

## Characterizing fungal inhibitors in drought-stressed switchgrass

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**Project goals: The overarching goal of this project is to identify fungal inhibitors from drought-stressed switchgrass using extraction techniques and use this information to develop strategies to overcome biomass variability and ensure consistent biofuel generation.**

Development of economically viable and greener pathways to synthesize renewable energy has been an important research theme in recent years. Lignocellulosic biomass is a major resource that can be used for biofuel production. Recent research has showed that biomass characteristics are altered by environmental growth conditions, and directly influence the extent of biomass conversion to fuels. Previously it was reported that drought experienced during the growth of switchgrass led to complete inhibition of yeast growth during fermentation<sup>[1]</sup>. In this project, we characterized specific compounds that led to this inhibition. Switchgrass harvested in drought and non-drought years were pretreated using Ammonia Fiber expansion (AFEX). Untreated and AFEX processed samples were extracted using solvents (i.e. water, ethanol, and ethyl acetate) to selectively remove potential inhibitory compounds and determine whether pretreatment affects the inhibition. A key goal of the project was to determine whether the microbial-inhibitors are plant-generated compounds, by-products of the pretreatment process, or a combination of both. High solids loading enzymatic hydrolysis was performed on all samples followed by fermentation using genetically modified, xylose consuming yeast strain *Saccharomyces cerevisiae* Y330. Cell growth (OD<sub>600</sub>), sugar consumption, and ethanol production were used to evaluate fermentation performance. Extracts were analyzed using liquid chromatography-mass spectrometry (LC-MS) to identify potential inhibitory compounds.

***This material is based upon work supported by the Great Lakes Bioenergy Research Center, U.S. Department of Energy, Office of Science, Office of Biological and Environmental Research under Award Number DE-SC0018409***

### References

[1] Ong, R. G., et al. (2016). "Inhibition of microbial biofuel production in drought-stressed switchgrass hydrolysate." *Biotechnol Biofuels* 9: 237.