



## Abstract

Polyhydroxyalkanoic acids (PHA) are biodegradable polymers synthesized by microorganisms which can serve as alternative to conventional petrochemical based plastics. In the present study, twenty one bacterial isolates from one hundred & twenty six samples taken from anaerobic digester were screened for the PHA accumulation ability using Sudan Black B dye. Biochemical tests including gram staining, catalase, citrate and starch hydrolysis tests were carried out on the bacterial isolates. The bacterial isolates were further subjected to submerged fermentation to estimate their relative PHA production capability. By 16S rRNA gene sequencing, the maximum PHAs yielding strain was identified as *Bacillus cereus* MUL-A. Effect of different physicochemical parameters such as fermentation media, composition of the modified PHA production media (PPMG), incubation temperature, incubation time, pH, inoculum size, inoculum age, agitation speed, carbon source, concentration of carbon source, nitrogen source and concentration of nitrogen source were studied on the polyhydroxyalkanoates (PHAs) yield. After 72 hours of submerged fermentation, a significantly high yield of PHAs i.e. 64.3% w/w were produced by the *Bacillus cereus* MUL-A strain by using PPMG medium having glucose (10 g/L) as carbon source and peptone (2 g/L) as nitrogen source, at 35 °C, pH 7.0, agitation speed 150 RPM and 24 h old 3 % inoculum size. Provision of agricultural residues i.e. corn cob (grinded), crude molasses and acid treated molasses as carbon source in fermentation media resulted in promising results regarding PHAs yield for *Bacillus cereus* MUL-A. For *Bacillus cereus* MUL-A, maximum PHAs yield of 44.6% was achieved when fermentation media was employed with acid treated molasses (6%) as carbon source, at an incubation time of 72 h, at 35 °C, pH 7.0, agitation speed 150 RPM and 24 h old 3 % inoculum size. Characterization of the purified PHA by Fourier transform infrared (FTIR) spectroscopy determined its purity and major functional groups. Fermentation parameters including incubation time, incubation temperature, and agitation speed and aeration rate were studied during upscale studies of PHA production by *Bacillus cereus* MUL-A. Maximum PHAs yield i.e. 49% was attained for *Bacillus cereus* MUL-A at 60 h of incubation time, 35 °C, agitation speed of 200 RPM and at an aeration rate of 2 vvm. Fermentation kinetics on the upscale studies further validated our results.