Grow experimental cultures to saturation. Set aside an aliquot to quantify cell count on a hemocytometer.

For each sample, harvest 2 x 1 ml of saturated culture and spin out @ 13,000 rpm for 1 min (*Saccharomyces*) or 3 min (*Hanseniaspora*). (1 tube will be no enzyme control).

Wash 2x in 1ml ice-cold H2O.

Resuspend pellet in 250 ul of 250mM Na2CO3.

Boil suspension for 3h @ 95°C. Vortex once per half hour.

Vortex vigorously to resuspend debris.

Add 10ul of amyloglucosidase-z to 1 tube per sample.

Incubate O/N at 54°C.

Dilute each sample 1:2 with dH2O and send 500ul volume upstairs for end-products metabolomics. *Final dilution performed because the facility requires a minimum volume of 500ul.*

***Media & Reagents***

*Amyloglucosidase-z 200 mMNaAcetate*

100 mg α-amyloglucosidase 820 mg NaAcetate

5 ml 200mM NaAcetate 50 ml H2O

HCl to 5.2