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**Progress Report**

PROJECT TITLE: Nutritional improvement of corn ethanol coproducts via yeast engineering

PROJECT NUMBER: 1081-16EU

REPORTING PERIOD: quarterly reports

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1.) PROJECT ACTIVITIES COMPLETED DURING THE REPORTING PERIOD. (*Describe project progress specific to goals, objectives, and deliverables identified in the project workplan.*)

As we mentioned before, the specific goal during this period is to figure out how to increase the total key amino acids such as lysine, arginine, and tryptophan during fermentation. We are continuing the method that expressing peptides rich in tryptophan, arginine, and lysine in yeast cells.

During this period , we test the PDA1-1xKR, RPL4-1xKR, SSA1-1xKR, HSP26-1xKR, and TDH3-1xKRstrain fermentation ability in corn mash medium and perform a 4 day mock fermentation media(YP medium with 150g/l glucose at initial time point with glucose feeding). Now, the ethanol, glucose and amino acid composition of the fermentation broth are being analyzed by HPLC and the best strain will be selected.

2.) IDENTIFY ANY SIGNIFICANT FINDINGS AND RESULTS OF THE PROJECT TO DATE.

In our secondary batch analysis on the specific amino acid profile, we found the arginine (R) content is increased after fermentation in the corn mash media. Comparing with the wild type strain (CEN.PK), the arginine (R) in SSA1-1xKR strain increased by 9%.

In the 4 day mock fermentation media, all the strains showed similar growth ability and final OD. The amino acid profile of 4 day culture is under analysis.

3.) CHALLENGES ENCOUNTERED. (*Describe any challenges that you encountered related to project progress specific to goals, objectives, and deliverables identified in the project workplan.*)

The overall goal is to significantly increase the feeding value of corn ethanol co-products. For the strategies involving tryptophan and arginine being developed in alternative ways, the major challenges up to now is the detection of cysteine and tryptophan in amino acid profile. Because of the acid hydrolysis will destroy all the cysteine and most of tryptophan. This may require other samples processing method to analysis these two kinds of amino acid.

4.) FINANCIAL INFORMATION (*Describe any budget challenges and provide specific reasons for deviations from the projected project spending.*)

5.) EDUCATION AND OUTREACH ACTIVITES. *(Describe any conferences, workshops, field days, etc attended, number of contacts at each event, and/or publications developed to disseminate project results.)*

Now the manuscript is being drafted for genetic engineering of lysine biosynthesis for improved lysine production.

A manuscript is also being drafted to disseminate research findings on the corn fungal secondary fermentation.

A manuscript will be drafted for genetic engineering for overexpression of arginine-rich protein for improved arginine content in DDGS.