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Research Article

Microalgae Biorefinery from Palm Oil Mill Effluent (POME): Bioremediation and Biomass Production

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ABSTRACT

Palm oil processing in Malaysia produces massive amounts of palm oil mill effluent (POME) every day. Effective treatment of POME is in necessity for compliance of regulations and for environmental sustainability. There is a new innovation in effluent treatment which is the application of microalgae for wastewater treatment and biomass production. The present study applied microalgae isolated from POME for the biomass production and bioremediation. The microalgae isolated are identified as Chlorella sorokiniana. Microalga was cultured in bold basal medium (BBM) for 20 days in a total volume of 250 mL, range of pH 6 to 8, with continuous of air supply and light illumination, as well as supplementation. The highest microalgal biomass concentration attained was 3.02 g L^{-1} at pH 7, white light illumination, and 25 mM of glycerol and urea supplementation. Furthermore, the study has found reduction in chemical oxygen demand (47%), total nitrogen (76%), and color (69%) with 50% POME addition into the microalgal culture medium. Overall, this study is expected to be able to provide information to stakeholders on wastewater technology improvement, the private sector for future investment, and the government sector in making decisions. With that, sustainable POME management can be achieved with environmental sustainability.

Keywords: Bioremediation, Microalgae, Palm oil mill effluent, Wastewater.

INTRODUCTION

The palm oil mill effluent (POME) generated from palm oil processing is at massive amount, at daily basis, as the Malaysia palm oil industry ranked second in the world palm oil export. The export of crude oil accounts USD14 billion in 2023, with the main destinations exported to India, China, Turkey, Kenya and Japan. POME generated contains various types of contaminants, including hazardous organics, inorganics chemicals, oil and grease and much more. These has caused arisen of environmental and health problems, and difficulties in industrial wastewater treatment. Microalgae has shown its potential to provide biological wastewater treatment as it assimilates organics, inorganics and heavy metals from the wastewater, for its own growth [1, 2]. Microalgae has been applied in secondary treatment for municipal and domestic wastewater, and industrial effluents, such as textile wastewater, industrial wash water, and mine effluents, which contain high heavy metal concentrations [3]. *Chlorella sorokiniana* CY-2, *Spirulina plantensis*, *Nannochloropsis Oculata*, *Chlamydomonas* sp., *Tetraselmis suecica* are the microalgae species which have shown promising in POME phycoremediation [2, 4-6]. Further, microalgae are as well serving as feedstock for pharmaceuticals, healthcare, food and biofuel industries of its biological active agents and its lipid, carbohydrate, and protein components [7, 8]. Palanisamy et al. (2023) claimed that *Spirulina plantensis* is highly commercialised due to its high value in protein and phycocyanin content, and the natural pigment for food and cosmeceutical industries [5].

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MATERIALS AND METHODS

1.0 Sampling

The raw Palm Oil Mill Effluent (POME) sample was collected from a Palm Oil Mill refinery located at Johor, Malaysia. The POME samples were collected directly from the drainage at the mill's final discharge point, as shown in Fig. 1. The samples were stored in Scott bottles, as shown in Fig. 2. All samples were stored at 4°C in the freezer, sealed with parafilm to prevent light exposure, for further treatments.

Figure 1. Final discharge point of POME from the industrial wastewater plant

Figure 2. The POME samples were stored in Schott Bottle

1.1 Isolation and identification of microalgae species

The agar used was Bold Basal Medium (BBM) agar for spread plate of microalgae species from POME sample. An amount of 1 ml sample was spread onto the BBM agar. The agar plates were incubated under continuous light for 7 days (Fig. 3). After 7 days, two types of microalgae were observed and further streak on BBM agar to obtain single colony. The microalgal colony was identified via 16s rRNA analysis.

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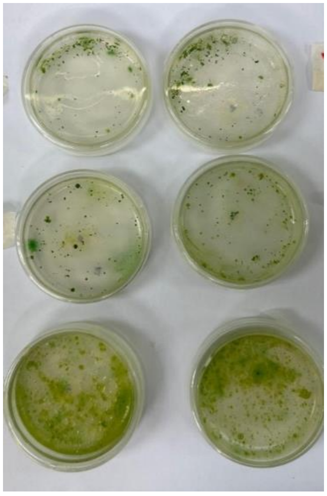

Figure 3. BBM agar plates incubated under continuous light for 7 days

1.2 Measurement of microalgae concentration in enhanced parameters and pollutants degradation

Microalgae were precultured at BBM liquid medium initially for 5 days. An amount of 20 ml culture was subsequently concentrated by centrifugation at 1500 rpm for 30 minutes. Concentrated medium was then transferred to main culture with 50% of POME, at conditions of varied pH of 6, 7 and 8, with continuous supply of light and aeration for 20 days (Fig. 4). The liquid samples were collected daily for each of the pH for analysis. Biomass concentration was measured using a spectrophotometer, at an optical density of 690 nm. Similarly, the parameters of white light and red-light of 1000 lux and 1300 lux unit, and addition of supplements which were 5mM, 15mM and 25Mm of glucose, urea and glycerol, were evaluated. The wastewater characteristics of chemical oxygen demand (COD), total nitrogen (TN) and color were determined following the American Public Health Association on Standard Methods for the examination of water and wastewater. The analysis of COD, TN and color of wastewater was carried out via a spectrophotometer model DR 1900 and Hach standard reagent.

Figure 4. The medium in different pH and light supply for all the medium were put under the vaired light with aeration continuously for 20 days.

RESULTS

The isolated microalgal colony was selected and sent to external laboratory for molecular study, which includes DNA extraction using Macherey-Nagel kit, electrophoresis, polymerase chain rection, DNA sequencing for algorithms analysis via BLAST. The microalgae isolated was identified as *Chlorella sorokiniana* with similarity percentage of 98.91%. Fig. 5 indicated the best concentration of biomass was obtained consistently at pH 7 culture, if compared to pH 6 and pH 8. By referring to Fig. 6, it can be seen that the highest biomass concentration of 3 g L^{-1} was attained at white light. Fig. 7 has indicated on the maximal biomass concentration attained was 3.02 g L^{-1}, in 50% POME and BBM medium, at condition of pH 7, continuous supply of air and white light, with 25mM of glycerol and urea. Further, the pollutants of COD, TN and color have been removed by 47%, 76%, and 69%, respectively.

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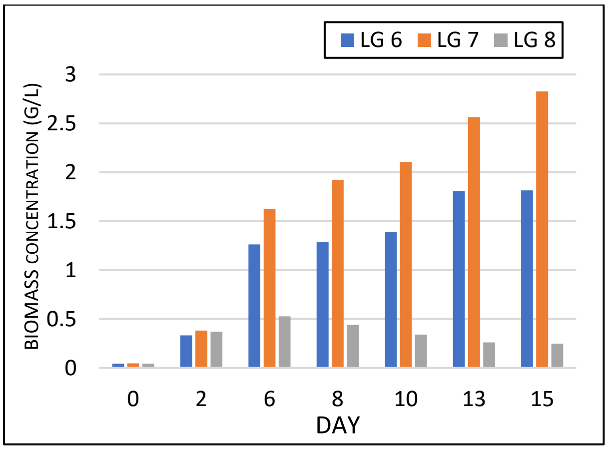

Figure 5. Biomass concentration of microalgae in 50% POME and BBM medium at varied pH

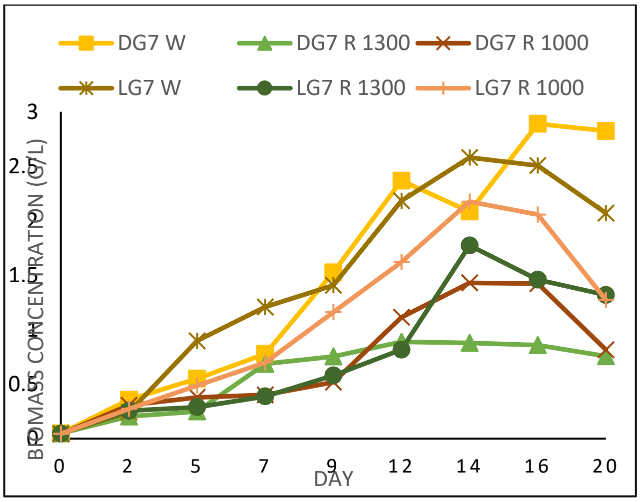

Figure 6. Biomass concentration of microalgae cultured in 50% POME and BBM in pH 7 at white light (W) and red light (R) at intensity of 1000 lux and 1300 lux unit for 20 days

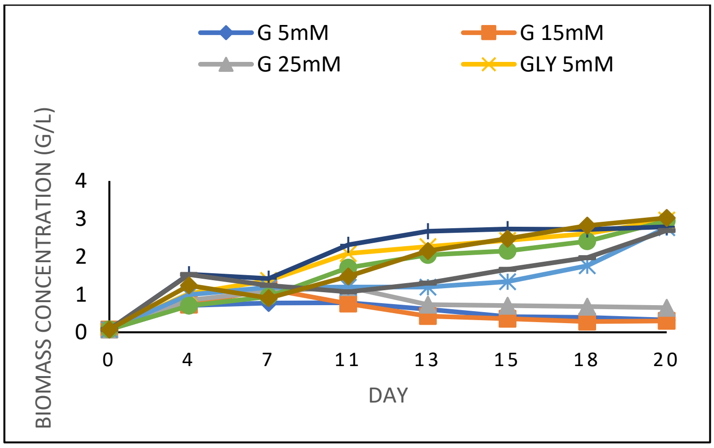

Figure 7. Biomass concentration of microalgae in varied types of supplements at varied concentration

DISCUSSIONS

The present study worked on microalgal-based phycoremediation on POME, to achieve both microalgae biomass production and POME treatment. Varied parameters have been tested, including the pH, light supply, supplementation addition, to boost microalgal biomass concentration, and pollutants degradation. It could be seen that the *Chlorella sorokiniana* isolated from POME was able to produce high biomass concentration, only at optimized parameters. This could be due to the color of the POME which block the penetration of sunlight for microalgal photosynthesis to take place. Similarly, Low et. al. (2021) have reported on POME pretreatment carried out prior to its utilization as microalgae culture medium, has improved microalgal biofuel production and POME treatment [4]. The pretreatment includes adsorption using activated carbon, coagulation, acid-het treatment and much more. Also, in our previous work, pretreatment using bacteria *Pseudomonas* sp. for POME decolorisation was found effective [9]. Thus, it could be seen that only 50% of POME in BBM is used and suitable for microalgae growth in the present study. POME generated typically exhibited low pH, therefore neutralization is also required prior to cultivation. Neutralization of POME up neutral (pH 7) is desirable, with consideration of 50% POME used in the culture medium. Further, highest biomass concentration of 2.82 g L^{-1} was produced from white light rather than 2.58 g L^{-1} from red light cultivation. This is in contrast of work from Esteves et. al. (2025) who stated with red light boosted microalgae growth, nutrient uptake and carbohydrate accumulation; while white light helps for nitrogen uptake, lipid and pigment production [10]. Supplementation of glucose, glycerol and urea are essential to boost microalgae growth for biomass production, before its lipid accumulation at nitrogen depletion stage [9]. Similarly, this study found that supplement addition has enhanced biomass production from 2.82 g L^{-1} without supplementation, to 2.97 g L^{-1} with 5mM glycerol addition and highest 3.02 g L^{-1} with 25mM of glycerol and urea. In addition, the reduction in COD (47%), total nitrogen (76%) and color (69%) have indicated in the findings. This has proven that the pollutants uptake taken place from 50% POME added in BBM medium, together for biomass production.

CONCLUSION

This present study is expected to be able to provide information to industrial stakeholders on the feasibility of applying microalgae to phycoremediate industrial wastewater, especially the palm oil industry, technology improvement with existing treatment system, as well as the private sector for future investment on microalgae industry, and the government sector in making decisions. With that, sustainable POME management can be achieved with environmental sustainability.

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