



Biofuels: Advanced Enzymes and Mixtures

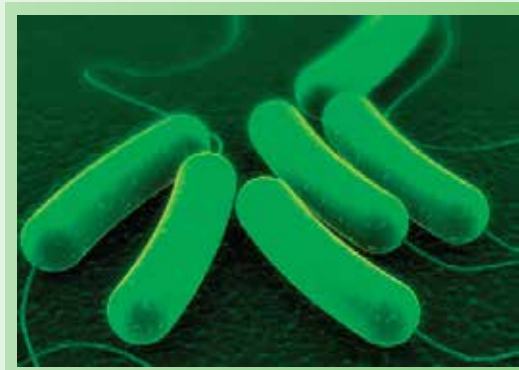
Optimizing enzyme mixtures to meet biorefinery conditions lowers conversion costs of lignocellulosic biomass to biofuel

Enzymes Key to Biofuels Production

Sandia National Laboratories is developing new enzymes and mixtures that can convert lignocellulosic biomass into sugar. Currently, commercially available enzymes are expensive, amounting to about 50% of the minimum ethanol selling price and not suited for the harsh conditions of biorefineries, which limits the economic viability of biofuels at scale.

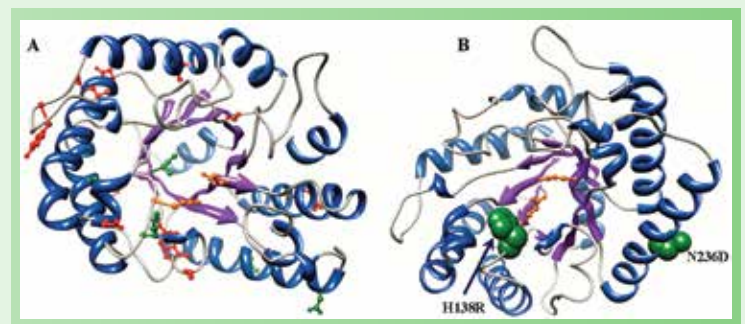
Characterizing and Refining Enzyme Mixtures

Researchers are approaching this problem by first characterizing enzymes for compatibility with biorefinery conditions and then optimizing the ratios of these enzymes



Genes are synthesized and expressed in an appropriate expression host, typically in E. Coli. Each enzyme is screened for activity across a range of temperatures, pH and biorefinery relevant conditions.

in the mixture to maximize sugar yields. Leveraging Sandia's expertise in genomics, researchers are mining databases for genes encoded with enzymes from organisms that live in environments with similar properties to those of biorefineries. These enzymes that meet desired activity are used to develop enzyme mixtures. By optimizing the enzyme ratios in mixtures, Sandia researchers can study the change in glucose yields to maximize the output while minimizing the total enzyme dose, resulting in reduced costs.



An enzyme mixture developed by Sandia researchers that functions optimally at 70 °C and 20% of the ionic liquid 1-ethyl-3-methylimidazolium acetate.

Enzyme research is focused on:

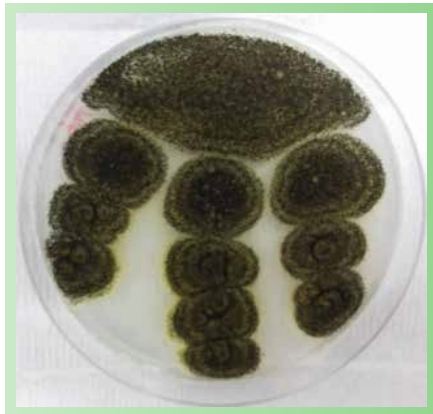
- Identifying and characterizing enzymes with a wide range of activity profiles
- Tailoring enzyme mixtures to meet any set of biorefinery conditions
- Analyzing enzyme activity to understand protein features that impart stability in the presence of ionic liquids at extreme temperatures and pH.

One Pot Conversion Process

Using enzymes tailored to survive in biorefinery conditions, researchers have developed several mixtures that catalyze the conversion of lignocellulose that is pretreated with ionic liquid. This approach shortens the conversion time and reduces the costs at scale. The ability of the enzymes to function under these conditions facilitated development of a “one-pot” simultaneous pretreatment and saccharification (breaking down carb into sugar) process in which the enzymes are added directly into the pretreatment reaction.

Enzyme Database

Sandia researchers at JBEI are developing a database of enzymes that have been characterized across a matrix of biorefinery conditions including temperature, pH, salinity and ionic liquid concentrations. This database will be used to understand the protein features that impart stability in the presence of ionic liquids at extreme temperatures and pH.

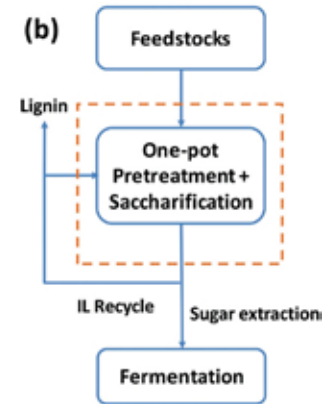


*Commercial enzyme mixtures are typically based on the secreted enzymes of fungi such as *Trichoderma reesei* (*Hypocrea jecorina*). These mixtures are limited to the conditions in which the organism lives.*

Current Process



New One Pot Process



Accelerating Innovation at JBEI

The Joint BioEnergy Institute (JBEI) is a U.S. DOE BioEnergy Research Center in Emeryville, California that is investigating the efficient conversion of lignocellulosic biomass into fuels. Sandia joined Lawrence Berkeley, Lawrence Livermore and Pacific Northwest national labs, the UC Campuses of Berkeley and Davis and the Carnegie Institution of Science in the formation of JBEI. Currently, Sandia researchers lead the deconstruction division that is focused on liberating sugars from biomass.

Partnership Opportunities Available

Sandia has a variety of current enzymes mixtures available for license. Customized enzyme mixtures tailored to meet the specific requirements of industry applications can also be developed in partnership with industry. Sandia’s research involves the entire enzyme mixture development process allowing for cooperative research from enzyme characterization to enzyme mixture optimization.

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