

ABSTRACT

A bioprocess utilizing *Candida bombicola* ATCC 22214 that extracellularly producing sophorolipids for the update of palm oil as a secondary substrate has been investigated. The biosynthesis of sophorolipids biosurfactant, constituting of 2 units of glucose molecules linked to a 16 units of carbon atoms long hydroxylated fatty acid chain, is associated with the microbial growth on glucose as the primary substrate. The objectives of this research project are to optimize the production of sophorolipid biosurfactant by studying the production medium formulations and respective yield in a fractional factorial design. In addition, this research project also studied the physical properties of sophorolipids in structural stability and emulsification.

High yield of sophorolipids was obtained when *Candida bombicola* was grown on production medium that with a higher concentration of palm olein and glucose, which is 100 ml/L and 100 g/L. The process optimization of sophorolipids production was first studied in shake flask and the fermentation cycle was studied in 5 L bioreactor. The highest sophorolipid yield obtained in shake flask is 28.05 g/L. With the optimized medium formulation, a total yield of 68.29 g/L of sophorolipids was obtained in a bioreactor. The recovered sophorolipids showed the same result of structural stability as glycerin in vanishing cream and also demonstrated significant emulsification effect in oil displacement test when compared with glycerin.

In this work, the residual nitrogen was determined using the Dumas method and the Kjeldahl method. The residual nitrogen in the medium had totally depleted after 36 hours of cultivation, from 100% to 22%. The residual glucose was determined using high-performance liquid chromatography (HPLC) analysis. The glucose in the medium was utilized by 24 hours of cultivation when the sophorolipids production began, from 87.2% to 11.4%. Both results indicated the conversion rate of substrates to metabolites and intake of glucose during microbial growth is high and efficient.

Thin layer chromatography (TLC), nuclear magnetic resonance spectroscopy (NMR) and fourier-transform infrared spectroscopy (FTIR) analysis were applied to further confirm the yield product is sophorolipids. A total of 8 anisaldehyde positive spots were detected in TLC. The FTIR data reveals that the absorption peak at 1020 cm^{-1} corresponded to the stretching of C-O-H groups

which is the sophorose moiety of sophorolipids, and thus the crude product obtained in this research project was confirmed are sophorolipids.

Keywords: *Candida bombicola*, sophorolipids, biosurfactant, fermentation cycle, palm olein.