Tailoring the performance of commercial cellulolytic cocktails towards the production prebiotic cello-oligosaccharides from waste forest biomass

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Supplementary Material

Table S1. Hydrolysis yields after 24 and 48 h of hydrolysis. Cellobiose (CB) and glucose (Glu) production is expressed in % w/w cellulose conversion. CB: Glu and total mg CB per gram of substrate are also described.

	24 h				48 h			
Conduritol B epoxide (mM)	**CB (% w/w)**	**Glu (% w/w)**	**CB: Glu**	**mg CB/g substrate**	**CB (% w/w)**	**Glu (% w/w)**	**CB: Glu**	**mg CB/g substrate**
0	6.6 ± 0.7	10.8 ± 0.3	0.6	*57.1*	8.0 ± 0.1	16.2 ± 0.4	0.5	*69.6*
0.49	18.4 ± 0.2	9.3 ± 0.5	2.0	*159.7*	20.3 ± 0.4	9.7 ± 0.5	2.1	*175.7*
0.99	23.6 ± 0.1	7.2 ± 0.1	3.3	*204.7*	24.3 ± 0.4	7.3 ± 0.1	3.3	*210.2*
1.98	19.7 ± 0.7	4.3 ± 0.1	4.5	*170.4*	20.9 ± 0.0	4.6 ± 0.1	4.5	*181*
2.96	20.9 ± 0.3	3.7 ± 0.3	5.7	*180.8*	21.7 ± 0.2	3.9 ± 0.0	5.6	*187.7*
3.95	17.7 ± 0.1	2.7 ± 0.0	6.7	*153.4*	19.5 ± 0.17	3.0 ± 0.1	6.5	*168.5*
4.94	16.0 ± 0.5	2.2 ± 0.3	7.1	*138.4*	17.9 ± 0.0	2.5 ± 0.0	7.1	*154.8*

Table S2. Effect of pH and enzyme loading on the % w/w cellulose conversion into cellobiose (CB) and glucose (Glu). CB: Glu and total mg CB per gram of substrate are also described.

Enzyme loading	Incubation time	pH	CB	Glu	CB: Glu	mg CB/g substrate
25 mg/g substrate	24 h	**4**	13.7 ± 1.2	2.6 ± 0.1	5.3	*119.1*
		5	17.6 ± 1.7	3.1 ± 0.3	5.7	*152.8*
		6	18.5 ± 0.7	2.6 ± 0.0	7.2	*160.6*
		7	12.1 ± 13	0.6 ± 0.2	21.6	*104.4*
	48 h	**4**	15.7 ± 0.2	4.1 ± 0.1	3.9	*136*
		5	19.5 ± 0.6	2.4 ± 0.1	8.3	*168.9*
		6	19.9 ± 1.1	3.5 ± 0.0	5.8	*172.5*
		7	13.1 ± 1.8	0.6 ± 0.2	22.5	*113.2*
50 mg/g substrate	24 h	**4**	15.7 ± 0.8	3.7 ± 0.0	4.2	*135.7*
		5	19.0 ± 0.6	4.4 ± 0.1	4.3	*165*
		6	20.3 ± 0.8	3.5 ± 0.1	5.9	*175.5*
		7	15.1 ± 0.5	0	-	*130.8*
	48 h	**4**	18.6 ± 0.4	5.9 ± 0.0	3.1	*161.1*
		5	20.5 ± 0.3	6.1 ± 0.0	3.4	*177.3*
		6	21.8 ± 0.4	4.8 ± 0.0	4.6	*189.1*
		7	15.4 ± 0.4	0	-	*133*

Table S3. Cellulose conversion (% w/w) to CB and Glu for different hydrolysis time and evaluation of buffer exchange after 72 and 96 h of hydrolysis. CB: Glu and total mg CB per gram of substrate are also described.

Incubation time/ condition	CB (% w/w)	Glu (% w/w)	CB: Glu	mg CB/ g substrate
24 h	12.6 ± 0.3	0.6 ± 0.0	21.8	*109.5*
48 h	13.2 ± 0.2	0.5 ± 0.0	27.9	*114.3*
72 h	14.2 ± 0.2	0.6 ± 0.0	23	*122.8*
72 h with exchange	14.3 ± 0.4	0.9 ± 0.0	15.9	*124.0*
96 h	15.1 ± 0.2	0.8 ± 0.0	19.4	*131.1*
96 h with exchange	14.9 ± 0.4	0.6 ± 0.1	23.6	*129.5*

Table S4. Effect of addition of various concentrations of conduritol-B-epoxide at pH 7.0 on the % w/w cellulose conversion into cellobiose (CB) and glucose (Glu). CB: Glu and total mg CB per gram of substrate are also described.

conduritol epoxide (mM)	CB	Glu	CB: Glu ratio	mg CB/g substrate
0	12.6 ± 0.3	4.4 ± 0.0	2.9	*109.2*
0.99	15.7 ± 0.6	0.8 ± 0.0	19.7	*135.9*
1.98	16.3 ± 0.6	0.6 ± 0.0	28.3	*141.7*
3.95	14.9 ± 0.1	0.7 ± 0.1	21.8	*129.7*

Table S5. Effect of buffer exchange and/or supplementation with additional enzyme loading or conduritol-B-epoxide on the cellulose conversion (% w/w) to CB and Glu. The ratio CB: Glu is also described for 8, 24 and 48 h of hydrolysis.

Supplementation conditions	Incubation time	CB	Glu	CB: Glu	mg CB/ g substrate
Buffer	8 h	9.9 ± 0.9	0.6 ± 0.3	17.7	*98.9*
	24 h	13.2 ± 0.4	0.7 ± 0.1	19.4	*132.2*
	48 h	13.8 ± 0.0	0.7 ± 0.2	18.6	*137.6*
Buffer, enzyme	8 h	9.4 ± 0.1	0.6 ± 0.0	16.8	*94.1*
	24 h	15.5 ± 0.2	0.4 ± 0.1	38.6	*154.5*
	48 h	16.3 ± 0.4	0.4 ± 0.0	39.8	*163.1*
Buffer, conduritol	8 h	9.9 ± 0.3	0.6 ± 0.1	16.7	*98.6*
	24 h	13.7 ± 0.6	0.6 ± 0.0	21.5	*137.4*
	48 h	14.1 ± 0.5	0.7 ± 0.1	20.4	*140.6*
Buffer, enzyme, conduritol	8 h	9.5 ± 0.2	0.6 ± 0.0	15.3	*94.7*
	24 h	16.4 ± 0.2	0.4 ± 0.0	45.6	*164.3*
	48 h	17.2 ± 0.2	0.4 ± 0.0	44.2	*172.2*

Table S6. Hydrolysis yields from birch and spruce substrates, described as % w/w cellulose conversion into cellobiose and glucose of at pH 7.0, upon the addition of 1.98 mM conduritol-B-epoxide, at an enzyme loading of 25 mg/g of substrate, with buffer exchange at 8 and 24 h. CB: Glu and total mg CB per gram of substrate are also described.

Substrate	Incubation time	CB	Glu	CB: Glu
B1	8 h	10.6 ± 0.3	0.6 ± 0.0	16.5
	24 h	12.2 ± 0.2	0.8 ± 0.0	15.8
	48 h	13.0 ± 0.0	0.6 ± 0.0	22.5
B2	8 h	6.4 ± 0.3	-	-
	24 h	8.3 ± 0.3	-	-
	48 h	9.78 ± 0.2	0.3 ± 0.0	37.5
S1	8 h	5.9 ± 0.4	0.7 ± 0.1	8.1
	24 h	7.3 ± 0.3	0.8 ± 0.2	8.9
	48 h	9.5 ± 0.0	1 ± 0.1	9.5
S2	8 h	3.2 ± 0.5	-	-
	24 h	4.3 ± 0.1	-	-
	48 h	5.4 ± 0.1	0.1 ± 0.0	33.9