

Production of Biofuels by Micro-, Lab-, Pilot- and Industrial Scale Based Biocatalytic Processes

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The need for the production of biofuels from various renewable sources is becoming increasingly interesting especially as the availability and accessibility of fossil fuels is significantly declining. Biodegradability, low pollution emissions and non-toxicity of raw materials are some properties making biogas, biodiesel and bioethanol more environmentally friendly fuels. Solid-state fermentation could be a suitable technology for the production of value-added products by utilization of the renewable waste materials, which makes it also economically feasible. So far, this technology was used for production of enzymes, organic acids, mushrooms, flavour and aroma compounds, pigments, polysaccharides, hormones, human food and animal feed. Different type of bioreactors have been developed and successfully used for solid-state fermentation of broad range of substrates and in production of value-added products. Solid-state fermentation will be demonstrated as part of anaerobic degradation on lab-, pilot- and industrial-scale of several waste materials such as brewer's spent grain, whey and cow manure, and corn silage and cow manure, respectively. The application of different microreactor systems in intensification of the biodiesel production process is widely studied. However, previous studies of the application of microreactor technology in the production of biodiesel were limited to the use of chemical catalysts. Mild reaction conditions, absence of by-products, reusability, simple separation and purification of the resulting biodiesel as well as lower energy consumption are some of the many advantages that make the enzyme lipase – a biocatalyst – a better choice than traditional chemical catalysts in the process of biodiesel production. Different microreactor systems utilising a commercially available lipase and a lipase produced by solid-state fermentation were used for transesterification of fresh and waste cooking oil while biodiesel was separated using integrated microseparation unit. Selected examples are clear demonstration of environmentally friendly and economic technologies used for efficient production of biofuels on micro-, lab-, pilot- and industrial scale.

Biodiesels are sustainable powers generally created from vegetable oils, including those from soybean, palm, sunflower, rapeseed, jatropha, and others. As of now, biodiesel commands are set in excess of 60 nations, huge numbers of which have a place

with high-burning-through areas like the European Union and North America. Worldwide biodiesel creation is extended to continue its fast increment and arrive at more than 40 billion liters by 2020. Tragically, the acknowledgment of biodiesel as an alternative fuel is affected by the presence of foreign substances that structure silt and cause the disappointment of motors. Steryl glucosides, present in different biodiesels at fixations going from 10 to 300, have been identified as the significant segment of such dregs. Accordingly, the particular evacuation of SGs could deliver biodiesels of a predominant quality, improving the probability that these sustainable energizers will be embraced by shoppers. As of now, the main accessible strategy able to do totally eliminating SGs from biodiesel is distillation, an energy-serious and costly process. Recently, we portrayed an efficient technique for removing SGs from biodiesel. The technique depends on the utilization of compounds with steryl glycosidase (SGase) movement. The most efficient SGase tried so far is a thermostable fl-glucosidase from *Thermococcus litoralis*. Since this hyperthermo-philic archaeon is difficult to develop, an engineered codon-improved form of the SGase quality was planned and effectively communicated in *E. coli*. The delivering strain was then advanced through broad design, which incorporated the utilization of different advertisers and the co-expression of atomic chaperones to significantly expand the creation of the SGase. Furthermore, a high cell thickness, taken care of cluster aging cycle has been created. Nonetheless, much remaining parts to be done to accomplish a cost-effective cycle for assembling SGase.

Presently, modern catalysts created by maturation are viewed as items. The carbon source utilized in the aging cycles can represent up to half of the all out assembling cost. Likewise, downstream supportive of cesses including a couple of basic activities can enormously lessen capital consumptions and working expenses. In this work, we break down the effect of different carbon sources and the post-enlistment taking care of procedure on the profitability of the SGase in *E. coli*, intending to build up a business fermentation measure for delivering this catalyst for an enormous scope. Also, a basic thermolysis technique is portrayed for encouraging the recuperation of the protein. The outcomes depicted here could be utilized to configuration measures for creating other thermostable

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chemicals in *E. coli* in a cost-effective manner. Growth was dictated by estimating optical thickness at 600nm (OD600) in a General Electric Novaspec III spectrophotometer. HM medium comprises of 20.8g/L KH₂PO₄, 3g/L (NH₄)₂PO₄, 3.25g/L K(OH), and 4g/L NaH₂PO₄. Microplates (Thermo Fisher Scientific) were set up by adding to each well, 190 µL of HM medium enhanced with 1% of the wellspring of carbon being tried and the necessary anti-microbials. Each very much was immunized with 10 µL of the centrifuged culture. The microplates were hatched at 37°C for 12h with ceaseless shaking in a microplate peruser. OD600 were recorded each 30min.

Bioprocesses dependent on *E. coli* generally use for their vehicle bon and fuel source glucose acquired from the hydrolysis of cornstarch. This glucose has a normal expense of \$800 F=X-0V0e(u.t)S0YX\$ per metric ton. To investigate cheaper other options, we chose to test sucrose, glycerol, and modern items containing these mixes, for example, molasses and biodiesel-determined rough glycerol—as feedstocks for the creation of SGase. While subordinates of *E. coli* BL21 can develop efficiently by using glucose or glycerol, these strains can't utilize sucrose as a carbon source, since they do not have the qualities for moving and using this disaccharide. *E. coli* W strains develop with sucrose as the sole carbon and fuel source utilizing the *cscB*, *cscA*, and *cscK* qualities, which intercede the vehicle and hydrolysis of sucrose and the phosphorylation of fructose, individually. Thusly, we first designed the BL21-based delivering strain by embeddings a sucrose utilization tape containing these three qualities into the *lacZ* locus of the chromosome. For this progression, the operon containing the qualities and the local advertiser was amplified by PCR utilizing oligonucleotides with expansions homologous to the objective grouping in the chromosome.