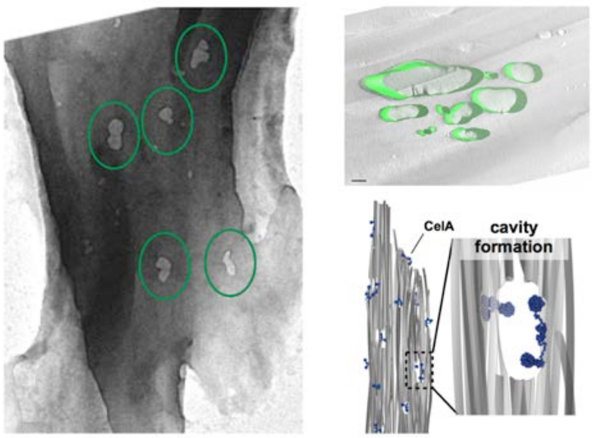
NREL
NATIONAL RENEWABLE ENERGY LABORATORY

Advantages of Enzyme Could Lead to Improved Biofuels Production

Highlights in Science

Cellulase *C. bescii* CelA, a highly active and stable enzyme, exhibits a new cellulose digestion paradigm promoting inter-cellulase synergy.



Transmission electron micrographs and schematic of partially digested small Avicel particles. Particles digested to approximately 65% conversion with CelA display surface cavities of various sizes. All scale bars are 500 nm. Image by Bryon Donohoe, NREL

C. bescii CelA, a hydrolytic enzyme with multiple functional domains, may have several advantages over other fungal and bacterial cellulases for use in biofuels production: very high specific activity, stability at elevated temperatures, and a novel digestion mechanism.

A research team from the U.S. Department of Energy's BioEnergy Science Center, which comprised scientists from the National Renewable Energy Laboratory (NREL) and the University of Georgia, isolated the thermophilic cellulase CelA from *C. bescii*. A comparison was conducted of its cellulolytic activity with that of a binary mixture containing both *T. reesei* Cel7A exoglucanase and *A. cellulolyticus* Cel5A endoglucanase on several substrates. The researchers also compared the cellulose digestion mechanisms of these two enzyme systems using electron microscopy and modeling.

CelA was shown to retain high activity at all temperatures tested, converting 60% of glucan at 85°C compared to 28% glucan conversion for the common exo/endo cellulase standard mixture, Cel7A/Cel5A, at its optimal temperature of 50°C. This difference in activity translates to a seven-fold increase in activity for CelA at the molecular level.

Transmission electron microscopy studies of cellulose following incubation with CelA suggest that CelA is capable of not only the common surface ablative mechanism driven by general cellulase processivity, but also of excavating extensive cavities into the surface of the substrate. Additionally, during the digestion experiments, CelA achieved 60% conversion of xylan in native switchgrass, showing its potential for industrial processes using mild or no pretreatment.

Technical Contact: Yannick Bomble, *yannick.bomble@nrel.gov*

Reference: Brunecky, R.; Alahuhta, P.; Xu, Q.; Donohoe, B.; Crowley, M.; Kataeva, I, Yang, SJ, Adams, M; Lunin, V.; Himmel, M.; Bomble, Y. (2013). "Revealing Nature's Cellulase Diversity: The Digestion Mechanism of *Caldicellulosiruptor bescii* CelA." *Science* (342:6165); pp. 1513–1516. DOI: 10.1126/science.1244273.

Key Research Results

Achievement

The research team isolated CelA, a highly active cellulase with a novel cellulose digestion mechanism. The X-ray structures of the primary protein components of CelA were also determined, advancing the understanding of the mode of action of this cellulase.

Key Result

CelA was shown to retain high activity at all temperatures tested, converting 60% of glucan compared to 28% glucan conversion for the more common exo/endo cellulase standard mixture, Cel7A/Cel5A, at its optimal temperature of 50°C.

Potential Impact

CelA and similar multi-functional cellulases represent a new and distinct paradigm for cellulose digestion. This mechanism is fundamentally different from conventional cellulases and could help increase inter-cellulase synergy in consolidated bioprocessing microorganisms, as well as in commercial cellulase formulations used for biofuels production.

NREL is a national laboratory of the U.S. Department of Energy, Office of Energy Efficiency and Renewable Energy, operated by the Alliance for Sustainable Energy, LLC.

15013 Denver West Parkway
Golden, CO 80401
303-275-3000 | www.nrel.gov

NREL/FS-2700-61022 | January 2014

Printed with a renewable-source ink on paper containing at least 50% wastepaper, including 10% post consumer waste.