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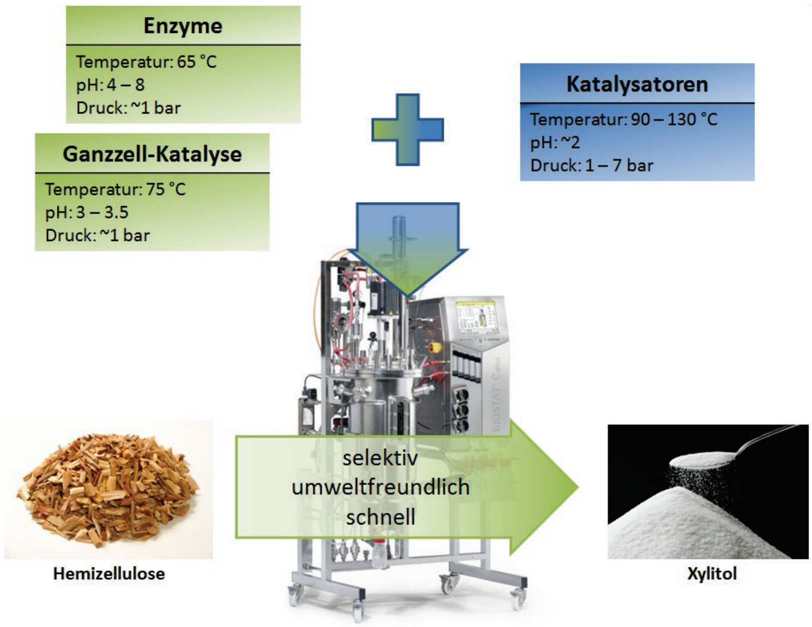
Hot and Sour

Symbiosis of chemical and biological catalysis in the biorefinery of the future

As basis for a sustainable industry and as a contribution to the independence of fossil raw materials, it is necessary to use current waste streams in an eco-friendly and cost-effective way. For example, hemicellulose, which incurs as a by-product of the pulp and paper industry, has only rarely been integrated into any further value chain so far.



Novel hybrid process for the eco-friendly conversion of hemicellulose into xylitol

Objective

The objective of the research has been to develop a new biorefinery hybrid process that combines chemical catalysts, enzymes and extremophiles (microorganisms / whole cells). This new process should be preferably realised in an one-pot reaction and enable simple re-use of valuable enzymes and cofactors, as well as a continuous generation of products.

A cost and resource efficient method for the production of the sugar substitute xylitol from the waste material hemicellulose will form the basis for the creation of a widely applicable platform technology. Xylitol is highly solicited as a sugar substitute in the food and chemical industry, because despite its high sweetness intensity, it only causes low insulin release and at the same time helps to prevent tooth decay. The conventional chemical production process of xylitol is rather ineffective and very expensive, which is why a more efficient procedure would also be interesting for economic reasons.

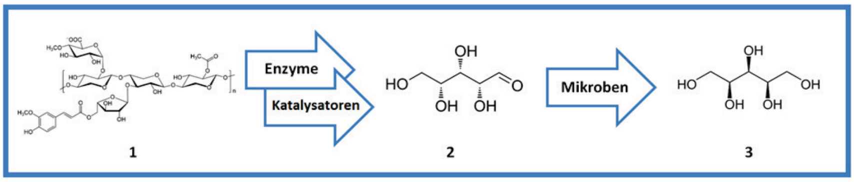
Approach

The transformation of hemicellulose into xylose is conventionally achieved by use of acid at high temperatures. This leads to the formation of inhibitors, which in turn requires large amounts of neutralising reagents. The conversion from xylose into xylitol is then accomplished with a catalyst made from Raney nickel, via a chemical hydrogenation at 50 bar pressure. Raney nickel, however, has a porous structure, which poses significant potential risk, e.g it is self-igniting when in contact with air. Overall, this is an energy-intensive and expensive process. In contrast, TU Wien uses the advantages of a chemical, enzymatic and whole cell catalysis in an one-pot reaction. The enzymes and cofactors necessary for the reaction can be generated in situ by purposeful strain development and by the use of extremophilic organisms, which enjoy ideal growth conditions at pH values of 3 and temperatures of 75 °C.

The genetic modification of the organisms necessary for the process was achieved by means of a plasmid-based system andthe CRISPR/Cas method.

Results

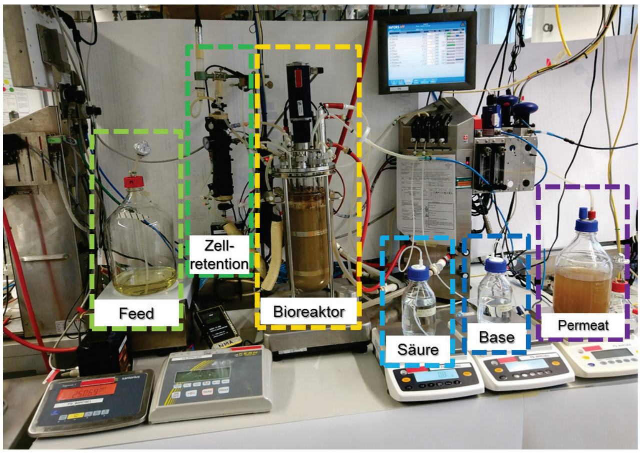
A reactor system for the controlled implementation of cultivation was designed, fully equipped with sensors for optical density, capacity and exhaust gas composition. This system allows the exact



Hemicellulose (1) is degraded to xylose (2) by means of enzymes and catalysts and converted into xylitol via whole cell catalysis (3).

determination of the biomass concentration in real time, and the purposeful readjustment of feeding conditions. To discharge substances from the system which inhibit the process, and to enrich the cells as well as to harvest the product, a cell retention system was included. Through exact characterisation of chemical, enzymatic and biological reactions it is possible to establish a controlled bioprocess for the conversion of hemicellulose.

It was possible to produce a 20 times higher cell density as hitherto reported in scientific literature. A setup in a 3 liter scale was built, which enables the determination and optimisation of process parameters for further tasks.



Bioreactor – with regulated gassing, stirring, temperature, pH value (by addition of acid and base).
Substrate is added continuously, cell retention allows products to be harvested and inhibiting substances to be discharged (permeate).

Benefits

For the first time, a cost and resource efficient one-pot method for the production of the sugar substitute xylitol from the waste material hemicellulose can be offered. Looking ahead, the transformation of other solid raw materials into other products by means of the developed platform technology is also on the agenda.

The particular characteristics of this one-pot method from TU Wien are:

- first hybrid bioprocess that transforms hemicellulose into valuable products in a highly energy efficient one-step process
- first-time combination of the advantages of whole cells, enzymatic and chemical catalysts
- first-time application of the species *Sulfolobus* as whole cell catalysts
- use of vectors systems and the CRISP/Cas method to generate recombinant variants of *Sulfolobus*
- from the hot acid acidulation directly to the production of valuable substance in an 'one-pot' reaction – no energy-intensive cooling of the substrate solution necessary
- designer microorganisms that can efficiently convert substrates into products under extreme conditions (70-80 ° C, pH 2.5-3.5)
- gentle treatment of waste to gain usable substrate solutions, preventing the formation of molecules that reduce the growth of the microorganisms (inhibitors)
- 'waste to value' platform technology, which enables the transformation of different types of solid organic waste into valuable products
- vast experience of researchers at this department of TU Wien with parametrization and scaling-up of industrial bioprocesses

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