Development of emerging model microorganisms: *Megasphaera elsdenii* for biomass and organic acid upgrading to fuels and chemicals

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The native ability to condense acetyl-CoA groups to efficiently generate C4 to C8 compounds makes *Megasphaera elsdenii* a compelling platform for the production of fuels and chemicals from lactate and plant carbohydrates. Our overall objective is to develop *Megasphaera elsdenii* as a platform for the conversion of lignocellulosic biomass sugars and organic acids into hexanol and other valuable chemicals. *M. elsdenii* produces organic acids as fermentation products when growing on lactate and glucose, including formation of butyric (four carbon), hexanoic (six carbon), and in some cases octanoic (eight carbon) acids as major fermentation products, likely via a chain elongation pathway using acetyl-CoA. As the carbon chain length increases, fuel properties improve, with the energy density increasing and hygroscopicity decreasing. Virtually nothing is known about the metabolic pathways in *M. elsdenii* that result in organic acid formation beyond predictions based on genome annotation. We have developed the first methods for DNA transformation of two strains of *Megasphaera elsdenii*, opening this organism to advanced physiological studies and bioengineering. We have begun the use of newly generated genome sequences to create a metabolic reconstruction of glucose and lactate conversion to hexanoic acid using the DOE KBase platform as the foundation of a metabolic model. We continue to develop genetic tools to enable more rapid and complex strain construction to develop strains capable of producing long chain carbon molecules at high yield and high titer. We will present a progress report on these efforts.

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