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ORAL SESSION 6

[12] SELECTION AND DOMESTICATION OF NOVEL ENVIRONMENTAL BACTERIA FOR THE VALORIZATION OF LIGNOCELLULOSIC BIOMASS

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Currently most fermentation products are produced from first-generation biomass by well characterized and genetically modifiable model strains, often referred to as cell-factories. To efficiently valorise second-generation biomass using fermentation, strains are needed that can grow on the diversity of sugars and tolerate the inhibitory compounds present in this substrate, traits that are often challenging to engineer in model organisms.

In this project we have isolated bacteria from soil samples that are tolerant to hemicellulosic biomass hydrolysate from beech wood. This has yielded several novel environmental strains that are able to grow on and tolerate this generally toxic substrate. The most interesting isolates are a novel species of *Pseudomonas* and a novel strain of the *Pantoea* genus. Both strains show relatively high tolerance to the inhibitors present and can grow on a wide range of different substrates. The genome sequences of these strains have been determined using nanopore sequencing and basic physiological features have been characterized. The *Pseudomonas* strain has been shown to grow in a wide pH range; from pH 3 up to a pH of 9, which could be an advantage depending on the substrate and product. Both strains showed great robustness and tolerance to high density fermentation in a bioreactor, indicating a large potential for industrial applications.

In order to use these novel strains to produce valuable compounds from lignocellulosic biomass they need to be modifiable and domesticated. To do this, a wide range of molecular tools must be identified and developed. We first established efficient transformation protocols for both strains. We have characterized a number of different constitutive and inducible promoters, a prerequisite for further development of efficient engineering tools. We are currently in the process of developing molecular tools for gene deletions and integrations to improve the industrial applicability of these novel strains.

The discovery, isolation, characterization and modification of these novel strains could help in enabling efficient production of chemicals from lignocellulosic biomass.