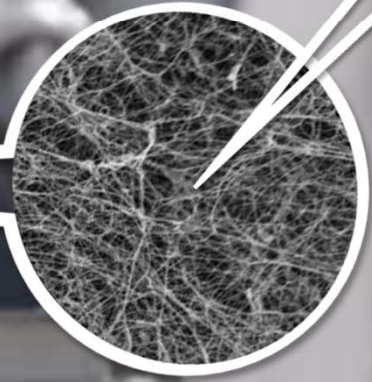
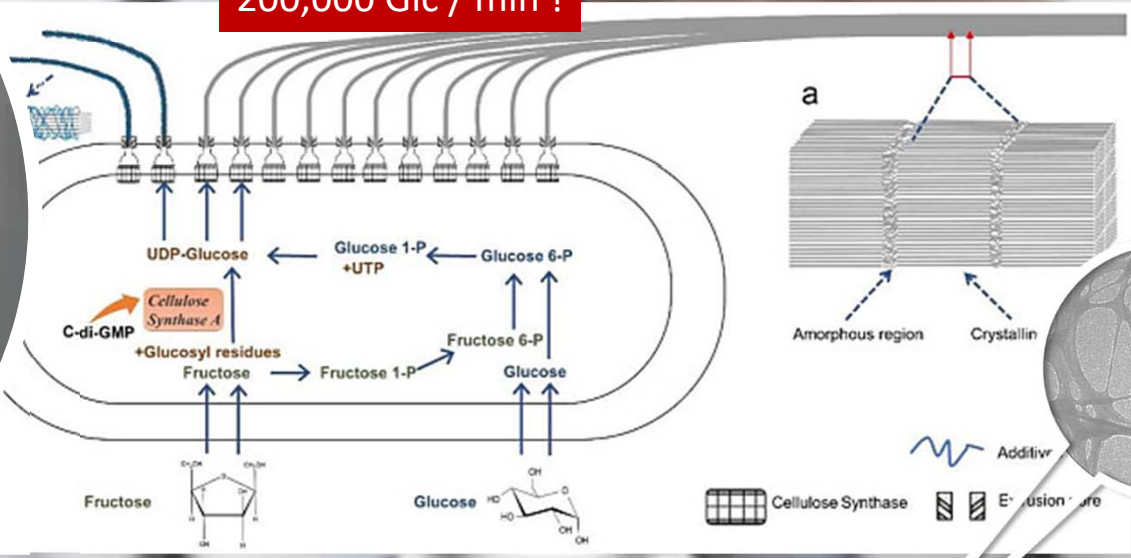


Economic & environmental assessment of bacterial cellulose production

Ana Forte, Fernando Dourado, Miguel Gama, Eugénio C. Ferreira

What is Bacterial Cellulose?

200,000 Glc / min !



Appealing properties

Properties		References
Degree of polymerization	up to 8000	(Wu et al., 2016)
Crystallinity	60–90%.	(Klemm et al., 2005)
Nanofibers' diameter	20–100 nm	(Wu et al., 2016)
Nanofibers' length	1–9 μm	(Foresti, Vázquez and Boury, 2017)
Density	1.50 $\text{g}\cdot\text{cm}^{-3}$ 1.25 $\text{g}\cdot\text{cm}^{-3}$	(Hervy et al., 2018) (Lee, Blaker and Bismarck, 2009b)
Surface area	2.7–37 $\text{m}^2\cdot\text{g}^{-1}$	(Kim, Nishiyama and Kuga, 2002)
Water absorption	610.5 - 320%	(Potivara and Phisalaphong, 2019) (Rathinamoorthy et al., 2019)
Water vapor permeability	765.95–1045.55 $\text{g}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$ 2.38 $\times 10^{-11}$ $\text{g}\cdot\text{m}^{-1}\cdot\text{s}^{-1}\cdot\text{Pa}^{-1}$ 151 $\text{g}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$ 1.26 $\times 10^{-10}$ $\text{g}\cdot\text{m}^{-1}\cdot\text{h}^{-1}\cdot\text{Pa}^{-1}$ 1–11 $\times 10^{-13}$ $\text{g}\cdot\text{m}^{-1}\cdot\text{s}^{-1}\cdot\text{Pa}^{-1}$	(Kamal, Misnon and Fadil, 2020) (Cazón, Velázquez and Vázquez, 2019) (Rathinamoorthy et al., 2019) (Jebel and Almasi, 2016) (Tomé et al., 2010)
Young's modulus	1044 MPa 5–17 GPa 93.8 MPa 138 GPa 114 GPa 15–35 GPa	(Cazón, Velázquez and Vázquez, 2019) (Potivara and Phisalaphong, 2019) (Jebel and Almasi, 2016) (Hu et al., 2011) (Lee, Blaker and Bismarck, 2009b) (Klemm et al., 2005)
Tensile strength	20.8 MPa 70–300 MPa 26.3 MPa 2 GPa 200–300 MPa	(Cazón, Velázquez and Vázquez, 2019) (Potivara and Phisalaphong, 2019) (Jebel and Almasi, 2016) (Hu et al., 2011) (Klemm et al., 2005)
Elongation	2.3% 0.5%–5.0% 6.1 % 1.5–2.0%	(Cazón, Velázquez and Vázquez, 2019) (Potivara and Phisalaphong, 2019) (Jebel and Almasi, 2016) (Klemm et al., 2005)

6886

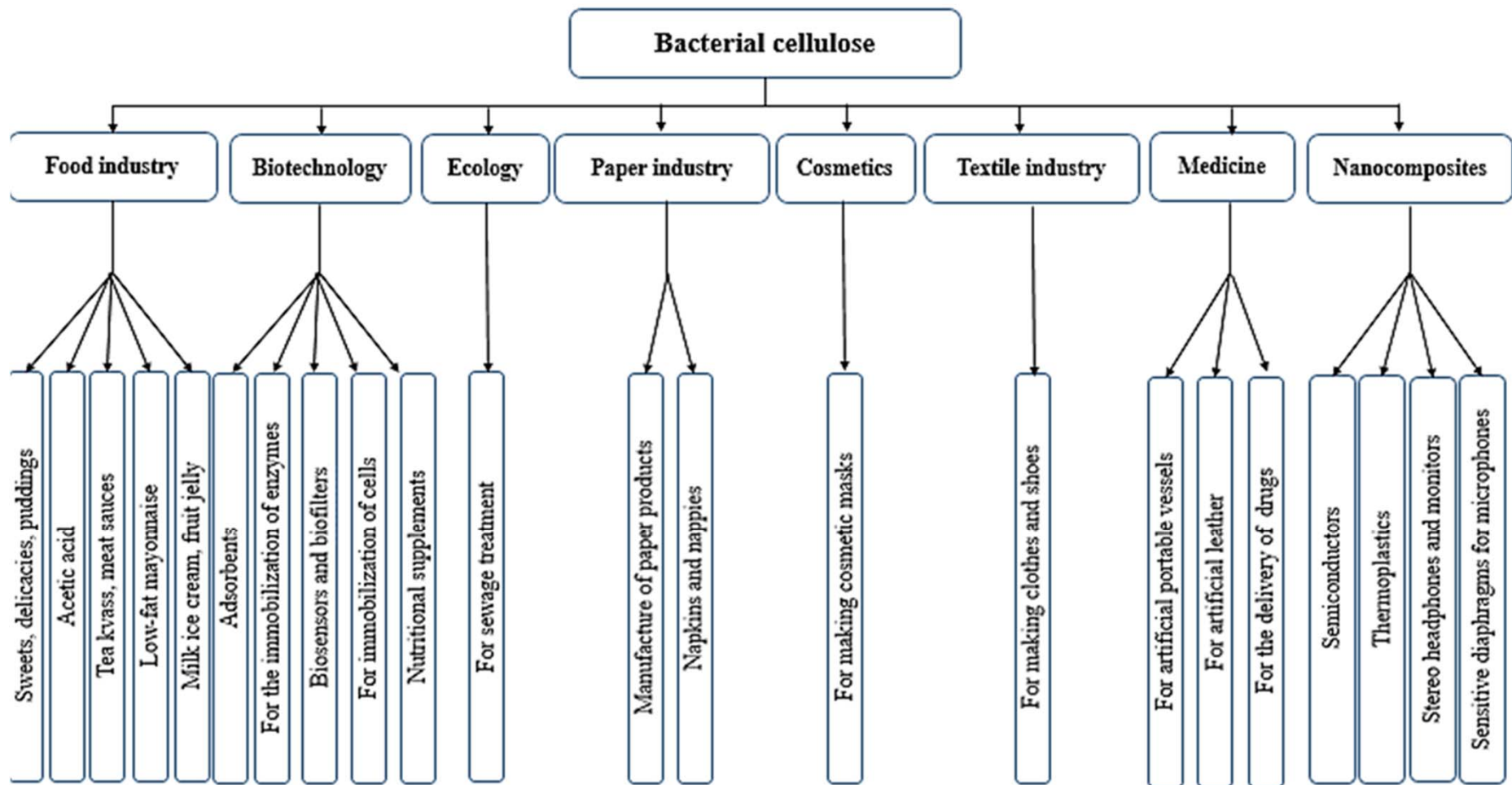
Appl Microbiol Biotechnol (2018) 102:6885–6898

Table 1 Bacterial cellulose productivity (g/L/days) by AAB in different conditions

Present name ^a	Carbon source/feedstock		System	Time (days)	Productivity (g/L/days)	Reference
	Type	Amount (g/L)				
<i>K. xylinus</i> AX2-16	Glucose	25	Static	8	1.47	Zhong et al. (2013)
<i>K. hansenii</i> PJK=KCTC10505BP	Glucose	10	Submerged	2	0.86	Jung et al. (2005)
	Acetic acid	1.5 mL				
	Succinate	2				
	Ethanol	10				
<i>K. xylinus</i> BRC5=KCCM 10100	CSL:	80	Submerged	2	7.65	Hwang et al. (1999)
	Glucose	20				
	Citric acid	1.15				
<i>K. rhaeticus</i> P1463	Apple juice		Static	14	0.68	Semjonovs et al. (2017)
	Glucose	3				
	Fructose	12.4				
	Sucrose	4.6				
<i>K. hansenii</i> B22	Apple juice:		Static	14	0.50	Semjonovs et al. (2017)
	Glucose	3				
	Fructose	12.4				
	Sucrose	4.6				
<i>K. xylinus</i> K2G30=UMCC 2756	Glucose	50	Static	15	1.31	Gullo et al. (2017)
	Ethanol	14				
<i>K. xylinus</i> ATCC 23767 ^T	CSL:		Static	7	0.41	Cheng et al. (2017)
	Glucose	3.87				
	Xylose	29.61				
	Mannose	1.84				
	Acetic acid	18.73				
<i>K. xylinus</i> BCRC 12334	Glucose	60	Static	14	0.52	Kuo et al. (2015)
<i>K. xylinus</i> ATCC 23770	HFS:		Static	7	1.57	Cavka et al. (2013)
	Glucose	14.1				
<i>K. hansenii</i> M2010332	Glucose	55	Static	7	2.33	Li et al. (2012)
	Citric acid	1				
	Ethanol	20				
<i>Gluconacetobacter</i> sp. st-60-12 and <i>Lactobacillus</i> sp. st-20	CSL:	40	Submerged	3	1.4	Seto et al. (2006)
	Sucrose	40				
<i>K. xylinus</i> BPR2001	CSL:		Submerged	2.5	3.2	Noro et al. (2004)
	Fructose	40				
	Inositol	0.002				
<i>K. xylinus</i> BPR2001	CSL:		Static	3	1.8	Bae and Shoda (2005)
	Sucrose	37				

CSL com steep liquor, HFS hydrolysate fiber sludges

^aThe present names of AAB species are reported, according to LPSN bacterio.net (Euzéby 1997)

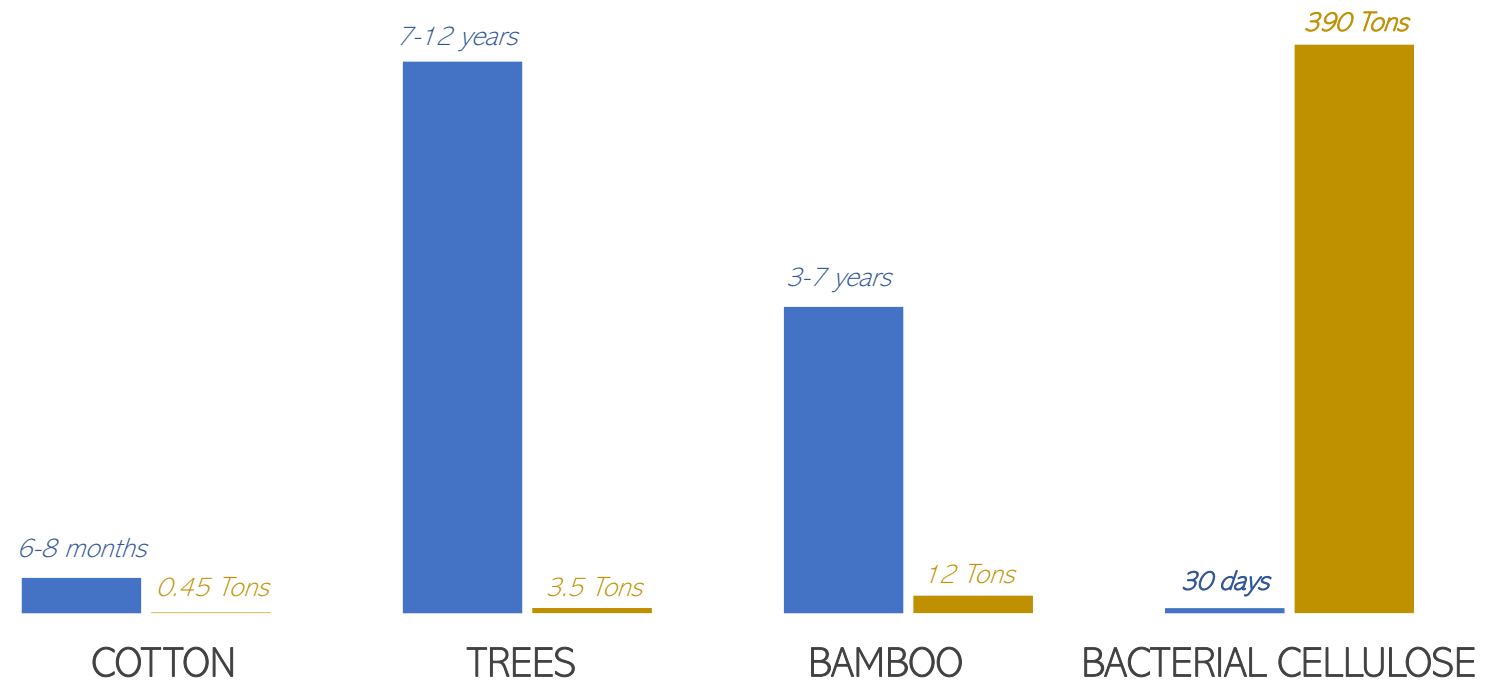




Advantages of bacterial cellulose fermentation technology

Reference area: football field


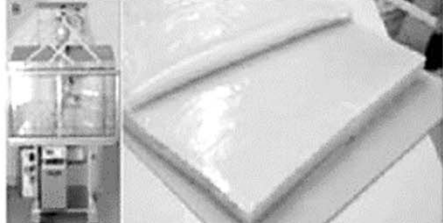
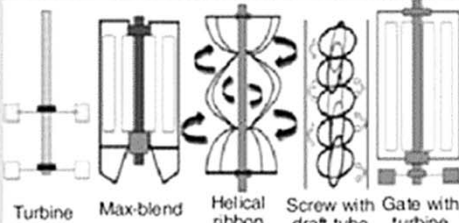
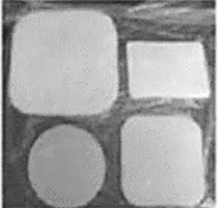

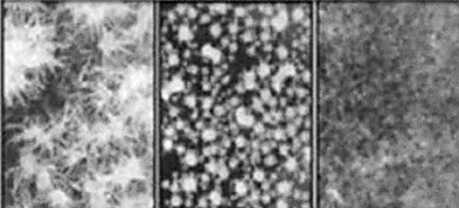
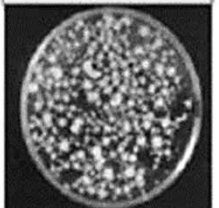
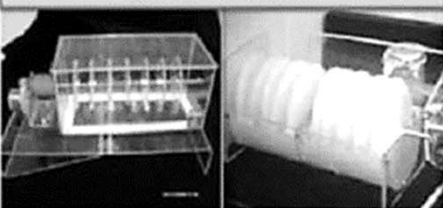
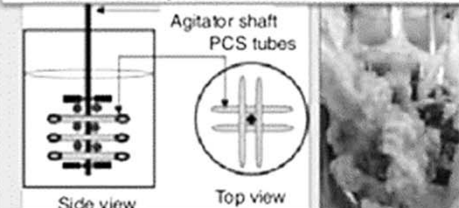
The combination of recyclability, sustainability, biodegradability and new end-use opportunities makes BC the potential super material of the future.



Efforts towards cost-effective BC production

Different strains
 Different media composition
 Different culture conditions

Low BNC yields

Bacterial cellulose production in laboratory	Bacterial cellulose production in reactors (static conditions)	Bacterial cellulose production in reactors (agitated conditions)
 <p data-bbox="875 651 1093 715">Production in flask</p>	 <p data-bbox="1144 651 1585 715">Horizontal lift reactor</p>	 <p data-bbox="1630 651 2087 715">Agitated configuration in bacterial cellulose production</p>
 <p data-bbox="875 938 1093 986">Mats</p>	 <p data-bbox="1144 938 1585 986">Aerosol bioreactor</p>	 <p data-bbox="1630 938 2087 986">Various shapes of bacterial cellulose</p>
 <p data-bbox="875 1193 1093 1241">Pellets</p>	 <p data-bbox="1144 1193 1585 1241">Rotary disc reactor</p>	 <p data-bbox="1630 1193 2087 1241">Plastic composite support biofilm reactor</p>

Dourado, F., et al. (2016). *Bacterial Nanocellulose*. Amsterdam, Elsevier: 199-214

Ullah, M.W., Manan, S., Kiprono, S.J., Ul-Islam, M. and Yang, G. (2019). *Nanocellulose* (eds J. Huang, A. Dufresne and N. Lin).

Appealing marketable applications (but confined mostly to health/biomedical)



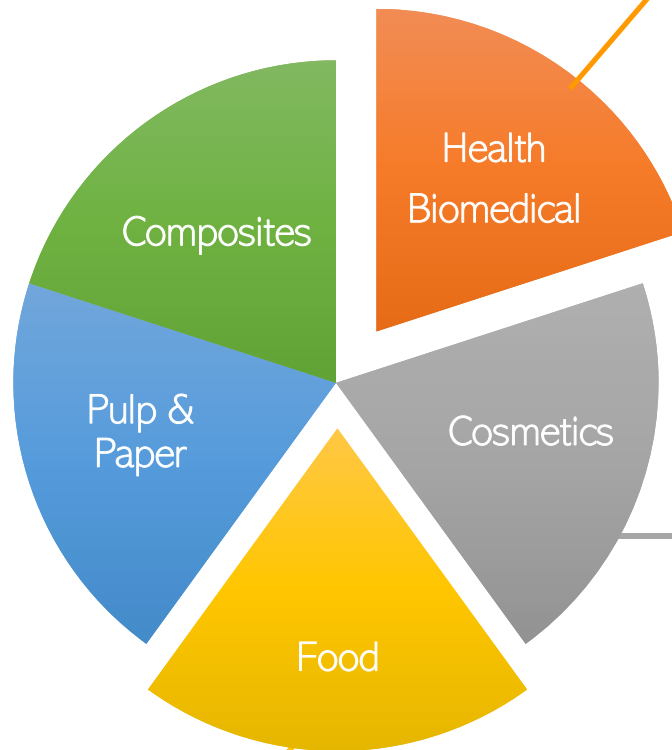
● Active companies

Low value-added
Large volume



Nata de coco:
Asian Countries
(several companies)

Cellulon®, Primacel®:
Cetus Co. (USA)
Weyerhaeuser Co. (USA)



High value-added
Nich markets

Xylos (USA)
Biofill (Brazil)
JenNaCell (Germany)
Hylomorph (Switzerland)

Gengiflex®
(surgery and dental implants)

Bioprocess®
XCell®
Biofill®
Dermafill®
Gengiflex®
(wound dressings)

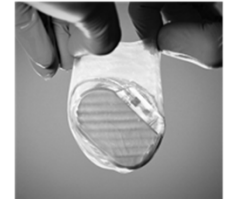
Asian Countries
(several companies)

Bowil Biotech (Poland)

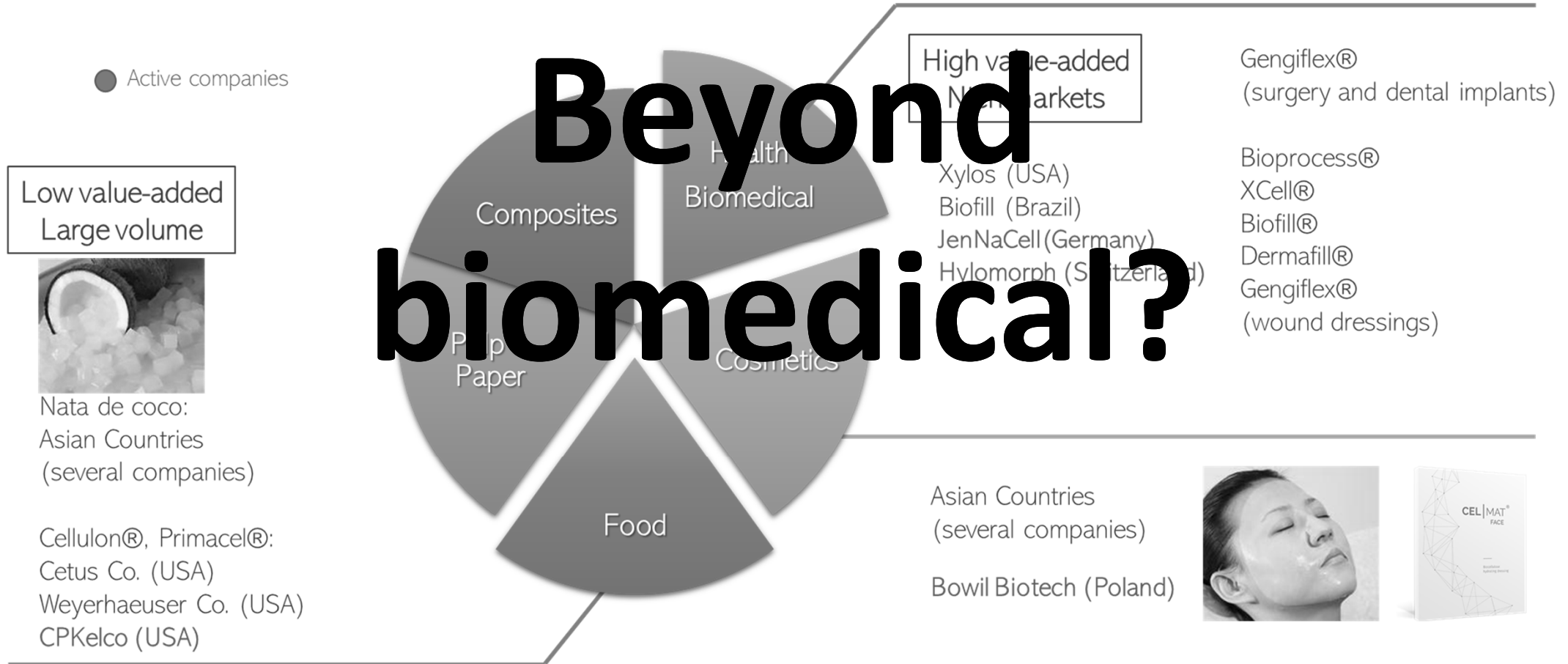


CPKelco (USA) AxCel®PX, AxCel®CG-PX, AxCel®PG, Cellulon™PX....

Appealing marketable applications (but confined mostly to health/biomedical)



Beyond biomedical?



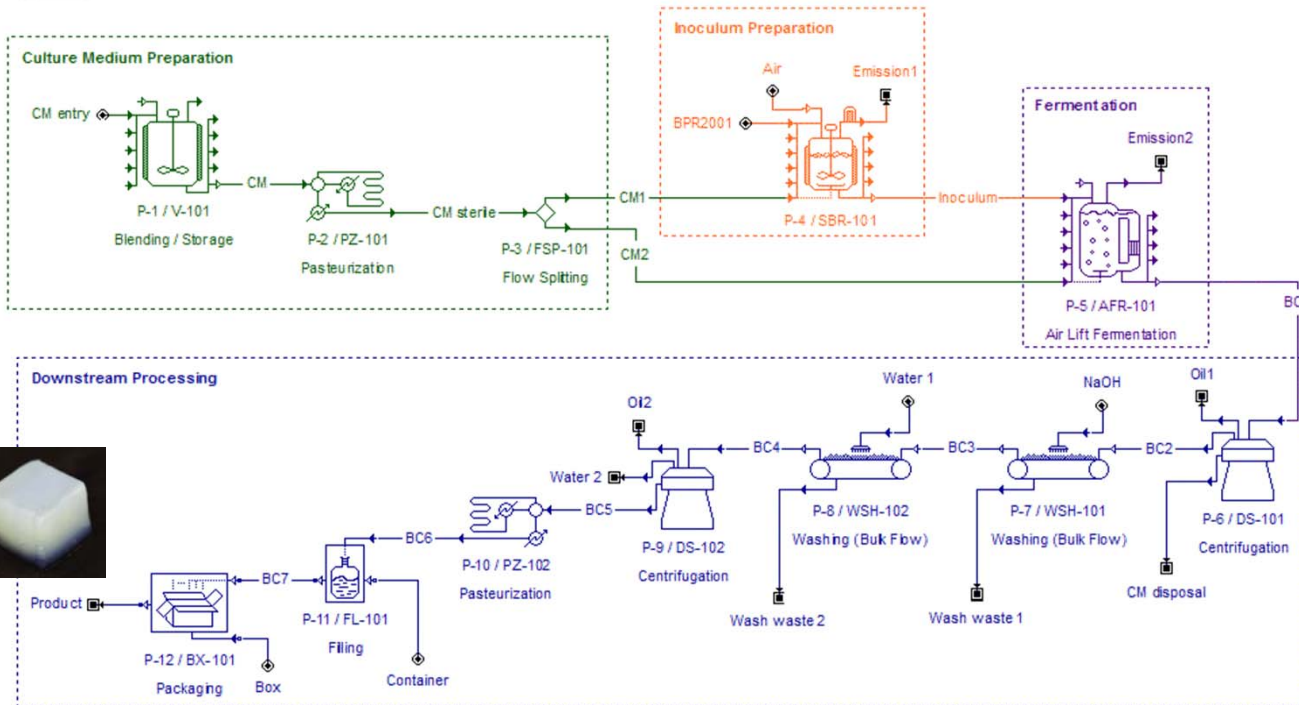
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Scaling up

How expensive?

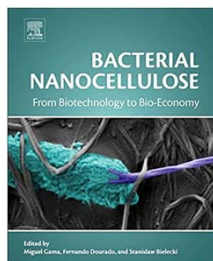
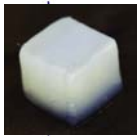
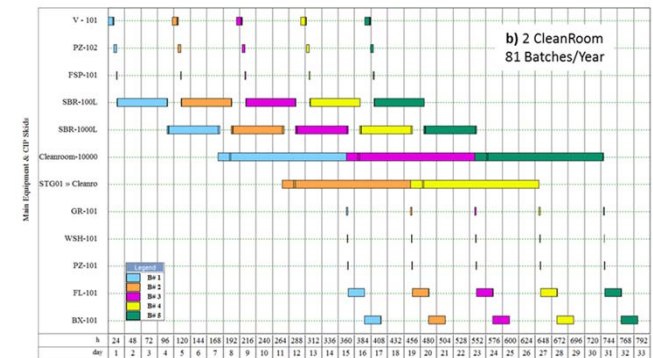


Economic assessment of the BC production



Static & agitated culture conditions

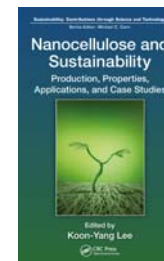
Super-Pro process flowsheet for BNC production
 ≈ 3.5 Ton/year, dried basis (500 Ton/year wet)



Process Modeling and Techno-Economic Evaluation of an Industrial Bacterial NanoCellulose Fermentation Process

Fernando Dourado, Ana Fontão, Marta Leal, Ana Cristina Rodrigues, Miguel Gama
 Minho University, Biological Engineering Department, Campus de Gualtar, Braga, Portugal

CHAPTER 12



1 Process Modelling and Techno-Economic Evaluation of an Industrial Airlift Bacterial Cellulose Fermentation Process

Fernando Dourado, Ana Isabel Fontão, Marta Leal, Ana Cristina Rodrigues and Miguel Gama

M.S. Peters, K.D. Timmerhaus, in: M.S. Peters, K.D. Timmerhaus (Eds.) Plant Design and Economics for Chemical Engineers, McGraw-Hill, Singapore, 1991, pp. 150-215.

Capital Investment Cost (M USD\$)	AGITATED	STATIC
Direct Costs (DC)	12.5	9.2
Indirect Costs (IC)	3.1	2.1
<i>Fixed Capital Investment (FCI)</i>	<i>15.6</i>	<i>11.3</i>
<i>Working Capital (WC)</i>	<i>2.3</i>	<i>1.7</i>
TOTAL CAPITAL INVESTMENT (TCI)	17.9	13.0

Estimation of the Annual Product Manufacturing Cost (M USD\$)		
<i>Manufacturing Costs (MC)</i>	<i>6.25</i>	<i>5.4</i>
Direct Production Cost (Variable costs)	2.4	2.2
Fixed charges	3.0	2.1
Plant overhead Costs (50-70% OL, DS & CL, and M & R)	0.85	1.1
<i>General Expenses (GE)</i>	<i>1.8</i>	<i>2.0</i>
TOTAL PRODUCT COST (TPC): MC + GE	8.0	7.4

Profitability (M USD\$) (500 MTon/year, at a selling price of 25 USD\$/Kg)		
Total income: Selling Price X Quantity of product	12.5	12.5
Gross income: Total Income - Total Product Cost	4.5	5.1
Taxes: 30-40% Gross Income	1.6	1.8
Net Profit: Gross Income - Taxes	3.0	3.4
Rate of Return: Net Profit*100/TCI	7%	11%
Payout Period: FCI/(Net Profit + Depreciation)	5 Years	4 Years



Dourado, F., et al. (2016). Process Modeling and Techno-Economic Evaluation of an Industrial Bacterial NanoCellulose Fermentation Process. Bacterial Nanocellulose. Amsterdam, Elsevier: 199-214

Dourado, F., et al. (2017). Process modelling and techno-economic evaluation of an industrial air-lift bacterial cellulose fermentation process. Nanocellulose and Sustainability: Production, Properties, Applications, and Case Studies. CRC Press *in press*.

And what about the wastewaters?

Biodegradation (2020) 31:47–56

<https://doi.org/10.1007/s10532-020-09893-z>

ORIGINAL PAPER

Study and valorisation of wastewaters generated in the production of bacterial nanocellulose

Francisco A. G. Soares da Silva · João V. Oliveira · Catarina Felgueiras · Fernando Dourado · Miguel Gama · M. Madalena Alves

Low cost substrates
(agroindustrial wastes)



High COD, BOD!



\$\$\$ Uphill/Downhill

Bacterial cellulose production

Strain: *Komagataeibacter xylinum* ATCC 700178

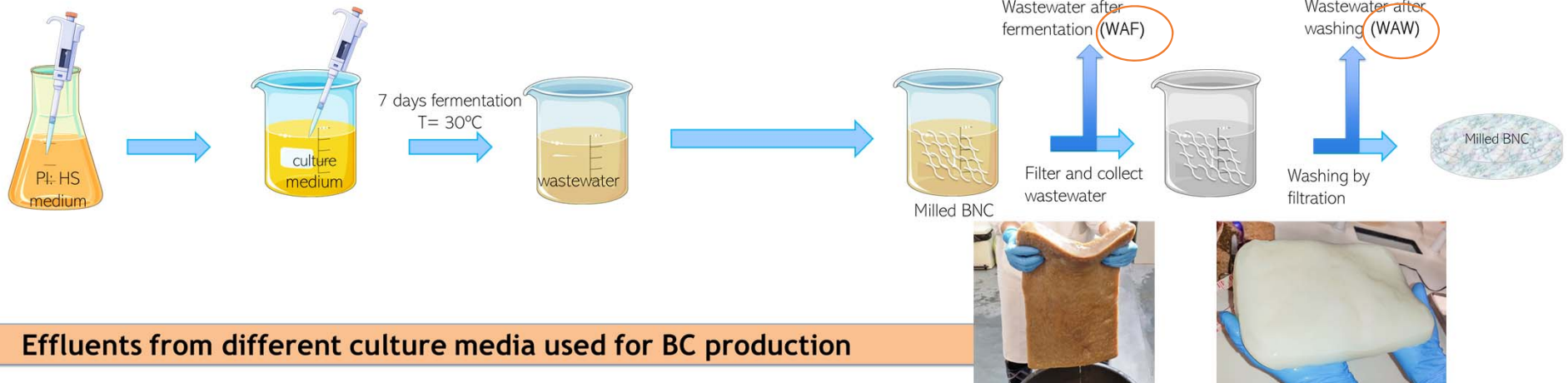
Culture media tested:

HS-EtOH - Glucose 40.0 g·L⁻¹; peptone 5.0 g·L⁻¹; yeast extract 5.0 g·L⁻¹;

Mol-HS - untreated molasse 40.0 g·L⁻¹; peptone 5.0 g·L⁻¹; yeast extract 5.0 g·L⁻¹;

Mol-CSL - untreated molasse 40.0 g·L⁻¹; CSL 7.0 g·L⁻¹; ammonium sulphate 0.5 % (w/v);

All formulations complemented ethanol 1.5 % (v/v), disodium phosphate di-hydrated 3.39 g·L⁻¹



Wastewater characterization

- Chemical oxygen demand
- Total and volatile solids
- Total nitrogen
- Volatile fatty acids

Anaerobic digestion:

- Biochemical methane potential assays
- UASB reactor

Effluents from different culture media used for BC production

Table 1. Characterization of effluents generated in the BC production.

Parameter	Units	Culture medium		
		HS-EtOH	Mol-HS	Mol-CSL*
TS	g·L ⁻¹	26.8 ± 0.4	53.1 ± 1.3	73.4 ± 1.6
VS	g·L ⁻¹	21.5 ± 0.4	43.7 ± 1.0	58.1 ± 1.4
COD	g·L ⁻¹	37.4 ± 5.7	54.5 ± 2.1	75.2 ± 2.5
TN	g·L ⁻¹	1.51 ± 0.11	1.21 ± 0.01	1.73 ± 0.08
BNC titre	g·L ⁻¹	1.79 ± 0.04	8. ± 0.29	5.95 ± 0.76

* This experiment generated the designated Wastewater after Fermentation - WaF

Table 2. Characterization of WaF and WaW as used in the batch assays.

Parameter	Units	WaF	WaW
TS	g·L ⁻¹	73.4 ± 1.6	20.6 ± 0.1
VS	g·L ⁻¹	58.1 ± 1.4	13.5 ± 0.1
CODt	g·L ⁻¹	75.2 ± 2.5	19.2 ± 0.2
CODs	g·L ⁻¹	74.3 ± 3.0	18.8 ± 0.3
TN	mg·L ⁻¹	1730 ± 80	900 ± 10
VFA	g L ⁻¹	nd	1.89

Biochemical methane potential of effluents from BNC fermentation of Mol-CSL

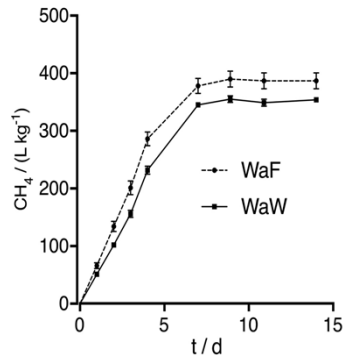
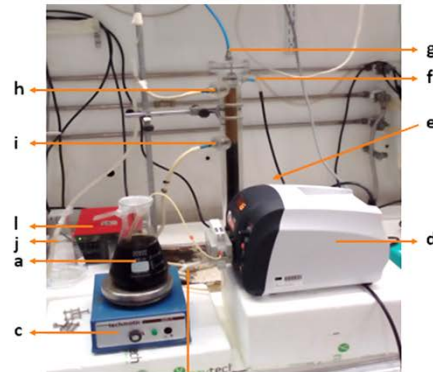


Table 3. Characterization of WaW after anaerobic biodegradability tests.

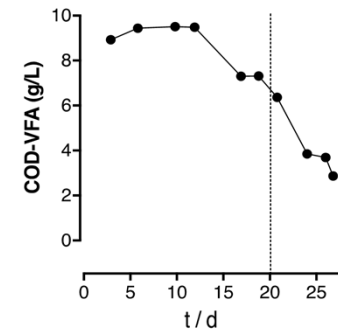
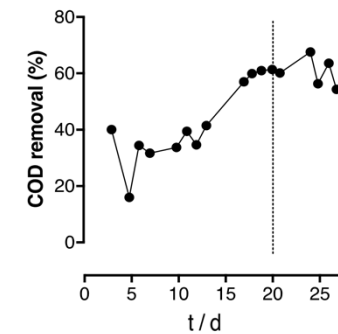
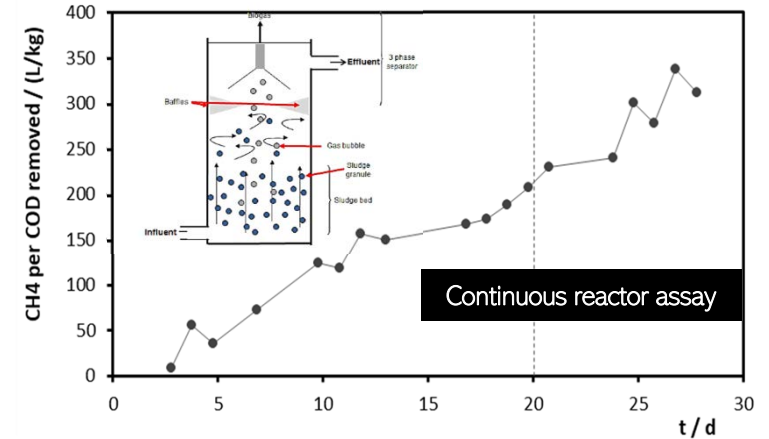
Parameter	Units	WaF	WaW
PM	%	86.9 ± 3.1	79.5 ± 0.9
CODs	g·L ⁻¹	0.72 ± 0.03	0.75 ± 0.10
VFA	g·L ⁻¹	n.d.	n.d.
NH ₄ ⁺	mg·L ⁻¹	312 ± 35	186 ± 10
NH ₃	mg·L ⁻¹	9.0 ± 1.0	8.0 ± 0.0

n.d. – non-detected;
Note: the BMP results presented were calculated after subtraction of blank value.

Fig1. Methane production (L.kg⁻¹ of VS) present in the biogas produced with WaF and WaW per day.



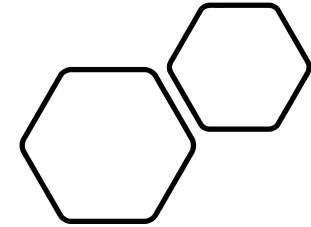
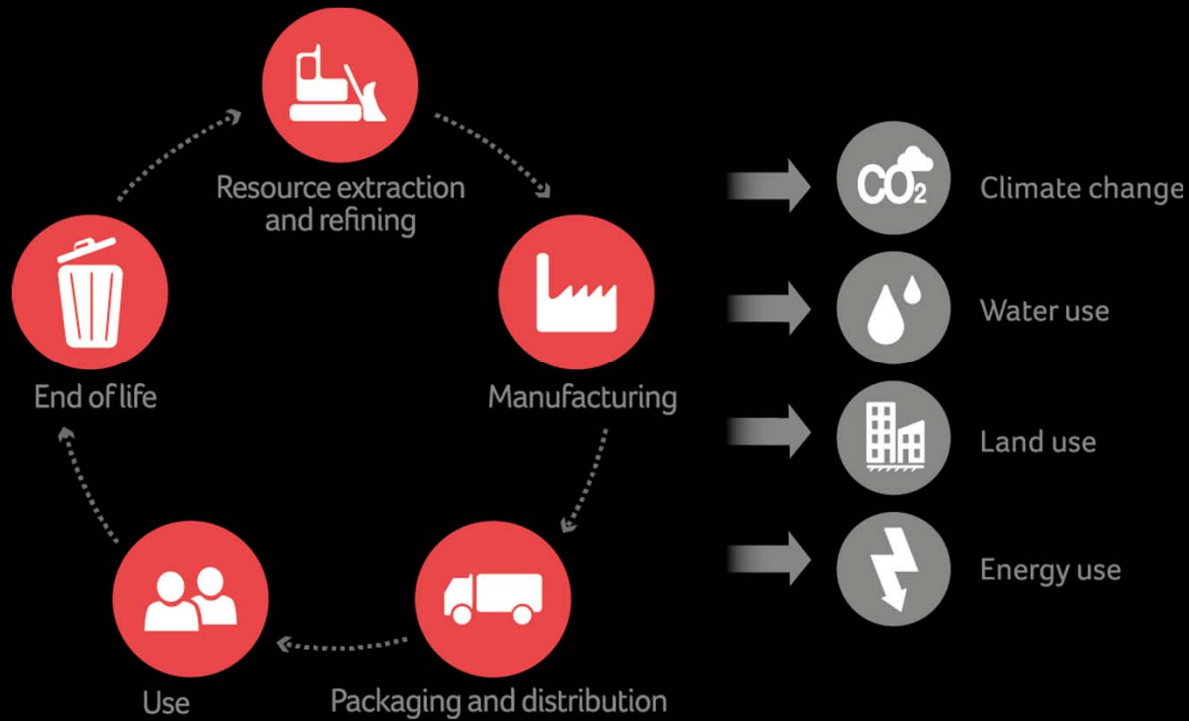
Upflow anaerobic sludge blanket reactor



Biodegradability tests: significant amount of biogas was obtained from WaF and WaW, along with reduced COD values.

UASB assay: while lower methane yield was obtained due to low HRT (40 h and 80h), this study showed the feasibility of using AD for the valorization of the wastewaters from the BC fermentation process.

LIFE CYCLE ASSESSMENT

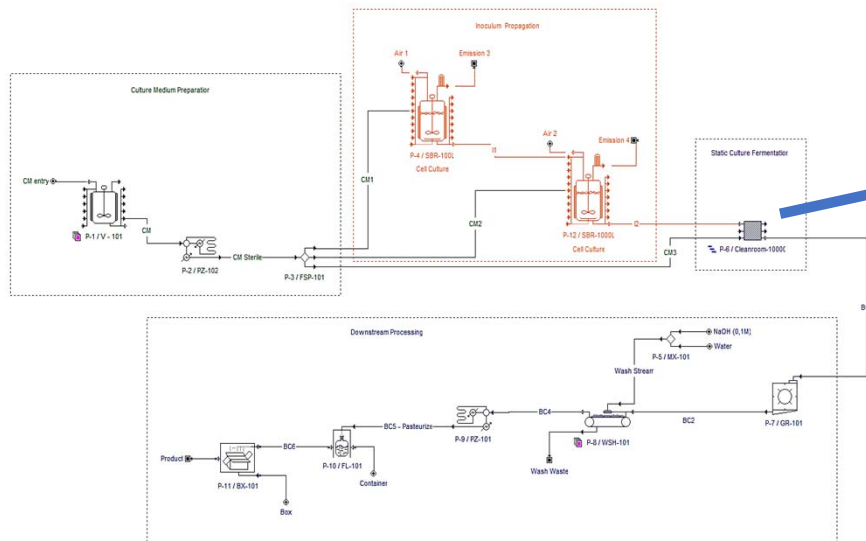


environmental,
health and
resource depletion

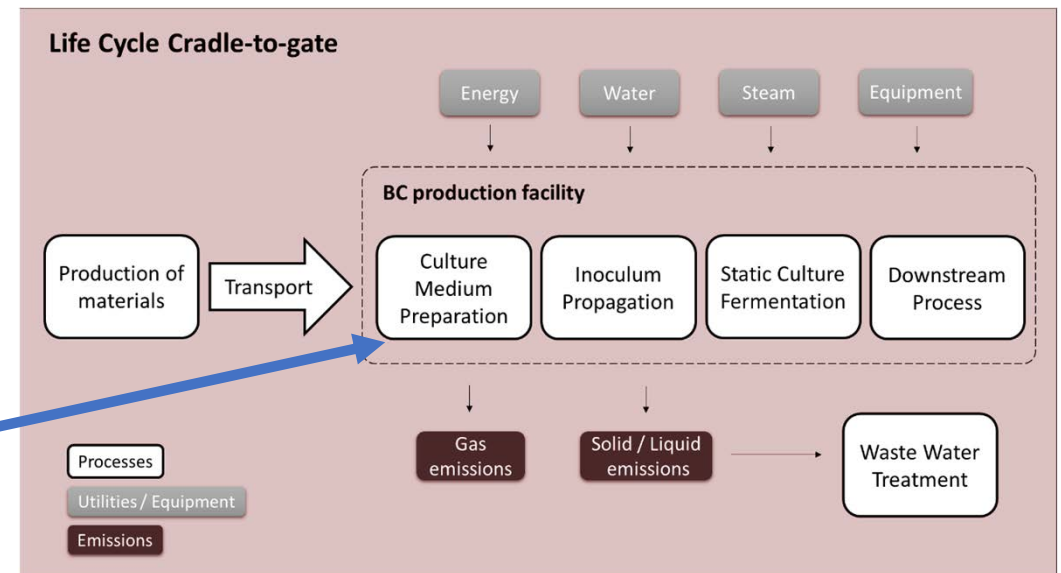
BACTERIAL CELLULOSE

Methodology: ISO 14040 and 14044

- Description of the BC production process
- Goal and scope definition and description of system boundaries
- Inventory Analysis
- Impact Assessment



Functional unit: 1 kg of BC (bone-dry mass), equivalent to 139 kg of final (wet) product



Impact Assessment



Gabi Pro software
ReCiPe 2016

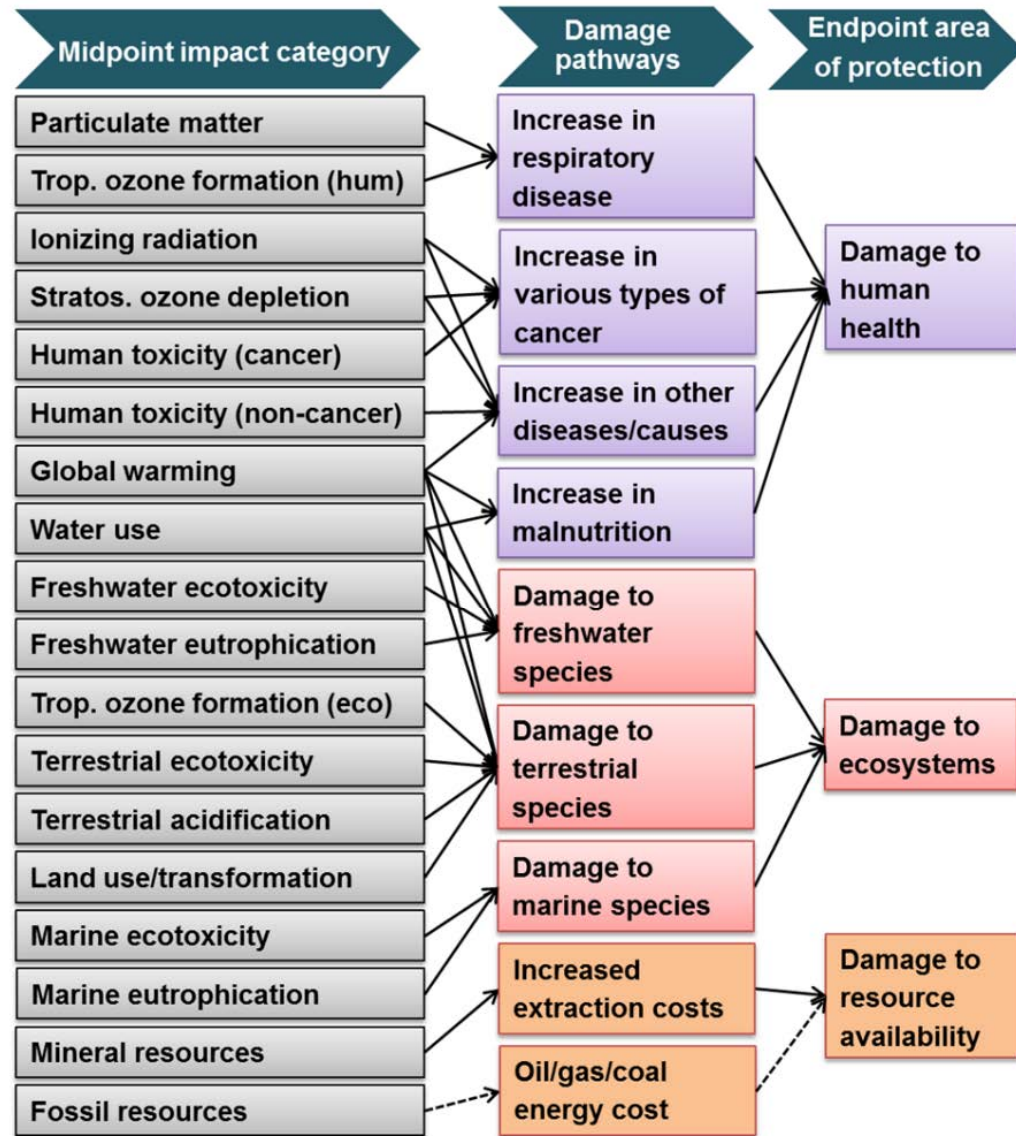
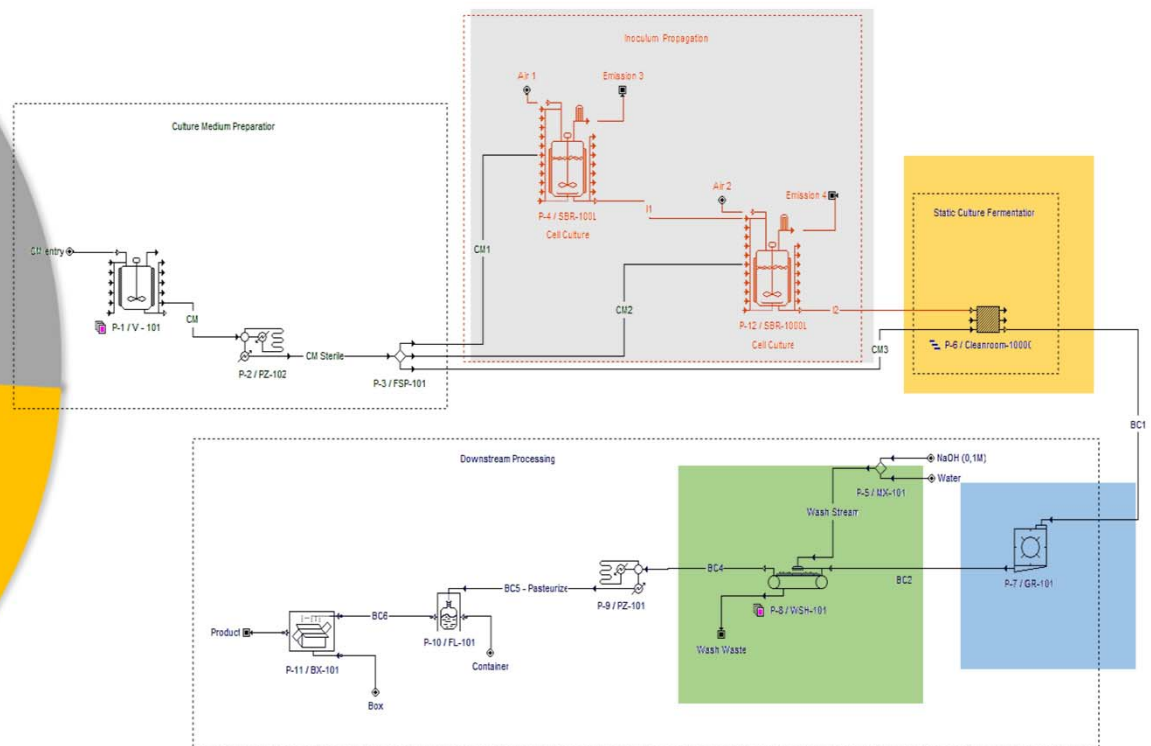
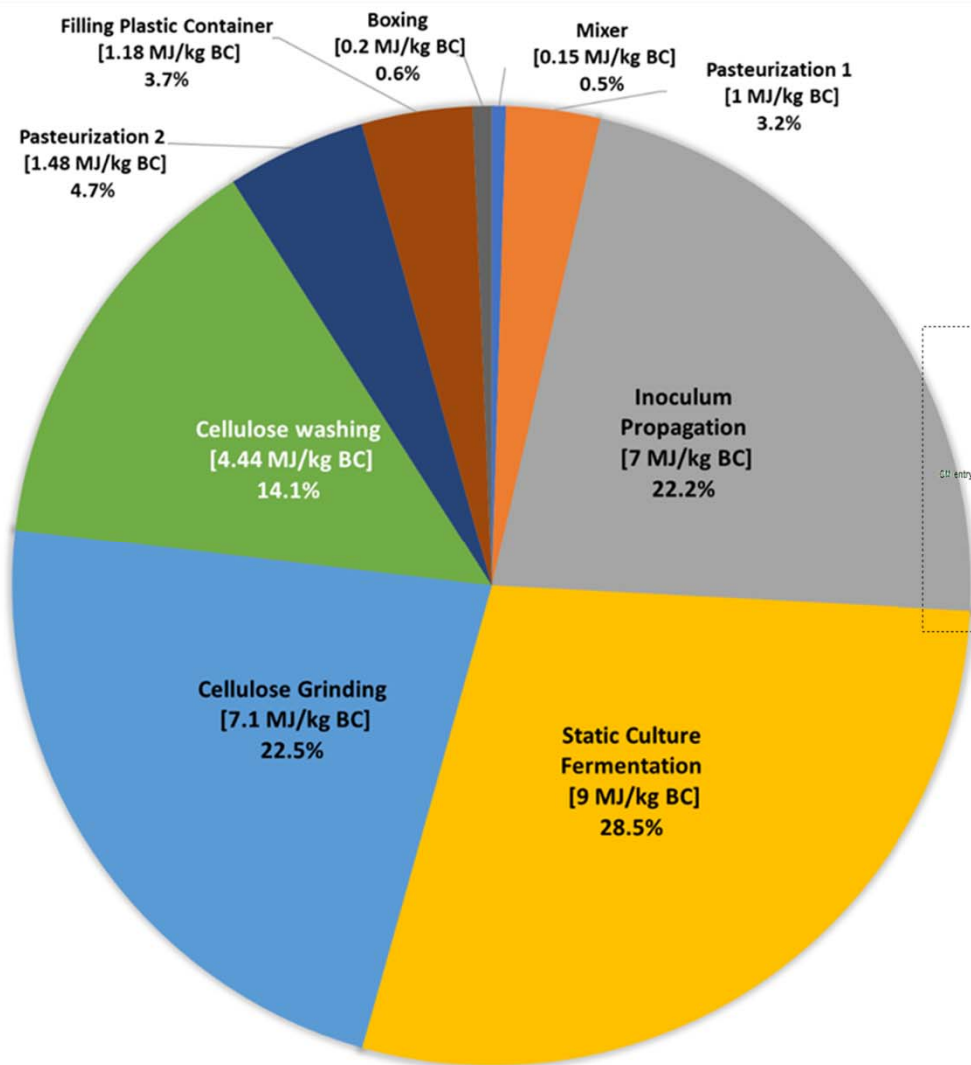


Figure 1.1. Overview of the impact categories that are covered in the ReCiPe2016 methodology and their relation to the areas of protection.

Mass balance of input (resources) and output (deposited goods and emissions) flows of the BC life cycle (1 kg BC, dry mass).

	Total (kg)	Production of materials (%)	Transport of materials (%)		Culture Medium Preparation (%)	Inoculum Propagation (%)	Stactic Culture Fermentation (%)	Downstream Process (%)		Wastewater treatment (%)
Resources (Total)	38,863.9	45.9	0.0		3.8	8.5	9.9	19.9		12.0
Energy resources	7.311	58.3	1.4		5.0	4.0	12.6	14.2		4.4
Material resources	38,856.5	45.9	0.0		3.8	8.5	9.9	19.9		12.0
Non renewable elements	0.4	81.9	0.0		5.7	0.6	0.9	3.1		7.8
Non renewable resources	20.6	31.8	0.1		8.1	5.6	11.0	19.4		24.1
Renewable resources	38,835.6	45.9	0.0		3.8	8.5	9.9	19.9		12.0
Water	36,129.8	49.3	0.0		4.0	9.1	10.7	21.3		5.7
Other renewable resources	2,705.8	0.8	0.0		0.8	0.3	0.5	1.5		96.1
Deposited goods	13.5	25.3	0.1		7.7	6.8	7.6	19.2		33.4
Emissions to air	2,96.0	29.9	0.1		5.4	11.2	18.2	27.2		8.1
Emissions to fresh water	35,861	47.9	0.0		1.6	8.8	10.6	18.1		12.9
Analytical measures to fresh water	0.2	39.3	0.0		0.5	0.9	1.1	2.2		56.0
ecoinvent long-term to fresh water	0.4	100.0	0.0		0.0	0.0	0.0	0.0		0.0
Inorganic emissions to fresh water	0.3	73.1	0.2		0.9	1.6	2.1	4.0		18.0
Other emissions to fresh water	35,554.3	47.9	0.0		1.6	8.9	10.7	18.2		12.8
Radioactive emissions to fresh water	305.8	50.3	0.3		2.1	4.2	5.0	10.2		28.0
Emissions to sea water	58.2	21.0	0.0		11.5	5.3	23.4	26.9		11.8
Emission (total)	72,089.7	47.8	0.0		1.7	8.8	10.7	18.2		12.9



Energy consumption for the whole life cycle of BC production. Values are based on the functional unit of 1 kg BC dry basis).

Environmental impacts of life cycle of BC using ReCiPe 2016 Midpoint (H) for 1 kg of dried BC, blue bars represent positive values while red bars represent negative values.

Midpoint Impact category	Units	Total	Production of materials	Transport of materials	Culture Medium Preparation	Inoculum Propagation	Stactic Culture Fementation	Downstream Process	Wastewater treatment
Climate change, default, excl biogenic carbon	[kg CO2 eq.]	16.774	7.3500	0.3200	1.0300	0.8330	2.6700	2.9400	1.6200
Climate change, incl biogenic carbon	[kg CO2 eq.]	16.729	6.4100	0.3200	1.0300	0.8310	2.6700	2.9400	2.5200
Fine Particulate Matter Formation	[kg PM2.5 eq.]	0.016	0.0123	0.0004	0.0003	0.0003	0.0008	0.0009	0.0006
Fossil depletion	[kg oil eq.]	6.565	3.7850	0.1050	0.3610	0.2340	0.8880	0.9380	0.2560
Freshwater Consumption	[m3]	0.470	0.7740	0.0000	0.8590	0.1170	0.0329	1.1800	-2.5000
Freshwater ecotoxicity	[kg 1,4 DB eq.]	0.086	0.0600	0.0000	0.0002	0.0001	0.0001	0.0003	0.0258
Freshwater Eutrophication	[kg P eq.]	0.004	0.0024	0.0000	0.0000	0.0000	0.0000	0.0000	0.0016
Human toxicity, cancer	[kg 1,4-DB eq.]	0.826	0.6930	0.0000	0.0013	0.0007	0.0013	0.0027	0.1280
Human toxicity, non-cancer	[kg 1,4-DB eq.]	13.765	7.6479	0.0021	0.0464	0.0173	0.0170	0.0808	5.9500
Ionizing Radiation	[Bq C-60 eq. to air]	0.342	0.3009	0.0001	0.0016	0.0025	0.0029	0.0066	0.0268
Land use	[Annual crop eq..y]	0.967	0.6260	0.0000	0.0137	0.0703	0.0842	0.1400	0.0332
Marine ecotoxicity	[kg 1,4-DB eq.]	0.123	0.0856	0.0003	0.0003	0.0002	0.0002	0.0006	0.0353
Marine Eutrophication	[kg N eq.]	0.004	0.0010	0.0000	0.0000	0.0000	0.0000	0.0001	0.0030
Metal depletion	[kg Cu eq.]	0.113	0.0639	0.0000	0.0240	0.0032	0.0012	0.0276	-0.0070
Photochemical Ozone Formation, Ecosystems	[kg NOx eq.]	0.030	0.0175	0.0024	0.0010	0.0011	0.0028	0.0033	0.0021
Photochemical Ozone Formation, Human Health	[kg NOx eq.]	0.029	0.0163	0.0024	0.0010	0.0011	0.0028	0.0032	0.0021
Stratospheric Ozone Depletion	[kg CFC-11 eq.]	0.000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Terrestrial Acidification	[kg SO2 eq.]	0.043	0.0323	0.0010	0.0008	0.0010	0.0027	0.0028	0.0020
Terrestrial Ecotoxicity	[kg 1,4-DB eq.]	15.625	14.2683	0.0317	0.1800	0.1530	0.1870	0.3660	0.3950

Culture medium:

Corn syrup production,
Disodium phosphate,

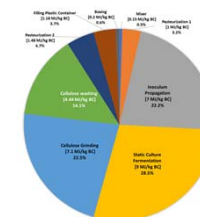
Washing BC:

Sodium hydroxide,

Plastic bags & Carton
production:

Polyethylene,
Carton production

Fermentation process: Energy consumption



Funcarb group

FUNctional andCARBohydrates
Nanobiotechnology



Miguel Gama

Associate Professor with
Habilitation
fmgama@deb.uminho.pt



Fernando Dourado

Assistant Researcher
fdourado@deb.uminho.pt



Universidade do Minho SPINOFF



SATISFIBRE
HYDROFIBRE SOLUTIONS

$\beta(1-4)$ -linked D-glucose units

2013



LABORATORY

Optimization of Bacterial Cellulose fermentation;

Proof of concept: food & cosmetic formulations containing BC

2014 – 2018



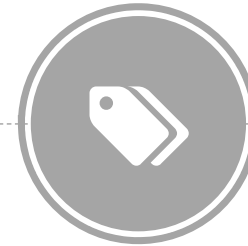
PILOT

Scale-up of BC fermentation;

Food product formulations containing BC

Cosmetic formulations containing BC

R&D



PROJECT

Design of the industrial BC production facility

Regulatory approval of BC as a novel food ingredient for human consumption in EU

Regulatory assesment of BC for cosmetic applications in EU

2020



FUTURE DIRECTIONS

Exploring several paths for the comercial exploitation of the BC production technology & products



THE
NAVIGATOR
COMPANY

Navigator Products & Technologies, S.A.

Intellectual property

inpi instituto nacional
da propriedade industrial

DESCRIPTION
BACTERIAL CELLULOSE COMPOSITES, METHODS AND USES THEREOF

DESCRIPTION
BACTERIAL CELLULOSE FORMULATIONS, METHODS AND USES
THEREOF





BUILD - Bacterial cellulose leather

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FCT FACULDADE DE CIÊNCIAS E TECNOLOGIA UNIVERSIDADE NOVA DE LISBOA



Bacterial Cellulose

Inpactus - Innovative Products and Technologies from Eucalyptus

THE NAVIGATOR COMPANY



KETI Korea Electronics Technology Institute

CRESYN

