Optimization of Bacterial NanoCellulose fermentation using lignocellulosic residues and development of novel BNC-starch composites

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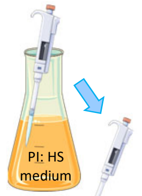
Introduction

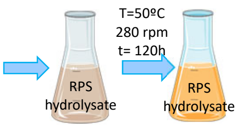
In the paper industry, significant fraction of fibers that cannot be re-utilized are wasted by the paper companies, which raise economic and environmental concerns. Unique properties of BNC have sustained promising applications in the biomedical field, pulp and paper, composites and foods. In order to ally the recycling of lignocellulosic residues and the production of BNC, recycled paper sludge (RPS) was enzymatically hydrolyzed to obtain sugar hydrolysates, which were used for BNC production. More specifically, culture medium was optimized to enhance BNC production by response surface methodology. Overall, the results suggest that RPS had potential to be an alternative carbon source for BNC production with a maximum BNC yield of 4.40 g/L. BNC as described above was used for the development of novel green thermoplastic nanocomposites, combined with starch. In this work, two approaches for composite production were assessed. Firstly, impregnation method and cast method. All nanocomposites manufactured were then characterized in terms of mechanical properties and permeability to water vapor (WVT). Overall, enhanced mechanical and barrier properties were obtained with composites composed by BNC membrane filled with TPS.

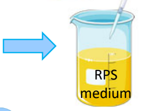
Bacterial nanocellulose production with RPS residues

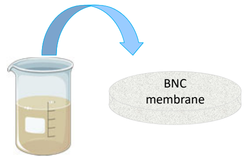
Pre inoculum preparation:
100 mL in E-Flask 1L ; HS medium with pH 5.5; 48 h; static culture;30 ºC
Strain:
Komagataeibacter hansenii ATCC 53582



BNC washing
1- NaOH 0.1M
2- Distilled water (until neutral pH)







Recycled paper sludge (RPS)
30 % of glucans, 16,11 % of xylans in the solids

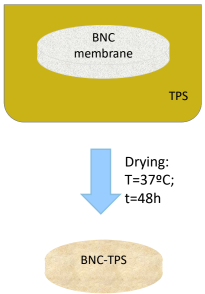
Neutralization (RPS)
Fibre suspension 16 % (m/v); HCL 4 %

Enzymatic hydrolysis
cellulase with b-glucosidase (cellicCtec2) 20 FPU 120 FPU/ml

Culture medium composition:
RPS medium(%,m/v): RPS hydrolysate (0.38– 4.62); yeast extract and peptone (0.50 – 1.71); citric acid (0.126) ; Na2HPO4 (0.339);
Fermentation parameters:
55 mL (height = 1.5 cm) of each run; 15 days , static culture, 30 ºC;

Nanocellulose-starch composite production

Impregnation method

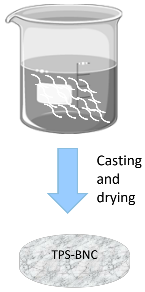


Parameters tested:
Wet thickness (mm)
- 1.00
- 3.00
- 5.00

TPS composition
- Starch 5%
- Glycerol 1.5%

Impregnation time (h)
- 0
- 24
- 72

Casting method



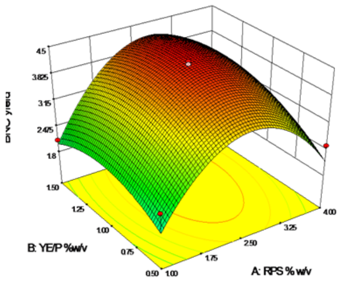
Parameters tested:
Type of cellulose:
- Nanofibrilated BNC
- Celluforce
- Avicel

Cellulose (% m/v)
- 0.25
- 0.50

TPS composition
- Starch 5%
- Glycerol 1.5%

Results

BNC production optimization with *K.hansenii 53582* using RPS hydrolysate



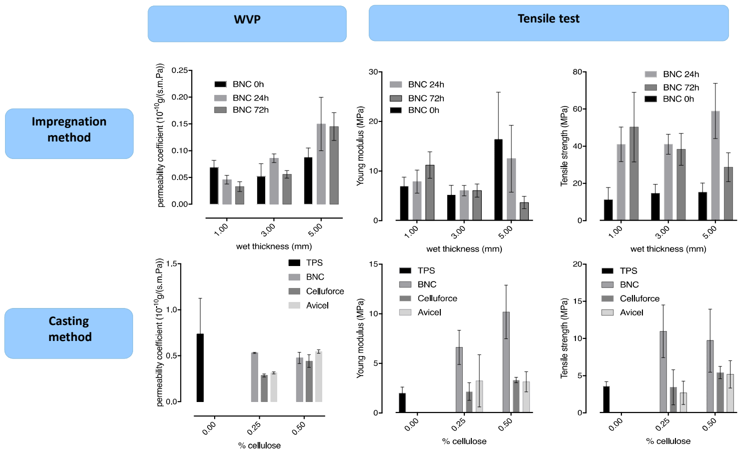
Model equation:

$$\text{BNC yield} = 3.52 * \text{Carbon source} + 3.79 * \text{nitrogen source} + 0.233 * \text{Carbon source} * \text{nitrogen source} - 0{,}705 * \text{Carbon source}^2 - 1.93 * \text{Nitrogen source}^2 - 2.51$$

Table 3: validation experiments of CCD

Validation runs	RPS (% w/v)	YE/P (% w/v)	Predicted BNC yield (g/L)	Experimental BNC yield (g/L)	95 %CL low	95 %CL high
1	4.00	0.50	2.30	3.03	1.38	3.21
2	1.00	0.50	1.74	3.36	0.82	2.66
3	2.50	1.00	4.38	4.40	3.85	4.90
4	2.61	1.20	4.46	4.85	3.95	4.97

Water vapour permeability (WVP) and mechanical characterization



Conclusions

1. *Response surface mothodology G. hansenii 53582*
   1. According to the model, carbon source concentration from RPS hydrolysate, is critical to maximize the BNC. Culture medium was successfully optimized (26.9 g/L of RPS hydrolysate and 11.4 g/L of yeast extract and peptone). However, BNC yield did not exceed 4.40 g / L. To achieve higher yields of BNC, other parameters should be optimized such as the depth of culture medium and incubation time;
2. *Composites characterization*
   1. Lower permeability was observed in composites produced by impregnation method. Permeability increased with higher thickness. However, similar values were observed between 24h and 72h impregnation time; Composites produced by cast method presented similar WVT with Avicel, BNC and celluforce at all cellulose concentrations, although being higher than TPS film;
   2. Higher young modulus and tensile strength were obtained with composites produced by impregnation method. No significant differences were seen in terms of tensile parameters between impregnation time; Improved tensile strength was seen with higher thickness; Composites produced by cast method presented enhanced mechanical properties when reinforced with BNC at all concentrations.

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Acknowledgements

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