BRIEFING NOTE

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Organic Waste: a bio-resource for production of novel cellulose nanocomposites

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KEY FINDINGS

Mango seed waste and wheat residues are potential feedstocks for multi-product biorefineries. Selectivity of bioproducts (hemicelluloses, lignin, polyphenols, and cellulose rich residues) from mango seed, and (hemicelluloses, *p*-coumaric acid, ferulic acid, lignin and cellulose-rich residues) from wheat residues during extraction can be achieved by strategic selection and optimization of processing conditions at each stage in multiproduct-biorefinery schemes. Optimized mild alkaline pretreatments (NaOH concentration= 1.92 M Temperature = 86.0 °C and Time = 3.84 h) of mango seed husks produced hemicellulose extracts with xyloglucan to xylan ratios which produced films with thermal stability (330 °C) higher than thermal stability (290 °C) of films made with xylan only. Treatment of the alkaline pretreated mango seed husks under optimized high shear homogenization-assisted organosolv (60% ethanol, 148.41 °C, and 15 min homogenization) produced lignin (70% yield and 96% purity), and cellulose-rich residues (crystallinity of 56 % and cellulose content of 78%) that suited the production of cellulose nanocrystals (CNCs). The hydrolysis of the cellulose-rich residues by formic acid without any catalyst, produced mostly spherical CNC with an average diameter of 25 nm and thermal stability of 361 °C higher than the thermal stability of CNCs (309 °C) produced by the sulphuric acid-based method.

A two-stage alkaline pre-treatment of wheat residues produced cellulose-rich residues with crystallinity >50% rendering them suitable feedstock for nanocellulose production under mild acid and enzymatic conditions. The nanocellulose from wheat residues produced with either hydrochloric acid (HCl) at optimal conditions, 4 M HCl, 7.0 h, and 110 - 115^{o}C or enzymatic treatments at optimal conditions, 4.64 h, FibreCare of 75 ECU/g and Viscozyme of 10.8 FBG/g, and 50 ^{o}C, had comparable yields (≈20%). However, the nanocellulose yields were less than the yield (34%*)* but with thermal stability over 40 °C higher than of nanocellulose obtained using sulphuric acid treatment. The morphology and surface charge (Zeta potential ≈ – 16 mV) of the nanocelluloses produced with HCL and the enzymes respectively, classified them as cellulose nanofibers and relatively stable colloids.

Biocomposites with varying mechanical properties and functional properties can be made with the hemicelluloses and nanocelluloses produced from the biorefinery schemes. Nanocomposites made with MHH with an addition of a plasticiser, and those made with acetylated mango seed husk hemicellulose with nanocellulose produced by formic acid had improved Young's modulus.

INTRODUCTION

Mango wastes and wheat residues are available in abundance and currently have very low economic value. Furthermore, the disposal of these organic wastes is a cause of environmental concerns. The chemical composition of the organic wastes shows potential for extraction of multiple high-value compounds. Nanocellulose is a potential bioproduct that has become an important engineering material in the fields such as food packaging, lightweight manufacturing, and medicine. Nanocellulose is generally produced from woody biomasses, however, these woody biomasses have many other competing uses. Alternatively, nanocellulose can be produced from organic waste such as mango waste and wheat residues.

Nanocellulose production requires cellulose-rich pulp as the precursor. The conventional method is to use controlled sulphuric acid hydrolysis of bleached cellulose-rich pulp. However, bleaching degrades most of the cell wall components (hemicellulose, hydroxycinnamic acids, and lignin). Furthermore, the sulphuric acid treatment lowers the thermal stability of the nanocellulose, limiting industrial applications such as the production of biocomposites via melt blending. However, the nanocelluloses produced disperse easily in water. Selective extraction and the introduction of certain functional groups can improve the nanocellulose functionality and the economic, and environmental impacts of the whole nanocellulose production process.

The challenge of extracting multiple products in a biorefinery setup is the different optimal extraction conditions of the biomass components. Thus, low product yields and qualities are obtained if the bioproducts are produced at sub-optimal conditions. The development of fractionation routes for obtaining cellulose-rich pulp for nanocellulose production while generating these other cell wall components at their optimal conditions as additional value-added products is inevitable. This study developed optimized sequential processes for fractionation of two types of organic wastes, mango seed and wheat residues into nanocellulose in multi-product biorefinery schemes. The ultimate goal was to ensure high selectivity, by optimizing the conditions at each stage for the recovery of the targeted bioproducts and the production of cellulose-rich residues (pulp) suitable for nanocellulose production. In the study, the mango seed waste, in particular the mango seed husk, was fractionated into hemicellulose, lignin, and cellulose-rich pulp which was further treated using organic acids without any

catalyst to produce nanocelluloses (Fig 1). The use of organic acids as alternative treatment methods was preferred over the sulphuric acid-based treatment owing to their ability to produce spherical and thermally stable nanocelluloses with a potential to recycle them after first use. The study further developed an integrated biorefinery process route for producing cellulose-rich pulp from wheat residues while generating hemicellulose, lignin, and ferulic/*p*-coumaric acid. The biorefinery route involved a two-stage alkaline treatment for the extraction of hemicellulose, lignin, and ferulic/ *p*-coumaric acid while producing a cellulose-rich pulp that was subjected to either mild acid (hydrochloric acid) or enzymatic treatments to produce nanocellulose. The potential of the hemicellulose extract as a raw material for biocomposite development of thermally stable and self-supporting films was investigated using the extracted hemicelluloses from mango seed husks.

METHODOLOGY

Mango seed waste biorefinery schemes: Mango seed waste contains the seed husks and the kernels (Fig 1). When separated, the mango seed husks were subjected to a multi-step sequential alkaline pretreatment (AP) and rotor-stator high shear homogenization-assisted organosolv (HSHO) [ethanol concentration (50–70%), temperature (130–150 °C), homogenizing time (10–20 min)] process to recover hemicellulose (xylan/xyloglucan), lignin and cellulose-rich fibers (Fig. 1) nanocellulose from the cellulose-rich fibers obtained from the multi-step process via optimized non-catalyzed organic acid-based treatment.

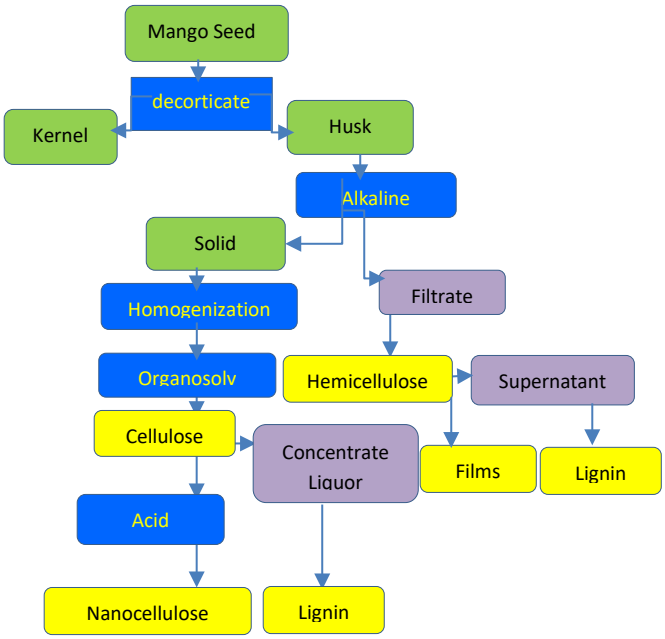

Figure 1: Developed integrated biorefinery route for multi-product generation from mango seed waste. Green blocks = starting materials, blue blocks = main treatment stages, yellow blocks = main products, and purple blocks = Effluents.

Wheat residues biorefinery schemes: The wheat residues were subjected to a two-stage alkaline pre-treatment involving a mild alkaline stage (20 - 60 ^{o}C and NaOH between 1.5. and 3.0 wt. %) to selectively extract hemicelluloses and phenolic compounds (*p*-coumaric acid/ ferulic acid) was followed by a severe alkaline delignification process (≈6 - 12 wt. % NaOH; at 121 ^{o}C) for 0.5-1.5 h to release lignin while retaining the cellulose-rich residues for nanocellulose production. The mild alkaline treatment was optimised for high yields of hemicelluloses and phenolic compounds while the second stage alkaline treatment was optimised for cellulose-rich pulp yield with minimal lignin content. The optimization was done statistically in central composite designed experiments.

Wheat residues nanocellulose production:

Mild acid nanocellulose production: The delignified cellulose-rich fibers were treated with 4 M HCl at fiber to acid ratio of 1:20 at varied times (4 – 8 h) and temperatures (80 – 120 ^{o}C) with agitation in a central composite design experiment. The reaction mixtures were washed with distilled water and centrifuged (3000×g for 10 mins) several times. The precipitate was re-suspended in distilled water and neutralized with 0.1 M NaOH. A fiber suspension (2-3%) was made from the recovered precipitate and subjected to a high-shear homogenization for 20 mins. The resulting mixture was freeze-dried to obtain the cellulose nanoparticles.

Enzymatic nanocellulose production: The two-stage alkaline pretreated wheat straw was subjected to enzymatic treatment. The extraction conditions were optimization in central composite designed experiments by varying time (4 – 8 h), FiberCare dosage (50 – 100 endo-1,4-β-glucanase unit/g), and Viscozyme (10 – 20 fungal β-glucanase units/g) for the enzyme-based treatment. The extraction conditions were selected to produce the cellulose nanoparticles while minimizing glucose formation. A 5 wt. % cellulose pulp suspension of the cellulose rich fibers in a 50 mM acetate buffer solution (pH 5) was treated at 50°C using a shaking incubator (MRC Orbital shaker TS600) at 150 rpm.

Sulphuric acid nanocellulose production: The cellulose-rich fibers were mixed in a fiber-to-acid solution ratio of 1:20 in a three-neck flask at 45°C for 60 min. The cellulose nanoparticles suspension was diluted with 10 volumes of distilled water and centrifuged at 3000×g for 10 mins before being dialysed against distilled water for 3 days and then neutralized with 0.1 M NaOH to pH 5 and dialysed again for a day before being centrifuged again and thereafter, freeze-dried to obtain the cellulose nanoparticles.

MAIN RESULTS

Mango seed multiproduct biorefinery

The yield and composition of the hemicelluloses extracted under varying alkaline pre-treatment conditions are shown in table 1. At the optimized AP process conditions (1.92 M NaOH, 86.0 °C, and 3.84 h) hemicellulose extract suitable for self-supporting and thermal stable films was obtained. The yield, xyloglucan/ xylan ratio, lignin content and uronic acid were 46%, 0.13, 17% and 12% respectively (Table 1). The composition at optimal conditions was predictable at a 95% confidence level by quadratic models (Table 1).

The maximum degradation temperature of the hemicellulose extracts and films made from them is shown in Table 2. The films' thermal stability was higher than the thermal stability of films made with xylan only.

Table 1: Hemicellulose yield and composition at optimized alkaline pre-treatment conditions

Responses	Predicted	[a] Observed	Deviation
Yield (%)	45.71	46.24	1.15
Xyloglucan/Xylan (%)	0.125	0.130	3.85
Lignin content (%)	18.79	16.73	10.96
Uronic acid (%)	15.08	12.02	20.29

[a] mean of triplicates determination. Note: Regression equations, Analysis of variance. Optimized extraction conditions were NaOH concentration= 1.92 M Temperature = 86.0 °C and Time = 3.84 h

Table 2: Maximum degradation temperatures (T_{max}) of Mango seed Husk Hemicelluloses and their corresponding films made with hemicellulose extracts with xyloglucan /xylan ratio 0.13

Hemicelluloses T_{max} [°C]		Films T_{max} [°C]
MSHH (optimum point) (XGN/XLN ratio = 0.13)	330	290
XLN (Commercial)	290	240
XGN (Commercial)	380	332
Commercial XGN/Commercial XLN (0.13)	333	290
MSHH (non-optimum point) XGN/XLN ratio (0.13)	332	290

MSHH= Mango seed husk hemicellulose, XGN =Xyloglucan, XLN = Xylan

The subsequent HSHO treatment of the solid residues produced lignin with a purity level of 96% whereas, the lignin produced without high shear homogenization had a purity level of 94%. The cellulose content of the solid residues was over 75% with hemicellulose contents that were within 10% (Table 3), making the residue obtained suitable for nanocellulose production.

Table 3: Yields and properties of lignin and cellulose-rich residues obtained after high shear homogenization-assisted organosolv process

Component	Lignin		Solid residue	
	yield (%)	purity (%)	Cellulose (%)	Hemicellulose (%)
Predicted	67.18	96.67	75.21	11.13
optimum conditions	70.43	96.18	77.84	10.98
Deviation	4.61	0.51	3.38	1.35
[a] Reference	68.58	94.74	76.75	10.16
Deviation	2.04	1.99	2.00	8.79

Optimum conditions (Ethanol concentration = 60%, Temperature = 148.41 °C and Homogenizing time = 15 min), Reference lignin (non-assisted process)

The properties of the lignin and cellulose-rich residues were predictable using quadratic models at a 95% confidence level. The hydrolysis of the remaining cellulose-rich residues by formic acids without any catalyst produced cellulose nanocrystals (CNCs). The CNCs produced by formic acid hydrolysis attained mostly spherical shapes (average diameters 25 nm) as shown in Fig. 2 and thermal stability of 361 °C, which is higher than the thermal stability of CNCs (309 °C) produced by sulphuric acid (Fig. 3).

The wheat straw cellulose-rich pulp obtained after the two-stage alkaline treatment had cellulose, hemicelluloses content, lignin content, and crystallinity of 76%, 3%, 5%, and 55%, respectively (Fig. 4) which rendered the residues suitable for nanocellulose production.

Table 4: Formic acid nanocellulose production at optimum conditions

Component	Yield [%]	[b]Average particle size [nm]	CrI [%]	Formate content
Predicted	65.	25	68	0.94
FCNC	64	24	66	0.92
SCNC	28	176	75	-

[a]FCNC: Spherical formic acid cellulose nanocrystals, SCNC= sulphuric acid cellulose nanocrystals [b]Represents particle size (length) obtained from several scanning electron microscopy images using ImageJ software, CrI= crystallinity index

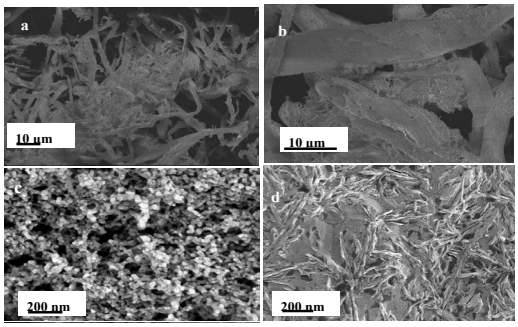

Figure 2: Scanning Electron Microscopy of (a) unbleached organosolv treated pulp (b) bleached pulp (c) formic acid cellulose nanocrystals, (d) sulphuric acid cellulose nanocrystals

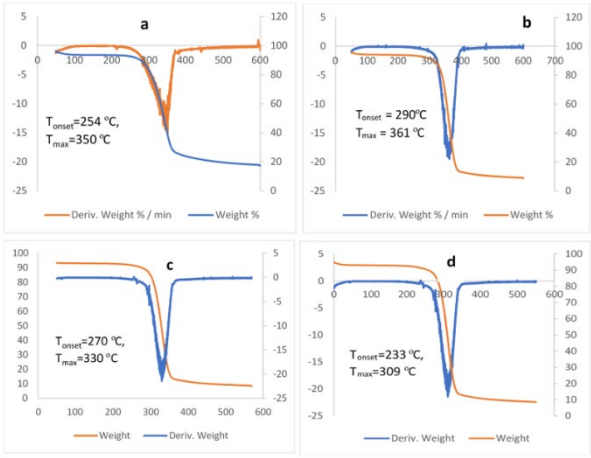

Figure 3: Maximum thermal degradation temperature (T_{max}) of (a) unbleached mango seed husk pulp, (b) formic acid cellulose nanoparticles (at the optimum point) (c) bleached mango seed husk pulp, and (d) sulphuric acid cellulose nanoparticles.

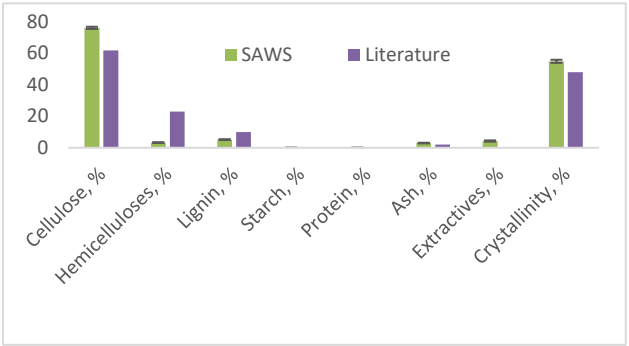

Figure 4: Composition and properties of wheat straw (WS) after the severe alkaline treatment (SAWS) compared to literature.

The nanocellulose production from delignified wheat straw using hydrochloric acid and enzymatic had comparable yields (≈20%) but the yields were less than the yield (34%*)* obtained using the sulphuric acid treatment (Fig. 5). The crystallinity of the HCL produced nanocellulose (HCN) was comparable (70%) to that of nanocellulose produced by the sulphuric acid (SCN) (75%) (Fig. 5).

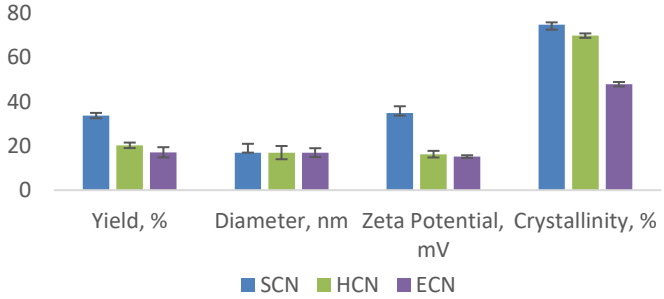

Figure 5: Yield, diameter, zeta potential, and crystallinity for commercial nanocelluloses, sulphuric acid produced nanocelluloses (SCN), hydrochloric acid produced nanocelluloses (HCN), and enzymatic treatment produced nanocelluloses (ECN)

The particle size of the HCN, SCN, and enzyme-produced nanocelluloses (ECN) was comparable (Fig. 5) but the polydispersity index and zeta potential (-16 mV) were lower than those of the SCN. However, the SCN had the lowest maximum thermal decomposition temperature (309^{o}C) as compared to both the HCN and ECN (378°C and 380°C, respectively) (Fig. 6). Based on the morphology and the zeta potential, the HCN and ECN were classified as cellulose

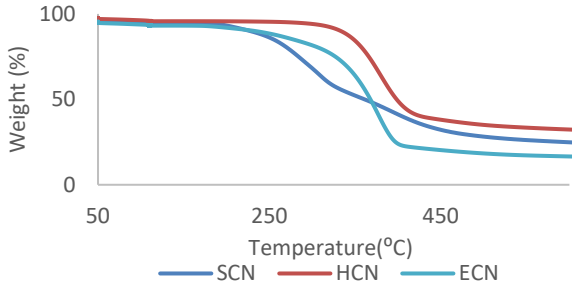

Figure 6: Maximum thermal degradation temperature of sulphuric acid nanoparticles (SCN), hydrochloric acid nanoparticles (HCN), nanofibrillated cellulose (CNF), cellulose nanocrystals (CNC), enzymatic cellulose nanoparticles (ECN

nanofibers and as relatively stable colloids. Various self-supporting biocomposite films were produced through the casting method from the MSHH and FCNC (Fig 7). The Young's Modulus improved by adding plasticizer and FCNC (Figs. 8a and b).

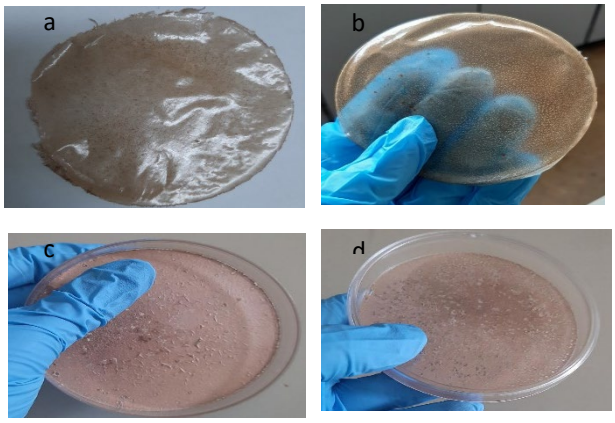

Figure 7: Biocomposite films (a) mango seed husk hemicellulose (without plasticizer), and b) mango seed husk hemicelluloses with plasticizer, (c) Mango seed husk hemicelluloses with 10% and (d) 40% formic acid generated nanocellulose

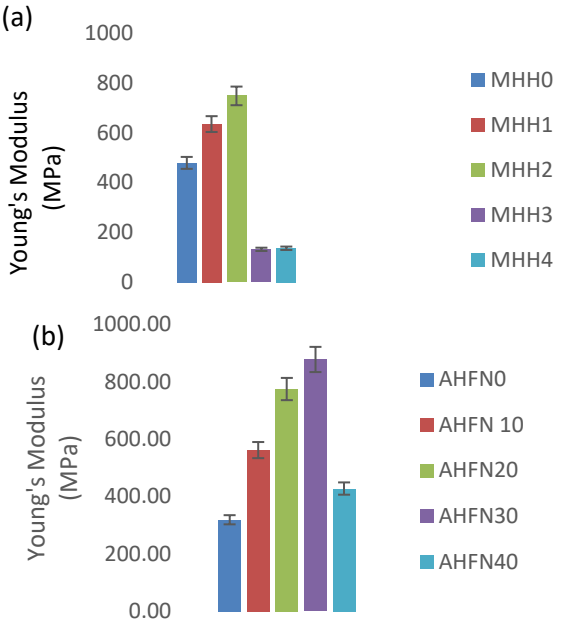

Figure 8: (a) Young's Modulus of mango seed husk hemicellulose (MHH) with plasticizer and (b) Young's Modulus of acetylated MHH with formic acid produced cellulose nanocrystals at dosage sof 0, 10, 20, 30 and 40%.

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