Creation of a functional hyperthermostable designer cellulosome

Background

- Understanding and improving biomass deconstruction remains an important research area to enable the cost competitive production of biofuels and biochemicals from recalcitrant lignocellulosic feedstocks.
- Thermophilic cellulolytic bacteria are among the most efficient biomass degraders. One, *Clostridium thermocellum,* uses a cellulolytic system relying on tethered cellulases forming a complex called the cellulosome.
- Cellulosomes from *C. thermocellum* are very efficient but have an optimal temperature of 60°C and quickly degrade at higher temperatures. No other cellulosomes have been shown to function at higher temperatures.

Approach

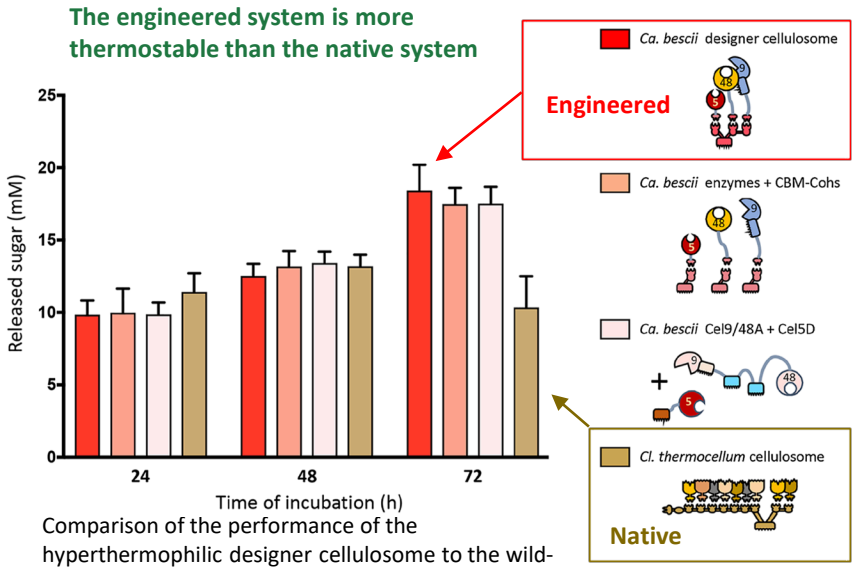
- We isolated divergent protein binding platforms and cellulase domains from hyperthermostable microbes to construct synthetic cellulosomes.
- These synthetic cellulosomes were compared to native cellulosomes and other synthetic cellulosomes using components from different clostridia.

Outcomes

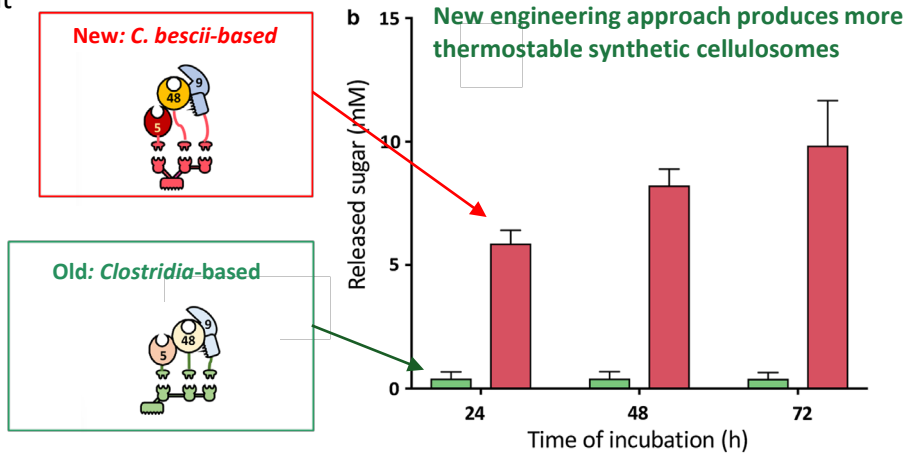
- The new synthetic cellulosomes are hyperthermostable and exhibit the optimal enzymatic activity at 75°C.
- These synthetic cellulosomes are more active than either previously reported designer cellulosome systems or native cellulosomes from *C. thermocellum.*

Significance

- This is the first example of engineered synthetic cellulosomes that can efficiently operate at 75°C.
- The efficiency of these synthetic cellulosomes at high temperature makes them good candidates for enhancing cellulolytic activity in hyperthermophiles, or thermophiles that could operate in harsh environments (e.g. low pH, high salt).



Comparison of the performance of the hyperthermophilic designer cellulosome to the wild-type system at 75°C.



Comparison of the activity of *Clostridia*-based designer cellulosome (green bars) versus hyperthermophilic *C. bescii*-based designer cellulosome (red bars) at 75°C

A. Kahn, S. Moraïs, A.P. Galanopoulou, D. Chung, N.S. Sarai, H. Neal, M.E. Himmel, Y.J. Bomble, and E.A. Bayer, "Creation of a functional hyperthermostable designer cellulosome", *Biotechnol. Biofuels* 12:44 (2019)

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