

Green Synthesis of Cellulose Nanocrystals from Banana Pseudo Stem and it's Characterization.

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Abstract

Cellulose nanocrystals were isolated from Banana Pseudo Stem, an under-utilized agriculture biomass. Raw Banana stem was collected from Abrama Farm, Kamrej, Surat. Which were then dried in the Hot Air Oven @ 105°C for 6 hours and after cooling and desiccating, crushed in a grinder and sieved to get fine powder particles. The chemical compositions of cellulose nanocrystals from banana stem after each treatment were determined according to Technical Association of the Pulp and Paper Industry (TAPPI) standard. The lignin content was determined according to the TAPPI norm T222 om-88. The Hemicellulose content was determined according to the TAPPI T257 om-09. Cellulose content was determined by extracting hemicellulose with the aqueous sodium hydroxide (17.5%) for 5 h before quenching the reaction with ice. The obtained white powder was washed with copious amount of water until filtrate (Whatman Filter Paper No. 42) becoming neutral. Cellulose nanocrystal (CNC) was obtained by sulfuric acid hydrolysis according to the reported method. The resultant mixture was first centrifuged at 1000 rpm for 10 min ice- cooling temperature (18°C) to remove large particles, and then centrifuged at 11,000 rpm for 15 min ice- cooling temperature (-15°C) to obtain cellulose nanocrystal. The obtained cellulose nanocrystal was washed and centrifuged repeatedly for 3 times before dialysis against distilled water for 2 days. The obtained CNC was processed by ultrasonic processor (VCX 500:500 W, Sonics & Materials, Newton, CT) to suspension better before further application. The composition of Banana Pseudo Stem found after research experiment was Cellulose - 69.9%, Hemicellulose - 19.6% and Lignin - 5.7%. The chemical pretreatment removed the non-cellulosic constituents of Banana Pseudo Stem was found to be an essential and fundamental procedure for the Cellulose Nanocrystal. The pretreatment process of Banana stem included Alkali Treatment, Delignification and Bleaching Treatment. FEG - TRANSMISSION ELECTRON MICROSCOPE (HR-TEM) was used to find the size of obtained CNC. The obtained Cellulose Nano Crystal exhibited a different Category of 500 nm, 200 nm, 100 nm, and 1µm. 500 nm scale observe the (331nm, 290 nm and 271nm) diameter size of Cellulose Nano Crystals. 200 nm scale observe the (95.2nm, 87.5nm, 81.6nm, and 67.9nm) diameter size of Cellulose Nano Crystals.

Keywords- CNC, Lignin, Cellulose, Hemicellulose, FEG-TEM.

I. INTRODUCTION

- The extraction of cellulose from agricultural wastes to produce green and clean products has received most attention in the world.
- The material is versatile in various applications such as optoelectronics, adsorption catalysis, sound and heat insulation, medical materials, aerospace materials, and many other fields [1, 2, 3, 4].
- There have been many publishes on the extraction and evaluation of the quality and efficiency of the extraction process from many different materials such as rice straw, corn, sugarcane bagasse, oil palm, starch [5].
- Among these raw materials, the banana stem was evaluated as a potential source of cellulose [6, 7, 8, 9, 10].
- Banana is planted in many areas in the world and it is one of main export product of many countries. Each hectare of bananas generates about 220 tons of waste, and makes the problem on landfills and polluting the environment [10].
- Therefore, making full use of waste from banana trees to create valuable products plays an important role in improving economic efficiency and moving towards sustainable development for banana cultivation as well in treatment of agricultural waste in general.
- Cellulose is the main component of the banana stem, it takes about more than 50% of the total banana stem, and this is a natural, renewable, and biodegradable polymer that can be used in many fields such as raw materials for construction, paper, insulation materials, adsorption materials, environmental treatment [8].
- The distinctive property of cellulose nanocrystals make it useful building block in various applications, for instance as reinforcing agents [13,14], biomedical implant [15], nanocomposites [12] as well as electronic components [16].
- Cellulose is one of the most important organic compounds on earth and is obtained from plants, algae bursal, animals, and bacteria.

Cellulose structures and names are clearly classified according to different geometric sizes [18].

- Micron to nano-scale applications differ and are determined by the cellulose geometric size [19].

II. REVIEW OF LITERATURE

- Nanocrystalline cellulose (NCC) has gained attention due to its versatile properties such as biocompatibility, sustainability, high aspect ratio, and abundance of –OH groups that favor modifications of NCC. The objective of this paper is to develop NCC by extracting and characterizing NCC prepared from banana peel powder (BPP).
- BPP was subjected to alkali and bleaching treatment to remove lignin and hemicellulose and then subjected to acid hydrolysis to prepare NCC.
- The particle size and zeta potential of the NCC were found to be 209 nm.
- All the parameters evaluated indicated that NCC was successfully prepared from BPP using alkali treatment, bleaching, and acid hydrolysis.
- Cellulose nanocrystals (CNCs) have attracted tremendous attention because of their excellent chemical and physical properties and due to their renewability and sustainability.
- Rice straw was selected as the raw material in this study. Initially, a large amount of lignin must be removed by an alkaline process to obtain a slurry.
- Thereafter, a green bleaching process can be used to remove the remaining lignin in the slurry. An UV-emitting diode with 365 nm wavelength assisted the oxidation reaction of the H₂O₂ solution without the use of chlorine-containing chemical bleach.
- The reaction required only 2.5 h to obtain high-purity cellulose and successfully enhanced the yield. Transmission electron microscopy images showed that the CNCs from rice straw were 100 nm long and 10–15 nm wide.

III. MATERIALS AND METHODS

3.1 Banana Stem Raw Material Collection & Treatment.

Banana Stem Sample: Raw Banana stem was collected from Abrama Farm, Kamrej, Surat. Which were then dried in the Hot Air Oven @ 105°C for 6 hours and after cooling and desiccating, crushed in a grinder and sieved to get fine powder particle.



Banana Stem



Oven-dried Banana Stem



Fine Powder Sample

Figure: 3.1 Raw Material Processes: Collection, Drying and Fine Powder

3.2 Determination of the chemical composition of cellulose nanocrystals from Banana Stem: ^[20]

3.2.1 Lignin

- The chemical compositions of cellulose nanocrystals from banana stem after each treatment were determined according to Technical Association of the Pulp and Paper Industry (TAPPI) standard. The lignin content was determined according to the TAPPI norm T222 om-88 [21,22]
- Briefly, 1 g of sample was added into the 15 mL H₂SO₄ solution (72 wt%), and maintained at room temperature (35°C) for 2 h. Then, the distilled water, 560 mL, was added and boiled the mixture for 4 h before the centrifuge to get the insoluble lignin. The obtained lignin was oven-dried and weighted. The lignin quantity was determined using following formula.

where M₁ was the obtained lignin mass, M was the initial sample mass.

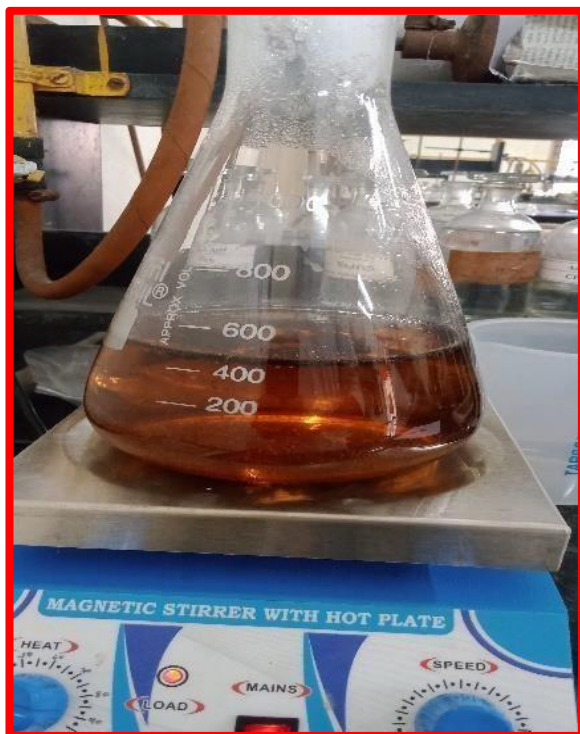
$$\text{Lignin (\%)} = \frac{M_1}{M} \times 100,$$



Sample + H₂SO₄



Oven-Dried Lignin



H₂SO₄ + Distilled Water

3.2.2 Hemicellulose

- Hemicellulose content was determined according to the TAPPI T257 om-09. Hemicellulose content was quantified with sodium chlorite treatment according to the reported procedure [23].
- Sample 1 g, was added into the solution, 30 mL, containing Acetic acid
- 0.25 mL, Sodium Chlorite 0.3 g and kept at (75 °C) for 1 h.
- The mixture is then cooled down and the residue is filtered (Whatman Filter Paper No. 42) and washed thoroughly with water. The residue was finally dried and weighted. The holocellulose content was calculated using following formula.

$$\text{Hemicellulose} = \frac{M_2}{M} \times 100$$

where M₂ was the obtained residue mass, M was the initial sample mass

Figure: 3.2.1 Extraction of Lignin from Banana Stem



Sample+CH₃COOH+NaClO₂



Filter + Wash Water



Hemicellulose

Figure:3.2.2 Extraction of Hemicellulose from Banana stem

3.2.3 Cellulose of Banana Stem

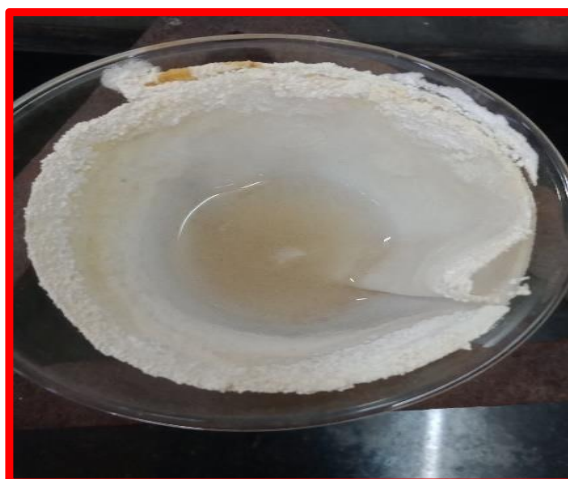
Cellulose content was determined by extracting hemicellulose with the aqueous sodium hydroxide (17.5%) for 5 h before quenching the reaction with ice. The obtained white powder was washed with copious amount of water until filtrate (Whatman Filter Paper No. 42) becoming neutral [24]. The cellulose content was calculated using following formula.

$$\text{Cellulose (\%)} = \frac{M_3}{M} \times 100,$$

where M₃ was the obtained white powder mass, M was the initial sample mass.



Hemicellulose + NaOH



Washed & Neutral



Cellulose



Distilled Water Wash

Figure: 3.2.3 Extraction of cellulose from Banana stem

3.3 Determination of the Pretreatment: Alkali Treatment

3.3.1 Alkali Treatment of Banana Stem Sample

- The pretreatment process of Banana stem included Alkali Treatment, Delignification and Bleaching Treatment [25,26].
- Firstly, the cleaned Banana stem powder 1 g, was immersed in NaOH solution (2 wt%), 100 mL, at room temperature (35°C) and stirred (80rpm) for 3 h to finish the alkali treatment to remove the impurities including pectin, wax. The alkali-treated sample was washed thoroughly till neutral. (Whatman Filter Paper No. 42).



Neutral pH



Sample + NaOH

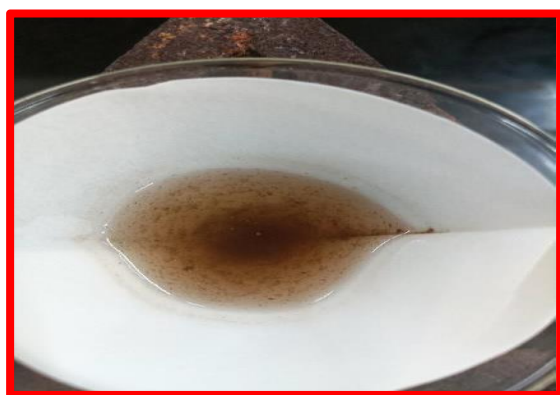
Figure: 3.3.1 Alkali Treatment of Banana Stem Sample

3.3.2 Delignification Process of Sample

The delignification process was carried out with dewaxed form the above sample, 1 g, suspended in acetic acid (93% v/v), 50 mL, and hydrochloride acid solution (0.3% v/v) under strong stirring (90 rpm) for 3 h at (90°C) and then washed till neutral pH value. (Whatman Filter Paper No. 42).



Sample+CH₃COOH+HCl



Wash till Neutral pH



Delignification

Figure: 3.3.2 The Delignification Process

3.3.3 Bleaching Process of Sample

The Bleaching Process was performed by adding the obtained delignification form the above sample into the mixture of H₂O₂ (5 wt%) and NaOH (3.8 wt%) at a ratio of 1:50 and stirred at (50 rpm) at room temperature (35°C) for 3 h before washing thoroughly. (Whatman Filter Paper No. 42).



Sample + H₂O₂ + NaOH



Washing Thoroughly



Bleaching Process

Figure: 3.3.3 The Bleaching Process

3.4 Extraction of Cellulose Nano Crystal

Cellulose nanocrystal (CNC) was obtained by sulfuric acid hydrolysis according to the reported method [27]. The pretreated Sample powder, 1 g, was slowly added into H₂SO₄ solution (63 wt%), 30 mL, under vigorously stirring at room temperature (35°C) for 1 h. After that, the hydrolysis was quenching by adding iced water, 300 mL, into the mixture.



Sample+H₂SO₄+ Iced water



Centrifuged Sample Holder



After Centrifuged

Figure:3.4 Extraction of Cellulose Nano Crystal

3.4.1 Cooling Centrifuge of Cellulose Nano Crystal

The resultant mixture was first centrifuged at 1000 rpm for 10 min ice- cooling temperature (18°C) to remove large particles, and then centrifuged at 11,000 rpm for 15 min ice-cooling temperature (-15°C) to obtain cellulose nanocrystal.



Cooling Centrifuged at 1000 rpm



Centrifuged at 11,000 rpm

Figure :3.4.1 Cooling Centrifugation of Cellulose Nano Crystal

3.4.2 Washing and Filtration

The obtained cellulose nanocrystal was washed and centrifuged repeatedly for 3 times before dialysis against distilled water for 2 days



Filter the Solution



Impurity of Solution



Clear Solution

Figure: 3.4.2 Washing of Cellulose Nano Crystal

3.4.3 Sonication Treatment of Cellulose Nano Crystal

The obtained CNC was processed by ultrasonic processor (VCX 500:500 W, Sonics & Materials, Newton, CT) to suspension better before further application



Figure: 3.4.3 Ultrasonic Processor (120W, 90 min)

IV. RESULT AND DISCUSSION

4.1 FEG - TRANSMISSION ELECTRON MICROSCOPE (HR-TEM)



- Description:
- This automation eases the learning curve for novice operators, reduces tensions in a
- multi-user environment, and improves time to data for the experienced operator.
- The 4K X 4K Ceta aCMOS camera with large field-of-view enables live digital

Instrument Name	FEG - TRANSMISSION ELECTRON MICROSCOPE (HR-TEM)
Model	Talos F200i S/TEM (HRTEM-200KV)
Make	Thermo Fisher Scientific
Accelerating Voltage	200KV
Electron Source	Schottky Field emitter (Field Emission Gun)
Resolution	Line Resolution 0.10 nm Point Resolution less than 0.25 nm or less
Magnification	TEM Magnification 50x to 1Mx STEM Magnification: 310X to 330Mx
Diffraction Mode	Diffraction Mode camera length 14 to 5700MM (1400 M in LAD)
STEM Detector	High Angle Annular Dark Field Detector (HAADF) STEM Resolution less than 0.16nm. STEM Magnification up to 330 Mx
EDS or EDAX	Bruker X Flash 6 30 EDS Detector Quantitative Elemental analysis and elemental Mapping Facility.

4.2 Synthesized cellulose Nanocrystals were found in Three Category.

Category 1: (Scale 200 nm)

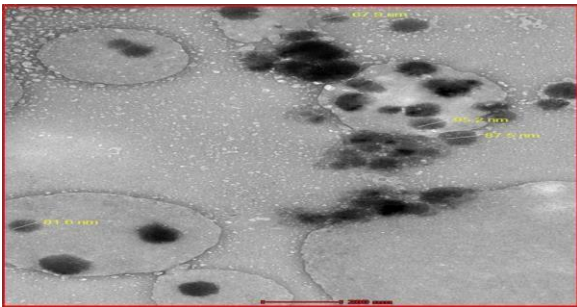
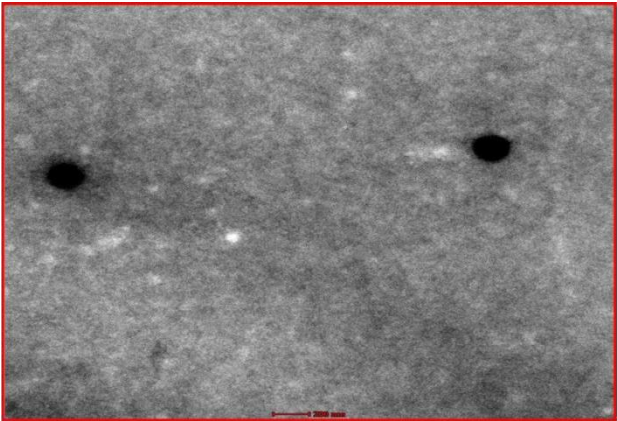


Table 4.2.1: Determination of Cellulose Nanocrystal by FEG-TEM.

Sr. No.	Scale	Specialization	Method	Observation (Scale – 65 to 90 nm)			
				(Diameter –Size of Cellulose Nanocrystal)			
				1	2	3	4
1	200 nm	FEG- TEM	MICROSCOPY	81.6 nm	87.5 nm	95.2 nm	67.9 nm

In this scale 4 different size of cellulose nano crystals are observed by FEG – TEM: 200nm Scale Observe the (81.6 nm, 87.5 nm, 95.2 nm and 67.9 nm) diameter size of Cellulose Nano Crystal. (All Nanocrystals size is < 100 nm)

Category 1: (Scale 200 nm)



Category 2: (Scale 500 nm)

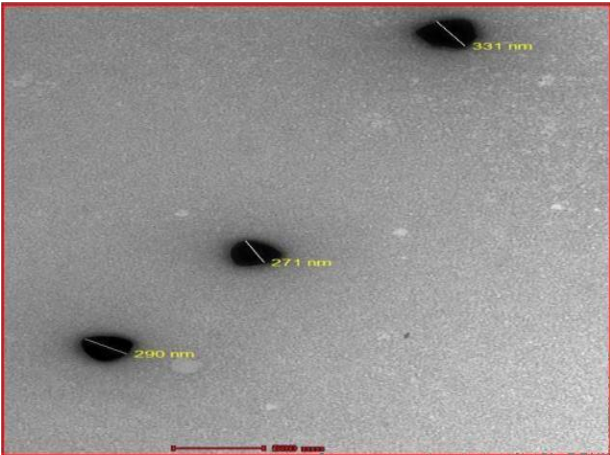
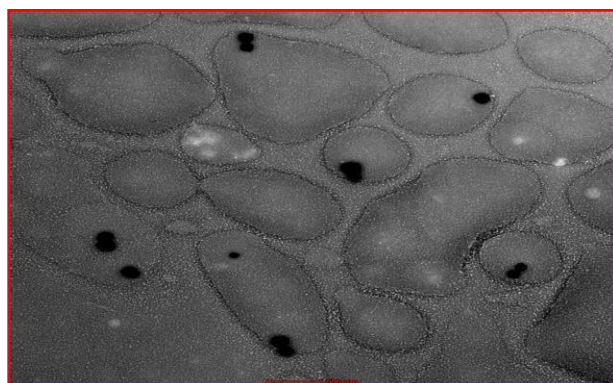


Table 4.2.2: Determination of Cellulose Nanocrystal by FEG-TEM.

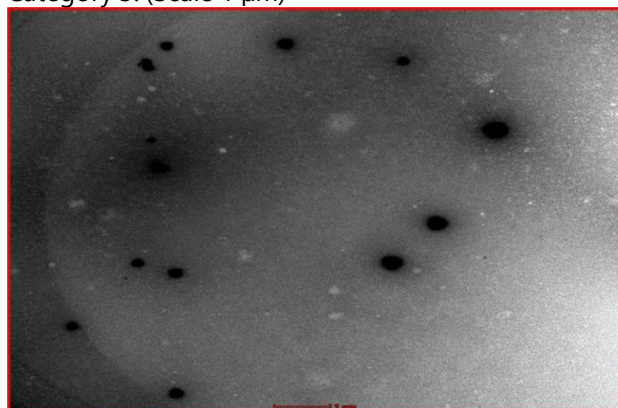
Sr. No.	Scale	Specialization	Method	Observation (Scale -250 to 350 nm) (Diameter - Size of Cellulose Nanocrystal)			
				Sr.No.1	Sr.No.2	Sr.No.3	Sr.No.4
1	500 nm	FEG- TEM	MICROSCOPY	290 nm	271 nm	331 nm	-

In this scale 3 different size of cellulose nano crystals are observed by FEG – TEM: 500nm Scale Observe the (290 nm, 271 nm and 331 nm) diameter size of Cellulose Nano Crystal. (All Nanocrystals size is < 350 nm).

Category 2: (Scale 500 nm)



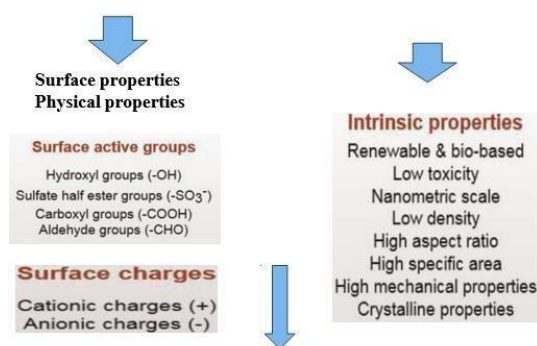
Category 3: (Scale 1 μ m)



4.3: Analysis of Banana Pseudo Stem Composition:

The Banana Pseudo Stem content of the Cellulose was 69.9%, Hemicellulose 19.6% and Lignin 5.7%.

Cellulose Nanocrystals



Market	Applications	Exploited CNC properties	References
Composites/films	Nanocomposites Flexible packaging Optical films	High mechanical properties Filmogenic properties Morphology	[28, 29]
Coatings/paints/adhesives	Coatings for flexible packaging	Morphology Rheological properties	[30, 31]
Filtration	Mesoporous films and membranes	High specific surface area High mechanical properties Hydrophilicity	[32]
Energy	Supercapacitors Flexible batteries	Strength Large surface area	[33]
Cosmetics	Hydrogels and foams	Colloidal stability Emulsion interfacial stabilization	[34, 35]

V. CONCLUSION

Cellulose nanocrystal can be isolated from Banana Pseudo Stem, an under-utilized agriculture biomass. The Banana Pseudo Stem content of the Cellulose was 69.9%, Hemicellulose 19.6% and Lignin 5.7%. The chemical pretreatment which removed the non-cellulosic constituents of Banana Pseudo Stem was

found to be an essential and fundamental procedure for the Cellulose Nano Crystal.

The obtained Cellulose Nano Crystal exhibited a different Category to 500nm, 200nm, 100nm, and 1µm. 500nm scale observe the (331nm, 290nm and 271nm) diameter size of Cellulose Nano Crystals. 200nm scale observe the (95.2nm, 87.5nm, 81.6nm, and 67.9nm) diameter size of Cellulose Nano Crystals.

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