Abstract

The present study deals with the isolation and optimization of lipase production from the new isolates of *Aspergillus terreus* and *Bacillus pumilus* and their application in the biodiesel production by using non-conventional feed stock. The isolation of lipase producing fungal and bacterial strains was carried out from the local habitats. Maximum lipase activity was achieved by using *Aspergillus terreus* and *Bacillus pumilus* when the culture media were supplemented with 1.0% and 1.5% olive oil, respectively. The culture media M6 and BM4 gave maximum lipase activity for *Aspergillus terreus* and *Bacillus pumilus*, respectively. Highest lipase yield by *Aspergillus terreus* was achieved after 72 hrs of incubation at 30°C at a pH of 6.0 while highest lipase activity by *Bacillus pumilus* was achieved after 48 hrs of incubation at 37°C at a pH of 7.5. Maximum lipase activity by *Bacillus pumilus* was achieved when 1.5% peptone and 1.0% glucose were used as nitrogen and carbon sources, respectively while *Aspergillus terreus* gave maximum lipase activity by using 1.0% peptone and 2.0% glucose as nitrogen and carbon sources. The partial purification of *Aspergillus terreus* and *Bacillus pumilus* lipases was achieved at 80% and 60% ammonium sulphate saturations while further purification of microbial lipases was carried out by using Anion exchange and cation exchange chromatography for *Aspergillus terreus* and *Bacillus pumilus* lipases, respectively and the purification folds of 2.600 and 1.835 were achieved, respectively. The Lewatit VPOC 1600 (Lanxess, Germany) was proved to be the best support for the immobilization of microbial lipases with the coupling yields of 76.220% and 89.731% for *Aspergillus terreus* and *Bacillus pumilus*, respectively. These microbial lipases were then used as catalysts with different non-conventional oils to produce biodiesel. Different algal oils, yeast oils, used cooking oil and non edible plant oils were subjected to chemical and enzymatic transesterification reactions. The percentage yield of Lewatit immobilized *Bacillus pumilus* lipase was 87% which was higher as compared to free lipase i.e., 66%. Different physico-chemical properties of non-conventional oils were also evaluated. Response surface quadratic models were used for the optimization of biodiesel production by using the Lewatit immobilized *Bacillus pumilus* lipase. The fuel properties and exhaust emission studies of enzymatically synthesized biodiesel showed that there was a significant decrease in

the exhaust emissions of all the biodiesel blends so biodiesel synthesized in the present study was more environment friendly than conventional diesel fuel.