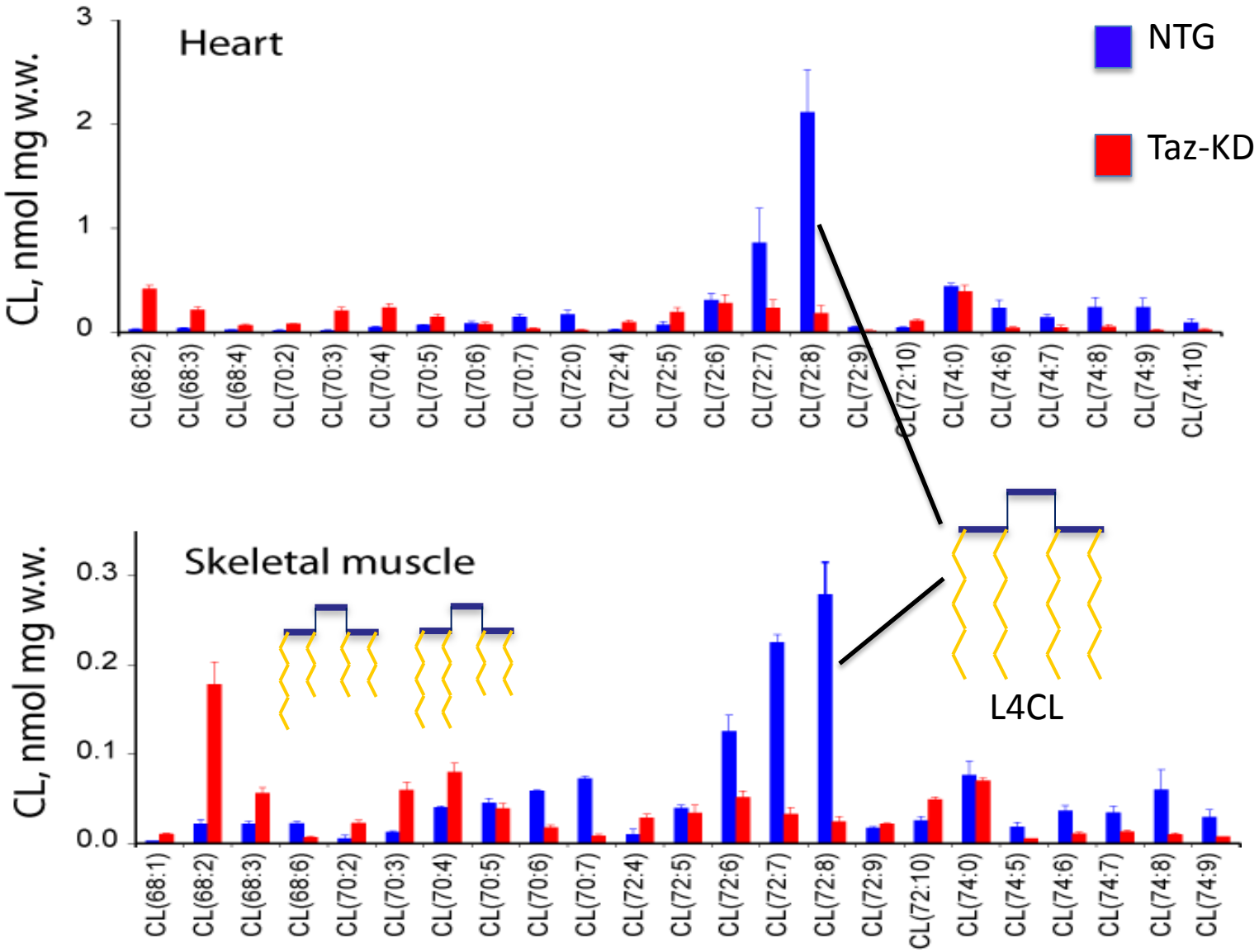
- Cardiolipin in physical interaction of fatty acid oxidation enzymes with mitochondrial complexes. 2D-Native electrophoresis.

Cellular Phospholipids

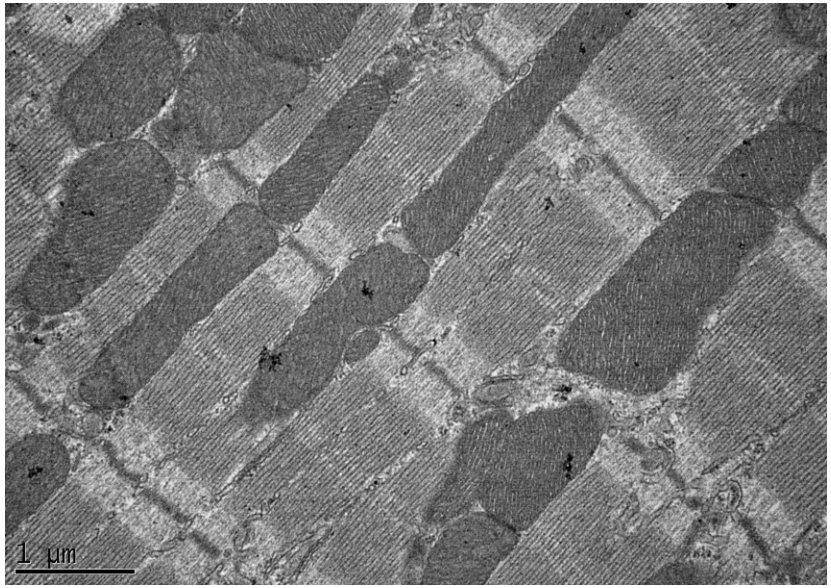


G. von Meer et al. Nature Reviews, 2008

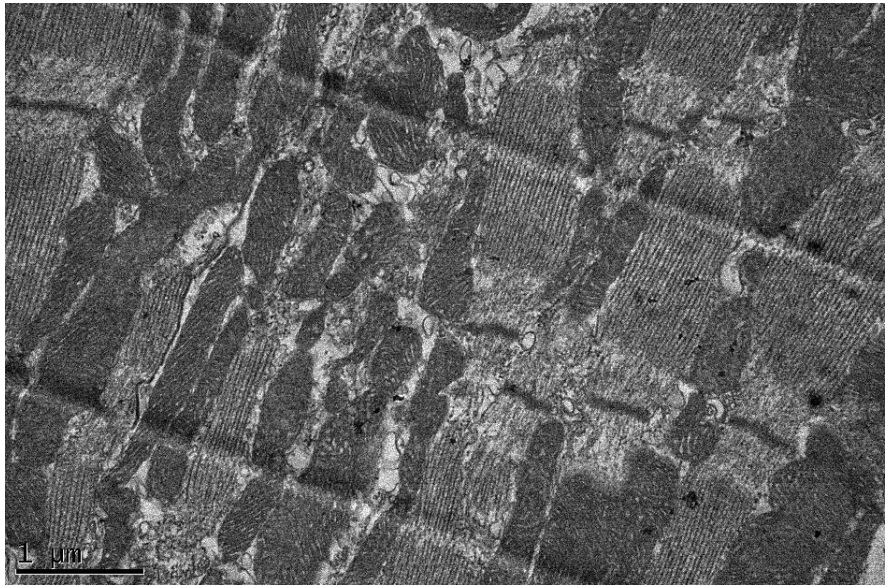
CARDIOLIPIN IN HEART AND MUSCLE



CONTROL HEART

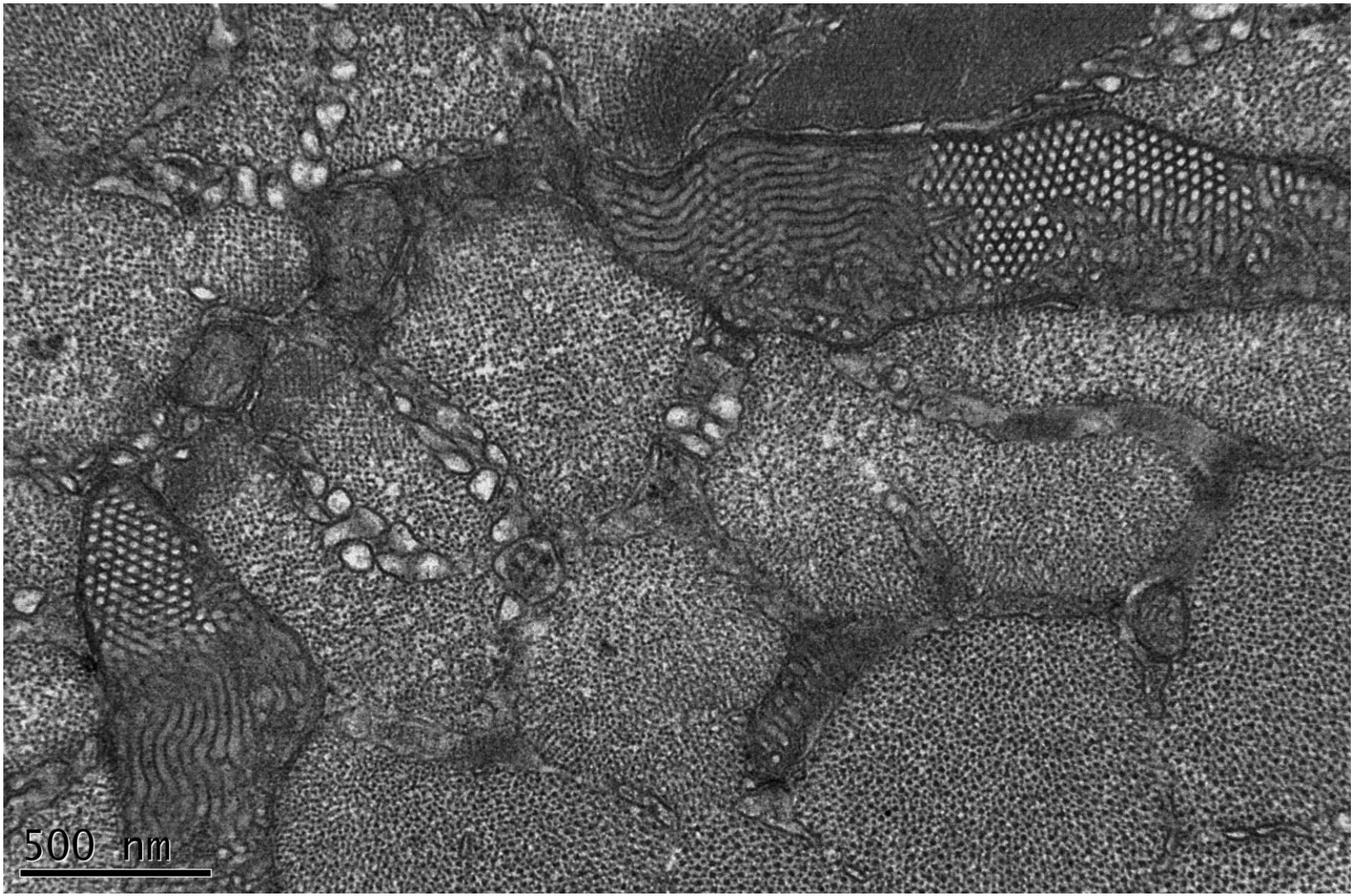


TAFAZZIN KNOCKDOWN HEART



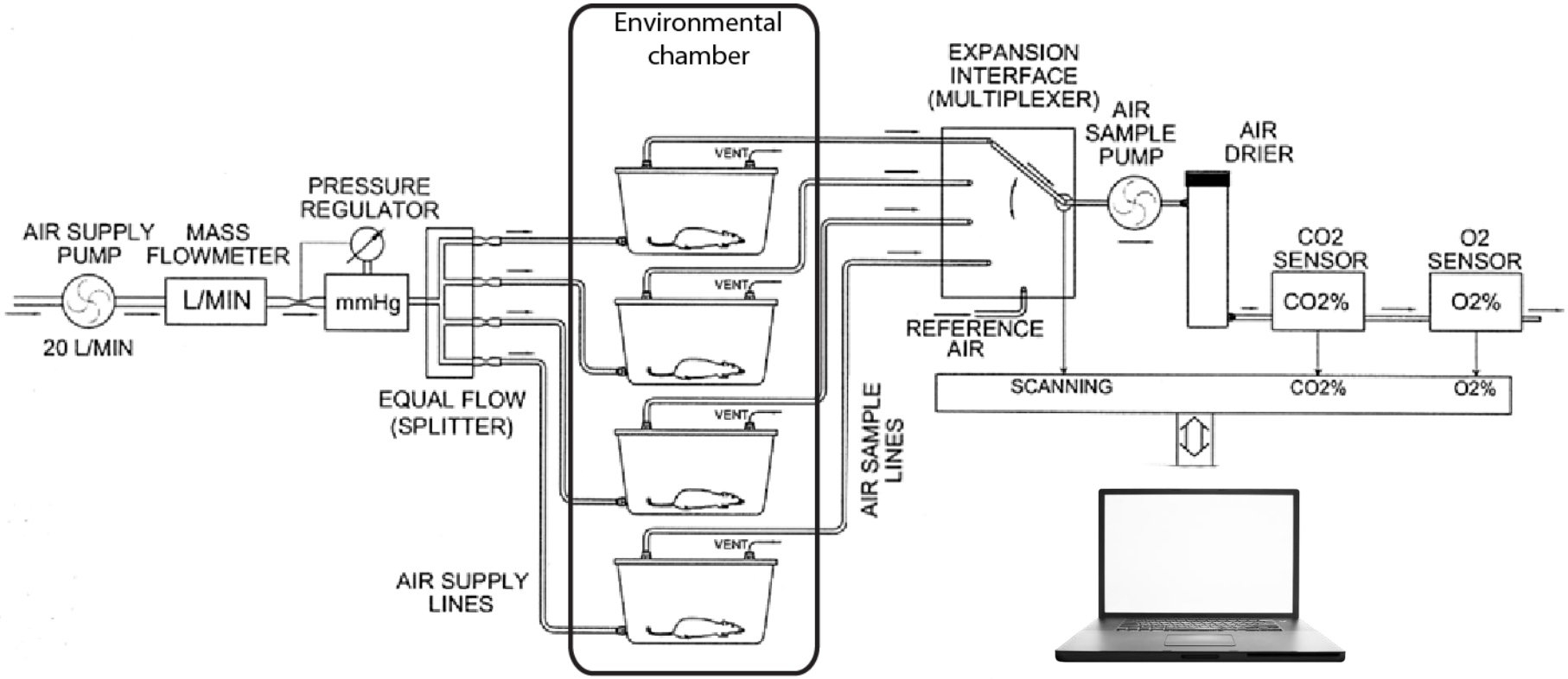
Acehan et al. JBC 2011

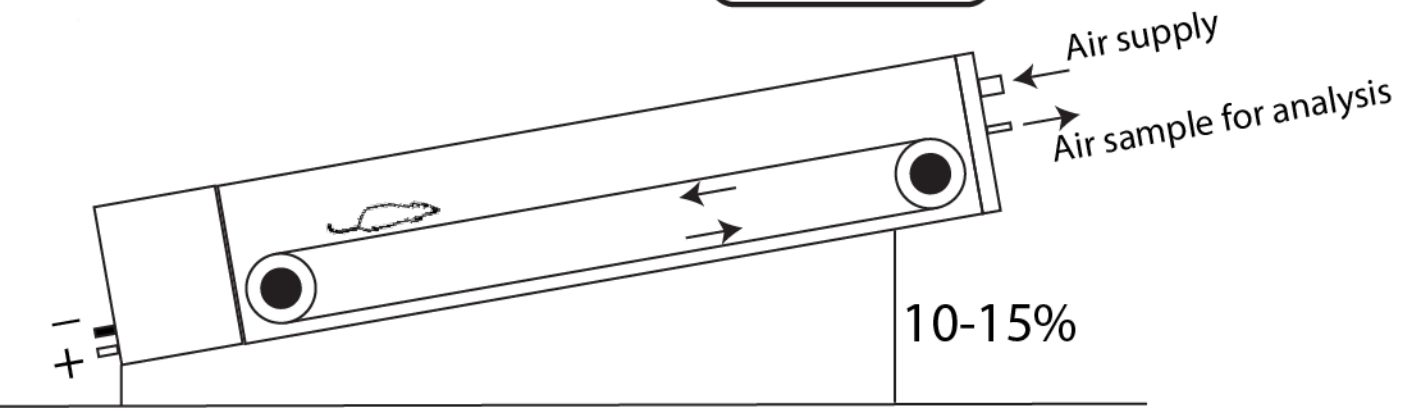
TAFAZZIN KNOCKDOWN SKELETAL MUSCLE

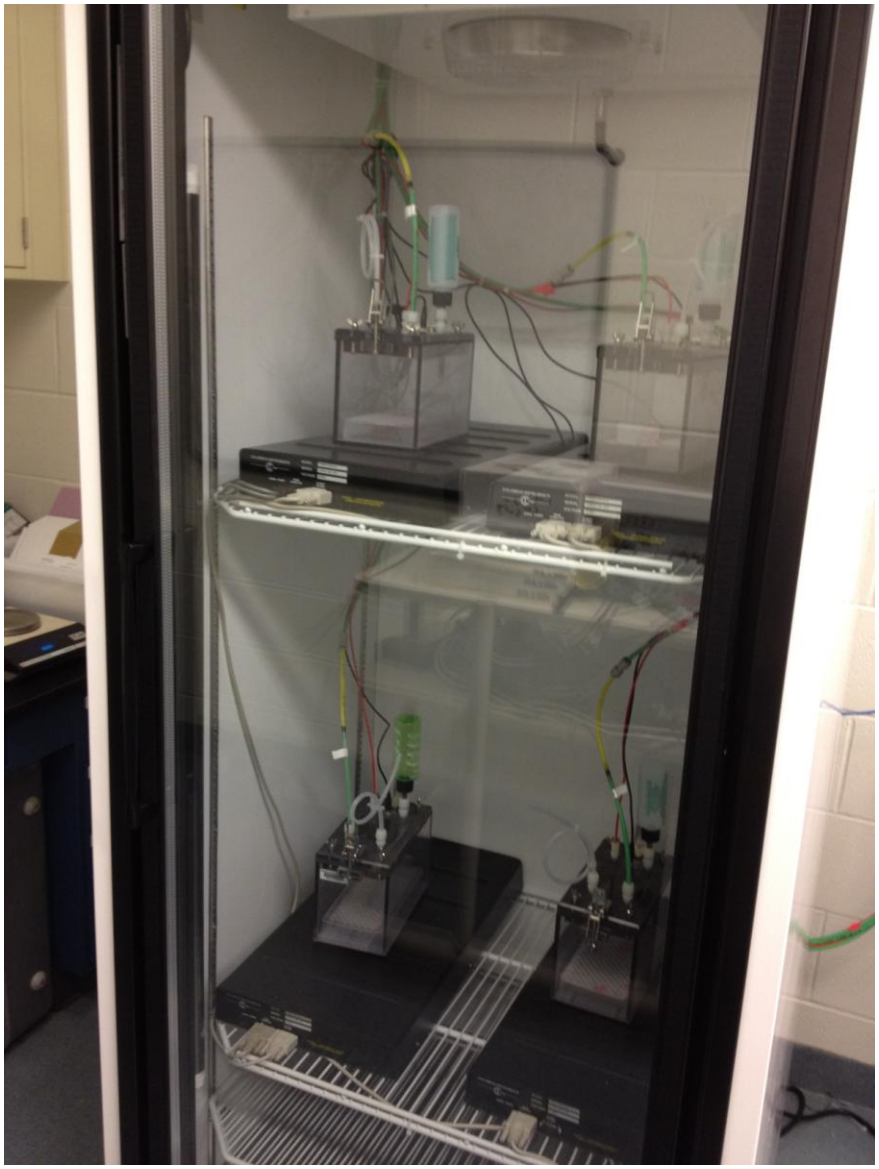


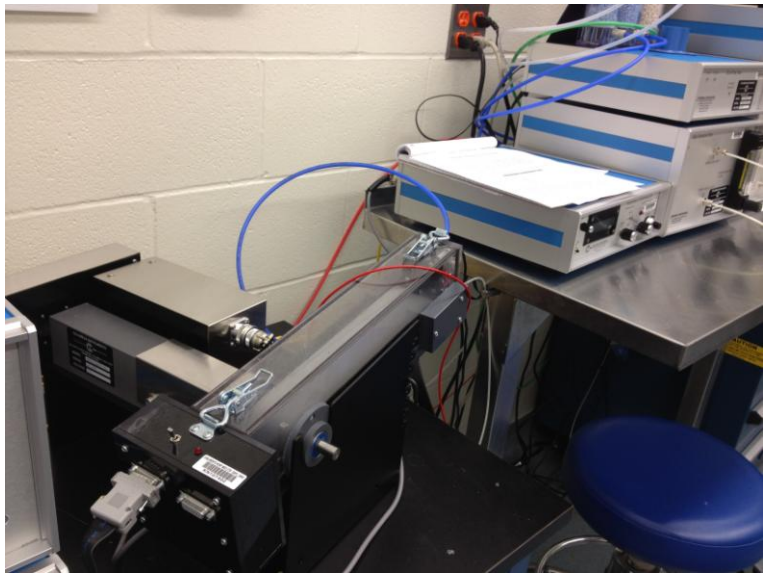
Acehan et al. JBC 2011

Indirect Calorimetry









Calorimetry
At room temperature

Calorimetry
cold (+5° C)

Calorimetry during
exercise on treadmill

Definitions & Abbreviations Used in Calorimetry

- **Reference O_2 Concentration** (O_2i)
- **Reference CO_2 Concentration** (CO_2i)
- **Sample O_2 concentration** (O_2o)
- **Sample CO_2 concentration** (CO_2o)
- **Fresh Air Flow**

- **$VO_2 = ViO_2i - VoO_2o$**
- **$VCO_2 = VoCO_2o - ViO_2i$**
- **Respiratory Exchange Ratio (RER) = VCO_2 / VO_2**
- **Heat = CV x VO_2, where CV = 3.815 + 1.232 x RER**

RESPIRATORY EXCHANGE RATIO (RER)

If **carbohydrate** is completely oxidised to CO_2 and H_2O then the relationships is as follows:

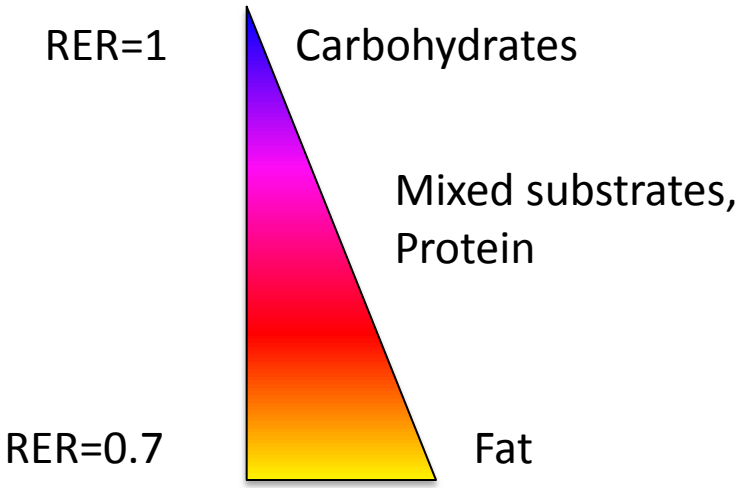
$$6O_2 + C_6H_{12}O_6 \rightarrow 6CO_2 + 6H_2O + 38ATP$$

$$RER = 6CO_2 \div 6O_2 = 1$$

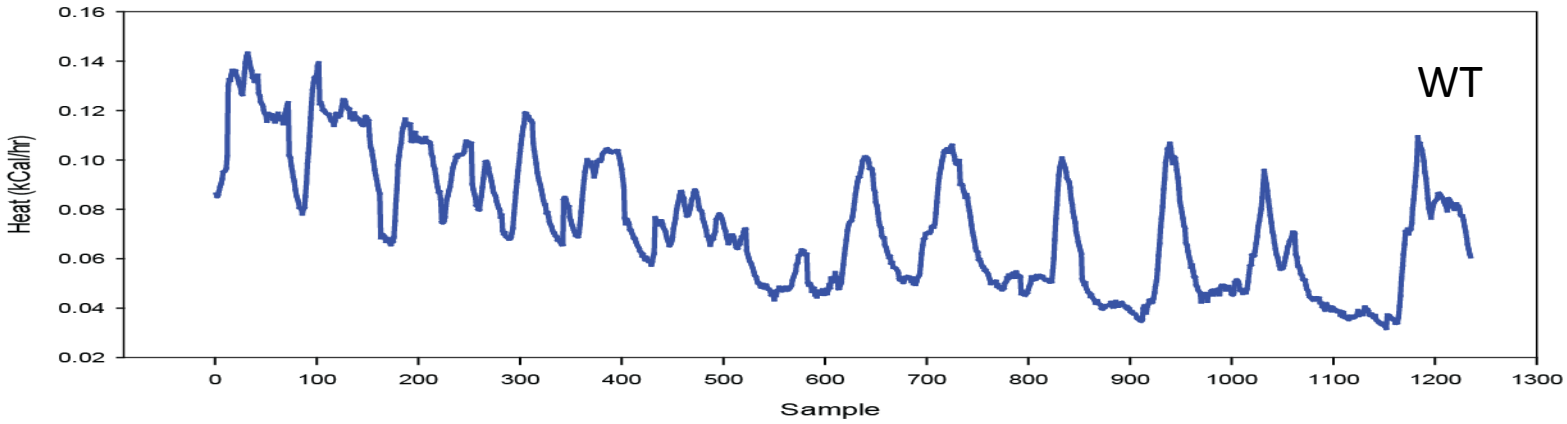
If **fat** is completely oxidised to CO_2 and H_2O then the relationships is as follows:

$$C_{16}H_{32}O_2 + 23O_2 \rightarrow 16CO_2 + 16H_2O + 129ATP$$

$$RER = 16CO_2 \div 23O_2 = 0.7$$



ENERGY EXPENDITURE
(Resting, Dark cycle)



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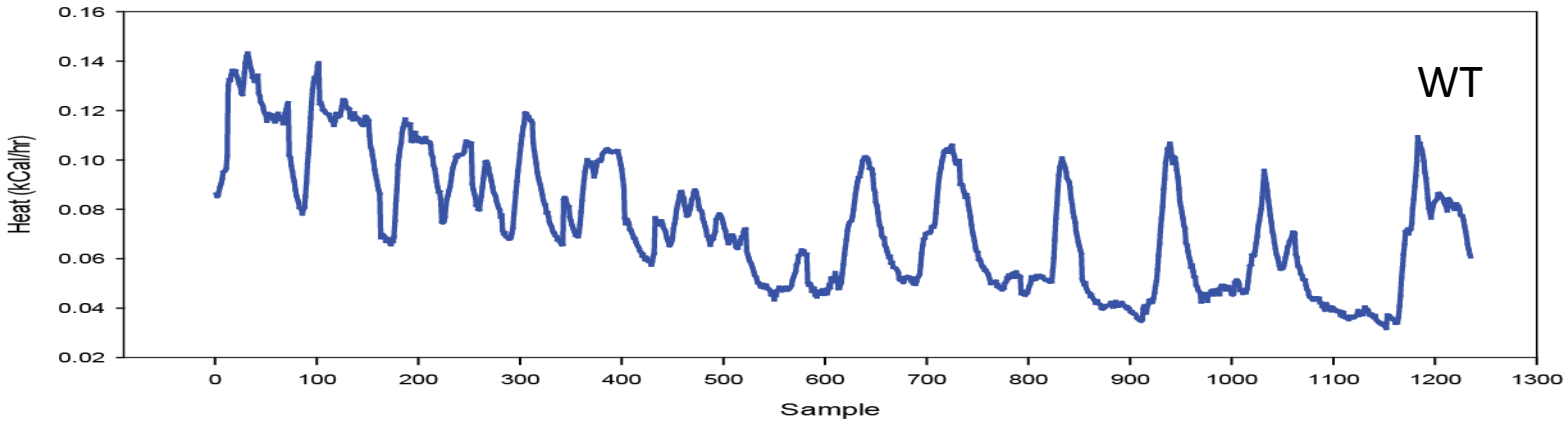
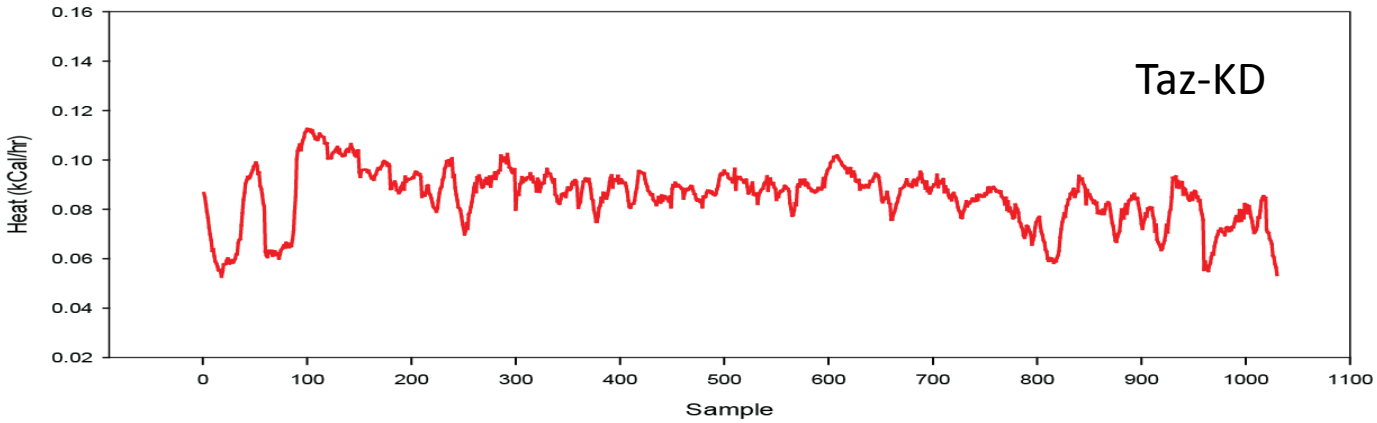
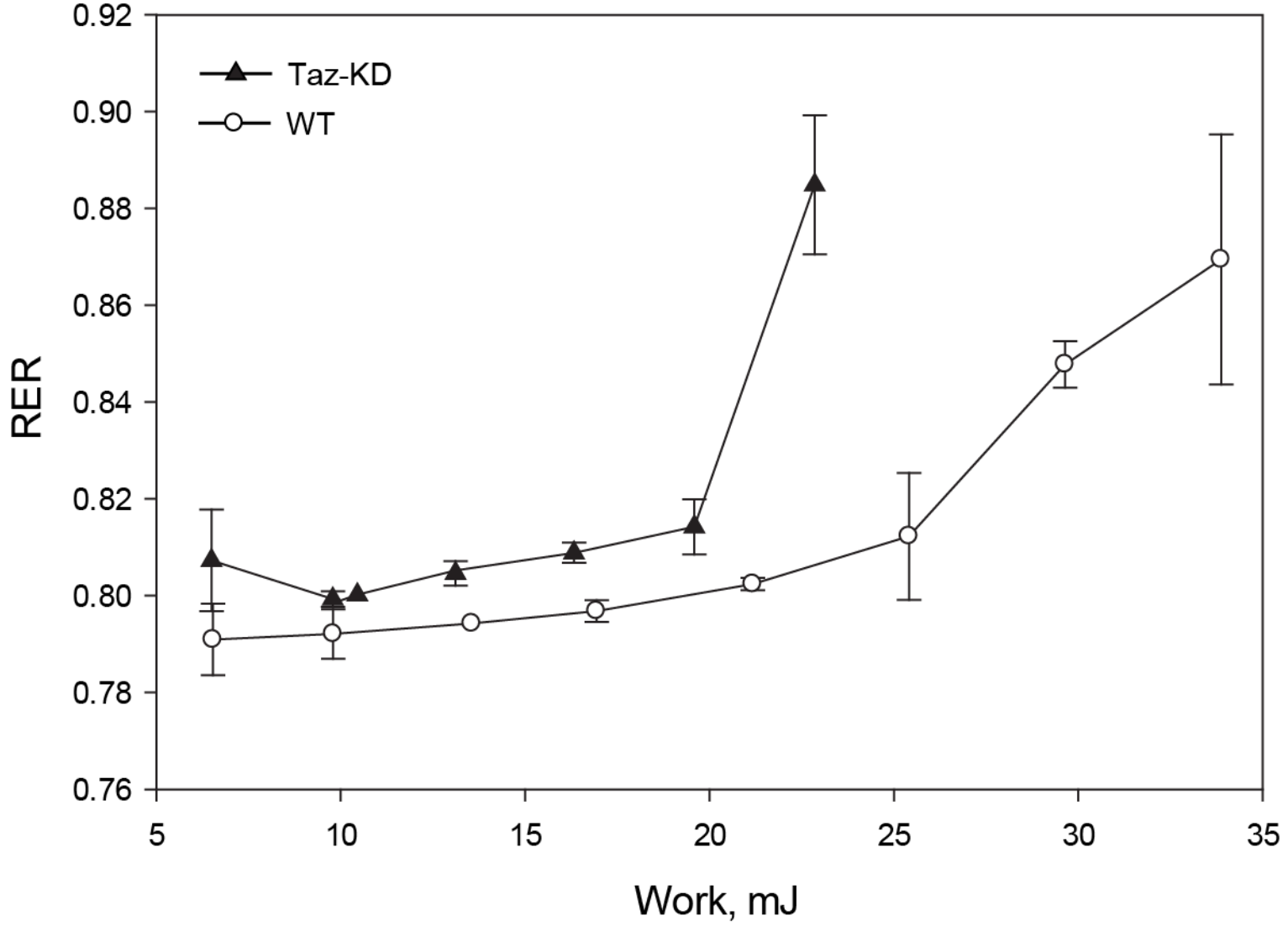
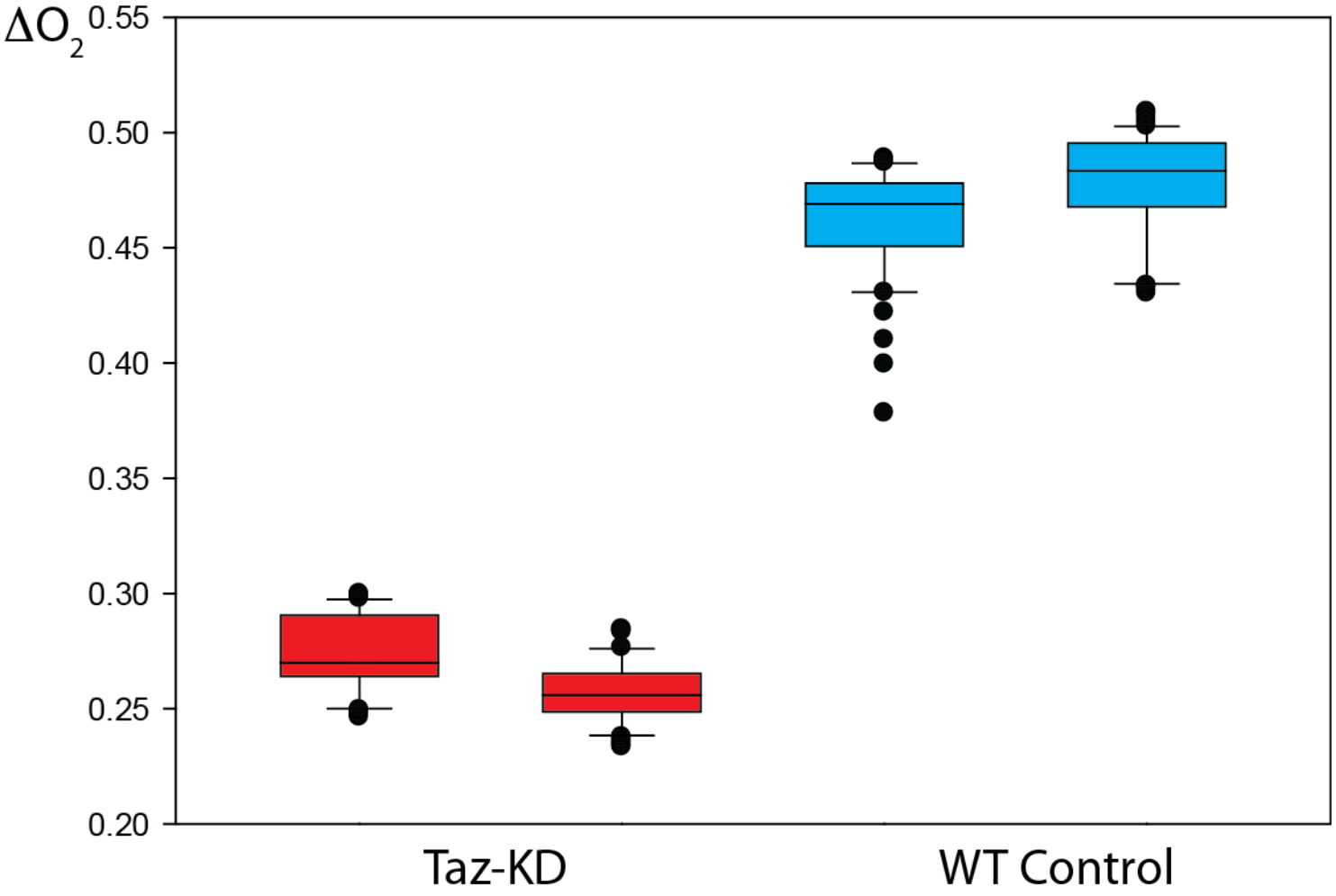




FORCED EXERCISE ON THE TREADMILL

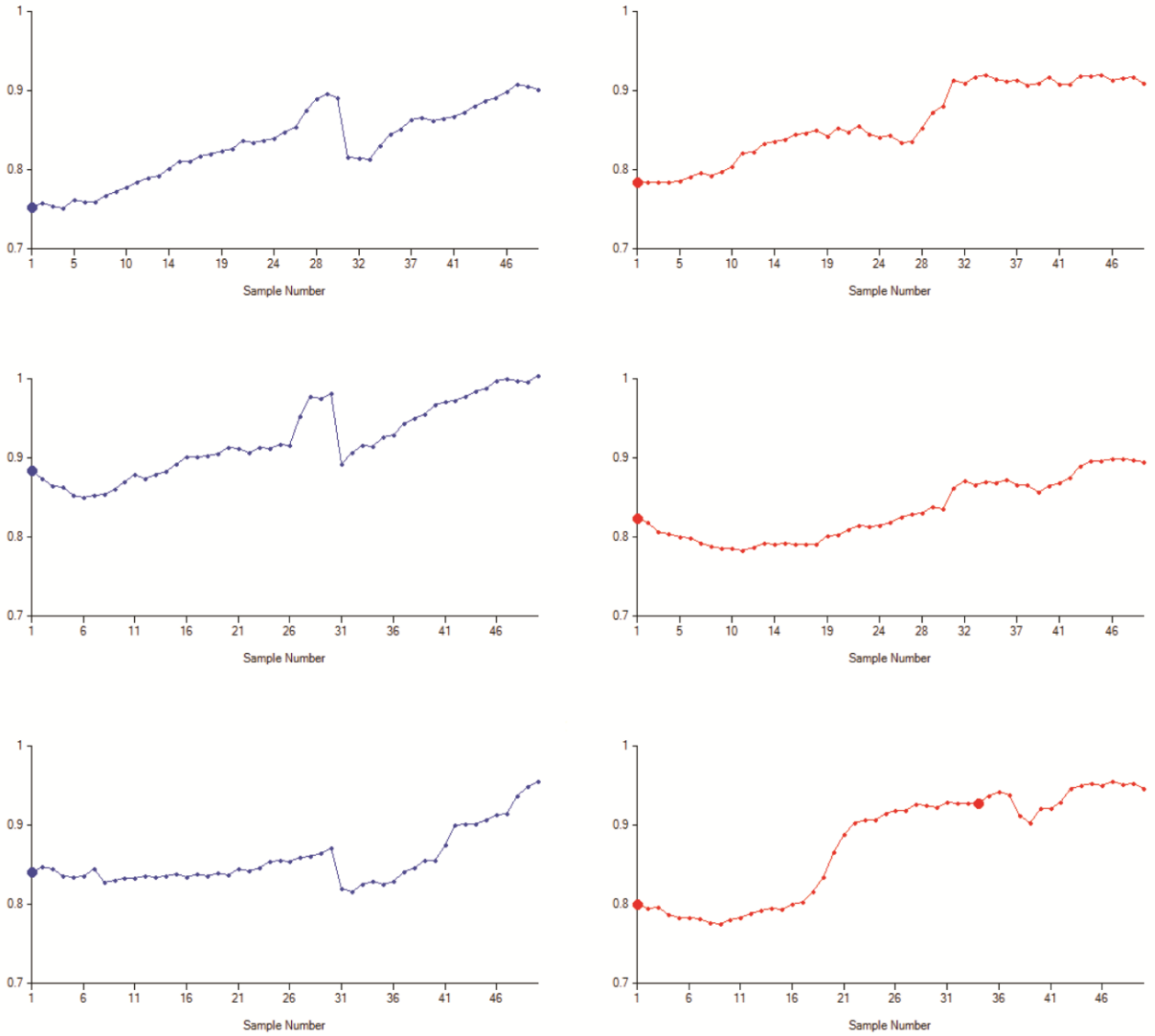


Oxygen consumption on the treadmill



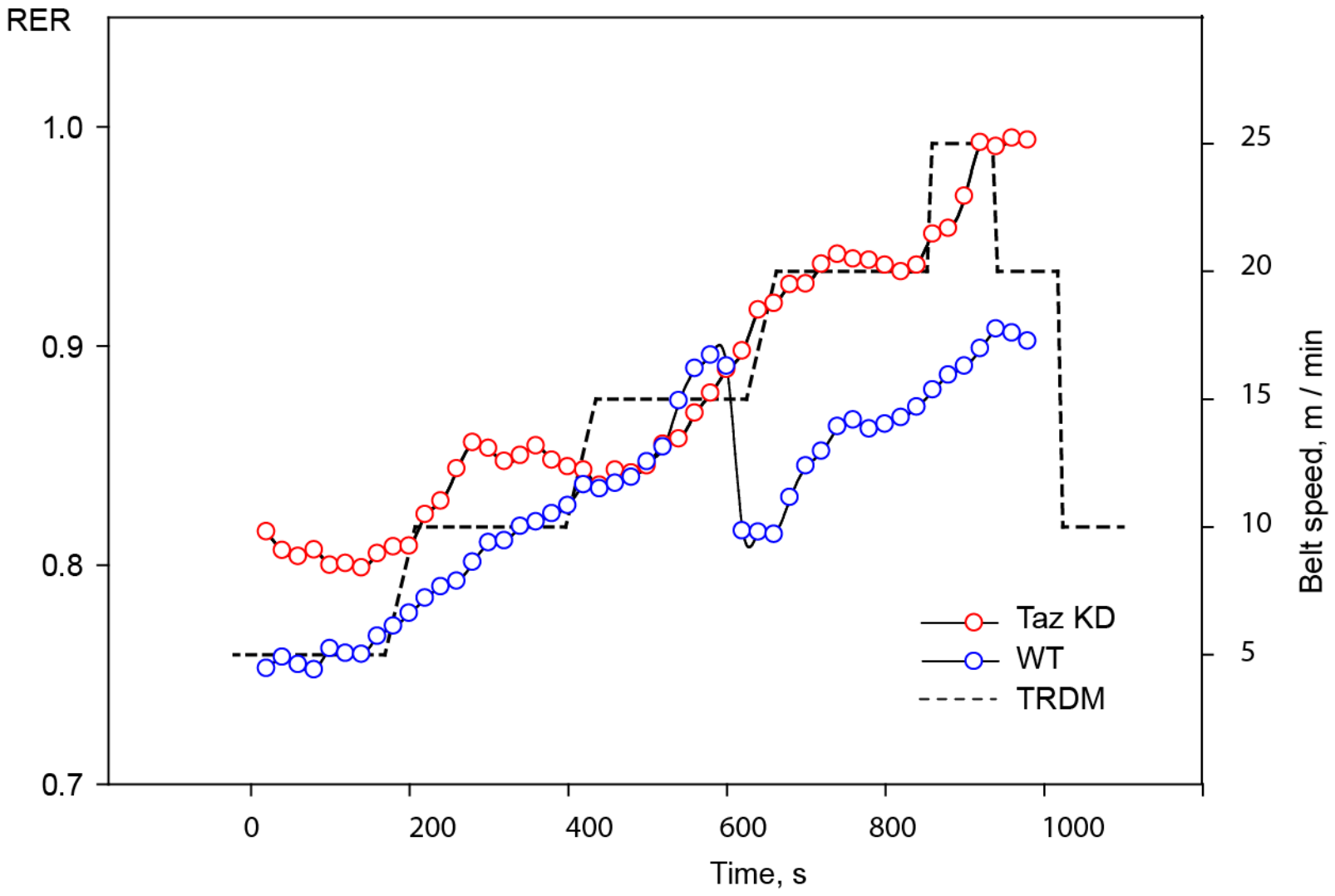
RER on the treadmill

RER

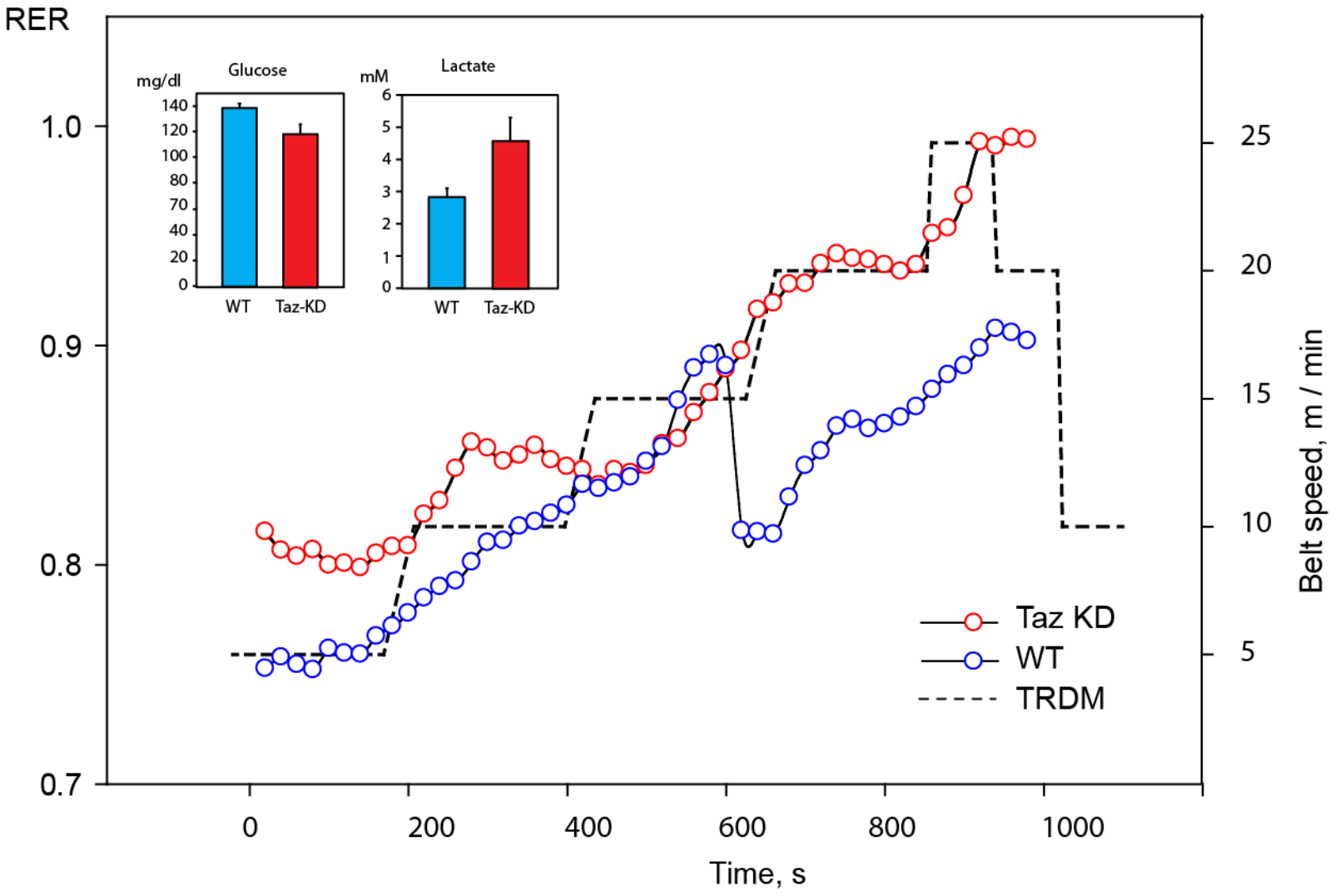


Exercise intensity

RER on the treadmill



RER on the treadmill



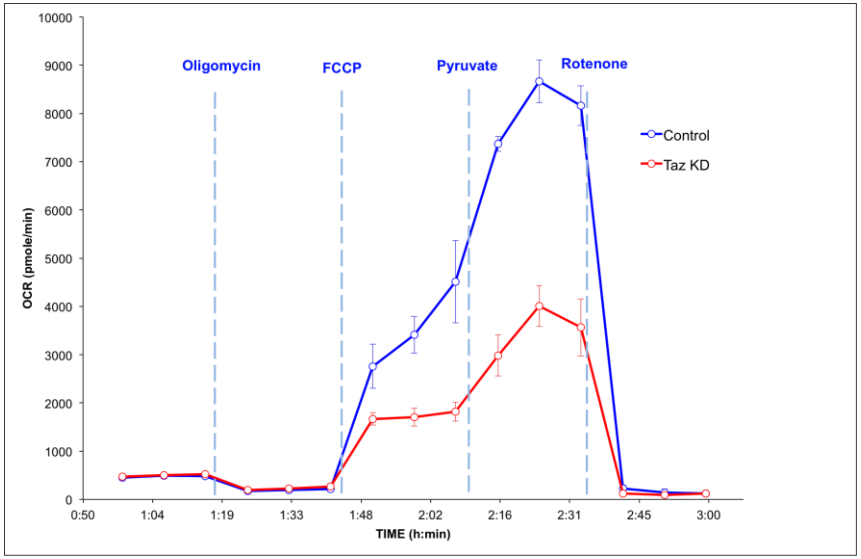
Summary (part one)

- ✧ Tafazzin-deficient mice demonstrate normal rates of energy expenditure at basal resting condition.
- ✧ When exposed to cold, energy expenditure in Tafazzin-deficient mice is severely impaired due to limited ability to consume oxygen.
- ✧ When subjected to moderate-intensity workload, Tafazzin-deficient mice exhibit reduced rates of oxygen consumption and fail to adapt to high-energy demands.

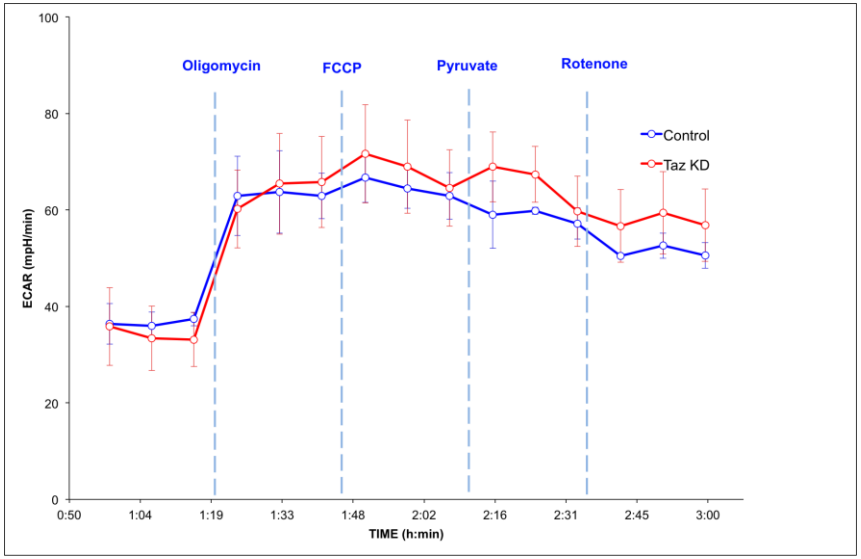
How mitochondrial function is affected in Tafazzin-deficient mice?

Metabolic profiling of tafazzin-deficient mouse neonatal cardiomyocytes

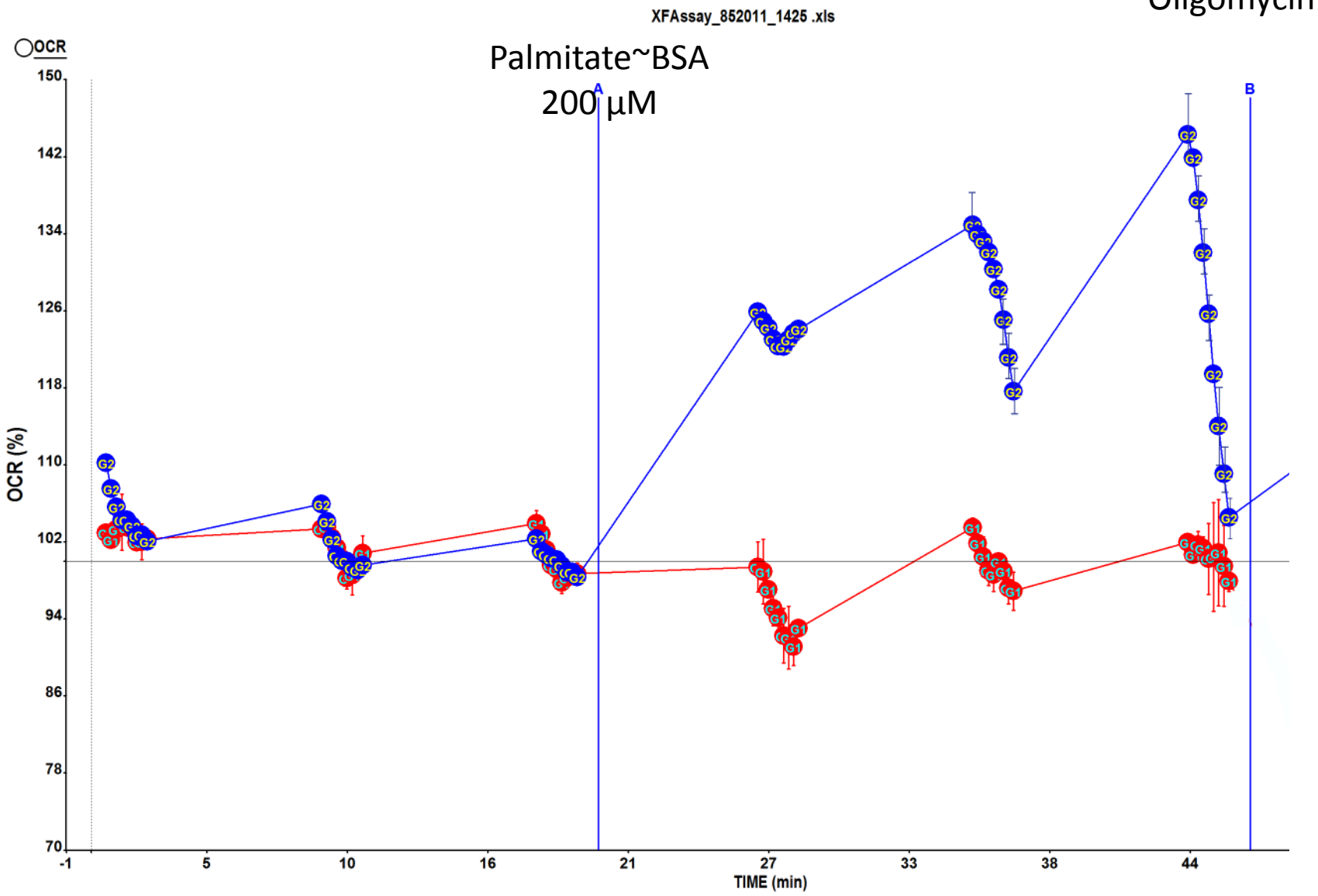
OXYGEN CONSUMPTION



GLYCOLYTIC FLUX



Palmitate-stimulated respiration in cardiomyocytes



LIMITED MITOCHONDRIAL PROTEOMICS

Lipid metabolism:

- Tri-functional protein, subunit β
- Acyl-Co A thioesterase 2

ETC and Metabolism:

- COX6
- ANT-1&2
- ATP synthase, subunit β
- NADP transhydrogenase
- Malate dehydrogenase

Structural proteins:

- Myosin-6
- Myosin LC 3
- Myosin regulatory LC 2
- α-Actin
- Tropomyosin α-1

Protein sorting and degradation:

- Lon protease homolog
- HSP60
- HSP70

Ca^{2+} homeostasis:

- SERCA-2a

Evidence of Physical association of FAO and OXPHOS complexes.

Y. Wang et al. JBC, 280(39) 2010

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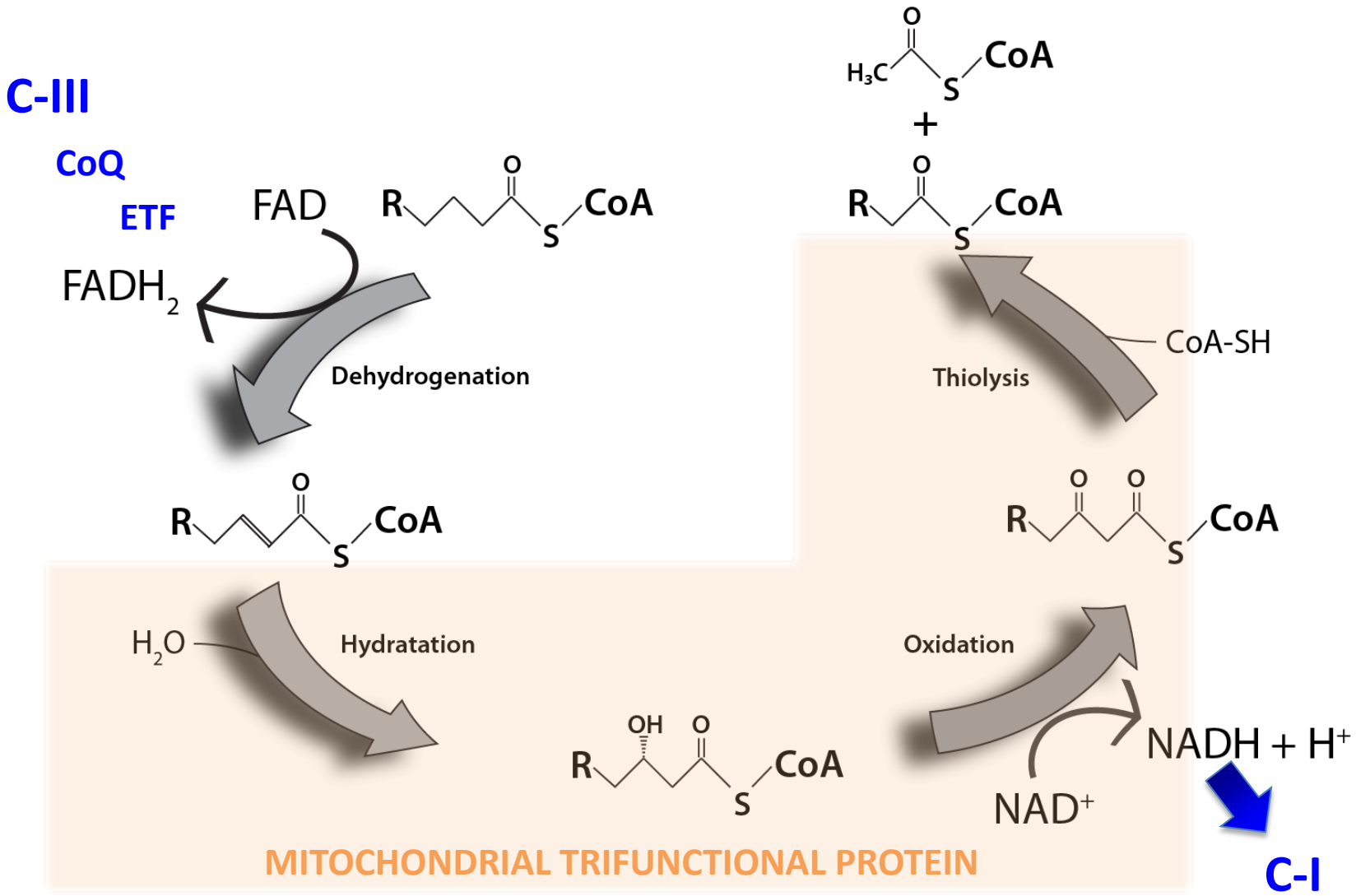
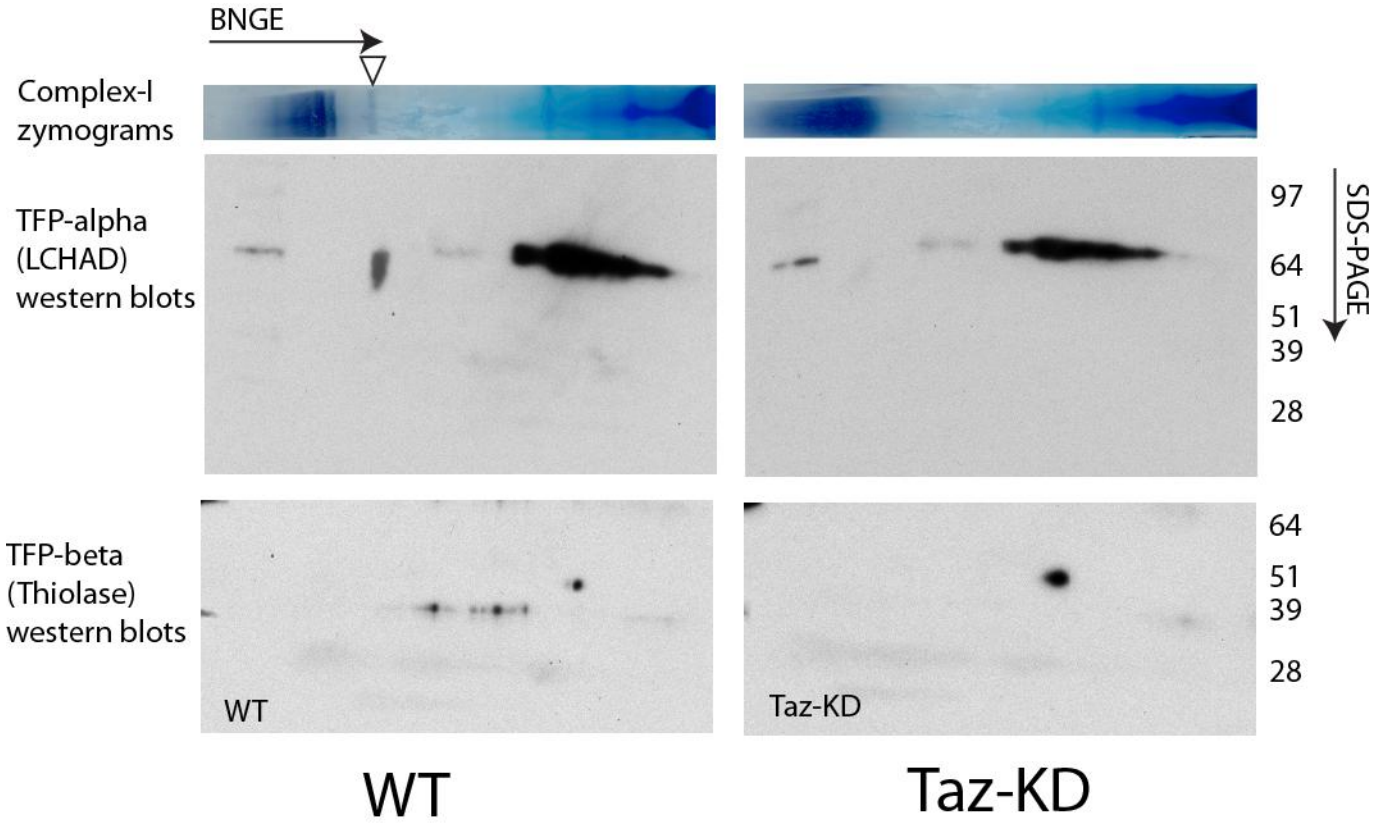
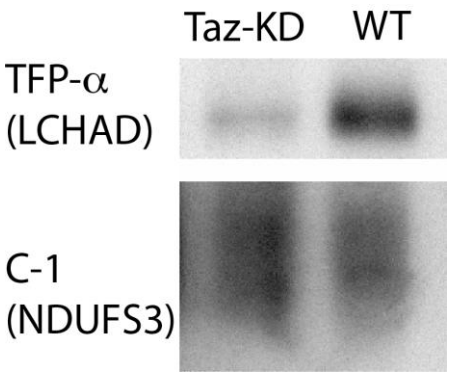
Is CL required for physical interaction of FAO system with OXPHOS complexes?

FATTY ACID OXIDATION



2D-Native gel electrophoresis





SUMMARY

- ✧ Under stress conditions, energy expenditure is severely limited in Tafazzin-deficient mice.
- ✧ Tafazzin-deficiency results in significant reduction of maximal mitochondrial oxygen consumption in neonatal mouse cardiomyocytes, while glycolytic activity is preserved.
- ✧ Oxidation of fatty acids is impaired in Tafazzin-deficient cardiomyocytes.
- ✧ In Tafazzin-deficient mitochondria physical interaction between C-I and TFP is destabilized. Evidence suggests that cardiolipin is required for interaction of TFP with ETC complex I.

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