Metagenomics Can Identify New Enzymes for Degrading Plant Biomass

UMB participates in an international network sequencing microbe populations in Svalbard Reindeer.

Fig. 1. Tammar wallaby.

The use of biofuels for energy production is an interesting topic worldwide because it has implications for energy security issues of nations and also because of concerns about CO2 emissions from burning of fossils fuels. There is a general agreement among experts that efficient industrial-scale processing of biomass will require new technologies with novel enzymes having higher efficiency and lower costs, especially enzymes for degrading recalcitrant energy sources such as plant cell walls composed of cellulose microfibrils, other polysaccharides and lignins.

This is where metagenomics comes into the picture. In a collaboration involving Max Planck Institute, the University of Tromsø, the University of Copenhagen and CSIRO in Australia, UMB is leading a metagenomics project that is characterizing the microbial metagenome of Svalbard Reindeer (Rangifer tarandus platyrhynchus), arctic animals that survive on a diet consisting of difficult to digest plant sources such lichens, mosses and shrub species (Fig. 1).

According to post-doc Phil Pope at UMB, 'The potential exists to find novel enzymes for processing biofuels by large-scale analysis of microbial DNA sequences in the guts of these animals. This is metagenomic bioprospecting for future biofuel technologies.' Pope is well placed to be a key member of this team. A native of Australia, he was first author on a Science paper (Science 333 (2011) 646) that reported on microbial species sequences in the Tammar wallaby (Macropus eugenii), an animal that produces considerably less methane compared to domesticated livestock during the digestion and processing of feed. Based on a total sequence length in the metagenome of 82.7 gigabases (82.7 x 109 bp), this study was able to assemble a nearly complete contig genome corresponding to 1,995,748 bp and containing several genes presumably involved in energy metabolism. Based on this knowledge, the team successfully developed a medium composed of starch and urea that was used to isolate this dominant microorganism in the wallaby gut and grow it in pure culture. The genome size of the microorganism grown in pure culture was 2,789,040 bp. A model for key aspects of metabolism used to isolate the low-methane producing microorganism from the Tammar wallaby is shown in Fig. 2.

In relation to progress on the metagenome of Svalbard Reindeer, Phil Pope reports that extensive sequence information has already been generated and analyzed together with Alice McHardy, a bioinformatics expert at the Max Planck Institute in Saarbruken. Alice was an invited Plenary Speaker at the 2012 NBS Contact Meeting. In an article accepted for publication in PLoS One, the team reports on metagenome sequencing using 454 pyrosequencing technology (approximately 80,000 reads) to generate assembled genes corresponding to polysaccharide utilization loci-like systems (PULs) as well as members of more than 20 glycoside hydrolase and other carbohydrate-active enzyme families for polysaccharides such as cellulose, xylan and pectin. Interestingly, the metagenome appears to be devoid of other enzymes thought to be involved in cellulose degradation, suggesting that novel processes are occurring in the guts of Svalbard reindeer.

Phil Pope

In addition to Phil Pope, Alisdair Mackensie, who is also from Australia, is working on the Svalbard Reindeer project in Vincent Eijsink's lab at UMB. Mackensie is contributing results on the biochemical function of enzymes discovered during the course of the project.

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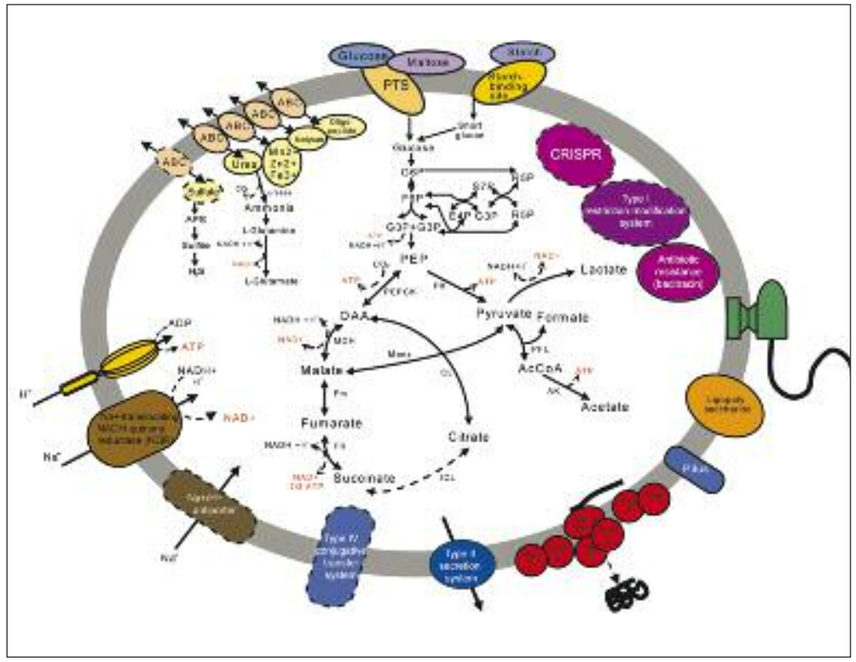

Fig. 2. Key aspects of metabolism. Metagenomic data predicted that starch would be the primary carbon source and that glucose would be metabolized via fermentation to succinate (via PEP and OAA), acetate, lactate and formate. Sequence information also indicated that the microorganism had a urease gene cluster encoding 13 genes required for urea transport and catabolism. Finally, it was predicted that a bacitracin resistance gene existed in the genome. The medium used to isolate this microorganism in pure culture contained starch and urea as the sole carbohydrate and nitrogen sources, respectively, as well as the antibiotic bacitracin. Abbreviations are as follows: AcCoA, acetyl-coenzyme A; AK, acetate kinase; APS, adenylylsulfate; CL, citrate lyase; E4P, erythrose-4-phosphate; F6P, fructose-6-phosphate; Fm, fumarase; FR, fumarate reductase; G3P, glyceraldehyde-3-phosphate; G6P, glucose-6-phosphate; ICL, isocitrate lyase; MDH, malate dehydrogenase; Menz, malic enzymes; OAA, oxaloacetate; PEPCK, PEP carboxykinase; PFL, pyruvate formate-lyase; PK, pyruvate kinase; PTS, phosphotransferase system; R5P, pentose-phosphates; S7P, sedoheptulose-7-phosphate.

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