Pretreatment and fiber content analysis of *Cannabis sativa*

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Abstract:

Cannabis sativa commonly known as hemp is one of the fastest-growing plants whose refined products have immense commercial value. Various products include refined hemp such as: biofuels, biodegradable plastics, textiles, dietary supplements, paper, clothing, and much more. Hemp fibers are also used in construction and manufacturing applications by strengthening their composite products. Hemp is a high yielding, sustainable, and environmentally friendly crop due to its various qualities, and has the potential to yield valuable raw materials for a great number of applications. Our research evaluates the pretreatment of hemp as well as the comparative analysis of the fiber content thereof. Our goal is to determine the suitability and the potential use of ionic liquid-based pretreatment (1-Butyl-3-methylimidazolium chloride) for the breakdown of hemp lignocellulosic biomass. The collected data is presented and discussed in the following sections.

Introduction:

Cannabis sativa, hemp, is a largely forgotten crop that was once popularly used as a major sustainable fiber for production. As a species of the Cannabis family, hemp contains 0.3% THC, compared to marijuana with 17.1% THC, allowing it to be a safe and compelling raw material. Hemp possesses natural properties that can replenish poor soil, thrive with little assistance, and grow without the need for pesticides, fertilizers, and much water. Therefore, hemp is considered a low input and low impact crop. The environmental impacts associated with the production of hemp fibers are smaller than those associated with most other crops; sugar beet, potato, which are considered high impact and high input crops. Hemp's adaptable species can be sustained in harsh environmental conditions. Additionally, hemp has a high and rapid yield due to its growth cycle of 108-120 days and its ability to grow four inches apart. These factors positively support hemp's resurgence in the textile, agricultural, pharmaceutical, and fuel industries.

Hemp is classified as second-generation biomass due to its composition of non-edible plant material such as lignocellulose and pectin. Lignocellulose comprises three polymers: cellulose, hemicellulose, and lignin. Together, these polymers account for the structural stability, high strength, and stiffness of hemp's cell wall. Due to the cell wall's structural composition ionic liquids were investigated as a dissolving agent. Ionic liquids (ILs) are a unique class of non-volatile, non-flammable chemicals. The properties of an ionic liquid can be altered by changing the identities of its constituent ions. Dissolving biomass in ILs has been reported to lead to a full release of all the functional groups and bonds from the matrix. These results have shown that lignocelluloses dissolved in ionic liquids are more susceptible to chemical attack by various reagents/catalysts. Therefore, this study aims to determine the effectiveness of ionic liquids for the pretreatment of hemp lignocellulosic material and evaluate the quality of fiber obtained thereof.

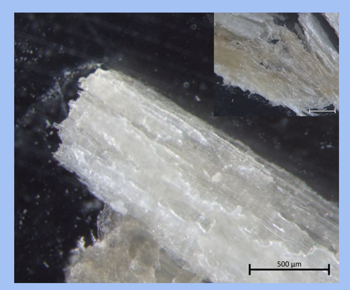


Figure 1. Raw hemp sample

Procedure:

I. Biomass Preparation

The hemp used in this study was donated by SUNYrf. The hemp was washed in deionized water, chopped into three sizes (Ground ¼", Short ½", and Large 1"), and placed in an oven at 70 degrees Celsius for a total of three days to dry. Once dried, the "Ground" samples were ground with mortar and pestle to resemble a fine powder. Then 0.3g of each sample size was weighed using an analytical balance and distributed into Erlenmeyer flasks for pretreatment. Nine flasks were prepared for each size group.

II. Ionic Liquid (IL) Pretreatment

The pretreatment method employed for the hemp samples was 1-Butyl-3-methylimidazolium chloride. Each flask received 3.0 grams of 1-Butyl-3-methylimidazolium chloride along with a magnetic stirrer. A 1:3 ratio of ionic liquid to biomass was maintained for each sample. Once prepared, flasks were heated in mineral oil baths in crystallizing dishes at temperatures between 75-80 degrees Celsius for either 3, 6, or 9 hours. Three flasks per size group were heated for each time interval. Once samples reached their target heating period, they were removed and left to cool.4

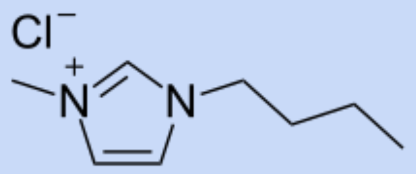


Figure 2. Chemical structure of 1-Butyl-3-methylimidazolium chloride

III. Acid Hydrolysis

After ionic liquid pretreatment, sample flasks were filled with 10.0 mL of 0.5 Hydrochloric acid and heated in mineral oil baths in crystallizing dishes for 3, 6, or 9, hours at 80 degrees Celsius. Once heated, samples were removed from the oil and left to cool for 30 minutes. After cooling, 10.0 mL of 0.5 M sodium hydroxide solution was added to each acidic sample as a neutralizer.

IV. Slide Preparation

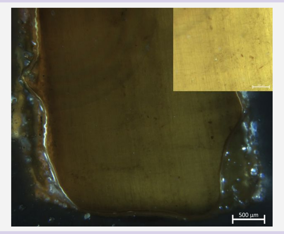
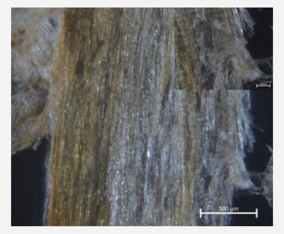
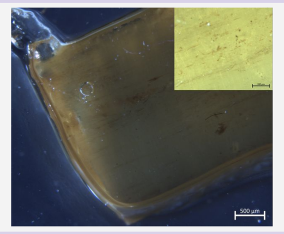
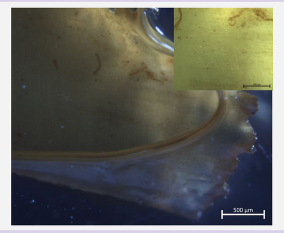
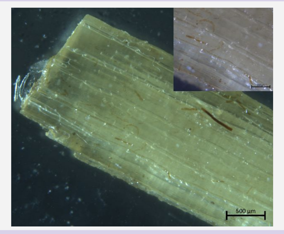
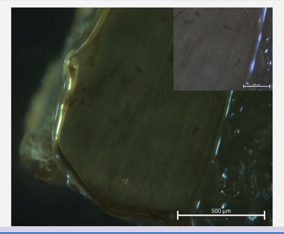
Hemp fragments were collected from each step: raw, ionic liquid, and acid hydrolysis, for fiber examination. Fragments were removed from each flask, rinsed with deionized water, and placed on glass slides to be analyzed. Slides were labeled appropriately for each treatment underwent: A for acid hydrolysis, IL for ionic liquid, and RAW for raw samples; numerical labels were also noted for hours spent in each treatment stage.

V. Stereomicroscope

Examination and imaging of sample slides were by the ZEISS SteREO Discovery.V20™ microscope. The objective lens applied for all images was the Achromat S 1.5x FWD 28mm. For optimal imagery, a z-stack was conducted based on the topography of each hemp fiber. Additionally, images underwent further processing methods such as "Extended View of Focus" to sharpen resolution, or "White Balance" to adjust light reflection or both. Displayed images demonstrate two fields of vision of each hemp sample, a half and zoomed view. Optimal images for each sample are presented.

Figure 3. Stereomicroscope

Table 1. Stereomicroscope images of hemp from ionic liquid, acid hydrolysis, and raw stages.

	Hours in Ionic Liquid	Hours in Acid Hydrolysis
Ground Hemp: 3hrs		Ground Hemp in Acid Hydrolysis 3-9 
Ground Hemp: 6hrs		
Ground Hemp: 9hrs		
Short Hemp: 3hrs		
Short Hemp: 6hrs		

Results and Future Directions:

Pretreatment of the hemp fibers results in more fibrous material obtained in each case. Based on these results, the ionic liquid (1-Butyl-3-methylimidazolium chloride) used in this case may serve as a good pretreatment candidate for further studies on hemp.

Future directions include imaging the remainder of the samples for a better understanding of differences in fiber breakdown after ionic liquid and acid hydrolysis treatment. Further data investigation is then possible with the use of Thermal Gravimetric Analysis, where the mass of the sample is measured over time as the temperature changes, providing information about the thermal decomposition/stability of hemp.

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