Preclinical evidence of efficacy of heptanoate and derivatives as anaplerotic therapy for medium-chain acyl-CoA dehydrogenase deficiency

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Imbalance in TCA cycle intermediates is postulated to be a key pathophysiologic mechanism in fatty acid oxidation disorders (FAODs). Dojolvi (triheptanoylglycerol; C7G) has shown efficacy as anaplerotic therapy for long-chain FAODs but has been contraindicated for MCAD deficiency as heptanoate, the active C7G’s drug form, has been assumed to be metabolized solely by MCAD. In this study, heptanoate was investigated as potential anaplerotic therapy for MCAD deficiency. ACADM G985A/G985A fibroblasts were incubated with heptanoate, or branched heptanoate derivatives, at various concentrations prior to monitoring oxygen consumption rate (OCR) using a Seahorse XFe96 Extracellular Flux Analyzer. A dose dependent increase in maximal respiration, spare capacity, and basal extracellular acidification rate (ECAR) was observed indicating heptanoate was readily utilized for energy and effective in improving these parameters. Intracellular metabolomics confirmed utilization of heptanoate with higher amounts of propionate and succinate generated compared to the other branched heptanoate derivatives. Cell-free extracts of MCAD-deficient fibroblasts were shown to utilize heptanoyl-CoA but not octanoyl-CoA to reduce ETF fluorescence. Purified human short-chain acyl-CoA dehydrogenase (SCAD) previously reported not to use octanoyl-CoA as substrate, utilized heptanoyl-CoA, indicating that SCAD should allow MCAD deficient patients to safely metabolize heptanoate. To further evaluate this possibility, Acadm-/- mice tolerance of C7G was assessed. Dose-dependent increases in plasma levels of heptanoyl-, valeryl-, and propionylcarnitine were observed that correlated with a dose-dependent reversal of hepatic steatosis and concomitant increase in glycogen content. This study provides the impetus to evaluate C7G as potential therapy for MCAD deficiency and investigate heptanoate derivatives as anaplerotic alternatives.