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PUMNAT CHUNCHOMRAT: ISOLATION AND CHARACTERIZATION OF BIOSURFACTANT PRODUCING MICROORGANISM. THESIS ADVISORS : ATTAWUT IMPOOLSUP, Ph.D., VITHAYA MEEVOOTISOM, Ph.D., PRANEE PHINYOCHEEP, Ph.D. 165 P. ISBN 974-663-148-9

The aims of this study are the screening of biosurfactant producing microorganisms and characterization of the chemical structures of the biosurfactants. From the stock cultures of two research projects, organism samples were obtained: Thailand oil degrading bacteria and lipase producing bacteria. Of 194 pure cultures, 95 strains (49%) showed clear zone around colonies on NA-Oil medium at 30 °C for 48 h of incubation. MR1/7-011 (*Bacillus pumilus*) showed the highest ratio of diameter of clear zone to diameter of colony (RZC). After cultivation in liquid medium (NB) at 30 °C, 200 rpm for 72 h, the surface tension of cell free culture broth of MR1/7-011 was reduced from 48.15 to 30.27 mN/m. This strain produced a lipopeptide biosurfactant (MR biosurfactant). The effects of aeration, medium composition and cultivation temperature on biosurfactant production were investigated. The suitable conditions for MR biosurfactant production were carried out in formula 2 medium with working volume of a half filled Erlenmeyer flask at 30 °C. The amino acid composition of MR biosurfactant was Asp-Glu-Val-Ile-Leu with the ratio of 1:1:1:1:3. After analysis by LC/MS and MS, the molecular mass of the major component of MR biosurfactant was 1050. It was higher than that of Surfactin with 14 units. The fatty acid methyl ester (FAME), the major component of MR biosurfactant, analyzed by GC/MS was $C_{13}H_{27}CH(OH)CH_2COOCH_3$. It indicated that MR biosurfactant had one more methylene group (CH_2) in hydrophobic moiety than did Surfactin. MR biosurfactant is composed of 10 derivatives including a Surfactin like structure and 9 other compounds with molecular masses between 1064 and 1176, each differing by 14 units. Evidence confirmed that medium composition had no influence on the structure difference between MR biosurfactant and Surfactin (*B. subtilis* IFO 3035).