



Mouse UCP-1 Quantification in Brown Adipose Tissue in University of Fribourg, Inst. of biochemistry

CONTEXT

Our research group is working on circadian rhythms and biological clock, especially on their regulations at the molecular level. In the field, Mouse Uncoupling Protein 1 (UCP-1) is a mitochondrial transmembrane channel protein involved in thermogenesis and only found in brown adipose tissue. This study compares UCP-1 quantity between control mice (that at room temperature during experiment) and mice that are exposed to cold (4°C) during a couple of hours.

PROTOCOL

Precellys® 24 Parameter :
5000rpm, 2x50 sec., 5 sec. Break

1. Western blot against mouse UCP-1
1st antibody: rabbit anti-UCP-1
2nd antibody: infrared (IR)- labeled anti-rabbit
2. Detection of bands with odyssey Infrared Imaging System

MATERIAL

- Precellys®24
- Precellys® kit CK28 (big ceramic beads)
- Sample : brown adipose tissue (between 30-50mg)
- Buffer : 25mM TrisHCl - 1mM EDTA - 1% Triton X-100 0.5% Na-deoxycholate : 500µl



30 mg of Brown Adipose Tissue from a 3-month old wild type female



RESULTS

The lanes correspond to 8 different concentrations of total proteins in order to control the resolution by using the infrared imaging system. UCP-1 quantification studies enable us to get more information about body temperature regulation. Results between control and cold-exposed mice remain confidential.



Detection of lanes with Odyssey Infrared System by using IR-labeled 2nd antibodies. The lanes correspond to 10-30-50-70-20-40-60-80mg concentrations of total proteins.

CONCLUSION

Precellys® is well adapted for easy extraction, as well as the homogenization of adipose tissue and transmembrane proteins. Sample preparation is not only easy but cross contamination free. Efficiency, saving time, and protein collection are the main benefits of the Precellys®.