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Continuous Production of 2,3-butanediol from CO_2 by *Cupriavidus necator* gas fermentation

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The world is still producing industrial chemicals from finite resources and emit CO_2 as by-product. To sustain our chemical production and avoid catastrophic climate change, alternative ways of producing chemicals must be implemented. Microbial gas fermentation of CO_2 has the potential of addressing the sustainability challenge of chemical industry. The process uses renewable raw material and is carbon neutral or negative [1]. The rapid advancement of synthetic biology provides tools and strategies to modify the host gas fermentation bacteria for producing various chemicals. This study demonstrates the production of 2,3-butanediol (2,3-BDO), a precursor to plastics and pesticides [2], using gas fermentation of *Cupriavidus necator* from CO_2.

C. necator, capable of chemolithotrophic growth on CO_2, H_2 and air, was modified to host the 2,3-BDO production pathway. The pathways, containing genes: alsS (acetolactate synthase), alsD (acetolactate decarboxylase) from *Bacillus subtilis* and either NADH-dependent BDH (butanediol dehydrogenase) or NADPH dependent sADH (secondary alcohol dehydrogenase) from clostridium, were constructed as an operon under arabinose inducible promoter and integrated to the chromosome.

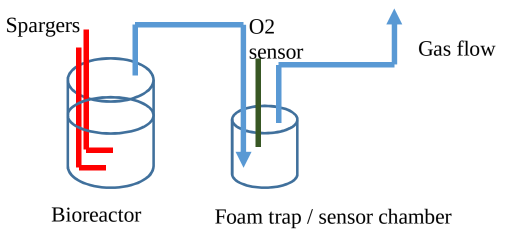

Figure 1. Schematic diagram of the gas fermentation process.

A phosphorous limiting chemostat gas fermentation process was developed to characterize the 2,3-BDO production strains at lab scale. The main features of the process are presented in Figure 1. In this design, H_2 was introduced to the bioreactor through a separate sparger. The headspace O_2 concentration was monitored and controlled at the set point of 4% (below the O_2 percentage corresponding to the upper explosive limit of H_2). A PID controller was set up to regulate the air flow rate by the headspace O_2 concentration. This improved the oxygen transfer rate, and allowed high cell density (maximum nearly 50 g dry cell weight/L) at steady state. 2,3-BDO concentration in the two different strain variants during the steady state was 7 g/L, corresponding to a productivity of 0.35 $g/L^{-1}h^{-1}$. This is the highest productivity of 2,3-BDO reported to date in autotrophic cultures on CO_2 of *C. necator*. And the process is still being optimized.

The results suggest the biochemical production of 2,3-BDO can be further optimized to a viable industrial process. More importantly, as a successful case study, producing chemicals from CO_2 through gas fermentation was demonstrated to be a promising alternative to conventional petrochemical processes. The understanding and ability to manipulate biological metabolism will continue to expand the product portfolio and market of chemicals produced from CO_2.

Acknowledgments

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References

[1] Md. S. Islam Mozumder, L. Garcia-Gonzalez, H. De Wever, E.I.P. Volcke, *Biochem. Eng. J* **2015**, 98, 107.
[2] X. Ji, H. Huang, P. Ouyang, *Biotechnology Advances* **2011**, 29, 351.