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<AT>Assessment of glycerol usage by five different purple non-sulfur bacterial strains for bioplastic production

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<ABS-HEAD>Highlights ► Phylogenetic tree along with already characterized *Rhodopseudomonas* sp. type strains ► Purple bacteria have been fed with glycerol for producing degradable bioplastics ► Starvation of N-S nutrients induces cumulative PHB in *Rhodopseudomonas* S16-VOGS3

<ABS-HEAD>Abstract

<ABS-P>Five purple bacterial strains were investigated by sequencing the 16S rDNA gene and by screening the strains for glycerol usage. The screening was carried out in order to identify the bacterial capacity to consume glycerol. Of the five examined, only three strains metabolized glycerol. A controlled cylindrical photobioreactor was operated in batch mode for testing the substrate consumption. A combination of glucose and glycerol was also used to increase the glycerol consumption rate (GCR). Using *Rhodopseudomonas* sp. S16-VOGS3, the highest GCR of 14.0 mg/L h was obtained. This most suitable bacteria (*Rhodopseudomonas* sp. S16-VOGS3) was next grown at two consecutive growth-steps: the first, under a standard batch-growth condition; the second was carried out under fed-batch growth mode, after having removed the exhausted medium and replaced it with a fresh one deprived of Nitrogen and Sulphur nutrients. The fed-batch growth mode caused a significant increase of glycerol concentration (>6.0 g/L) in the medium, which decreased when the glycerol addition was stopped. The Nitrogen and Sulphur starvation condition caused a stress in *Rhodopseudomonas* cells, which stimulated poly-3-hydroxybutyrate accumulation. During both steps, no bioH₂ was produced. Nevertheless, when the bacterium was tested under nutrient starvation, the poly-3-hydroxybutyrate content increased by up to 18%, demonstrating that a degradable plastic material could be efficiently produced with the aforementioned two-step procedure. In conclusion, *Rhodopseudomonas* sp. S16-VOGS3 is a potential candidate for employing crude glycerol coming from biodiesel industry, as a suitable feedstock for the production of degradable products such as bioplastics.

<KWD>Keywords: glycerol; bioplastics; photofermentation; purple non sulfur bacteria; substrate consumption rate.

<H1>1. Introduction

Glycerol is the main by-product of oleo chemical production (i.e., fatty acids, surfactants, and soaps) and adds competitive value to oleo chemical processes; glycerol is currently produced in large amounts as a by-product during fat splitting and biodiesel fuel production [1]. Biodiesel production has been growing worldwide over recent years, since it represents one of the promising alternatives to fossil fuels. It is biodegradable, non-toxic and has low pollutant emissions (especially SO_x). However, despite these attractive characteristics, biodiesel production is increasing at a slow

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