



Citation for published version:

Hamley-Bennett, C, Lye, GJ & Leak, DJ 2016, 'Selective fractionation of sugar beet pulp for release of fermentation and chemical feedstocks; optimisation of thermo-chemical pre-treatment', *Bioresource Technology*, vol. 209, pp. 259-264. <https://doi.org/10.1016/j.biortech.2016.02.131>

DOI:

[10.1016/j.biortech.2016.02.131](https://doi.org/10.1016/j.biortech.2016.02.131)

Publication date:

2016

Document Version

Peer reviewed version

[Link to publication](#)

Publisher Rights

CC BY-NC-ND

The published version is available via: <http://dx.doi.org/10.1016/j.biortech.2016.02.131>

University of Bath

Alternative formats

If you require this document in an alternative format, please contact:
openaccess@bath.ac.uk

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

Take down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Table 1: Summary of DoE factors and responses in the fractionation of SBP. The total monomeric and oligomeric sugars in the soluble fraction are shown as the percentage of each sugar released from the total available in the starting material. Experiments are shown in standard order.

Exp	Factors		Insoluble fraction recovered (g)	Soluble fraction recovered (ml)	Responses					
	Time (A)	Pressure (B)			% soluble arabinose released		% soluble galacturonic acid released		% soluble glucose released	
	(min)	(Bar)			Monomer	Oligomer	Monomer	Oligomer	Monomer	Oligomer
1	1.00	4.00	9.60	275	0.1%	9.7%	0.0%	38.9%	0.2%	2.1%
2	30.00	4.00	5.04	670	4.8%	74.9%	0.0%	46.9%	1.5%	2.3%
3	1.00	8.00	8.61	435	0.6%	33.0%	0.0%	47.6%	0.4%	3.2%
4	30.00	8.00	4.00	690	36.8%	59.7%	8.1%	10.7%	2.3%	7.8%
5	-5.00*	6.00	50.00	-	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
6	36.00	6.00	4.76	585	19.1%	61.9%	6.4%	10.3%	2.1%	4.3%
7	15.50	3.00	9.13	295	0.5%	27.6%	0.0%	46.3%	0.5%	2.2%
8	15.50	9.00	4.45	570	12.3%	69.6%	1.8%	23.8%	2.0%	2.7%
9	15.50	6.00	5.04	470	5.2%	59.6%	0.0%	29.7%	1.4%	1.7%
10	15.50	6.00	4.93	560	5.1%	73.8%	0.0%	44.6%	1.7%	2.1%
11	15.50	6.00	5.15	410	7.5%	76.4%	0.0%	35.1%	2.0%	4.6%

*This value was adjusted to zero minutes for the purpose of the experiment.

Table 2: Factors influencing arabinose release during pre-treatment. ANOVA table for Response Surface Reduced Quadratic model based on the response data reported in Table 1.

Source	Sum of Squares	Df	Mean Square	F Value	p-value Prob > F
Model	10921.75	4	2730.44	31.33	0.0001
<i>A-Time</i>	<i>9416.14</i>	<i>1</i>	<i>9416.14</i>	<i>108.05</i>	<i>< 0.0001</i>
<i>B-Pressure</i>	<i>1613.48</i>	<i>1</i>	<i>1613.48</i>	<i>18.52</i>	<i>0.0036</i>
<i>A^2</i>	<i>1386.47</i>	<i>1</i>	<i>1386.47</i>	<i>15.91</i>	<i>0.0053</i>
<i>B^2</i>	<i>331.45</i>	<i>1</i>	<i>331.45</i>	<i>3.80</i>	<i>0.0921</i>
Residual	610.00	7	87.14		
<i>Lack of Fit</i>	<i>412.82</i>	<i>5</i>	<i>82.56</i>	<i>0.84</i>	<i>0.6232</i>
<i>Pure Error</i>	<i>197.18</i>	<i>2</i>	<i>98.59</i>		
Cor Total	11531.75	11			

Table 3: Factors influencing galacturonic acid release during pre-treatment. ANOVA table for Response Surface Reduced Quadratic model based on the response data reported in Table 1.

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	1341.29	2	670.65	4.57	0.0426
<i>AB</i>	<i>1023.36</i>	<i>1</i>	<i>1023.36</i>	<i>6.98</i>	<i>0.0268</i>
<i>A^2</i>	<i>685.51</i>	<i>1</i>	<i>685.51</i>	<i>4.68</i>	<i>0.0589</i>
Residual	1319.61	9	146.62		
<i>Lack of Fit</i>	<i>1201.58</i>	<i>7</i>	<i>171.65</i>	<i>2.91</i>	<i>0.2796</i>
<i>Pure Error</i>	<i>118.03</i>	<i>2</i>	<i>59.01</i>		
Cor Total	2660.91	11			

Table 4: Factors influencing glucose release during pre-treatment. ANOVA table for Response Surface Reduced Quadratic model based on the response data reported in Table 1.

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	56.07	3	18.69	10.01	0.0044
<i>A-Time</i>	<i>39.93</i>	<i>1</i>	<i>39.93</i>	<i>21.38</i>	<i>0.0017</i>
<i>B-Pressure</i>	<i>14.78</i>	<i>1</i>	<i>14.78</i>	<i>7.91</i>	<i>0.0227</i>
<i>AB</i>	<i>9.42</i>	<i>1</i>	<i>9.42</i>	<i>5.05</i>	<i>0.0549</i>
Residual	14.94	8	1.87		
<i>Lack of Fit</i>	<i>7.82</i>	<i>6</i>	<i>1.30</i>	<i>0.37</i>	<i>0.8565</i>
<i>Pure Error</i>	<i>7.12</i>	<i>2</i>	<i>3.56</i>		
Cor Total	71.00	11			

Table 5: Summary of statistical analysis performed on the models for arabinose, galacturonic and glucose release.

	Arabinose	Galacturonic Acid	Glucose
Std. Dev.	9.34	12.11	1.37
Mean	59.06	34.53	4.21
C.V. %	15.81	35.07	32.47
PRESS	2038.18	2702.79	35.24
R-Squared	0.9471	0.5041	0.7896
Adj R-Squared	0.9169	0.3939	0.7107
Pred R-Squared	0.8233	-0.0157	0.5037
Adeq Precision	16.265	4.207	9.689

Fig.1 Legend

Surface response diagram showing percentage of arabinose solubilised as a function of pressure (Bar) and time (min). The optimum conditions and % solubilisation of arabinose and glucose as predicted by the model (5.3Bar for 24.4min) are highlighted in bold. The red data points represent actual experimental design points. Response surface plotted according to Equation 1. Optimisation criteria were to maximise the yield of arabinose (>70%) and minimise the yield of glucose (<6%) in the soluble fraction, thereby retaining the cellulose in the insoluble fraction.

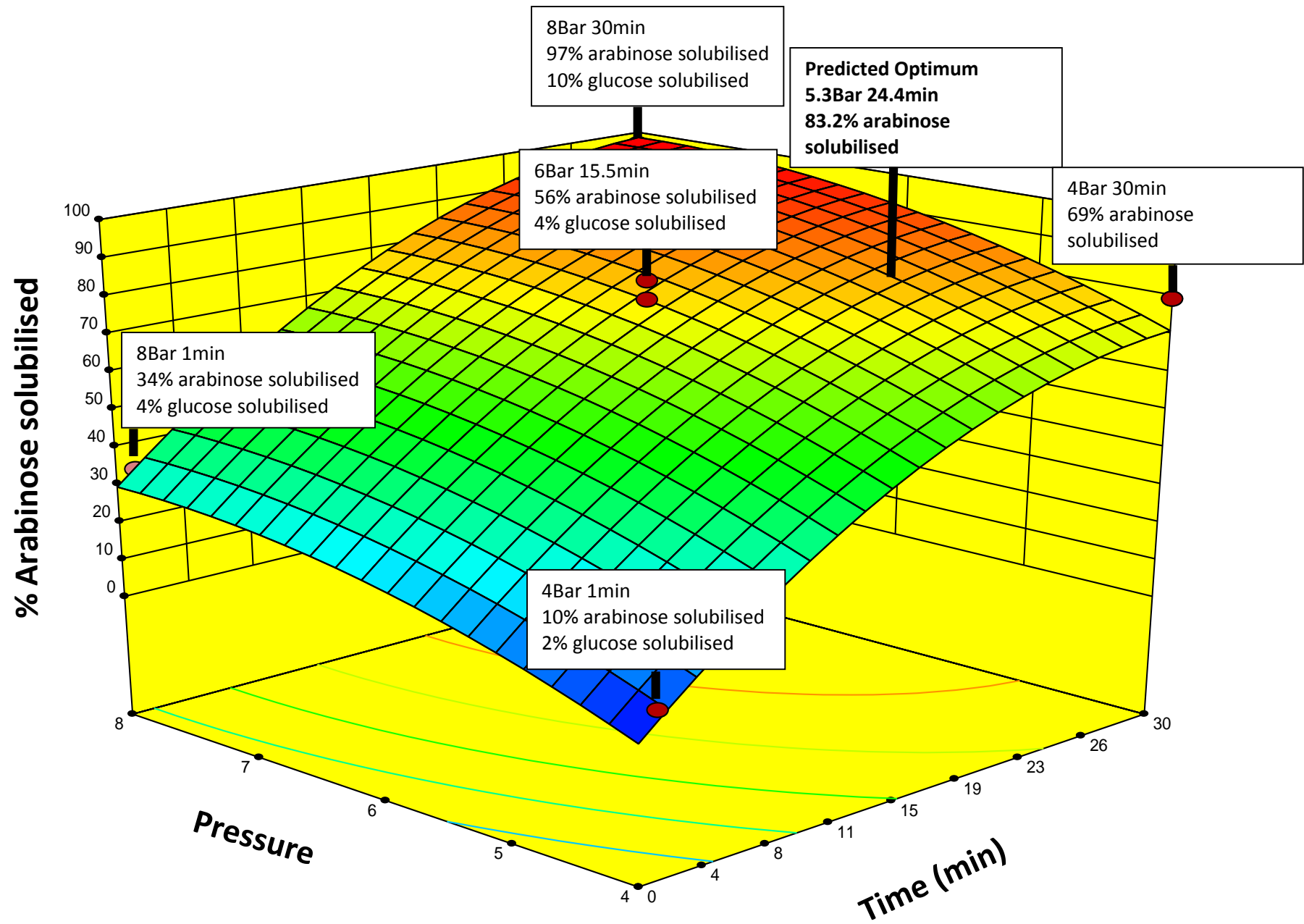


Figure 2: Analysis of SBP fractions produced under predicted optimal conditions

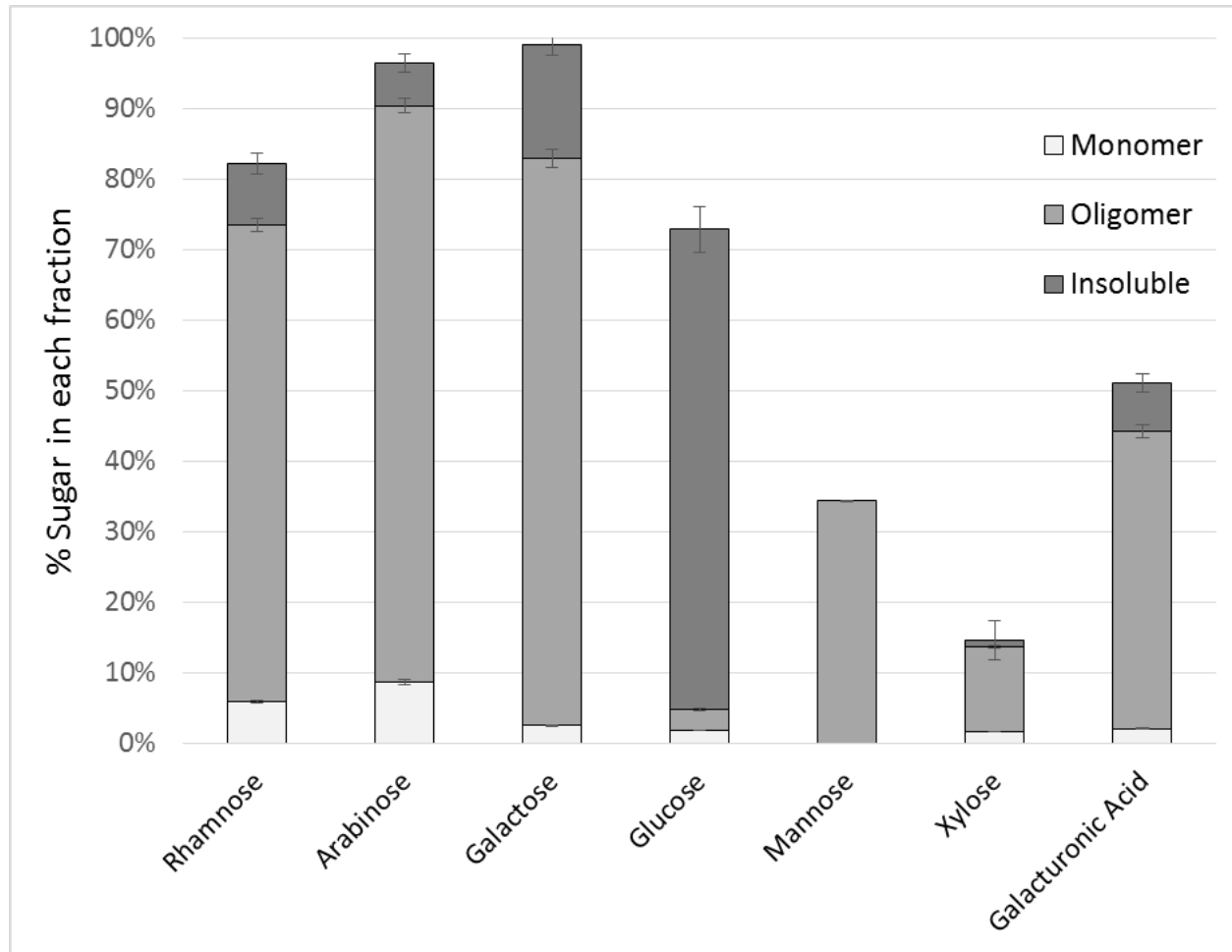


Table 6: Optical density of yeast cultures after 24hrs growth measured at 600nm. The residual carbohydrate and ethanol concentrations were determined by ion chromatography and HPLC as described in section 2.3.2. The conversion yield of glucose to ethanol is also shown.

	OD₆₀₀	Glucose consumed (mg)	Ethanol produced (mg)	Conversion g/g
SBP	5.28 (±0.10)	155.87 (±1.0)	75.26 (±0.12)	0.48 (±0.008)
YNBG	5.98 (±0.12)	134.39 (±4.0)	51.43 (±0.18)	0.38 (±0.013)

Table 7: Results of protein extraction performed on samples of raw untreated SBP, the insoluble residue after optimised steam pre-treatment and the residue after cellulase hydrolysis as described in section 2.4. Steam pre-treatment results in a loss of around 1% w/w protein, but the final insoluble product from cellulase hydrolysis is enriched in protein as the cellulose fraction is removed.

Sample	Dry weight of sample (mg)	ug protein in sample	%w/w
Washed SBP	106.9	8.69	8%
Steam pre-treated SBP	103.6	7.55	7%
Cellulase hydrolysed	119.0	18.02	15%