

Drosophila homogenization for lipid analysis

Department of Developmental Biology, University of Regensburg, Germany

CONTEXT

Our research aim is to establish *Drosophila* as a model for Friedreich's ataxia. This human neurological disorder is produced by the lack of the mitochondrial protein frataxin. Frataxin depletion results in a mitochondrial dysfunction and metabolic problems. We wanted to study weather reduction of frataxin in *Drosophila* also induced some metabolic responses such as loss of lipid homeostasis.

In this work we have found that ubiquitous and glial-targetted reduction of frataxin expression leads to an increase in fatty acids [1].

MATERIAL

- Precellys®24 homogenizer.
- Precellys[®]lysing kit: 03961-1-002 (ceramic beads 2.8mm)
- Sample: Drosophila L3 larvae (15) or *Drosophila* adult heads (80).
- Buffer: Water.

PROTOCOL

- Precellys®24: 5500 rpm, 2x25 sec, 10s break.
- Centrifugation steps: 5000 rpm 60s.
- Analysis of lipid content: Samples were delipidated according to Bligh and Dyer, 1959 for thin layer chromatography studies and gas chromatography coupled with mass spectrometry (GC/MS) was carried out after FA methyl ester derivatization according to Ecker et al., 2010

[1] J.A. Navarro et al., Altered lipid metabolism in a Drosophila model of Friedreich's ataxia, Human Molecular Genetics, 2010 1–13 doi:10.1093/hmg/ddq183.

RESULTS

Quality and quantity of extracted lipids from *Drosophila* samples using Precellys®24 technology was sufficient on the one hand to have reliable and reproducible results from different biological replica (not illustrated) and on the other hand to observe clear differences between control flies and frataxindeficient individuals (Figure 1).

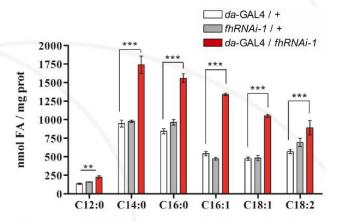


Figure 1: GC/MS analysis of fatty acids from Drosophila L3 larvae (Myristic acid (C14:0), palmitic acid (C16:0), palmitoleic acid (C16:1), oleic acid (C18:1) and linoleic acid (C18:2)).

Frataxin deficiency increases the amount of each fatty acid.

In conclusion, loss of frataxin affects lipid metabolism and catabolism provoking an accumulation of fatty acids. Moreover, triacylglicerides and other neutral or phospholipids are not so affected.



CONCLUSION

Precellys®24 provided us a complete fly homogenate containing lipid in the right range of both amount and purity, in order to carry out our experiments.

Sample preparation is not only easy but cross contamination free.

Problem

Solution







For more details, please contact precellys@bertin.fr



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http://www.technosaurus.co.jp

■東京

〒162-0805 東京都新宿区矢来町 113番地 TEL(03)3235-0661(代) FAX(03)3235-0669

■大阪

〒532-0005 大阪市淀川区三国本町2丁目12番4号 TEL(06)6396-0501(代) FAX(06)6395-2588

■福岡

〒812-0054 福岡市東区馬出 1 丁目 2 番 23 号 TEL(092)631-1012(代) FAX(092)641-1285

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