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HARVESTING OF MICROALGAL BIOMASS USING MORINGA OLEIFERA AS NATURAL COAGULANT: A COST-EFFECTIVE APPROACH

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ABSTRACT

Microalgae has been considered one of the most promising 3rd generation biofuel sources. Nevertheless, the commercialization of this biomass has not been successful in developing countries due to the energy-intensive harvesting process. The aim of the present study is to find a suitable coagulant which is locally available, cost-effective, non-toxic, and environmentally friendly. In this regard, Moringa Oleifera (MO) seed powder, locally known as Shojne in Bangladesh, is used as natural coagulant to fulfil the present crisis. However, the harvesting efficiency and coagulant properties of MO were evaluated through jar-test, where wastewater grown microalgae, cultured in a photobioreactor (PBR), was used. This study optimized the harvesting parameters such as coagulant dose, mixing rate, and mixing time. As those parameters have a significant impact on the microalgae harvesting process. Low mixing rate and mixing time have better performance with coagulant dose varied from 10-70 mg/L. The highest harvesting efficiency of 83% microalgae recovery was achieved at 20 rpm for 10 minutes with coagulant dose of 70 mg/L. The MO seeds are recommended to be prospective coagulants for both water treatment and microalgae harvesting.

Keywords: Coagulation, Microalgae, Moringa oleifera, Optimum dose, Recovery rate.

1. INTRODUCTION

Worldwide future predictions reveal that by 2050, the world's population will need 70% more food, 50% more fuel, 50% more water, and a 50-80% reduction in carbon dioxide (CO2) emissions to maintain public, political and climate protection (Yang et al., 2016). To meet up the fresh water requirement, biological wastewater treatment has been widely adopted in low income and lower income countries (Khalekuzzaman et al., 2018; Hasan et al., 2018) . But, the major challenge in biological treatment (e.g. aerobic/anaerobic) process is nutrient-rich effluents (Alimahmoodi et al., 2013). Again if this partially treated effluent is discharged to the aquatic environment without proper removal of nitrogen and phosphorus, it will lead generally to symptomatic changes such as eutrophication, increased algae production and other aquatic plants, degradation of fisheries and deterioration of water quality (Barrado-Moreno et al., 2016).

In addition, together with the rapid rise in global energy demand, the decline of petroleum supplies makes it necessary to develop new renewable energy sources (Uggetti et al., 2014). The development of innovative ways of producing biofuels using microalgae has the potential to meet these challenges (Rawat et al., 2011). Because, microalgae based biofuels have multiple economic and ecological advantages compared to terrestrial based crops such as continuous growth in waters with a wide range of salinities, growth in any area without the need of pesticides, high specific yield and photosynthetic efficiency (Khalekuzzaman et al., 2019). Microalgae could play an important role in the bioremediation of wastewater and carbon dioxide sequestration (Khalekuzzaman et al., 2020). Furthermore, these photosynthetic microorganisms are considered a potential renewable energy source (Hasan et al., 2021). Although microalgae have a huge application opportunity, the production is still not economically viable due to the difficulties of the harvesting process (Kabir et al., 2022).

Nevertheless, there has been no extensive study into microalgal recovery and the biochemical characteristics of biomass obtained from Photobioreactors (PBRs). Ultimately, a minimal cost harvesting method should be examined since this production phase reflects 20-30% of the biomass production costs (Barros et al., 2015). Different harvesting methods applied to microalgae biomass such as filtration, centrifugation, coagulation/ flocculation, gravity sedimentation, flotation, and electrical based process (**Figure-1**). However, centrifugation and filtration are extensively used in the commercial harvesting of microalgae. But those technologies are very expensive and energy-intensive operations due to the energy required for the machinery (Lananan et al., 2016). Among all of these methods, chemical coagulation-flocculation with sedimentation is the most simple and fast

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method. But, the major disadvantage of this method is chemical coagulants may be expansive and toxic to microalgal biomass. One more disadvantage is recycling of culture medium is limited and high concentration of metal salts in the wastewater effluents caused effect not only on fish but also birds or other higher animals eating the infected fishes and animals (Abdul Hamid et al., 2016). An effective environmentally friendly harvesting method must therefore be fully developed not only to promote the recovery of microalgae biomass but also to protect our natural environment.

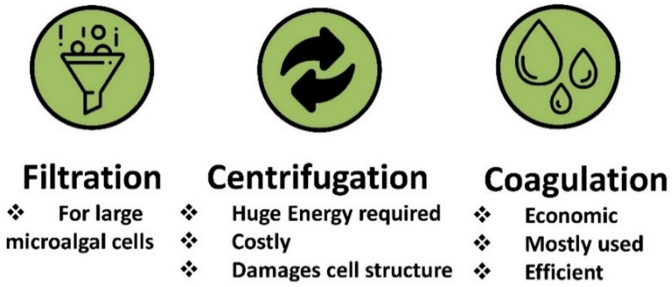

Figure 1: Microalgae recovery methods.

To resolve this entire problem, natural coagulant such as *Moringa oleifera* (MO) seeds flour may be used in order to harvest microalgal biomass. MO is tropical tree from the genus moringaceae which originates from sub-Himalayan valleys and is currently distributed throughout the tropics and subtropics around the world (Khalekuzzaman et al., 2018). In addition, it has high coagulation properties, low cost and low toxicity, making it very promising to be used as an alternative coagulant to recover algal biomass from its suspension system. The seeds of this tree have a high amount of proteins that function as cationic polyelectrolytes (Sharma et al., 2006). Proteins that destabilize colloids and remove them by sedimentation are those that act as cationic polyelectrolyte and neutralize suspended materials, as most of them are negatively charged like microalgal biomass, which have a negative electrical charge in their cell walls (Hasan et at., 2021). Hence, the use of natural coagulants would be an alternative solution (**Figure-2**). Thus, Moringa Oleifera (locally known as Shojne in Bangladesh) could be used instead of chemical coagulants. Moreover, it is locally available, cheap, non-toxic, and environmentally friendly (Keogh et al., 2017).

Figure 2: Benefits of using natural coagulant.

However, microalgal recovery and the biochemical characteristics of biomass harvested from PBRs by using natural coagulant such as MO seed powder have not been extensively investigated. The main objective of this research is to optimize the harvesting parameters such as coagulant dose, mixing rate, and mixing time. As those parameters have a significant impact on the microalgae harvesting process. The optimum dose was also determined based on the highest recovery rate and efficiency of the system.

2. MATERIALS AND METHOD

2.1 Microalgae Cultivation

A photobioreactor (PBR), made of transparent water container having a capacity of 8 L, was used to cultivate microalgal biomass (**Figure-3**). In order to use natural sunlight instead of artificial light, the experimental setup

was installed at the rooftop of the Civil Engineering Department, KUET, Khulna. As clear andtransp arent reactor was used so iteasily received the natural ,sunlight proves thewhich im microalgal growth. On the other hand, nutrients-enrich wastewater effluent (Hasan et al., 2021) instead of fresh water was used as a culture media to cultivate microalgae. All these things will significantly reduce the cultivation cost. The reactor was inoculated with Chlorella Vulgaris as it has good tolerance in saline water. Moreover, the research area (Khulna) is located in a coastal zone with high salinity. An air pump was installed to supply CO_2 in PBR formicroalgae .growthOther operating parameters and details of microalgae process was described in the previous study done by Islam et al. (2022).

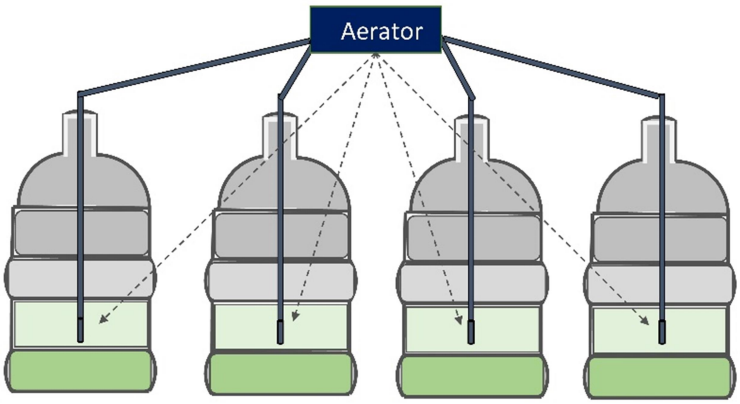

Figure 3: Microalgae cultivation in a photobioreactor (PBR).

2.2 Preparation ofCoagulant

The seed pods of MO were collected at the KUET (Khulna University of Engineering & Technology, Khulna, Bangladesh) campus. The seeds were separated and dried in natural sunlight at temperature 35±3 °C for three days. After careful removal of seed coats and seed wings, the white kernels were reduced to powder by a mechanical grinder. Then the powder was sequentially sieved through No #300 sieve to obtain the seed flour (**Figure-4**). Finally, the obtained powder was stored in an air-tight container and protected from light and moisture to avoid oxidation and degradation of its active properties (Abdul Hamid et al., 2014).

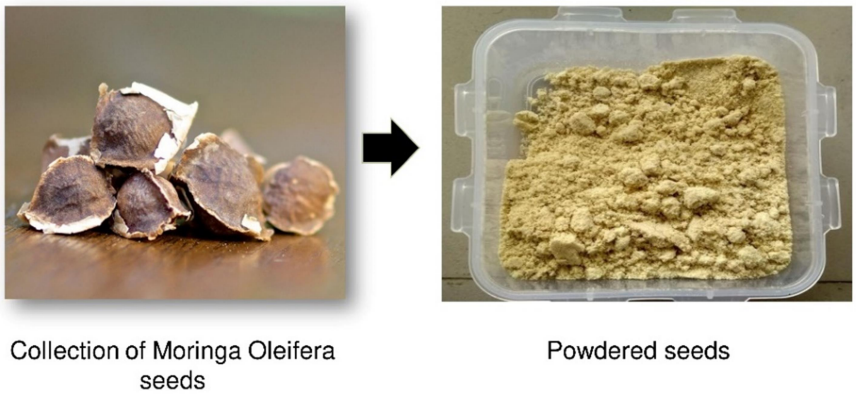

Figure 4: Preparation of coagulant.

2.3 Experimental and Analytical Methods

After the cultivation of microalgae and preparation of coagulant, the jar test was used for mixing the microalgae suspension and coagulant (Okoro et al., 2019). The overall research methodology is presented in Figure-5 as a flowchart.

The experiment was carried out in nine batches, each batch containing five jars of microalgae suspension as mentioned in Figure-6. The concentration of coagulant were 10, 25, 40, 55, and 70 mg/l in each batch. The

suspension was mixed for three different time sets 10, 30, and 50 mins in a mixing rate of 20, 60, and 100 rpm. The sedimentation time was 30 mins and temperature was 28°C. At the end of the sedimentation process, the supernatant liquid samples were collected from each beaker, and the microalgae cell concentrations were measured at 680 nm using a HACH DR 3900 spectrophotometer to determine the microalgae biomass recovery. Microalgae biomass recovery was calculated based on the initial (C_i) and final (C_f) microalgae cell concentration measurements. The microalgae removal efficiency was determined using the following equation (1):

$$Microalgae\ recovery = \frac{C_i - C_f}{C_i} * 100 \quad (1)$$

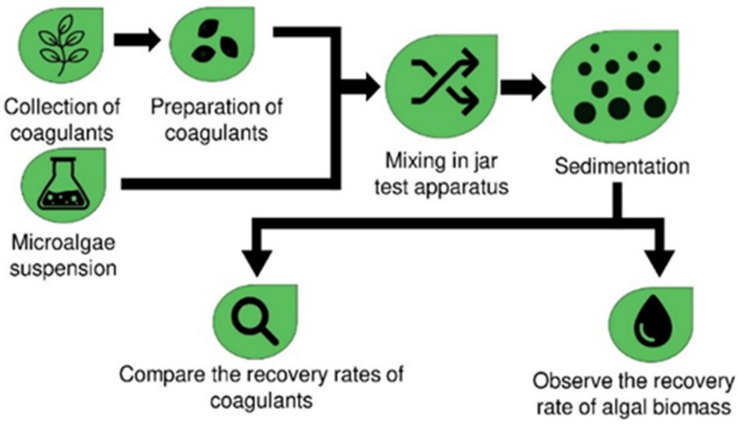

Figure 5: Research methodology applied for using *Moringa Oleifera* as biocoagulants.

Figure 6: Jar test of microalgae coagulation and flocculation process.

3. RESULTS AND DISCUSSION

3.1 Microalgal Growth

Four PBRs were was used to cultivate microalgal biomass and during the cultivation period, the microalgal growth rate was monitored as mention in Figure-7. The microalgal concentration was increasing slowly up to

day-3 that indicated the biomass was in the lag phase, where microalgae cells were growing slowly and adapting to the new environment. After day-3, the rapid growth was observed up to day-5 and maximum growth was ranged between 0.58 to 0.75 g/L. After then, the concentration of was decreasing due to the limitation of nutrients and also low light availability arising from self-shading at high microalgae density (Uggetti et al., 2014). Finally, the cultivated microalgae was harvested using MO seed powder.

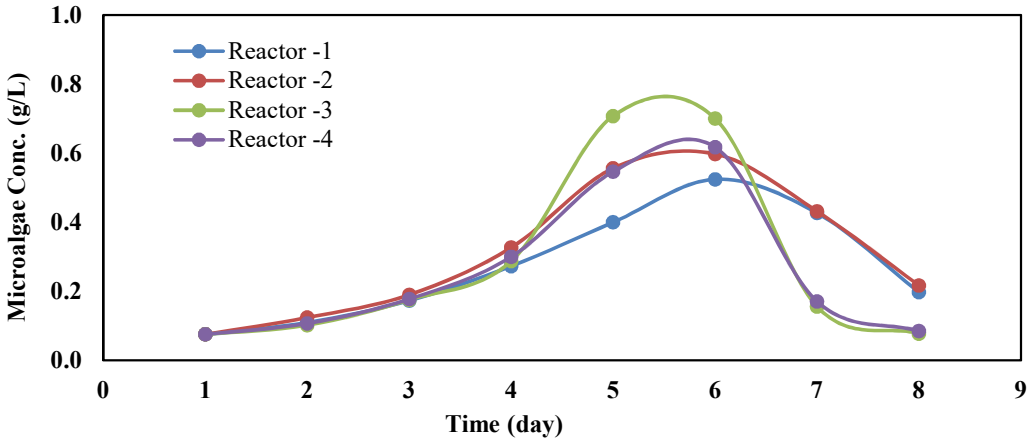

Figure 7: Microalgal growth in PBRs.

3.2 Coagulant Dose

Gorin et al. (2015) reviewed the importance of the optimization of coagulant dose in the coagulation flocculation process. In Figure-8, the recovery rate observed to be increased with the higher dose. The rate of microalgae recovery is proportional to the coagulant dose for any mixing time and mixing rate. This means a higher dose of MO seed powder will harvest a higher number of microalgae. Kurniawan et al. (2022) reviewed that MO seeds contain chemical compounds ofAmides group, Ester group, and Hydroxyl group that helps to form bridging structure that enhances the microalgal adsorption capacity. In this context, a higher dose of 70 mg MO seed per litre of microalgae solution is recommended for an effective harvesting process.

3.3 MixingRate

The mixing rate can be an important parameter for an efficient treatment. For 10 mins mixing time (**Figure-8**), the increasing mixing rate had no significant impact on the recovery rate. On the other hand, for 30- and 50-mins mixing time, the removal rate is reduced with higher rpm. 60 and 100 rpm mixing rate resulted in similar results for higher mixing time. Best results obtained at 10 minutes mixing time at every mixing rate. Nonfodji et al. (2020) investigated that MO seed powder showed high performance at mixing rate of over 100 rpm. So, higher 60-100 rpm is recommended to harvest microalgae with MO seed powder.

3.4 Mixing Time

Three mixing times were tested by varying three mixing rates to evaluate the impact of mixing time. As shown in Figure-9, higher mixing time does not increase the removal rate. Relatively low mixing time shows better results.

3.5 Optimum Dose

The higher dose of MOseed powder resulted in a higher removal rate. The mixing rate has no significant impact if the mixing time is low. However, for higher mixing time, the mixing rate should be kept low to reach optimum. If the mixing time is low, any mixing rate can be chosen. But in the case of higher mixing time, an intense mixing rate will give the best results. Based on the results, the highest microalgae were recovered (i.e., 83%) using a coagulant dose of 70 mg/L.

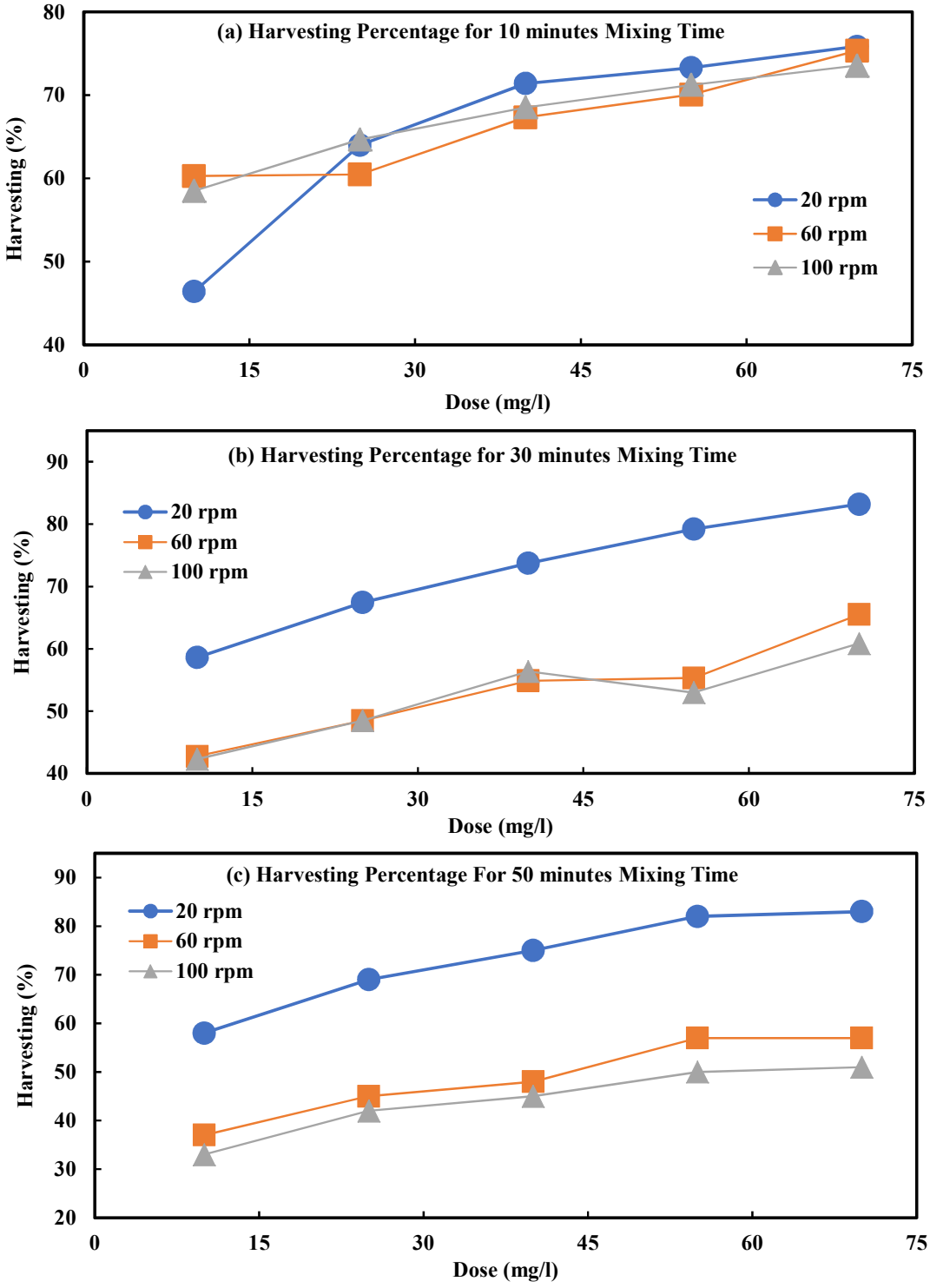

Figure 8: Influence of dosing rate on microalgae harvesting for varying mixing rate (20 rpm, 60 rpm and 100 rpm) and mixing time of (a) 10 mins, (b) 30 mins, and (c) 50 mins.

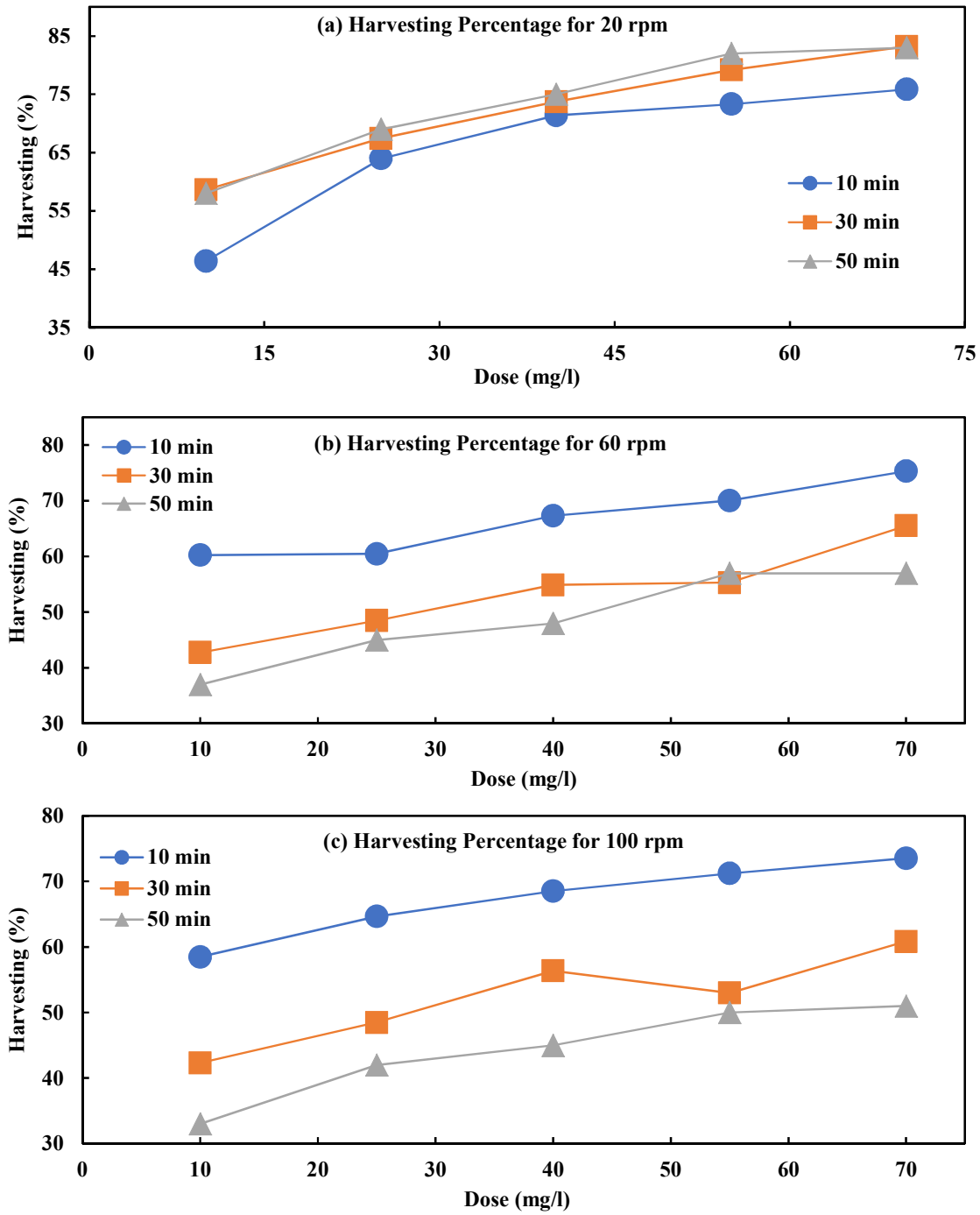

Figure 9: Influence of mixing time (10 mins, 30 mins, 50 mins) on harvesting efficiency for varying mixing rate of (a) 20 rpm, (b) 60 rpm, and (c) 100 rpm.

4. CHALLENGES AND PROSPECTS OF *MORINGA OLIFERA* BIOCOAGULANT

The use of toxic chemicals for microalgae harvesting is growing day by day which possess a potential threat to the environment. These chemicals are non-biodegradable and the environmental exposure of these chemicals hampers the ecological balance of nature (Kabir & Khalekuzzaman, 2022). In this context, biocoagulants play an important role to reduce the environmental impacts by replacing the toxic chemical coagulants (Kabir et al., 2021). Teixeira et al., (2012) recommended *Moringa oleifera* as an economic biocoagulants for microalgae harvesting for processing biodiesel. Again, the toxicity of biomass could be removed by using natural

flocculants. Mubarak et al. investigated *Moringa oleifera* to harvest microalgae and observed the removal of toxicity from harvested biomass (Mubarak et al., 2019). To conclude, the economic and environment friendly coagulant for microalgae harvesting is *Moringa oleifera*. Again, the harvested biomass could be efficiently converted to biofuels.

5. CONCLUSION

This study explored the coagulant efficiency of *Moringa oleifera* for harvesting microalgal biomass. Increasing coagulant dose has a positive impact on the microalgae removal rate. However, a higher dose of coagulant results in higher harvesting cost. In such circumstances, higher mixing rate & lower mixing time can be adopted, where a low dose of coagulant gives an almost similar result as a high one. Low mixing time & mixing rate always had a positive impact on a better economy. To conclude, the coagulant dose of 70 mg/L, mixed at a rate of 20 rpm for 10 minutes, is recommended for microalgal economic recovery (i.e., 83%) from wastewater. The recovered biomass has the potentiality of biofuel conversion for an economic biorefinery approach.

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