Eicosapentaenoic acid (EPA) production in silico by *Pythium irregulare*, as value-added product, using sugarcane vinasse as carbon source

Bruna S Fernandes, Oscar Dias, Marcelo Zaiat, José G C Pradella, Isabel Rocha

1 Centre of Biological Engineering, Universidade do Minho, 4710-057 Braga, Portugal
2 Brazilian Bioethanol Science and Technology Laboratory, 13083-970 Campinas, SP, Brazil
3 Biological Processes Laboratory, University of São Paulo, 13563-120 São Carlos, SP, Brazil

This study aims to assess in silico the production of Omega-3, mainly Eicosapentaenoic acid (EPA), as a value-added product, by *Pythium irregulare*, using sugarcane vinasse as carbon and nitrogen source. EPA is a 20-carbon polyunsaturated fatty acid with five cis double bonds, with its first double bond at the third carbon from the omega end, as an Omega-3 fatty acid. The Food and Agriculture Organization of the United Nations recommends ingestion up to 500 mg per day of EPA in the early years of life and for prevention of cardiovascular diseases, since it is not naturally synthesized in humans. EPA is an important dietary supplement, highly expensive ($600 – $4000 per kg of omega-3), with a promising market. The expected Omega-3 revenue is US$ 2.7 billion for 2020, with a Compound Annual Growth Rate (CAGR) of 17.5% (2014-2020), only in the pharmaceutical market.

Sugarcane vinasse is low cost carbon source produced in large amount. In 2019, it is estimated that 413 Brazilian sugarcane and bioethanol mills will produce more than 588 billion litters of vinasse, equivalent amount of the total of sewage produced by the world population (based on COD and Volume).

*Pythium irregulare* is an oleaginous Oomycete, a microscopic Stramenopile, able to accumulate large amounts of lipids, including Eicosapentaenoic acid (EPA). Previous studies have highlighted the promising production of EPA by *P. irregulare*, exploiting diverse low cost carbon sources, which include wastewaters as vinasse from corn-meal bioethanol production, glycerol and several sugars, but not from vinasse of sugarcane bioethanol plants. Moreover, there is still a lack of knowledge about its biosynthetic pathways.

For this propose, the genome-scale metabolic model will be constructed using Merlin, user-friendly software, in order to evaluate and validate in silico EPA and biomass production. Finally, OptFlux software will be applied in order to maximize *Pythium irregulare* Eicosapentaenoic acid and biomass production pathways using sugarcane vinasse as carbon and nitrogen source.

References
