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Waste to wealth: Chlorine free extraction of nanocellulose from waste husk of rice

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Abstract
This work was done to extract cellulose from a sustainable source, i.e. rice husk, which is an agro-waste of rice industries. Chlorine free pretreatments were done on powdered rice husk for removing non-cellulosic components like silica, lignin and hemicellulose. Alkali treatment (5% NaOH) followed by bleaching with hydrogen peroxide was done. Finally, acidic treatment with nitric acid followed by steam explosion was done to obtain nanocellulose. Nanocellulose yield was calculated. Cellulose content was also found out by Anthrone method. It varied from 1550.9 to 2730.3mg/g in raw and bleached rice husk, respectively. Nanocellulose yield was approximately 65-70%. This study was basically done to utilize huge amounts of industrial waste in the form of rice husk. Moreover, the extracted cellulose can be used for various food based applications such as an edible coating, as a thickener/emulsifier, as an anti-caking agent, as a fibre supplement, as a calorie reducer.

Keywords: Chlorine free extraction, nanocellulose, waste husk, rice

1. Introduction
In recent times, there is an increased focus on "sustainable development" due to growth in replacement of synthetic products with environmentally degradable and bio-based products. This enables undisturbed and reliable supply of materials for the future generation. With the increase in green methods and use of natural materials, the greater safety of both present and future generations is ensured. Moreover, it is a known fact that synthetic composites are toxic, non-biodegradabe, costly, and nonrenewable (Vahabi *et al.* 2021) [18].
Agricultural wastes such as coconut husk fibres (Rosa *et al.* 2010), cassava bagasse (Pasquini *et al.* 2010) [13], banana rachis (Zuluaga *et al.* 2009) [22], mulberry bark (Li *et al*., 2009), soybean pods (Wang and Sain, 2007) [19], wheat straw, and soy hulls (Alemdar and Sain, 2008a) [2], and corn stalks (Reddy and Yang, 2005) [17] have been studied as a resource in the production of crystalline cellulose fibres.
Rice is the largest cereal crop in the world. Rice husk is an important agricultural waste that can be easily found in some states of Malaysia. The rice milling industry generates a huge quantity of rice husks during the paddy milling process from the fields. It accounts for 20% of the 500 million tons of paddy produced worldwide. Societies, especially among the rice millers, often dispose of the rice husk waste using open burning. This situation directly leads to environmental concerns and becomes a great environmental threat causing damage to the land and the surrounding area in which it was dumped. Although rice husk is a very rich source of cellulose which has various uses.
Cellulose is the most abundantly available biopolymer and its unique properties has diverse applications. It is made up of D glucopyranose units that are linked together by 1,4-glycosidic linkages (Midhun Dominic *et al.* 2022) [11]. Plant cell walls contain cellulose as microfibrils that are coupled with some non-cellulosic components thereby providing cellulose molecules more rigidity (Chirayil *et al.* 2014) [6]. Native fibers contain non-cellulosic constituents like silica, hemicellulose, lignin, pectin, and waxes. Lots of pretreatments are needed for extraction of cellulose. Although a variety of natural fibres were investigated in detail, the use of rice husk as a natural source for the production of cellulose nano crystals has not been widely explored yet.
Green methods are gaining popularity because they are safe and biodegradable. Highly pure cellulose nanofibers were isolated from wheat straw by Liu *et al* 2021 [10]. using green process having steps such as steam explosion, microwave-assisted hydrolysis, and micro fluidization. Cellulose nanofibers were extracted from sugar beet pulp by following hydrogen peroxide

bleaching, high-speed blending, and ultrasonic treatment (Wang *et al.* 2007) [19]. Ravindran *et al*., 2019 [15] reported the from pineapple leaf fibers were subjected to green process for isolation of cellulose nanofibers. Some other such work was carried out by few other researchers following environmentally friendly procedure.

Acid hydrolysis is a straightforward and cost-effective process for preparing CNFs that has gained widespread acceptance in recent years. CNFs can be extracted from agro-waste using both strong and mild acids. By dissolving the amorphous portion in cellulose, acid hydrolysis en- hances the crystalline domain. Cerqueira *et al.* 2017 [4] evaluated the reinforcing efficacy of nanocellulose produced by sulphuric acid hydrolysis from coconut fibers in biodegradable starch composites. Chen *et al*. 2016 [5] demonstrated that metal-catalyzed sulphuric acid hydrolysis may produce nanocellulose with a high degree of crystallinity index. They also found that the thermal stability of the developed fibers was considerably lower than that of the native form. Bahloul *et al.* 2021 [3] employed sulphuric acid as well as phosphoric acid to convert eggplant residues to nanocellulose. Abraham *et al.* 2011 [5] used oxalic acid hydrolysis along with steam explosion to develop nanocellulose from banana, jute, and pineapple leaf fibers. They showed that the aforementioned technique yields nanocellulose with better crystallinity index and thermal stability than the corresponding lignocellulosic fibers. Chirayil *et al.* 2014 [6] demonstrated that steam coupled acid hydrolysis would enhance the rate of depolymerization and defibrillation which is a vital phase in the isolation process. After a thorough investigation of the literature, it can be concluded that mild acid hydrolysis assisted with steam explosion and ultra-homogenization is an efficient way to extract CNFs from agro-waste residues. Environmental friendliness, recrystallization possibility, low corrosion on apparatus, and good yield, are some of the advantages of the aforementioned method (Xie *et al.* 2018) [20].

Even though numerous works have been reported for the isolation of CNFs from various biomasses, only limited works outline the chlorine- free extraction of nanocellulose. The limited uses of hazardous chemicals fall off the threat of pollution as well. As rice husk is the precursor for nanocellulose isolation, their large availability and cultivation promote the agrarian economy. Moreover, the application of millet husk in nanofiber isolation is not reported yet. Also, from the compositional analysis it is found that the rice husk contains higher amount of cellulose whereas it possesses lower lignin content. This further enhances its potential as a perfect precursor for cellulose.

The conversion of agro-wastes to future promising materials will be a sustainable waste management solution. According to the knowledge of the authors of this work, very limited investigations have been conducted on the chlorine-free extraction of CNFs from rice husk. Thus, the current study aims to extract CNFs from rice husk, an agro-waste by using a green approach. Alkylation at a lower concentration of alkali (NaOH, 2%) and chlorine-free bleaching (using hydrogen peroxide) is adopted for the pre-treatment of the samples. Commonly used strong acid hydrolysis is replaced by oxalic acid hydrolysis along with steam explosion for the isolation of nanocellulose. The adopted chlorine free methodology for nanocellulose isolation from rice husk intends to significantly reduce the environmental impact of using hazardous chemicals. The proposed study is also an attempt to convert 'waste to wealth'.

2. Materials and Methods

2.1. Materials

Rice husk was procured from Rice Mill of Chaman Lal Setia Exports Limited, Karnal, Haryana, India. Oxalic acid, concentrated hydrochloric acid (HCl), concentrated sulphuric acid (H_2SO_4), sodium hydroxide (NaOH), hydrogen peroxide (H_2O_2), and anhydrous magnesium sulfate were of analytical grade, and bought from Helix Chemicals Private Limited, New Delhi, India.

2.2. Method

2.2.1. Preparation of cellulose nanofibers from rice husk

The rice husk was collected, washed, and dried before being used. Winnowing was used to separate the rice husk from the dried sample. About 100 g of the raw rice husk (RRH) was subjected to the steam explosion in an autoclave with 2% NaOH solution in the ratio of 1:15 g/ mL at a temperature of 120 ◦C and a pressure of 15 psi. Two more subsequent cycles are done under similar conditions. The alkali-treated rice husk (ARH) was washed well with distilled water. About 100 g of the ARH sample was bleached with 1000 mL of 15% H_2O_2 and 100 mL of 10% NaOH solution for 2 h. The sample was bleached three times until it was completely white. The bleached rice husk sample (BRH) was washed well with distilled water and dried. About 100 g of BRH was treated with 1000 mL of 5% nitric acid in an autoclave with a sudden pressure release of 15 psi. The acid hydrolysis was repeated 3 times. The sample was then removed from the autoclave and properly washed to eliminate the acid. CNFs were obtained by homogenizing the acid hydrolysed sample for 30 min (D lab homogenizer model D-500).

2.2.2 Cellulose content determination

Cellulose undergoes acetolysis with acetic/nitric reagent forming acetylated cyclodextrins which gets dissolved and hydrolyzed to form glucose molecules on treatment with 67% H_2SO_4. This glucose molecule is dehydrated to form hydroxymethyl furfural which forms green colored product with anthrone and the color intensity is measured at 630nm. Fig.1 shows the schematic representation of cellulose content in rice husk.

2.2.4 Nanocellulose yield

The yield of isolated cellulose nanofibers was determined by following the Eq. (1) explained by Yadav *et al*. 2019 [21]

$$\text{Yield of extracted cellulose (\%)} = \left(\frac{W2}{W1}\right) X100 \quad (1)$$

where W_2 is the weight of extracted cellulose nanofibers and W_1 is the weight of native cellulose fibers.

3. Results and Discussion

3.1 Cellulose content

The cellulose content is evaluated to find out total cellulose present during various stages of cellulose extraction. The cellulose content at each stage of cellulose shows the increase in cellulose. Table 1 completely illustrates the findings.

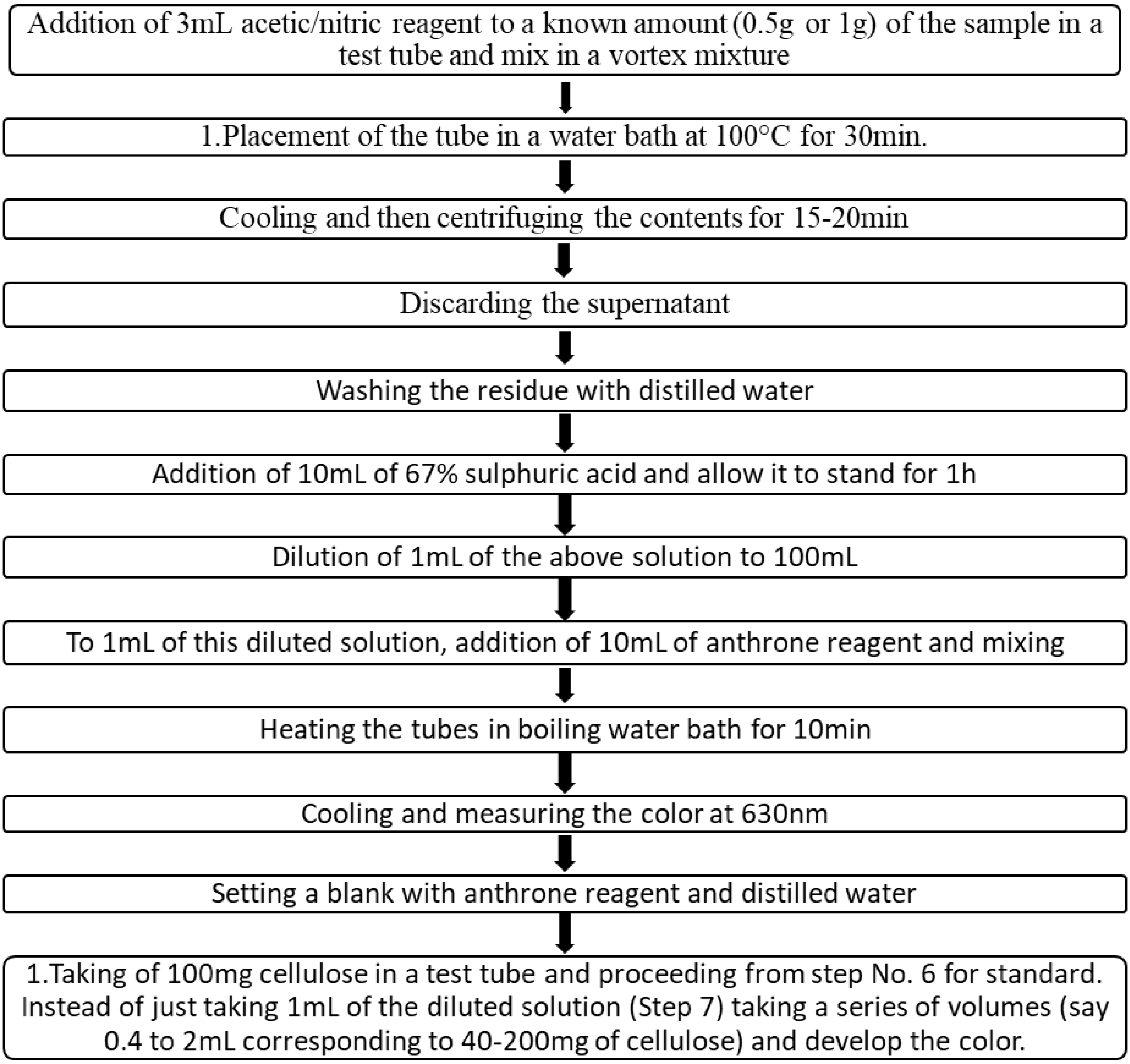

Fig 1: Schematic representation of cellulose content in rice husk

Table 1: Cellulose content analysis at each stage of treatment

Sample	Cellulose content (mg/100 g)
RRH	1550.9
ARH	1991.7
BRH	2730.3

From the table, it is clear that due to pre-treatments there is a rise in cellulose content. The raw sample (RRH) has approximately 35% cellulose with an increase to 52% after undergoing alkali treatment. Furthermore, the bleaching step elevates the cellulose level to approximately 65%. Due to pre-treatments, non- cellulosic components are removed or reduced. The non-cellulosic components are removed mostly based on their molecular weight and intermolecular bonding. The breaking of hemicellulose-lignin linkages and partial delignification occurs when steam explodes in the presence of alkali. The breakdown of cellulose molecules may be caused by an increase in NaOH concentration (Nie *et al.* 2018) [12]. According to the chemical analysis, ARH has higher cellulose content (52%) than RRH (35%). According to Chirayil *et al.* 2014 [6] the increased availability of –OH groups after the steam explosion process in the presence of an alkali medium causes a rise in cellulose content.

The major strategy of pre-treatment is to disrupt the lignin structure in the lignocellulosic biomass and thus improve the susceptibility of the remaining polysaccharides for other treatments. The hydrophilic nature of fiber and NaOH will result in the enrichment of the reaction rate and increase the cellulose exposed to the surface. The alkali pre-treatment will also help in increasing internal surface area and decreasing the degree of polymerization. It also destroys the structural linkages between lignin and carbohydrate by saponification of intermolecular ester bonds. The lignin structure is disrupted because of the breaking of its glycosidic ether bond.

Bleaching is usually done for delignification and to remove the remaining hemicellulose content after alkali treatment which occurs as a result of the ester bond cleavage (Fareez *et al.* 2018) [7]. On bleaching, the lignin content decreases significantly from RRH to BRH.

As the study emphases the utility of chlorine-free pre-treatments in the extraction process, it is critical to look at the effect of peroxide bleaching separately. According to Qasim *et al.* 2020 [14], it was concluded that the time and energy required in acidified sodium chlorite treatment (ASC) was greater than that of alkaline hydrogen peroxide (AHP) treatment. Though both the nanofibers synthesized have same crystallinity the ASC treated samples was better in providing thermal stability.

On the contrary, AHP treatment was more environmentally benign since it is a chlorine-free extraction procedure. Joy *et al.* 2016 [9] found that extracting CNFs from *Helicteres isora* plant through peroxide bleaching had a greater yield than preparing CNFs from the same precursor by chlorite bleaching. An increase of 10% in yield was obtained via chlorine-free bleaching treatment. Following extensive literature, it has been shown that peroxide bleaching (15%) at minimal temperature is favorable for delignification in an environmentally responsible manner. Hence chlorine-free bleaching can be used as a promising and sustainable method for the production of cellulose nanofibrils. Thus, the chemical composition analysis showed that the proposed pre-treatments were successful in removing non- cellulosic components such as lignin and hemicellulose from the raw millet sample, allowing for the extraction of CNFs.

3.2 Nanocellulose yield

The yield of CNFs was calculated and found to be 60–65%. This was similar to the yield of Bambusa rigida, a work reported by He *et al*. 2015.

While choosing an isolation process, environmental safety is also quite important. As previously mentioned, the study uses mild conditions (mild alkali hydrolysis and peroxide bleaching) for the isolation of the nanocellulose. For cellulose purification, the adopted methodology might also readily replace the conventionally used chlorine based bleaching agents, especially sodium chlorite. The usage of chlorine based bleaching agents might lead them to linger in the environment for extended period of time. This may eventually result in contribution of consequences to the global warming. Exposure to sodium hypochlorite can cause headache, dizziness, nausea and vomiting. Being completely chlorine free, the method is found to be safer.

4. Conclusion

This work effectively extracts very clean cellulose nanofibers from rice husk, an agro waste, using a chlorine free/green technique. The technique uses hydrogen peroxide bleaching, mild acid hydrolysis, steam explosion and homogenization. The CNFs were extracted from the rice husk with a good yield, 60–65%. The cellulose content is evaluated to find out total cellulose present during various stages of cellulose extraction. Overall nanocellulose yield reveals the effectiveness of the green method used. As a result, it would be concluded that the cellulose nanofibers obtained from rice husk can be used for a variety of operations such as an edible coating, as a thickener/emulsifier, as an anti-caking agent, as a fibre supplement, as a calorie reducer. This research proposes a long-term waste management plan for agro- industries, as well as a way for “waste-to-wealth conversion”.

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