## De-emulsification capabilities of a *Microbacterium* sp. for breaking water-in-crude oil emulsion

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An aerobic and gram positive strain belonging to *Microbacterium* sp. was isolated from an oily sludge sample from Siri Island in the south of Iran. *Microbacterium* was able to grow on glucose as a sole carbon source and produced microbial de-emulsifier only in the presence of yeast extract. Production of the microbial de-emulsifier by the *Microbacterium* sp. was optimized using the Taguchi experimental design statistical method. De-emulsification activity of the culture broth reached a maximum value (>96%) at the end of the exponential growth phase (approximately 24 h) and did not change during the stationary phase which was stopped after 48 h. The temperature and inoculum concentration were the most significant interactive factors in the production of the de-emulsifier. The optimum values of temperature, inoculum concentration, pH, and culture age were 25 °C,  $10^8$  CFU mL<sup>-1</sup>, pH 7 and 24 h, respectively.

The stability of different volumetric ratios of the W/CO emulsions (20:80, 30:70 and 40:60) was studied. The W/CO ratio of 20:80 exhibited the highest emulsion stability. Stability increased as pH increased from 5 towards the higher pHs and the best results obtained at pH 6.3. Concomitantly, the pH of the W/CO emulsion was adjusted to 6.3 in all experiments. The uninoculated control emulsion was stable for 30 d at 80 °C and 240 d at 25 °C.

The best de-emulsification activity by the culture broth of *Microbacterium* sp. growing on glucose (3% w/v) and yeast extract (1.0 g L<sup>-1</sup>) was 96.4 % ( $t_{1/2}$ =36 h) (in flask) at W/CO deemulsification assay temperature of 80 °C and pH 6.3. With addition of 4.33 mg mL<sup>-1</sup> of purified microbial de-emulsifier, half-life value ( $t_{1/2}$ ) of the W/CO model emulsion was 3 h at 80°C. De-emulsification of water-in kerosene system (30:70 v/v) using 24 old culture broth was also examined and the half-life value ( $t_{1/2}$ ) was reduced to 1 h at 25 °C. Conclusively, *Microbacterium* sp. de-emulsifier promises potential biotechnological application for use in the oil industry and for environmental remediation.