

## Balancing act

### improving the environmental and economic performance of industrial-scale basic oxygen furnace gas fermentation for isopropyl alcohol production

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hydrophilic sophorose moiety. Despite the gaining interest for and the research performed on this yeast, *S. bombicola* still is an unconventional organism, with rather limited engineering possibilities. Therefore in this work, standardized parts were developed and tested to enable streamlined engineering of this yeast. These parts include promoters, terminators, signal peptides and anchors amongst others. Several strategies to enable inducible expression of genes in *S. bombicola* were also developed. Using this approach, hydrolase expression to enable growth on different substrates was achieved and pathway balancing is underway to obtain higher sophorolipid productivities. This will pave the way to easier engineering of *S. bombicola* and enable competitive production of biosurfactants with traditional surfactants.

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IB-016

Balancing act: improving the environmental and economic performance of industrial-scale basic oxygen furnace gas fermentation for isopropyl alcohol production

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The iron and steel industry emits 30% of all industrial CO<sub>2</sub> emissions, mainly due to hot Basic Oxygen Furnace (BOF) gas (CO, H<sub>2</sub>, CO<sub>2</sub>) from steel production. BOF gas can be converted via gas fermentation into valuable chemicals such as the platform chemical isopropyl alcohol (IPA), currently only produced through petrochemical cracking. This study investigated the economic and environmental trade-offs to improve the plant-level performance of the industrial-scale BOF gas fermentation to IPA. The fermentation is based on the LanzaTech process with *Clostridium autoethanogenum*, and IPA is purified through extractive distillation with glycerol. The effect of eleven process parameters on the economic (total investment costs, minimal selling price) and environmental (global warming potential, water consumption, energy consumption, waste production) KPIs were studied. These process parameters are: the product selectivity (YIPA/CO), CO volumetric mass transfer rate (VMTCO), CO conversion, reactor dilution rate (D), temperature off-gas condenser, biomass separation liquid loss, extractive distillation glycerol mole fraction and molar reflux ratio, purge of glycerol and broth recycle, and anaerobic digestion efficiency. Our analysis highlights the gas fermentation parameters (CO conversion, VMTCO, YIPA/CO, and D) and the biomass filtration liquid loss as the most influential on the economic and environmental process performance. Here, increasing CO conversion, VMTCO, YIPA/CO, whereas decreasing D and biomass filtration liquid loss led to increased economic and environmental performance. But, combining them to increase performances causes trade-offs. These findings can be used to optimise both the sustainability and economic viability of the BOF gas fermentation to IPA process.

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IB-017

Strategies for the scale-up of the bioproduction of the biodegradable herbicide D-DIBOA

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The benzohydroxamic acid DIBOA is an allelochemical with herbicidal properties, high biodegradability, and a low environmental impact but its low natural production is a handicap for practical application. D-DIBOA, a chemical analog, can be synthesized in two consecutive reactions. However, the second step, which uses an expensive catalyst and is an exothermic reaction with hydrogen release and 70% molar yield, hinders the scale-up of this synthesis.

The use of an engineered *E. coli* strain overexpressing the autologous nitroreductase enzyme NfsB as a whole-cell biocatalyst can get over the drawbacks of the chemical synthesis and is in line with the principles of green chemistry. For that, several strategies have been implemented to increase the feasibility of scaling up the bioproduction of D-DIBOA.

Firstly, we screened *E. coli* single mutants overexpressing of NfsB, to investigate the most appropriate genetic background. This strategy allowed us to construct a double mutant strain that increased the biotransformation yield (%BY) from 60% to 76%. Secondly, the study of the culture conditions (temperature, % inoculum, addition of thiamine, and precursor) increased to 90% BY. Additionally, we developed a D-DIBOA quantification method to monitor the biocatalysis in real-time and to design feeding strategies to reach higher % BY and D-DIBOA titers. We achieved 90-100% BY and 7 mM D-DIBOA. These strategies made it possible to approach D-DIBOA production at the bioreactor level.

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IB-018

Green physical processes for the extraction of organic dye betacyanin from dragon fruit (*Hylocereus purpurii*) peels

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Production of plant-based extracts rich in bioactive compounds using green technology has been one of the core applications of biotechnology with emphasis on the valorization of growing agricultural and food wastes. Dragon fruit is known to be a promising super fruit rich in bioactive compounds and nutritional content such