Control Number_0001916-1548_Xylome Corporation_Summary:

Biodiesel and higher value products from stillage fiber

A proposal in response to DOE/EERE/BEEPS FOA Number DE-FOA-0001916

Summary Abstract: Biodiesel and higher value products from stillage fiber

The broader impact/commercial potential of this project is to produce, economically, the next generation of sustainable, renewable, clean burning, high energy density, transportation biofuels. Xylome will do this by producing *Biodiesel and higher value products from stillage fiber*. Xylome's principle investigator is Dr. Thomas Jeffries, who has 30+ years of experience with yeast genetic engineering and renewable fuels experience. He will lead the Xylome team of experienced molecular biologist and scale-up scientists to achieve this proposal's objectives. We will also engage some of the largest producers of ethanol and ethanol plant designers to help validate the technology and commercialize the production of a cellulosic lipid that suitable for biodiesel. Xylome's technology will enable existing biofuel producers to reduce their costs while increasing the value and diversity of their current byproducts. The proposed technology will convert wet organic stillage fiber, a waste-product containing cellulosic and hemi-cellulosic fiber and cellulosic hydrolysates into tailored fatty acids suitable for biodiesel. The technology will be compatible with and complementary to ethanol producer operations, and it could potentially double the amount of biodiesel produced in the U.S. today. The 203 domestic ethanol plants range in size from less than 50 to more than 150 million gallons, and they have a total annual capacity of ~15-billion gallons. Every gallon of ethanol also yields 1.9 pounds of soluble organics that must be evaporated or disposed. Xylome's technology has the potential to convert of the total 24 billion pounds into biodiesel with a value of $4 billion. The technology could open a path to expanded biodiesel production using other low-value cellulosic feedstocks and cellulosic hydrolysates. Xylome's idea will increase the efficiency of existing ethanol plants by increasing the economical production of an oil suitable for biofuel production. This type of diversification is essential for the long-term sustainability of the fuel ethanol industry.

The objectives of this grant are to enhance the technical readiness of *de novo* oil production from cellulosic and hemi-cellulosic fiber found in wet waste stream called thin stillage. The proposed work will develop fermentation of wet stillage organics into lipids at a value proposition that is s equivalent to current ethanol production. Yeast lipids normally accumulate under nitrogen limiting conditions after replication has stopped. These lipids are not excreted from the cells, so recovery does not require distillation. In prior development, Xylome scientists identified fermentation conditions and over-expressed genes that increase lipid accumulation by more than 4-fold under high nitrogen conditions. Also, they used mating, selection, screening, evolutionary adaptation and synthetic biology to combine the best of these modifications. Xylome scientists now want to incorporate cellulase and hemi-cellulase enzymes into our high lipid-producing yeast to improve the conversion of wet organic wastes and cellulosic fiber found in stillage along with other wet organic wastes to that same yeast-oil. Xylome plans to optimize cellulosic biodiesel from lipid production through metabolic engineering and from the "outside in" through bioprocess design and cultivation conditions for cellulosic biodiesel production.