Матеріали XXIII Міжнародної науково-практичної конференції
«Екологія. Людина. Суспільство» (м. Київ, Україна, 7 грудня2023 р.)

Handbook of the XXIII International Science Conference
«Ecology. Human. Society» (December 7, 2023 Kyiv, Ukraine)

ISSN (Online) 2710-3315
DOI: https://doi.org/10.20535/EHS2710-3315.2023.290930

UDC 57.043:044:620.951

PROSPECTS OF PRODUCING BIOBUTANOL USING MICROALGAE RESIDUE AFTER LIPID EXTRACTION

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Introduction. Biobutanol is a renewable biofuel chemically similar to gasoline but produced from biomass rather than fossil fuels. It is typically produced through fermentation, usually using strains of bacteria from the class Clostridia, which convert sugars derived from biomass into butanol via acetone-butanol-ethanol (ABE) fermentation [1].

Biobutanol can become a severe competitor to other types of biofuels. For instance, one advantage of biobutanol over ethanol is its higher energy density, which means it contains more energy per unit volume. This makes biobutanol a more attractive alternative to gasoline as a transportation fuel. In addition, biobutanol has a lower vapour pressure than ethanol, which can reduce evaporative emissions and make it easier to blend with gasoline.

Biobutanol can also be used in existing gasoline engines without significant modifications. It can be blended with gasoline at higher concentrations than ethanol without causing engine damage or performance issues [1]. Therefore, it has the potential to be used as a "drop-in" replacement for gasoline, which would make it easier to integrate into existing fuel infrastructure and vehicle fleets.

Discussion. Several feedstocks can be used for biobutanol production, including agricultural residues, energy crops, lignocellulosic materials, food waste, and algae [1]. The choice of feedstock depends on various factors, including availability, cost, and sustainability.

The production of first-generation biobutanol is largely made possible by the fermentation of primarily hexose sugars, which is a reasonably straightforward process. These sugars are produced by hydrolysing crops high in starch, like maise, wheat, rice, and cassava. Biobutanol yield for such raw materials is usually the highest; for example, Iyyappan et al. (2022) studied biobutanol production from black strap molasses using *Clostridium acetobutilicum* MTCC11274. The results show the maximum butanol production as 4.15 ± 0.48 g/L [2]. These materials are often readily available and may cost less than other feedstocks. However, they may also face competition from other industries that use them as feed or fertiliser.

Second-generation biobutanol is represented using agricultural residues such as corn stover, wheat straw, or energy crops, such as switchgrass and miscanthus. The last ones can be grown on marginal land, do not compete with food crops, and provide high biomass yields per unit area. These materials are abundant and widely available, but their high lignin content makes them more challenging to break down and convert into biofuels than other feedstocks. For instance, Pratto et al. (2020) used sugarcane straw and *C. acetobutylicum* NRRL B-527 for biobutanol production. The findings suggest that the butanol production peaked at 7.6 ± 0.7 g/L [3].

The third-generation biobutanol uses microalgae as a primary feedstock. Microalgae can be grown in various environments, including wastewater and brackish water, producing high biomass yields per unit area. Today algae-based biobutanol production is an area of active research and development, and several studies have shown that it is possible to convert algal biomass into biobutanol through fermentation. Melih Onay (2020) studied biobutanol production from microalgae *Chlorella zofingiensis* CCALA 944 by *C. acetobutylicum.* In that article, the results show that maximum butanol production reaches up to 3.1 ± 0.3 g/L [4].

Nevertheless, the primary focus of research on microalgae has been their capacity to accumulate lipids suitable for producing biodiesel, constituting a significant portion of their dry weight. Therefore, using microalgae residues after lipid extraction (e.g. after biodiesel production) for butanol production rather than whole microalgae cells could be more reasonable. Moreover, that strategy has several advantages.

Firstly, by utilising the residues after lipid extraction, we can potentially use a broader range of microalgae species for biobutanol production. Secondly, microalgae residues are typically rich in carbohydrates and proteins, which can be converted into other biofuels, such as biobutanol and bioethanol, or valuable co-products, such as animal feed, fertiliser, or bioplastics. Finally, utilising microalgae residues after lipid extraction can help improve microalgae biofuel production's sustainability. Microalgae are often grown using large quantities of water and nutrients, which can have negative environmental impacts if not appropriately managed. By utilising the residues, we can reduce the environmental footprint of microalgae-based biofuel production and move towards a more sustainable and circular economy [5].

Ionic liquid extraction is a solvent extraction that uses ionic liquids as solvents to extract target compounds (e.g. lipids) from various sources such as plants, microorganisms, or industrial wastes. This procedure involves treating microalgae with methanol and 1-ethyl-3-methylimidazolium ethylsulfate ($C_2mimEtSO_4$) at a mass ratio of 1:2 for one hour. Ionic liquid extraction is characterised by several advantages, such as high selectivity, good solubility, low toxicity and a sustainable approach to various compounds, making it an attractive alternative to traditional extraction methods, such as hexane extraction.

The study compared two types of substrates for direct ABE fermentation (without pre-treatment) by *C. saccharobutylicum*: *C. vulgaris* after ionic extraction and *C. vulgaris* after hexane/2-propanol extraction. The study shows that butanol concentration reached 4.99 ± 0.2 and 6.63 ± 0.3 g/L retrospective. However, hexane-extracted microalgae also require detoxification, while ionic liquid-extracted microalgae do not. The study showed that it was possible to produce both butanol and biodiesel from the same feedstock, which could assist in lowering the cost of feedstock for each process separately, and that ionic liquid pre-treatment can improve the efficiency of the process [5].

Conclusion. Biobutanol can be produced from various feedstocks, including corn, sugarcane, wheat, barley, switchgrass, and algae residues. Each feedstock has advantages and challenges depending on availability, cost, and sustainability.

Corn and sugarcane are currently the most widely used feedstocks for biobutanol production due to their high sugar content and availability in many parts of the world. However, these feedstocks can have negative environmental impacts, such as competition with food crops and land use change.

Alternative feedstocks such as switchgrass and algae residues have shown promise for biobutanol production due to their lower environmental impact and potential for use on marginal lands. However, further research and development are needed to optimise the production processes and reduce the costs associated with these feedstocks.

In general, the choice of feedstock for biobutanol production will depend on various factors, including availability, cost, sustainability, and local regulations. With continued research and development, it is possible to produce biobutanol from a range of feedstocks sustainably and cost-effectively, helping to reduce our dependence on fossil fuels and mitigate the impacts of climate change.

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