Abstract

Nutrient stress is a conventional approach used for increasing lipid accumulation in microalgae, but the reduced growth under such conditions eventually affects biodiesel production through reduced lipid productivity. To overcome this drawback, mixotrophy can be preferably considered for increasing the microalgal growth. In the present study, the effect of mixotrophy with six different exogenous carbon sources was assessed for growth and lipid accumulation in a green microalga, *Scenedesmus obliquus* (Trup.) Kutz. (SAG 276-3a). The results of the individual and interactive supplementations of the carbon sources demonstrated maximum lipid accumulation up to 29% of dry cell weight (dcw) in the test microalga under the combined supplementations of 0.16% citrate and 0.16% acetate in N 11 medium. The multifactor optimization study using Response Surface Methodology further boosted the lipid content to 56.4% (dcw). The biodiesel (transesterified lipids) obtained was found to be predominated with saturated and monounsaturated fatty acid methyl esters.

To economize the process, various industrially important co-products were also produced from the test microalga along with biodiesel. Under the optimized condition, although accumulation of β-carotene, omega-3 fatty acids and carbohydrates were not found to be significantly stimulated, a fivefold rise in glycerol recovery from the transesterified lipids was observed. 60% of the total carbohydrate present in the microalgal biomass was found to be converted into bioethanol under the optimized condition. Extraction of protein from *S. obliquus* biomass was standardized, following which, the extracted protein was formulated into a diet containing standard fish feed and whole *S. obliquus* biomass and the extracted protein in a ratio of 25:25:50 that induced maximum growth in the freshwater fish varieties, rohu, mrigal and catla.

The algal refinery thus designed by sequentially extracting the above microalgal components, was able to convert 70% of the microalgal biomass into industrially valuable products by yielding 0.6 g of β-carotene, 10 g of protein, 38 g (43 mL) of biodiesel, 1 g of omega-3 fatty acid, 3 g (2.4 mL) of glycerol and 18 g (23 mL) of bioethanol from 100 g of dry *S. obliquus* biomass under the optimized condition.

Keywords: Algal refinery, β-carotene, Biodiesel, Bioethanol, Glycerol, Mixotrophy, Omega-3 fatty acids, Protein-rich algae meal, *Scenedesmus obliquus*