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Dark Fermentation for Sustainable Biohydrogen Production from Food Waste

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ABSTRACT

The escalating global energy crisis, coupled with the generation of over 1.3 billion tons of food waste annually, necessitates sustainable solutions that address both environmental and energy challenges. Dark fermentation of organic waste represents a promising biotechnological approach for biohydrogen production a carbon-neutral fuel that releases zero emissions during combustion. This study investigates biohydrogen production via anaerobic dark fermentation of food waste using clostridium spp as the biocatalyst. The research focuses on comprehensive feedstock characterization and systematic optimization of batch fermentation parameters to maximize hydrogen yield. Food waste samples were collected from hostel mess facilities and preserved at 4 °C. Following thawing, the substrate underwent mechanical homogenization, size reduction through mechanical grinder, and analytical characterization including total solids (TS), volatile suspended solids (VSS), carbohydrate content via phenol-sulphuric acid method, and alkalinity determination through titrimetric analysis according to APHA standard protocols. The clostridium spp inoculum was cultured in Luria-Bertani broth, acclimatized in synthetic medium (pH 7.0), and harvested via centrifugation. Batch fermentation experiments were conducted in 500 mL conical flasks containing 120 mL pretreated food waste, 120 mL mineral salt medium, and 0.6 g dry cell mass. The pH was adjusted using NaOH/HCl, followed by nitrogen purging to establish anaerobic conditions. Cultures were incubated at 35-37 °C with agitation at 150 rpm for 48-72 hours. Biogas evolved was collected in Tedlar bags and analyzed using gas chromatography-mass spectrometry (Agilent 6890N, nitrogen carrier gas, m/z 29, thermal conductivity detector).The food waste substrate exhibited pH values ranging from 4.2 to 6.0, which facilitated effective pretreatment and fermentation initiation. Hydrogen production was successfully confirmed through mass spectrometric detection of protonated nitrogen signals, validating the dark fermentation process. This investigation demonstrates the viability of clostridium spp -mediated dark fermentation as a dual-benefit strategy for food waste conversion and renewable energy generation. These findings underscore the critical role of dark Fermentation in sustainable resource management and offer promising pathways for bioenergy development.

Keywords: *Bioenergy, Waste-to-Energy, Organic waste, Dark Fermentation, Microbial Inoculum*

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