RESEARCH ARTICLE

Chemical characterization of bamboo leaves (Gigantochloa albociliata and Dracaena surculosa) by sodium hydroxide treatment [version 1; referees: 1 approved with reservations]

Nadiah Ameram, Muhammad Afiq Che Agoh, Wan Farhana W. Idris, Arlina Ali
Faculty of Bioengineering and Technology, Universiti Malaysia Kelantan, 17600, Jeli, Kelantan, Malaysia

Abstract

Background: Dracaena surculosa and Gigantochloa albociliata leaves are different in morphology and appearance. Sodium hydroxide (NaOH) is widely used in pulping of wood for making paper or regenerated fibers. NaOH is used to separate lignin from cellulose fibers, and this treatment is needed to identify the characteristics of leaves. This study was conducted in order to investigate the characteristics of D. surculosa and G. albociliata species under NaOH treatment.

Methods: NaOH was applied to the leaves for 8 hours. Treated and untreated bamboo leaves were analysed using FTIR analysis, in order to identify the presence of functional groups in the leaves.

Results: It was observed that these two species not only differ physically, but also chemically. The presence of OH, CH and alkynes functional groups in the leaf sample indicates that the species share similar properties but have a slight difference in the molecular bonds. From the morphological observation of D. surculosa and G. albociliata leaves, they are slightly different in terms of leaf appearance and characteristics. G. albociliata have thicker leaves compared to D. surculosa, and NaOH treatment shows that D. surculosa leaves are harder to dissolve into the solvent. Scanning electron microscope (SEM) analysis of these two species shows the initial structure of fibres in the leaves are intact but after NaOH treatment, the fibres are ruptured and appear in non-uniform shapes.

Conclusions: The initial morphology of G. albociliata and D. surculosa is different in color and appearance. However after NaOH treatment, the color becomes almost the same. Regarding SEM analysis, after NaOH treatment the morphology of the bamboo leaves completely changes. Therefore, it can be concluded that the process of hemicellulose removal had occurred during treatment. The results show that lignin has been removed by NaOH treatment to enhance the characteristics of the bamboo leaves from different species.

Keywords
Bamboo leaves, Sodium hydroxide, Chemical treatment, FTIR, SEM
Introduction

Bamboo is one of the most common plants available in Malaysia and is known for its multiple uses for construction, transportation and cultural purposes. About 70 species of bamboo (50 in Peninsular Malaysia, 30 in Sabah and 20 in Sarawak) can be found in Malaysia\(^1\). Being abundant throughout the whole country, bamboo can be treated as a source of income. Its leaf is claimed to have medicinal benefits among traditional practitioners\(^2\). Bamboo leaves, also known as Lophaterum, and bamboo shavings are commonly used as relievers for stomach aches, cooling effects and also help to counter the negative flow of qi\(^3\).

Asian populations include bamboo in their dishes, especially Chinese individuals. In China and Southeast Asia, bamboo had been used as source of food and medicine for a long time\(^4\). Bamboo grows one third faster than the fastest growing tree\(^5\) and some species can grow up to 60 meters high. Bamboo can grow up to 1 meter per day and therefore it is easily accessible in a minimal amount of time. The optimum age for bamboo to be harvested is at 3–5 years old. For thousands of years, bamboo had been part of human’s diet.

In addition, the Indian elephant and the Giant Panda of China also utilize bamboo leaves as their main food source, which become their exclusive meal. The skeletal system of the panda is incredibly strong, and flexible and it can be related to their diet of bamboo leaves, which contains a great amount of silica\(^6\). Usage of silica includes silica as dessicants, as a cement, and in the production of tires. Silica gel desiccants are used as pharmaceutical-dessicants/silica gels to promote drying by absorbing water vapor and gases, like oxygen and hydrogen in humidity. It is very important in pharmaceutical industries as humidity and moisture can ruin pharmaceutical products\(^7\). Silica has been utilized as a cement substitution, as silica exists as quartz in the silica sand. It has also been used as a fractional substitution of cement during solid development\(^8\). In tires, mixing silica with carbon improves the tire performance on the road\(^9\), as silica helps to provide flexibility through low-heat build-up.

*Dracaena surculosa* is a fast growing species of bamboo and is therefore preferable as an indoor plant. This species also prefers partial shade for growth. Exposure to bright lights may cause the plant to lose or shed its leaves. A full grown *D. surculosa* leaf could be as long as 7 – 12 cm long. The surface of the leaf is leathery and smooth with obvious patterns of yellow or gold spots (Figure 1).

Popular for its shoots, *Gigantochloa albociliata* is commonly planted and harvested as a food source and decoration. The shoots are edible and in Japan canned bamboo shoots are used. This bamboo species can easily grow in dry tropical mixed forest from low to medium elevations. The leaves are linear - lanceolate with the width of 15–20cm × 2–25cm (Figure 2). In the dry season, the leaves are shed\(^10\).

The abundance of bamboo in Malaysia may give a great advantage to the nation. However when harvested, not all parts of the bamboo are fully utilized, the leaves being especially discarded.
This research aims to determine the organic compounds in bamboo leaves in two species, *D. surculosa* and *G. albociliata* after sodium hydroxide (NaOH) treatment. Along with sodium sulfide, NaOH is used to separate lignin from cellulose fibers in the craft process of making paper. Thus, this treatment is needed to separate lignin from fibers in order to identify the characteristics of leaves and was therefore used in the present study.

**Methods**

**Materials**

Sodium hydroxide (Quality Reagent Chemical [QREC], 1.0 M), nitric acid (QREC, 1.0 M) and toluene (QREC, 0.1 M) were utilized in this study. Bamboo leaves were collected with special permissions from Lundang Committee of Kg Lundang (*Dracaena surculosa*: 6°06'00.0"N 102°15'33.1"E) and Director of UMK Jeli Campus for UMK Agropark Kelantan (*Gigantochloa albociliata*: 5°44'46.0"N 101°51'58.3"E). All other chemicals and reagents used were of the highest commercially available purity.

**Solvent extraction method**

The bamboo leaves from *G. albociliata* and *D. surculosa* (5 g sample) was stirred in 125mL of 1.0M NaOH for 8 hours using a magnetic stirrer. The solution was filtered (filter paper grade 1, 11 µm) to separate the solute (bamboo leaves) and the aqueous solution (sodium silicate). The solution was then titrated with 1.0M nitric acid to obtain less than 3 pH in order to obtain the gel of suspended silica. Subsequently, this solution was centrifuged at 4000 rpm for 10 minutes. Then, the solution was left for 4 days to allow the silica gel to form. The silica gel were then filtered (filter grade 1, 11 µm) and extracted using 50mL of toluene. Finally, the solution obtained was dried over anhydrous sodium sulfate (QREC 1.0 M). This was then left overnight at room temperature. If any liquid remains, this should be collected and discarded. The final product will be analyzed using Fourier-transform infrared spectroscopy (FTIR). FTIR analysis is used to identify functional groups in a molecule by producing an infrared absorption spectrum. The spectrums were identified using Perkin Elmer Spectrum 100 FT-IR Spectrometer with range spectrum (wavelength): 4000–400/cm. Scan rate: 16, 32. Universal ATR sampling accessory (not using KBr) was employed to prepare the sample.

**Scanning electron microscopy (SEM)**

Each bamboo leaf sample was analysed using SEM to characterize the morphology of the specimen before and after treatment with NaOH. 30 g of each bamboo leaf was left untreated and ground to obtain a powder. In addition, 30g of bamboo leaves were treated with NaOH (30 mL) by stirring for 8 hours using a magnetic stirrer. The final solution was filtered using filter paper to obtain the solutes. The solutes were dried thoroughly before being ground into a powder to be analysed using SEM. The voltage used was between the ranges 1.5kV to 2.0kV. Secondary electron images were obtained.

**Results**

**Treatment with NAOH**

During NaOH treatment, *D. surculosa* leaves exhibit a rapid change in colour and texture. The initial colour for this sample is light green and yellowish. After being stirred with NaOH, the colour started to change to dark brown, and after 8 hours of treatment, the colour turned to very dark brown. A reason for the rapid colour change is due to the thickness of the leaves, as any pigment inside the leaf would be quickly released as the leaves are thin. In addition, after 8 hours of treatment, the leaves almost completely dissolved into the solution of NaOH. This makes it hard for the leaves to be used in the next treatment of solvent extraction.

For *G. albociliata* leaves, there was a slower reaction during treatment of NaOH. After a few hours of treatment, the colour of the leaves started to change to dark green (original color light brownish green) and after 8 hours, the colour turns to almost black. In contrast to *D. surculosa*, the leaves did not dissolve into the solution.

**FTIR analysis**

FTIR spectra of changes in functional group of both untreated and treated (NaOH) bamboo leaves of *G. albociliata* is illustrated in Figure 3.

The absorption band at 3293.32 cm\(^{-1}\) indicates that there is an O-H stretching vibrations between cellulose and hemicellulose\(^{11}\). Meanwhile the 2913.51 cm\(^{-1}\) for treated and untreated bamboo leaves demonstrate the presence of CH\(_2\) groups. The band also contains functional group of alkynes from the peak at 2117.32 cm\(^{-1}\). In accordance to the presence of this peak, it also indicates that before the treatment with NaOH, pectin and waxes are present in the bamboo leaves sample of *G. albociliata*\(^{12}\). Different results are achieved with bamboo leaves treated with NaOH where the peaks have almost disappeared. This shows that hemicellulose is present and is gradually removed from the sample during treatment. The lists of functional groups are shown in Table 1.

FTIR spectra of changes in functional group of untreated and treated *D. surculosa* leaf samples using 1.0M NaOH is illustrated in Figure 4.

The absorption band at 3294.48 cm\(^{-1}\) in *D. surculosa* leaves indicates that there is a O-H stretching vibrations between cellulose and hemicellulose\(^{13}\). Whereas the 2885.64 – 2895.44 cm\(^{-1}\) for treated and untreated bamboo leaves sample show the presence of CH\(_2\) groups. The band contains functional group of alkynes with triple bonds from the peak at 2117.32.51 cm\(^{-1}\). Different results are achieved with bamboo leaves treated with NaOH where the peaks had disappeared. The list of functional groups are shown in Table 2.

**SEM analysis**

The study of morphology requires SEM analysis before and after treatment is done, as the treatment may affect the morphology of the species\(^{14}\). Figure 5 shows a 400x magnification of *G. albociliata* leaf surface before treatment with NaOH.

From this figure, it can be observed that the bonding between fibers in the leaf are still intact as shown by the arrows in Figure 5.
Fibers play an important role in improving strength, providing higher initial modulus and reducing extensibility by improving the interfacial adhesion between them. From Figure 6, it can be observed that the leaves had no particular shape and the morphology changes completely after being treated with NaOH. The degradation of hemicellulose may occur during treatment, which causes the leaf to be dissolved in the solvent itself. From this observation, it can be concluded that NaOH is one agent that can degrade the molecular structure of bamboo leaves. Furthermore, high concentration of NaOH used in this test (1.0M) may contribute to the rapid degradation of the hemicellulose. Another observation using SEM analysis shows the structure of the fibres are in non-uniform shape.

The leaves of untreated D. surculosa still have bonds between the hemicelluloses. After the grinding process, the leaves are crushed but remains in solid form, as seen in Figure 7, and are not powdery like G. albociliata.

After treatment with 1.0M NaOH, the bamboo leaves become partially dissolved into the solvent, as can be seen in Figure 8. The final colour of the solution also changes to dark brown. This change of colour is caused by the removal of hemicellulose in the leaf itself.

Dataset 1. Replicates of FTIR analysis; bamboo leaves after NaOH treatment
http://dx.doi.org/10.5256/f1000research.15036.d208859

Discussion
From the FTIR analysis, the functional group found in the specimens can be identified based on the presence of the bands in the graphs. It can be concluded that the process of treatment and extraction had contributed to the organic compounds present in the samples of different species. Among the two samples, G. albociliata and D. surculosa, the initial morphology is different in two factors, colour and appearance. The initial colour of G. albociliata and D. surculosa is yellowish green and dark green, respectively. However after the treatment, the colour becomes almost the same. As for the SEM analysis, the treatment shows the morphology of the bamboo leaves completely changes due to the treatment with NaOH. Due to change in colour of NaOH, it can be concluded that the process of hemicellulose removal had occurred during the treatment. Bamboo is truly interesting to study as it has a unique anatomy and a
Figure 4. FTIR analysis on extracted Dracaena surculosa leaf samples. (Graphs are representative of 2 repeated experiments.)

Table 2. Functional groups in Dracaena surculosa leaves before and after treatment with sodium hydroxide.

<table>
<thead>
<tr>
<th>Before treatment with NaOH</th>
<th>After treatment with NaOH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak</td>
<td>Functional group</td>
</tr>
<tr>
<td>3294.48</td>
<td>OH</td>
</tr>
<tr>
<td>2895.44</td>
<td>CH</td>
</tr>
<tr>
<td>2117.32</td>
<td>Alkyne (triple bond)</td>
</tr>
</tbody>
</table>

Figure 5. SEM analysis of untreated Gigantochloa albociliata leaf sample at 400x magnification. White arrows show fiber bonding in the leaf.
Figure 6. SEM analysis of *Gigantochloa albociliata* leaf after treatment with NaOH at x400 magnification.

Figure 7. SEM analysis of untreated *Dracaena surculosa* leaves at 100k magnification. The white ring shows that the fiber bond which is intact but not clearly shown.

Figure 8. SEM analysis of *Dracaena surculosa* leaves treated with NaOH at 100k magnification.
superproductive behavior. Rhizomes are an essential part of bamboo anatomy. This study will be able to provide new data regarding the characteristics of *G. albociliata* and *D. surculosa* bamboo leaves after NaOH treatment.

**Data availability**

Dataset 1: Replicates of FTIR analysis; bamboo leaves after NaOH treatment. DOI, [http://dx.doi.org/10.5256/f1000research.15036.d208859](http://dx.doi.org/10.5256/f1000research.15036.d208859)

**Competing interests**

No competing interests were disclosed.

**Grant information**

We thank UMK research short term grant SGJP for funding (number R/SGJP/A13.00/00692A/001/2018/000498).

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

**References**

Open Peer Review

Current Referee Status: ?

Referee Report 18 July 2018
doi:10.5256/f1000research.16370.r35854

Noorfatimah Yahaya
Integrative Medicine Cluster, Advanced Medical and Dental Institute, Universiti Sains Malaysia, Penang, Malaysia

Ref: Chemical characterization of bamboo leaves (Gigantochloa albociliata and Dracaena surculosa) by sodium hydroxide treatment

This article describes a chemical characterization of bamboo leaves (Gigantochloa albociliata and Dracaena surculosa) through sodium hydroxide treatment. The topic is interesting, but it requires a revision according to the following comments:

1. Introduction: The authors should state the novelty and significant of the study in their manuscript.
2. Methods: ‘All other chemicals and reagents used were of the highest commercially available purity’. Instead, authors should state purity of each chemical and reagent used in the study.
3. Methods: FTIR should be in separate section from solvent extraction method. Details description of FTIR and SEM instruments should be included in text.
4. Results: FTIR spectrum should be improved. Only important peaks should be labelled in the spectrum. The spectrum should be improved in terms of resolution.
5. Results: SEM images should be combined together, so a clear comparison could be made.
6. Discussion: More discussion and explanation should be added in text to increase scientific values of the article.
7. CONCLUSIONS should be included in the text.

Is the work clearly and accurately presented and does it cite the current literature?
Partly

Is the study design appropriate and is the work technically sound?
Yes

Are sufficient details of methods and analysis provided to allow replication by others?
Partly

If applicable, is the statistical analysis and its interpretation appropriate?
Not applicable

Are all the source data underlying the results available to ensure full reproducibility?
Yes
Are the conclusions drawn adequately supported by the results?
No

**Competing Interests:** No competing interests were disclosed.

**Referee Expertise:** Analytical Chemistry

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

---

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com