Characterisation of Macrophyte \textit{Eleocharis dulcis} for potential selected bioproducts

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Abstract. Overpopulation of macrophytes in drainage ditches can lead to a problem in maintenance and reduce the efficiency of the system. Therefore, this study aims to evaluate the feasibility of \textit{Eleocharis dulcis} (\textit{E. dulcis}) for bioproduct potential by chemical composition analysis. \textit{E. dulcis} samples were collected from Parit Raja drainage ditch and sent to Malaysian Agricultural Research and Development (MARDI) for fibre content (cellulose, hemicellulose and lignin) and heavy metals (Cu, Pb, Zn) concentration. From the result, the fibre content of \textit{E. dulcis} show the potential for bioethanol with an average value of 37.62\% cellulose, 21.31\% hemicellulose and 12.25\% lignin. Meanwhile, Cu, Pb and Zn concentrations were 2.78, 0.63 and 2423.51 mg/kg, respectively. These values were low compared to WHO/FAO (2007), EN 13432 and Malaysia Food Regulations 1985, except for Zn, which has a slightly higher value indicating that it is suitable to be used as a bio-based straw. In conclusion, \textit{E. dulcis} had the potential as biomaterial straws and bioethanol based on the chemical compositions. Therefore, the productions are recommended for further analyses of the bioproduct performances.

1 Introduction

Parit Raja is one of the towns located in the district of Batu Pahat, Johor that is equipped with an open-drainage ditch as its irrigation system. The role of a drainage ditch is to direct the surface flow to the catchment area [1] and the drainage system should not be restricted by any means and be well maintained to avoid flooding [2]. However, the overgrowth of macrophytes in the drainage leads to difficulty for maintenance and reduces the system’s efficiency. According to the Department of Drainage and Irrigation Malaysia (DID), disrupted drainage water flow due to the high paced growth of macrophytes was a prolonged problem and needed to be overcome [3]. Therefore, the production of bioproducts can be seen as sustainable use of macrophytes. A bioproduct is a product produced for commercial purposes and from biomass, including biological materials (from agriculture, forestry, and marine sources), biological and microbiological waste, and by-products from processing and wood-based products [4]. The benefits of bioproducts are not just on the environmental aspect but also the health and economy, which can also be a

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source of income for the local community [4]. Hence, the production of bioproduct from macrophytes can benefit the local villagers of Parit Raja and its natural environment in the future.

*Eleocharis dulcis* is a macrophyte that belongs to the Cyperaceae species originally from South East Asia [5]. It is an aquatic plant and dominant weed, especially in swamplike areas. *E. dulcis* can grow rapidly in acidic environments and low nutrients soils [6]. *E. dulcis* consist of 40% of cellulose that can be used for various useful materials [7]. Production of bioproduct using *E. dulcis* can be a problem solver towards environmental issues especially involving plastic. However, research on *E. dulcis* application is very limited. In this study, three heavy metals, Cu, Pb and Zn, were considered as harmful. The heavy metals were chosen based on their toxicity level towards plants, animals and human health. High-dose of heavy metals exposure, particularly Pb and Cu, may induce severe complications such as abdominal colic pain, bloody diarrhoea, and kidney failure [8, 9]. This research aims to analyse the chemical compositions of *E. dulcis* for bioproduct production. The focused bioproducts were biomaterial straws and bioethanol for the conversion from overpopulated macrophytes.

2 Materials and methods

2.1 Study area and samples collection

*E. dulcis* was collected along 10 meters of the drainage ditch at Parit Raja in front of UTHM. The area was chosen due to the high density of the *E. dulcis* population present. A total of three samples were collected. For this study, the plant sampling method adopted was line transect. Figure 1 shows the layout for plant sampling. The sampling points were established at 0 m, 5 m and 10 m. The plant that lies within the points were collected, and each sample was weigh to 450 g.

![Sampling method layout](image1)

(a)

![Schematic diagram of sampling points](image2)

(b)

**Fig. 1.** Sampling method layout: a) Sampling at the site, and b) Schematic diagram of sampling points
2.2 Characterisation of E. dulcis

Samples of *E. dulcis* were sent to a commercial and accredited lab, MARDI Lab at Serdang, Selangor, to identify the composition of chemical compounds to deem the potential of *E. dulcis* bioproducts. The analysis conducted were categorised into two main categories, heavy metals (Cu, Pb and Zn) and fibre (cellulose, hemicellulose and lignin). Testing for the concentration of heavy metals present in *E. dulcis* was to ensure the *E. dulcis* is free from hazardous and toxic substances for the production of bio-based straws. Furthermore, the value of heavy metals shows the degradability of the macrophyte. Meanwhile, the testing for fibre content, cellulose, hemicellulose, and lignin proved *E. dulcis* as lignocellulosic biomass and a potential feedstock for bioethanol conversion. The value of the fibres content helped establish the theoretical yield of the biomass.

2.3 Estimation of theoretical Ethanol yields.

The cellulose and hemicellulose values were used to express hexose (HEXT) and pentose (PENT) sugar yield in the calculation since the conversion of the sugar (hexose and pentose) from biomass was assumed to convert 100%. The hexose and pentose sugar yields equations were obtained from Equations 1 and 2 [10].

\[
HEXT \ (kg/ton) = b \times Y_h \times HEX
\]

\[
PENT \ (kg/ton) = b \times Y_p \times PEN
\]

where,

- \(HEX\) = theoretical hexose (\(\Delta\)-glucose) yield from hexosan (C6 =1.111)
- \(PEN\) = theoretical pentose (\(\Delta\)-xylose) yield from pentosan (C5 = 1.136)
- \(Y_h\) = % cellulose in sample
- \(Y_p\) = % hemicellulose in sample
- \(b\) = weight of substrate (1000 kg)

Meanwhile, the equations developed for theoretical ethanol yield (ETOHTLT) based on simultaneous saccharification and fermentation with an assumption of 100% fermentation yield from hexose (HEXTEL) and pentose (PENTEL) were expressed as Equations 3, 4 and 5 as follows [5]:

\[
HEXTEL \ (kg/ton) = HEXT \times Y_{eh}
\]

\[
PENTEL(kg/ton) = PENT \times Y_{ep}
\]

\[
ETOHTLT(L/ton) = (HEXTEL + PENTEL)/d
\]

where,

- \(Y_{eh}\) = theoretical ethanol yield from hexose (0.511)
- \(Y_{ep}\) = theoretical ethanol yield from pentose (0.511)
- \(d\) = ethanol density (0.789 kg/L)

2.4 Permissible limit of heavy metals

To make credible and quality findings, a comparison to establish standards was made. The concentration of heavy metals in *E. dulcis* were compared to the permissible limit by
WHO/FAO (2007), EN 13432 (Standard for compostable and biodegradable packaging) and Malaysia Food Regulation 1985.

3 Result and discussion

3.1 The concentration of heavy metals

The concentration of Cu, Pb and Zn in *E. dulcis* was analysed and the result is given in Table 1. Those heavy metals are clear indicators of environmental contamination [11, 12]. The results of the heavy metals concentration in *E. dulcis* showed the following order of content Zn > Cu > Pb. The average concentration of Zn in this study was 2,423.51 mg/kg. The highest concentration of Zn was in Sample 1 with 2,963 mg/kg, while the lowest was 2,000.3 mg/kg. The concentration of Zn also had a huge difference between the concentration of Cu and Pb in the plant sample. The heavy metal concentration in the soils needs to be controlled to ensure the heavy metal concentration in *E. dulcis* does not exceed the standard limits, especially involving Zn concentration. One way to control the heavy metal in the soil is using the stabilisation method, which adds chemicals to the soils and causes the mineral's formation that contains the form of heavy metals that are not easily absorbed by plants, animals, or humans [13]. This method also, fortunately, did not disrupt the environment or produce hazardous waste. The average Cu content in *E. dulcis* was 2.78 mg/kg with the highest value of 2.88 mg/kg in S1, 2.81 mg/kg in S2 and 2.65 mg/kg in S3. The concentration of Pb in this study had an average of 0.63 mg/kg in all samples. The highest concentration was found in S1, followed by S3 and S2 with 0.92 mg/kg, 0.59 mg/kg and 0.37 mg/kg, respectively. There was a difference in the concentration of heavy metals in the tissues of the *E. dulcis* and slight contrast of each heavy metal between the three samples. These situations resulted from several factors that affect the uptake mechanisms of macrophytes, such as plant species, environmental conditions, and chemical properties of the contaminant [14].

<table>
<thead>
<tr>
<th>Heavy metals</th>
<th>Concentration (mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S1</td>
<td>S2</td>
</tr>
<tr>
<td>Cu</td>
<td>2.88&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Pb</td>
<td>0.92&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Zn</td>
<td>2963.00&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Table 1: Heavy metals concentration of *E. dulcis*

Convert from the unit<sup>a</sup> ppm and<sup>b</sup> ppb, S = Sample 1,2,3

3.2 Fibre content

Table 2 presents the cellulose, hemicellulose and lignin contents in *E. dulcis* from Parit Raja drainage. The average contents for each fibre are cellulose (37.62%), hemicellulose (21.31%) and lignin (12.25%). S3 has the highest cellulose value of 39.44%, followed by S2 and S1 of 37.62% and 35.8%, respectively. There is only a slight difference in the hemicellulose content between the three samples, with S2 having the highest hemicellulose contents of 22.69%, S1 of 21.34%, and only 19.90% of hemicellulose in S3. Furthermore,
lignin content is 12.63% in S1, 12.44% in S2, and 11.99% in S3. These values show that the fibre content (cellulose, hemicellulose and lignin) of *E. dulcis* are within the range of lignocellulosic content (cellulose: 9–80%, hemicellulose: 10–50%, lignin: 5–35%) [15]. This proved that *E. dulcis*, primarily found in the Parit Raja drainage, could be a promising carbohydrate source and feedstocks for bioproducts. The high composition of cellulose, hemicellulose and lignin will assure the suitability of becoming alternative feedstock for bioethanol and extraction of cellulose for biomaterials binder.

**Table 2:** Fibre content of *E. dulcis*: cellulose, hemicellulose and lignin (%)

<table>
<thead>
<tr>
<th>Fibre</th>
<th>S1</th>
<th>S2</th>
<th>S3</th>
<th>Average value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cellulose</td>
<td>35.80</td>
<td>37.62</td>
<td>39.44</td>
<td>37.62</td>
</tr>
<tr>
<td>Hemicellulose</td>
<td>21.34</td>
<td>22.69</td>
<td>19.90</td>
<td>21.31</td>
</tr>
<tr>
<td>Lignin</td>
<td>12.63</td>
<td>11.99</td>
<td>12.14</td>
<td>12.25</td>
</tr>
</tbody>
</table>

### 3.3 Biomaterial straw

The main aim of production straws from *E. dulcis* is that they should be safe for the consumer. In this study, the concentration of Cu, Pb and Zn were compared to WHO/FAO (2007), EN13432 and the Malaysia Food Regulation 1985 for toxicity level *E. dulcis* assessment. Figure 2a shows that the Cu concentration of all samples (2.88, 2.81 and 2.65 mg/kg) did not exceed the permissible limit. The limit set for Cu concentration by WHO/FAO (2007), EN13432 and the Malaysia Food Regulation 1985 is 40 mg/kg, 50 mg/kg and 10 mg/kg, respectively. The concentration limit set for Pb was 5 mg/kg by WHO/FAO (2007), 50 mg/kg in EN 13432 standard and 2 mg/kg for Malaysia Food Regulation 1985. All samples (0.97 (S1), 0.37 (S2) and 0.59 (S3) mg/kg) in this study are below the limit set for Pb, as shown in Figure 2b. However, the concentration of Pb (12.09 mg/kg) of the previous study exceeds all except the permissible limit by EN 13243. Next, the determined concentration of Zn (2963 (S1), 2000.3 (S2) and 2307.23 (S3) mg/kg) in this study was higher than the limit set by WHO/FAO (2007), EN13432 and the Malaysia Food Regulation 1985, fixed at 60 mg/kg, 150 mg/kg and 100 mg/kg, respectively, based on Figure 2c.

Even though Zn is considered an essential metal element for plants and non-toxic, extreme levels of concentration can cause Zn toxicity [16]. The excessive concentration of Zn in *E. dulcis* possibly due to the location beside the main road in Parit Raja. The anthropogenic sources of Zn were associated with brake and tyre wear, vehicle emission, traffic and street industrial activities [16]. Based on the chemical composition of *E. dulcis*, the production of biomaterial straw is feasible. *E. dulcis* is a suitable source of cellulose due to its fibre content. Meanwhile, in terms of toxicity, *E. dulcis* does not exceed the limit set by the established limit except for the Zn. However, it is unlikely to ingest enough zinc to cause harmful effects from the bioproduct. Therefore, the selection of the method for lowering the concentration should be considered for a safe bioproduct. A low concentration of heavy metals ensures the safety and increases the biodegradability of the products.
Fig. 2. a) Cu, b) Pb and c) Zn in *E. dulcis* samples compared with permissible limit by WHO/FAO (2007), EN 13432 and Malaysia Food Regulation 1985
3.4 Bioethanol

To investigate the feasibility of *E. dulcis* for bioethanol conversion, the theoretical ethanol yields from cellulose and hemicellulose of all samples were calculated. Furthermore, the theoretical ethanol yields from this study were compared to the previous studies of different biomass in Figure 3. The highest calculated ethanol yield in this study was 437.6 L/ton from S2, followed by a slight difference in the total ethanol yield in S3 with 430.2 L/ton. The lowest theoretical ethanol yield obtained was 414.6 L/ton from S1 of *E. dulcis*. In this study, the obtained theoretical yield of *E. dulcis* can be categorised as a high-level ethanol yield [17].

Compared to other lignocellulosic biomasses, *E. dulcis* ethanol yields were higher than *C. cyperoides*, *S. dulcis* and Switchgrass with 394.0 L/ton, 402.6 L/ton [17], and 399.6 L/ton [18], respectively. Meanwhile, the highest calculated theoretical ethanol yield was obtained from Napier grass with 566.3 L/ton [19]. From the obtained results, the correlation between cellulose, hemicellulose and ethanol yields can also be observed. A high yield of ethanol depends on the high amount of cellulose and hemicellulose. However, in a real production process, the efficiency of ethanol yields depends on several other factors discussed in the literature review. Overall, the theoretical ethanol yield results showed that *E. dulcis* could be a potential feedstock of biomass source for bioethanol conversion.

![Theoretical yield from various biomass](image_url)

**Fig. 3:** Theoretical yield from various biomass

4 Conclusion

Overall, the heavy metals concentration and fibre content in *E. dulcis* had shown a favourable value for bio-based straw and bioethanol conversion. The low value of heavy metals indicates a low toxicity rate and high biodegradability, ensuring safety for both humans and the environment. Several remediations can be taken to reduce the concentration of heavy metal, as suggested in the discussion. Meanwhile, the high value of cellulose,
hemicellulose and low lignin content assures the potential to become a feedstock for bioethanol production.

*E. dulcis* had the potential as biomaterial straws and bioethanol based on the chemical compositions. Therefore, the productions are recommended for further analyses of the bioproduct performances. More testing for the concentration of heavy metals present in *E. dulcis* needs to be done to ensure the *E. dulcis* is free from hazardous and toxic substances to produce bio-based straws. Meanwhile, the high value of cellulose, hemicellulose and low lignin content in *E. dulcis* assure the potential to become a feedstock for bioethanol production.

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