Algae Growth in Natural Water Resources

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Abstract

Algae have attracted much interest for production of foods, bioactive compounds and biofuel recently. We utilized natural water medium to grow algae, to understand the algae niche in ecosystem and to demonstrate the possibilities of production algae of water body in laboratory. The photo-reactors were designed to base on the universal bioreactor principles. The parameters of the medium and reactor were monitored during growth period. The study showed that natural water resource had suitable nutrients for microalgae to grow. The macro nutrients such as nitrogen and phosphorus were utilized by algae enormously and the system performed well in nitrogen and phosphorus removal. The pH and dissolved oxygen was raised up with the algae growth during the photosynthesis. The increase of chemical oxygen demand in reactor was contributed by the algae sludge which was decomposed through bacteria. We also find the possible CO₂ absorption from air. The long-term continuous operation of the culture process was expressed the reliability of the photosynthetic performance. Algae biomass by Total suspended solids (TSS) as 0.1 g/L and Chlorophyll-a resulted 1.1 mg/L. We proved the production of algae by the natural water is potentially feasible in ecological wisdom. The study will be helpful to understand the algae growth activity in water body better.

(Keywords: Algae, Bio-reactor, Water body, Water resources, Eco-tech.)

Introduction

Algae are playing an important role in this world and it is the predominant primary producer in any aquatic habitats (Oswald, 2003). It is very important ecologically because they are involving in symbiosis with bacteria in various ecosystems (Rebecca et al., 2009; Cole, 1982). It provides food and oxygen for many species in the aquatic environment and it’s vitally crucial to keep CO₂ of carbon cycle via photosynthesis (Anesio et al., 2009) to balance the CO₂ concentration in atmosphere (Sawayama et
Microalgae growth rate is the highest compared with the other plants (Andersen, 2005; Demirbas, 2009). Since it is an excellent biomass producer (Hsieh and Wu, 2009), the biomass is broadly extracted to obtain various biochemical used as medicine (Shimizu, 2000), nutrition (Oswald, 1962), food (Pulz and Wolfgang, 2004) etc. For energy crisis, algae biomass provide an innovative function as a renewable energy source, so called “bioenergy”, (Chisti, 2007; Sharif and Salleh, 2008) and turn algae as the most efficient bio-oil maker (Shay, 1993; Sheehan et al., 1998; Walker, 2009). It is one of the most important bio-technological species currently.

At present, ecological design has been applied to an increasingly diverse range of technologies and innovative solutions for the management and utilizations of resources. Algae are a highly diverse group of organisms that have important functions in ecosystem. Our thoughts owe a great debt to the ecologists who saw their science having a role to play in future. Since there are great potential of algae utilizations, algae application are broadly applied. Recently, most of studies use chemical medium which is nutrition enriched to grow algae. That method could get great algae production quickly, but possibly come out some problems like nutrition pollution, high cost, etc. Therefore, natural medium is another alternative which could accomplish both the goal of algae production and ecological function. We used natural water medium with mixing algae culture to mimic the natural system. Consequently, the purpose of this study was to evaluate the natural water medium with algae growth conditions in water body.

Materials and methods

1 Photoautotrophic cultivation systems

The mixed culture microalgae were collected from SRSE-LAB (Sustainable Resources and Sustainable Engineering research lab), Department of Soil and Water Conservation, National Chung-Hsing University, Taichung, Taiwan. The algae were grown in autotrophic conditions of algae/bacterial units with 10 days detention time, for 3 year’s period with the batch-fed in 4litres continuously stirred tank reactor (CSTR) under room temperature and illumination through fluorescent lamps. We used triplicate production units: P1, P2, P3 and the systems were shown in figure 1.

2 Medium preparation and analysis of environmental parameter

Neighboring river water was collected at Fu-Te Dao temple in Green river and the collection site is shown in figure 2. After the collection, the water was filtrated by 0.45 μm filter paper as feed. All the feed and growth condition indexes including of pH, alkalinity, dissolved oxygen (DO), chemical oxygen demand (COD), ammonia (NH₄⁺-N), Kjeldahl nitrogen (TKN), nitrite (NO₂⁻-N), nitrate (NO₃⁻-N), total nitrogen (TN), total phosphorous (TP) in addition of algae biomass of total suspended solids (TSS) etc. were continuously monitored according to the standard method (APHA, AWWA and
WPCF, 1985) and specific methods were listed in table 1. Chlorophyll- $a$ (chl- $a$) was extracted with acetone by boiling (Becker, 1994). The entire experiments were done in triplicate. Dominant algal species included *Anabaena*, *Chlorella*, *Oedogonium* and *Oscillatoria*.

Table 1. Chemical Analysis

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Analysis Method</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>SUNTEX Digital pH Meter, Model: SP-7</td>
</tr>
<tr>
<td>DO</td>
<td>TOA-DKK DO Sensor, Model: WQC-24</td>
</tr>
<tr>
<td>COD</td>
<td>Method 508B</td>
</tr>
<tr>
<td>NH$_4^+$-N</td>
<td>Method 417A with Method 417B for final ammonia</td>
</tr>
<tr>
<td>TKN</td>
<td>Method 420A with Method 417B for final ammonia</td>
</tr>
<tr>
<td>NO$_3^-$-N</td>
<td>Method 418A</td>
</tr>
<tr>
<td>NO$_2^-$-N</td>
<td>Method 419</td>
</tr>
<tr>
<td>TN</td>
<td>$= [TKN] + [NO_2^-N] + [NO_3^-N]$</td>
</tr>
<tr>
<td>TP</td>
<td>Method 424E following sulfuric acid-nitric acid digestion of Method 424C</td>
</tr>
<tr>
<td>TSS</td>
<td>Method 209C with Whatman GF/C filter paper</td>
</tr>
<tr>
<td>Chlorophyll $a$ (mg L$^{-1}$) = $12.7A_{663} - 2.69A_{645}$ (Becker 1994)</td>
<td></td>
</tr>
</tbody>
</table>

Results and discussion

1 pH

pH was measured from feed and reactor; the results were shown in figure 3. Feeding pH was 6.5 with the range of 7.1 - 8.1, while the reactors were 10.3 and ranged in 9.7 - 10.7. In general, the pH of the reactor increases during the period of
photosynthesis as a consequence of nutrient uptake to consume the bi-carbonate (Ai et al., 2008). The CSTR was around 10 which were obviously higher than feeding. The rationale to explain that phenomenon might be the reactor type. The CSTR with ideal mixing could optimize algae to access nutrients and to uptake dissolved inorganic carbon easily. According to Le Chatelier's principle, the dynamic equilibrium provided driving force to grab CO₂ from air and the alkalinity system would express in hydroxide ion at that high pH to reinforce the drag force of the absorption.

According to the carbonate equilibrium chemistry, algae growth with CO₂ uptake is permitting the growth unit because of carbon limitation to follow the equations bellowed:

a) \[ \text{CO}_2 + \text{H}_2\text{O} \rightleftharpoons \text{H}_2\text{CO}_3 \rightleftharpoons \text{HCO}_3^- + \text{H}^+ \]  
\( \text{(pKa-1)} \)

b) \[ \text{HCO}_3^- \rightleftharpoons \text{CO}_3^{2-} + \text{H}^+ \]  
\( \text{(pKa-2)} \)

c) \[ \text{CO}_3^{2-} + \text{H}_2\text{O} \rightleftharpoons \text{HCO}_3^- + \text{OH}^- \]  
\( \text{(pKa-3)} \)

Over the high range of pH, algae consumed bicarbonate according to pKa-3, and relieved OH⁻ to keep high pH in system. That is why the pH was increased in the reactor.

2 Alkalinity

Alkalinity was to measure the carbon conditions in feed and reactor; the results were shown in figure 4. Feeding was 98.1 mg/L as CaCO₃ with the range of 52.9 - 140 mg/L as CaCO₃, while the reactor were 52.7 mg/L and ranged in 32.7-74.7 mg/L. Figure 4 expressed the medium alkalinity was higher than reactor’s. Alkalinity changes were generated by algae growth (Brewer and Goldman, 1976). In the ecosystem, alkalinity is an important measurement of buffering to keep stable pH and to maintain a fairly optimal growth range in the water body (Godfrey, 1996).

![Figure 3. pH in feed and reactor](image-url)
3 Dissolved oxygen (DO)

Figure 5 indicates the Dissolved oxygen changes. Feeding DO was 5.4 with the range of 3.3-8.2 mg/L, while the reactor was 7.4 mg/L and ranged in 6.3-9.5 mg/L. During the photosynthesis, the algae released oxygen into the reactor, the content of the DO in the solution increased (Ai et al., 2008). Our CSTR is an algal bacterial unit which is the simplest symbiotic system to demonstrate the photosynthesis process increasing the DO level.

4 Chemical oxygen demand (COD)

COD was measured the nutrition carbon in feed and reactor; the results were shown in figure 6. Feed was 5.6 mg/L with the range of 1.7-14.0 mg/L, while the reactor was 13.3 mg/L and ranged in 8.4-22.0 mg/L. After algae growth the concentration of COD increased significantly about 2 times. The algae were grown autotrophically in algae/bacterial units which are the simplest ecosystem. Accordingly all age variety of algae existed and the old algae could be decomposed by bacteria to raise the COD up in reactors.
5 Total nitrogen (TN)

Total nitrogen was measured nitrogenous nutrition; the results were shown in figure 7. Feed TN was 7.5 mg/L as N L, while in reactor was 2.8 mg/L. Nitrogen was one of three important macro nutrition of algae growth (Becker, 1996). The medium TN was higher than effluent and nitrogen were utilized by algae vastly.

Algae require nitrogen to make amino acids to build proteins, DNA and RNA. Total nitrogen was containing ammonia, nitrite, nitrate and organic nitrogen in water. Our algal growth unit has symbiotic function. The bacteria play an important role in nitrogen cycle. Nitrosomonas bacteria convert ammonia into nitrite and then nitrobacter convert nitrite to nitrate (Peter, 2006). Algae may take up them as table 2 and our results demonstrated well about the nitrogen uptake of algae. From our results 1 g algae production needs 0.035g of nitrogen.

6 Total phosphorus (TP)

The concentration of phosphorus present in water medium has a direct influence on algal growth (Li et al., 2010). TP was measured phosphorus nutrition; the results were shown in figure 8. Feed TP was 0.6 mg/L with the range of 0.2-1.2 mg/L, while in reactor was 0.1 mg/L and ranged 0.01-0.4 mg/L. Phosphorus is an essential nutrient for algal growth, since it participates in intracellular energy transfer, nucleic acid synthesis, and special reactions associated with cell division (Martínez et al., 1999). It is generally considered to be the limiting nutrient of algal growth in fresh water body (Elser et al., 1990). The results of this study show phosphorus was utilized by algae enormously.

From our results 1 g algae production needs 0.004 g of phosphorous.

7 Biomass production

7.1 Total suspended solids (TSS)

The algal biomass was determined by dry weight according to “standard method” (Claudio et al., 2004). Since the feed was filtrated by 0.45 μm filter paper, there are no algae in feeding and the results were shown in figure 9. Our average biomass
Figure 7. Total nitrogen of feed and reactor

Figure 8. TP in feed and reactor

<table>
<thead>
<tr>
<th>Nitrogen concentrations</th>
<th>Feeding</th>
<th></th>
<th></th>
<th>Reactor</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Min</td>
<td>Max</td>
<td>Av</td>
<td>Min</td>
<td>Max</td>
<td>Av</td>
</tr>
<tr>
<td>NH₄⁺-N (mg/L)</td>
<td>0.0</td>
<td>5.1</td>
<td>1.2</td>
<td>0.0</td>
<td>2.3</td>
<td>0.8</td>
</tr>
<tr>
<td>NO₃⁻-N (mg/L)</td>
<td>0.3</td>
<td>8.9</td>
<td>3.5</td>
<td>0.0</td>
<td>2.2</td>
<td>0.5</td>
</tr>
<tr>
<td>NO₂⁻-N (mg/L)</td>
<td>0.0</td>
<td>5.7</td>
<td>1.3</td>
<td>0.0</td>
<td>0.4</td>
<td>0.1</td>
</tr>
<tr>
<td>Org-N (mg/L)</td>
<td>0.0</td>
<td>2.2</td>
<td>0.9</td>
<td>0.3</td>
<td>5.7</td>
<td>1.2</td>
</tr>
<tr>
<td>TKN (mg/L)</td>
<td>0.0</td>
<td>10.3</td>
<td>2.4</td>
<td>0.4</td>
<td>6.9</td>
<td>2.2</td>
</tr>
<tr>
<td>TN (mg/L)</td>
<td>4.1</td>
<td>15.1</td>
<td>7.5</td>
<td>0.6</td>
<td>7.6</td>
<td>2.7</td>
</tr>
</tbody>
</table>

production was 0.13 g/L and ranged 0.07-0.21 g/L.

The biomass of algae is a good source of nutrients and biologically active ingredients (Plaza et al., 2009). Most microalgae biomass are a rich source of omega-3 and omega-6 fatty acids, essential amino acids and carotene (Sánchez et al., 2002). Algal biomass became very important in the field of aquaculture (Muller-Feuga, 2000) and algae have attracted a great attention to produce fine chemicals as useful supplements of humans and animals (Chetsumon et al., 1994; Dallaire et al., 2007),
to absorb heavy metals (Lodeiro et al., 2005; Karthikeyan et al., 2007) and to fix carbon dioxide which is the current main issue of bioenergy (Sung et al., 1999; Chae et al., 2006).

7.2 Chlorophyll-a

Chlorophyll-a (Chl-a) as an algal biomass measurements was very popular (Ruley and Rusch, 2002; Ferrier et al., 2005). It is the most widely used proxy measurement (Todd et al., 2008). Their determination is relatively simple and straightforward. The results were shown in figure 10. The average value was 1.1 mg/L with the range from 0.8 to 1.5 mg/L. Chlorophyll is one of the valuable bioactive compounds (Luisa et al., 2010) and chlorophyll-a is the principal photo-chemically pigment which functions as a receiver of light for photosynthesis (Tremblin et al., 2000; MacIntyre et al., 2002; Carlota et al., 2004). It has a wide range of applications such as an additive in pharmaceutical (Hendry, 1996), cosmetic products and natural food coloring agent (Carlota et al., 2004). Additionally, it has antioxidant and antimutagenic properties (Aris et al., 2010). Chlorophyll is similar in chemical structure to hemoglobin and, as such, is predicted to stimulate tissue growth in a similar fashion through the facilitation of a rapid carbon dioxide and oxygen interchange (Tatyana et al., 2008).

![Figure 9. TSS in feed and reactor](image)

![Figure 10. Chlorophyll-a in feed and reactor](image)
Conclusion

This study was undertaken as an effort to develop a technique which could provide methods and information regarding algae growth to mimic a natural environment. Photo-bioreactor was implemented to grow the mixed natural algae species culture. The long-term continuous operation of this setup was expressed the reliability of the photosynthetic performance. Our lab experimental investigation was partially explained the microalgae growth in natural water body to understand the primary production of algae niche in ecosystem better and to make natural resource application more sustainable.

References


