

## Abstract

This research aims to isolate and screen microorganisms that capable degrading hydrocarbons which contaminated in environment. The hydrocarbons such as crude oil and weathered crude oil contaminated soil samples were collected from lower Southern region including Nakhonsrithammarat, Phatthalung and Songkhla Provinces. The result showed 56 isolates (38 bacteria and 18 yeasts) that can grow in MSM medium containing 1 percent Waste Lubricating Oil (WLO) as a carbon source. In addition, 6 selected isolates can degrade WLO more than 50 percent. 5 bacteria strains, namely WK1, WK2, WK3, WK4 and WK5 whereas 1 yeast strain, namely WK6 were identified by morphology and DNA sequencing analysis. The result of morphology of colonies on agar medium showed round shape, off white color, shiny and smooth, wavy and shuttle with margin. When Gram staining and view under compound microscope, the result indicated WK1 and WK2 are long rod whereas WK3 and WK4 are short rod and produce endospores. All of 4 strains (WK1, WK2, WK3 and WK4) are Gram positive but WK5 is negative Gram and short rod. Moreover, WK6 is a yeast strain. It has oval shape and reproduction by budding. The identification result of 6 selected strains, WK1, WK2, WK3 WK4, WK5 and WK6 were indicated *Bacillus cereus*, *Bacillus altitudinis*, *Bacillus amyloliquefaciens*, *Bacillus subtilis*, *Serratia* sp. and *Issatchenkia orientalis*, respectively. The factors effecting for hydrocarbon degradation were also studied by the selected consortium microorganisms with weight loss method. The result of optimum condition for the WLO degradation the consortium contained 10% WLO as a carbon source, 3 g/L ammoniumsulfate as a nitrogen source with an initial pH 7.0. When they were cultivated in this medium in shake-flask (50 ml medium in 250 ml flask) at 150 rpm and 37°C, it showed maximum growth and the highest degrading activity was at the 52.60% weight loss at 48 h. In addition, the consortium can produce biosurfactant. Determination of Oil Displacement Area test (ODA), the result showed area of displacement as 11 cm<sup>2</sup>. Emulsification activity (%EA) of supernatant with kerosene, soy bean oil, palm oil or rice bran oil, the result exhibited the best %EA with rice bran oil. From these results, the consortium not only can degrade WLO but also produce biosurfactant. The consortium was used for bioremediation of WLO-contaminated soil. They had 4 treatments, treatment 1 abiotic treatment (sterile waste lubricating contaminated soil + 10% WLO +consortium), the result of treatment 1 showed the amount of WLO-degrading microorganisms was increased from 0 day while

treatment 2 abiotic treatment (sterile waste lubricating contaminated soil + 10% WLO), exhibited the lower amount of WLO-degrading microorganisms than treatment 1 whereas treatment 3 and 4 were biotic treatment. The result of treatment 3 and 4 exhibited amount of WLO-degrading microorganisms were decreased. Finally, the application of the selected consortium to WLO degradation of the 4 experiments showed treatment 1 was the high potential, 48.67% weight loss followed by treatment 4 which had 41.33% weight loss while the control, treatment 2 was 16.50% weight loss. The conclusion of this research showed that this selected consortium which isolated from hydrocarbon contaminated soil can apply in xenobiotic contaminant effectively management and environmental friendly.