GLYCEROL BIOCONVERSION FOR 3-HYDROXYPROPIONIC ACID PRODUCTION THROUGH AN INTEGRATED PROCESS INCLUDING IN SITU PRODUCT RECOVERY

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Abstract Submission

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GLYCEROL BIOCONVERSION FOR 3-HYDROXYPROPIONIC ACID PRODUCTION THROUGH AN INTEGRATED PROCESS INCLUDING IN SITU PRODUCT RECOVERY

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Submit your abstract below (400 words): Within the framework of the development of the bioeconomy, there is an increasing drive towards the production of chemicals from renewable biomass resources. In this respect, interest in the sustainable production of biobased building blocks, such as the bifunctional carboxylic acid 3-hydroxypropionic acid (3-HP), is growing. The development of a biotechnological process to convert glycerol (by-product of biodiesel industries) into the platform chemical 3-HP is a key issue. Among its various applications, the 3-HP can be used for the synthesis of biobased polymers and acrylic acid[1].
Lactobacillus reuteri is known as a natural producer of 3-HP from glycerol although at low yield and productivity, due to the inhibitory effects of the product and of its metabolic intermediate 3-hydroxypropionaldehyde, as well as the formation of 1,3-propanediol as co-product[2].
The first aim was to improve the performance of 3-HP production by regulating operating parameters as pH (free or controlled at 6, 5, 4 or 3), stirring (100 or 250 rpm) and bacterial concentration. The pH control allowed maintaining the cell physiological state compared to a free-pH bioconversion as evaluated by flow cytometry. In fact, the *L. reuteri* viability (esterase activity assimilated to enzymatic activity) was assessed by cell staining with the fluorescent probe cFDA (carboxyfluorescein diacetate) and the bacterial membrane integrity was checked using PI (propidium iodide). No 3-HP production was observed at pH 3. Between pH 6 and 4, the production remained low probably due to inhibition phenomena, with only slight differences between the three conditions. We proposed to perform a controlled glycerol fed-batch to avoid 3-HPA accumulation.
Moreover, original results about the 3-HP inhibition mechanisms were obtained thanks to flow cytometry analyses and Fourier-Transform InfraRed (FTIR) spectroscopy, the latter allowing to study the cell biochemical composition. For the first time, we demonstrated that 3-HP induces damages on the secondary structure of cell proteins (among which enzymes) and alterations of nucleic acids.
In situ product recovery (ISPR) was also performed to extract 3-HP along its production. The ISPR approach has been successfully applied to the production of some solvents, organic acids, aromas and fine chemicals[3]. We used a membrane-assisted reactive extraction with long-chain tertiary and quaternary amines and decanol as diluent, a promising technique that drew a lot of attention for the extraction of hydroxypropionic acids[4]. A challenging bottleneck that has to be addressed deals with this process biocompatibility, studied by flow cytometry. The quaternary amines were shown to induce the highest toxicity compared to decanol with or without tertiary amines.
Our work thus underlines the interest of a smart coordination of the upstream reaction and downstream technologies.

Type of presentation :: No preference

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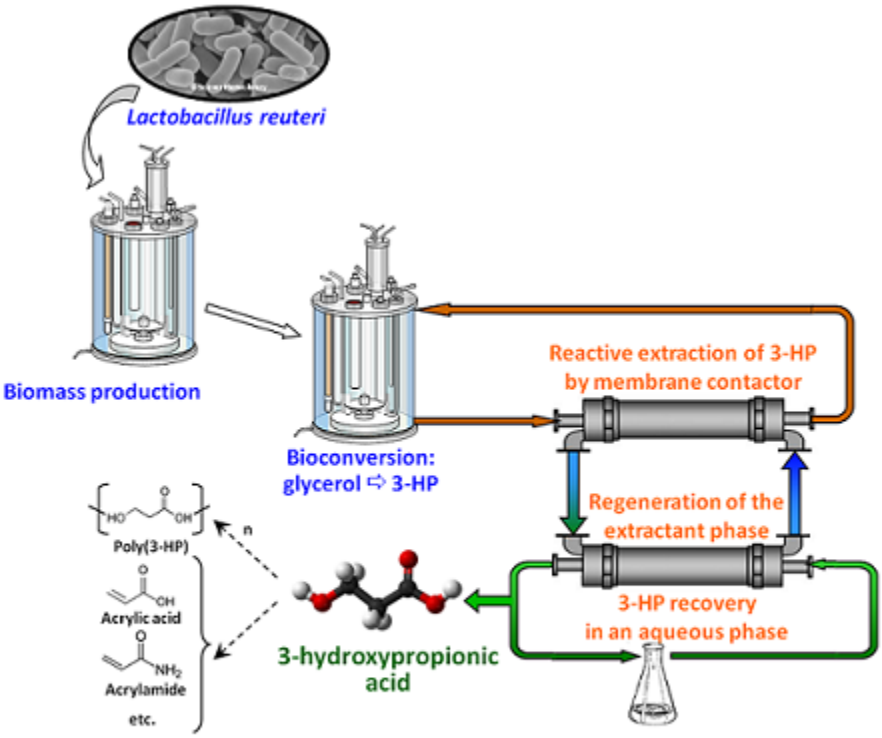

Figure: Integrated process for 3-hydroxypropionic acid biotechnological production

Highlight 1: Bioprocess control to increase the 3-hydroxypropionic acid (3-HP) production.

Highlight 2: In situ extraction to avoid product inhibition of bacteria.

Highlight 3: Physiological state of Lactobacillus reuteri subjected to 3-HP and to extraction process.

Reference 1 :: Choi s. Song cw. Shin jh. Lee sy. (2015). Biorefineries for the production of top building block chemicals and their derivatives. Metabolic engineering, 28:223–239

Reference 2 :: Dishisha T, Pereyra L, Pyo S, Britton R, Hatti-Kaul R (2014). Flux analysis of the Lactobacillus reuteri propanediol-utilization pathway for production of 3-hydroxypropionaldehyde, 3-hydroxypropionic acid and 1,3 propanediol from glycerol. Microbial Cell Factories, 13:76-85.

Reference 3 :: John M Woodley, Marc Bisschops, Adrie J J Straathof and Marcel Ottens (2008). Future directions for in-situ product removal (ISPR). Journal of Chemical Technology and Biotechnology, 83(2):121–123.

Reference 4 :: Kyuchoukov G and Yankov D. (2012). Lactic acid extraction by means of long chain tertiary amines: a comparative theoretical and experimental study. Industrial and Engineering Chemistry Research, 51(26): 9117-9122.

Keywords: Bacteria, Biomolecules, Bioprocess, Extraction