

H₂-Oxidizing Bacteria for Single Cell Protein production and sustainable nitrogen cycling

Silvio Matassa^{1,2,*}, Nico Boon², Jan B.A. Arends², Willy Verstraete^{1,2}

¹ Avecom NV, Industrieweg 122P, 9032 Wondelgem, Belgium

² Laboratory of Microbial Ecology and Technology (LabMET), Ghent University, Coupure Links 653, 9000 Gent, Belgium

* Corresponding author. E-mail: Silvio.Matassa@Ugent.be; Phone: +32 468165973

Abstract

A generic mixed microbial community was selectively enriched in order to obtain a H₂-oxidizing bacteria microbiome. The microbiome was characterized in terms of biomass yield, volumetric productivity and gas consumption. The functionality of the microbiome was studied in terms of single cell protein production starting from mineral reactive nitrogen (NH₄⁺-N) and gaseous CO₂, H₂ and O₂. The results obtained in this phase will be used to support the implementation of SCP production directly on-site of WWTP for the recovery and upcycle of used reactive nitrogen into valuable single cell protein.

Keywords

Hydrogen-oxidizing bacteria, single cell protein, nitrogen upcycle.

Introduction

H₂-oxidizing bacteria offer the possibility to autotrophically capture NH₄⁺-N and CO₂ by using H₂ as electron donor and O₂ as electron acceptor. Hydrogen produced by green energy sources such as wind and solar energy powered water electrolysis can potentially support a shift from resource dissipation to resource recovery and upcycle in wastewater treatment. Indeed, instead of dissipating the reactive nitrogen contained in wastewater by means of conventional nitrification-denitrification, the latter can be instead recovered and upcycled back to high quality edible microbial protein suitable as protein-rich feed additive: single cell protein (SCP). The process offers also the possibility of capturing the CO₂ contained e.g. in the biogas produced during anaerobic digestion, therefore reducing overall the greenhouse gas emissions (GHG) of wastewater treatment plants (WWTP). The present research focuses on enriching, characterizing and implementing in practice (SCP production from lab to pilot scale) a microbiome where autotrophic H₂-oxidizing bacteria constitute the core (primary consumers), supported in its functionality by other heterotrophic bacteria. As already demonstrated for methane oxidizing bacteria (Ho et al., 2014), the presence of heterotrophic satellites (secondary consumers) can help increasing the functionality of the microbiome by e.g. removing inhibitory metabolites or regulating oxygen levels.

Main results and observations

The aim of this first experimental phase was to selectively enrich a mixed microbial community with H₂-oxidizing bacteria. A first set of experiments was carried out in a lab-scale setup implementing a closed 0.5 L fermenter operating in batch conditions. The closed-gas recirculation system was set and operated at the optimal physical-chemical conditions reported in literature for H₂-oxidizing bacteria.

The main parameters affecting the microbial growth were studied, aiming at the highest biomass yields. The parameters studied were: sludge retention time (SRT), O₂ concentration, gas recirculation rate, macro and micro nutrients concentration; mixotrophic conditions. This first experimental phase allowed to have the proof of concept that a mixed culture can be enriched in H₂-oxidizing bacteria, able to capture NH₄⁺-N and CO₂ into new cell material. The efficiency of the process was monitored mainly in terms of biomass yield, defined as the cell dry weight (CDW) produced per gram of hydrogen COD (H₂-COD). The yield varied from a minimum of 0.10 g to a maximum of 0.25 g CDW/g H₂-COD, approaching the maximum yield of 0.30 g CDW/g H₂-COD

reported in literature for axenic cultures of H₂-oxidizing bacteria (Matassa et al., 2015). Once the mixed microbial community became highly enriched in H₂-oxidizing bacteria, further studies were conducted by implementing a 5 L Sartorius Biostat-A reactor operating under continuous gas feeding. This was done in order to study the behaviour of the enriched community under different operating conditions and to estimate the first important biotechnological process parameters for SCP and biopolymer production. Hydrogen gas was supplied by a GC-grade hydrogen generator directly connected to the reactor. Carbon dioxide and oxygen were fed respectively by CO₂ gas bottles and compressed air. In this phase maximum biomass concentration and volumetric productivity of the H₂-oxidizing bacteria microbiome were investigated. In summary, biomass concentrations as high as 15 g CDW/L were achieved, with volumetric productivities of the order of 2-3 g CDW/L·d and a specific biomass yield of about 0.2 g CDW/g H₂-COD. The produced biomass was also characterized by an average crude protein level of about 70% CDW, whereas the average PHA (polyhydroxyalkanoate) content of the biomass ranged between 20 and 25% of the cell dry weight. In order to assess the quality of the protein accumulated by the microorganisms, the biomass was harvested, dried and analysed for essential amino acids compositions.

The amino acid profile of the single cell protein produced approached the high-quality profile of fishmeal, which can be regarded as the higher standard animal protein commercially available. Moreover, the prebiotic properties of the biopolymers accumulated within the cells increase the nutritional value of the produced microbial biomass.

In conclusion, the production of edible microbial protein by the HOB microbiome developed so far can be regarded as a mean to supply high value protein while incorporating reactive nitrogen (possibly recovered from wastewater) and capturing carbon dioxide emissions from the treatment facilities.

References

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