Characterization and directed evolution of carbohydrate-binding modules (CBMs) for biomass conversion

by Vincent Eijsink — last modified 2011-07-26 09:26
History

Action	Performed by	Date and Time	Comment
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Vista 6505, "Characterization and directed evolution of carbohydrate-binding modules (CBMs) for biomass conversion" Progress report July 1, 2011

Report VISTA, Jan – June 2011

Project title: Characterization and directed evolution of carbohydrate-binding modules (CBMs) for biomass conversion

Project director:	Eijsink, Vincent; Norwegian University of Life Sciences (UMB)
Post-doc/ scholar:	Forsberg, Zarah
Project duration:	01.01.10 – 31.12.13 (75 % position)
Technical contact person in Statoil:	Kotlar, Hans-Kristian
Division head:	Kotlar, Hans-Kristian
Project number:	6505

Objectives:

Major objective: Development of non-hydrolytic helper proteins, in particular CBP21-like proteins, for more effective enzymatic conversion of lignocellulosic biomass.

Sub-goals:

1. Finding, expressing and characterizing naturally occurring proteins and domains that act synergistically with cellulases during enzymatic degradation of lignocellulose.

2. Generate CBMs that act synergistically with cellulases during enzymatic degradation of lignocellulose by protein engineering and directed evolution of proteins such as CBP21.

3. In-depth characterization of promising natural or engineered CBMs, and evaluation of their potential in industrial enzymatic conversion of lignocellulosic biomass.

Nb. ”CBM” stands Carbohydrate-Binding Module (“module” = “domain”); CBP21 is a one–domain protein consisting of one CBM, classified as family 33 (CBM33).

The overall goal is to make enzymatic conversion of lignocellulosic materials more effective. We are currently focusing on characterization of natural proteins (CBMs) and only a little on protein engineering and directed evolution, compared to the original plans. This is due to recent developments and discoveries (see below, under publications), which indicate that there are high-potential natural CBM proteins out there. We have had major breakthroughs in this area that are currently being investigated further.

Status:

In the past half year, we have only been working on Sub-goals 1 and 3. We have overexpressed several CBP21-like proteins selected from bacterial sources. Some of these are one-domain proteins comprising only a CBM33 module/domain. Others are two-domain proteins containing a (putative) cellulose-binding domain in addition to the CBM33 domain. Characterization of the properties and potential of these proteins is currently in progress. Most importantly, we have found several CBM33-containing proteins that work on cellulose. These proteins (enzymes) cleave cellulose using the oxidohydrolytic mechanism described in our 2010 Science paper and they act synergistically with cellulases. This is of major importance for the bioenergy field. As planned, two of the most promising CBMs have been selected for more in-depth characterization, including studies of the redox mechanism, catalytic efficiencies on various substrates and metal dependencies, as well as structural studies and studies on the importance of additional domains. The Vista scholar may have a research stay at the University of Minneapolis later during the project, but this has become less logical due to our recent findings (which are real breakthroughs in the field). Other options for research stays abroad are currently being considered.

Publications:

There are no publications yet, but the research group published a paper in Science in October 2010 about the type of proteins that the Vista scholar is working on: “An oxidative enzyme boosting the enzymatic conversion of recalcitrant polysaccharides.”, by Vaaje-Kolstad G, Westereng B, Horn SJ, Liu Z, Zhai H, Sørlie M, Eijsink VGH, published in Science, 2010, 330:219-222. The Vista scholar is further developing these findings and a first (breakthrough) paper, on the cellulose-activity of CBM33s, has been submitted for publication. The work has been and will be presented at several meetings in 2011, among which is the highly important Gordon Conference on Cellulosomes, Cellulases & Other Carbohydrate Modifying Enzymes

in July 2011. Here the scholar will present a poster whereas one of her PhD supervisors, Gustav Vaaje-Kolstad, will give an invited talk.

Relevant presentations in the first half of 2011:

· 23.2.2011, Gustav Vaaje-Kolstad, Novel enzymes in biotechnology - an academic approach. Invitert foredrag. Pre-conference Workshop 1, Bioprosp 2011, Feb 23, Tromsø.

· 28.3.2011, Vincent Eijsink, Novel insights into the enzymatic degradation of recalcitrant polysaccharides: The role of processivity and discovery of oxidative enzymes boosting the conversion process; Invited talk at Genencor, Palo Alto, CA

· 29.3.2011, Vincent Eijsink, Novel insights into the enzymatic conversion of recalcitrant polysaccharides; invited talk at the Energy Bioscience Institute, Berkeley, CA

· 30.3. 2011, Vincent Eijsink, Novel insights into the enzymatic degradation of recalcitrant polysaccharides: The role of processivity and discovery of oxidative enzymes boosting the conversion process; Invited talk at Novozymes, Davis, CA

· 15.4.2010, Svein Horn, Gustav Vaaje-Kolstad, Vincent Eijsink; Biomass enzymology and biomass processing at the Norwegian University of Life Science; meeting with IN-NNFCC, Trondheim

· 16.06.2011, ”Team 4 – Lignocellulose to Biofuels”; briefing meeting for the UMB-UMN transatlantic collaboration, Ås.