World Academy of Science, Engineering and Technology
International Journal of Biotechnology and Bioengineering
Vol:15, No:02, 2021

Exploration of in-situ Product Extraction to Increase Triterpenoid Production in Saccharomyces Cerevisiae

Authors : Mariam Dianat Sabet Gilani, Lars M. Blank, Birgitta E. Ebert

Abstract : Plant-derived lupane-type, pentacyclic triterpenoids are biologically active compounds that are highly interesting for applications in medical, pharmaceutical, and cosmetic industries. Due to the low abundance of these valuable compounds in their natural sources, and the environmentally harmful downstream process, alternative production methods, such as microbial cell factories, are investigated. Engineered Saccharomyces cerevisiae strains, harboring the heterologous genes for betulinic acid synthesis, can produce up to 2 g L-1 triterpenoids, showing high potential for large-scale production of triterpenoids. One limitation of the microbial synthesis is the intracellular product accumulation. It not only makes cell disruption a necessary step in the downstream processing but also limits productivity and product yield per cell. To overcome these restrictions, the aim of this study is to develop an in-situ extraction method, which extracts triterpenoids into a second organic phase. Such a continuous or sequential product removal from the biomass keeps the cells in an active state and enables extended production time or biomass recycling. After screening of twelve different solvents, selected based on product solubility, biocompatibility, as well as environmental and health impact, isopropyl myristate (IPM) was chosen as a suitable solvent for in-situ product removal from S. cerevisiae. Impedance-based single-cell analysis and off-gas measurement of carbon dioxide emission showed that cell viability and physiology were not affected by the presence of IPM. Initial experiments demonstrated that after the addition of 20 vol % IPM to cultures in the stationary phase, 40 % of the total produced triterpenoids were extracted from the cells into the organic phase. In future experiments, the application of IPM in a repeated batch process will be tested, where IPM is added at the end of each batch run to remove triterpenoids from the cells, allowing the same biocatalysts to be used in several sequential batch steps. Due to its high biocompatibility, the amount of IPM added to the culture can also be increased to more than 20 vol % to extract more than 40 % triterpenoids in the organic phase, allowing the cells to produce more triterpenoids. This highlights the potential for the development of a continuous large-scale process, which allows biocatalysts to produce intracellular products continuously without the necessity of cell disruption and without limitation of the cell capacity.

Keywords : betulinic acid, biocompatible solvent, in-situ extraction, isopropyl myristate, process development, secondary metabolites, triterpenoids, yeast
Conference Title : ICYBB 2021 : International Conference on Yeast Biotechnology and Biology
Conference Location : Lisbon, Portugal
Conference Dates : February 08-09, 2021