

## **Study Design: Brandon Higgins 7/8/2016**

**Platform:** Primary metabolism (GCTOF MS)

**Samples:** 72, *Auxenochlorella protothecoides* cells, 0.15-1 mg

**Sample Prep:** 1 ml algae cell culture was collected, 1 ml 70% MeOH (30% dH<sub>2</sub>O) at -80 C was added to the algae sample. Tubes were spun at 12,000 rcf to pellet for 2 min at 0 C. Supernatant was decanted and pellets stored at -80 before freeze drying at -45 C. Freeze dried samples were stored at -20 C until submission for extraction.

**Treatments:** 4, 3, 3 control cultures with no thiamine metabolites, 3 cultures received thiamine, 3 received HMP, and 3 were grown on residual medium from another algae species - *Chlorella sorokiniana*. Samples were taken daily from each of these 12 cultures over a 5 day period resulting 72 samples total (including time zero).

**Interest:**

**Notes:**

**Study Design Abstract:** The purpose of this study is to determine how thiamine metabolites impact central metabolism in *Auxenochlorella protothecoides* when grown in the presence of acetate. We hypothesize that thiamine metabolites alleviate bottlenecks in the TCA cycle and gluconeogenesis, thus allowing for greater starch production when they are present. Cells were grown in bioreactors: 3 control cultures with no thiamine metabolites, 3 cultures received thiamine, 3 received HMP, and 3 were grown on residual medium from another algae species - *Chlorella sorokiniana*. We suspect that this residual medium also contains thiamine metabolites. Samples were taken daily from each of these 12 cultures over a 5 day time course so that we can observe build-up of metabolites over time.