Untargeted Lipidomics reveals glycerolipid compositional changes in fasted, cold-exposed MCAD KO mice

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1. Introduction

Hepatic mitochondrial fatty-acid β-oxidation plays a central role in ATP production during fasting. Medium-chain acyl-CoA dehydrogenase (MCAD) oxidizes medium-chain fatty acids (C6-C12). MCAD-deficient children present with life-threatening low blood glucose concentrations or remain asymptomatic[1]. In order to understand the etiology, we subjected MCAD knockout (KO) and wild-type (WT) mice to fasting and cold (4°C) .

2. Approach

Male MCAD KO and WT mice on a C57BL/6J background were individually housed under standard conditions. The mice were fasted for 14 hrs and subsequently exposed to 4°C for 4-6 hrs until termination. Liver and blood were collected for analysis. We applied the untargeted lipidomic method on hepatic lipid extracts according to Gill et al[2] with slight modifications. In addition, we measured acyl-carnitines according to Derks et al[3] and liver TG with a commercially available kit.

3. Results

Abnormally elevated level of medium-chain acyl-carnitines (C6,C8,C10:1,C10) were detected in blood of MCAD KO compared to WT mice. In contrast, hepatic acyl-carnitine profile was similar in both the WT and KO mice. Liver TG was significantly higher in the MCAD KO compared to the WT mice.

More than 800 single lipid species were detected and identified from the extracted hepatic samples. Principle Component Analysis (PCA) of the hepatic lipidome showed clear differences between the MCAD KO and WT mice. The OPLS-DA revealed a list of lipids, which contributed the most to the differentiation between the WT and KO mice. This showed that medium to long chain (C12-14) triglycerides (TGs) (n=65) and diglycerides (DGs) (n=7) were most significantly upregulated in the livers of MCAD KO compared to WT mice (representative examples are shown in Fig.1). Interestingly, medium-chain TGs (e.g. TG(8:0/8:0/8:0) were very low in both MCAD KO and WT mice.

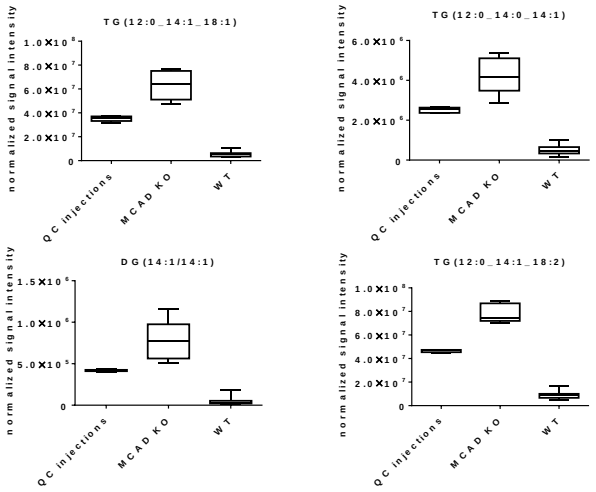

Figure 1. Normalized signal intensity detected from representative single lipid species on the VIP list. Intensity measured in quality control (QC) injections (n=5) suggested good reproducibility of the measurements.

4. Discussion

The results suggest significant differences in glycerolipid metabolism (majorly TGs) but hardly in other lipid species between MCAD KO and WT mice under stress conditions. we cannot assign the results to either of the treatments, but only to the combination. The elevated level of median to long chain TGs in MCAD KO mouse liver (fold change>5, $p<0.05$) suggests that only a few rounds of chain elongation occurred on medium-chain fatty-acids. Subsequently, the elongated median- to long-chain fatty acids were incorporated only into DGs and TGs to prevent toxicity in the liver.

Overall, the above result suggests an adaptation strategy in MCAD KO mice to prevent accumulation of medium-chain fatty acids in the liver under stress conditions.

References

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2. Gil A, Zhang W, Wolters J C, et al. PMID: 29968103
3. Derks T G J, Boer T S, Van Assen A, et al. PMID: 18188679