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# Quantitative method applicable for various biomass species to determine their chemical composition

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ABSTRACT

A quantitative method applicable for various biomass species to determine their chemical constituents was explored. The widely used wood analytical method was found to be not entirely applicable to different biomass species. It was then demonstrated that by incorporating protein and starch determinations, by ash-correcting the Klason lignin and holocellulose and also by protein-correcting Klason lignin and holocellulose of high protein content species, reliable summative results that enable comparison between different types of biomass materials were achieved. Thus, an analytical method with starch and protein determinations as well as ash and protein corrections was proposed for quantitative assay of chemical composition of various biomass species.



## 1. Introduction

Although several efforts [1–3] were accomplished in the past to settle a universal method to determine the chemical components of biomass species, evidence with reasonable explanation to support the choice of a method or the addition of a given step in one procedure has never been provided. As wood is quantitatively predominant among biomass species [4], the method to determine its chemical composition is well established [5–7]. However, wood analytical procedure is not necessarily adequate for other kinds of biomass resources, especially for the herbaceous ones such as wheat straw and rice husk mostly used as feed and forage. Their chemical characterization is usually made, based on crop analysis for animal feeding, and terms and methods used to characterize herbaceous plants [8–10] differ from those to characterize wood [5].

Therefore, in order to determine biomass chemical composition on the same basis, an analytical method applicable to various biomass species has been studied in this work. For that purpose, widely used wood analytical method was discussed in its advantages and disadvantages, and a revised new analytical procedure applicable for different biomass species was established.

## 2. Materials and methods

Based on taxonomical classification of the vascular plants, six selected biomass species were analyzed on their morphological parts as exposed in Table 1 which shows their taxonomical classification, age, parts studied, sampling location and time, storage condition before delivery to the laboratory

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**Table 1 – Taxonomical classification of the biomass species and their details.**

| Classification | Species | Age (years) | Parts studied | Sampling location | Sampling time | Storage condition before delivery to the laboratory (Temp; humidity) | Sample condition during delivery | Remarks |
|---|---|---|---|---|---|---|---|---|
| Gymnosperm | Japanese cedar (*Cryptomeria japonica*) | 50 | Trunk | Kyoto, Japan (35°08′N, 135°66′E) | 12/2000 | 0–10 °C; 50–60% | Air-dried, thinned | |
| Angiosperm | | | | | | | | |
| Dicotyledon | Japanese beech (*Fagus crenata*) | 45 | Trunk | Kyoto, Japan (35°22′N, 135°82′E) | 11/2000 | 0–10 °C; 50–60% | Air-dried, thinned | |
| Monocotyledon | Bamboo (*Phyllostachys heterocycla* f. *pubescens*) | 5 | Culm | Kyoto, Japan (35°01′N, 135°46′E) | 06/2008 | 10–20 °C; 60–70% | Undried, cut into pieces | |
| | Rice (*Oryza sativa* var. *Japonica*) | 0.5 | Straw, Husk | Aichi, Japan (35°23′N, 136°87′E) | 10/2007 | 20–40 °C; 70–85% | Air-dried, husk separated from straw | Husk was detached from grain using a mechanical paddy de-husker |
| | Corn (*Zea mays* cv. Yumeno-corn) | 0.4 | Leaves, Cob | Aomori, Japan (40°49′N, 140°45′E) | 09/2008 | 10–20 °C; 60–70% | Undried, cob together with grains | Corn cob was separated from grains using a knife |
| | Oil palm (*Elaeis guineensis* Jacq.) | 25 | Trunk | Johor Bahru, Malaysia (1°46′N, 103°75′E) | 03/2006 | 25–35 °C; 70–85% | Air-dried, cut into blocks | |

and condition during delivery in accordance with the biomass sample definition checklist recommended by Barton [11].

Upon arrival in the laboratory, the samples were air-dried, milled with Wiley mill (1029-C, Yoshida Seikakusho Co., Ltd.), and sieved to retain particles of 150–500 μm, 35–100 mesh according to ASTM E11-01 [12] in size. They were kept at room temperature, with 10–30% humidity before analyses. These samples were oven-dried and the traditional wood analytical method [6] illustrated in Fig. 1 was firstly applied to characterize their chemical composition.

The oven-dried samples were incinerated at 600 °C [6] for 4h to determine ash content on the non-extracted samples. The non-extracted samples were then Soxhlet-extracted [13] with acetone until the solvent was clear of any color to determine the content of extractives. For these extractives-free samples, Klason and acid-soluble lignins were determined by using 72% sulfuric acid through a modified Klason method [14]. In brief, 15 ml of 72% $H_2SO_4$ was added to 1 g of extractives-free sample and left to react at room temperature for 2 h. Then, the mixture was diluted to 3% $H_2SO_4$, autoclaved at 121 °C for 30 min and filtered to obtain Klason lignin gravimetrically and acid-soluble lignin from UV absorbance at 205 nm, using absorptivity value of 110 $lg^{-1}.cm^{-1}$. Holocellulose was quantified with sodium chlorite treatment according to the procedure of Wise *et al.* [15] as adapted by

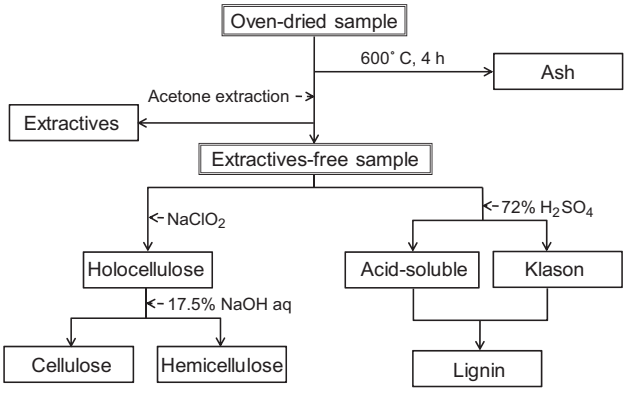


**Fig. 1 – Traditional wood analytical method to quantify its chemical composition [6].**

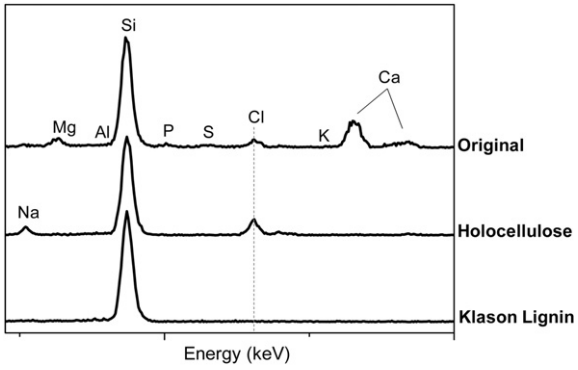


**Fig. 2 – Relative comparisons of the energy-dispersive X-ray spectra on the ashes for original sample of rice straw, its holocellulose and Klason lignin as determined by EDX analysis [23].**