

Article

Screening the Lipid Production Potential of Oleaginous Yeast *Yarrowia lipolytica* under Wood Hydrolysates

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Supplementary Materials

Table S1 Factors and responses

Factor 1	Factor 2	Factor 3	Response 1	Response 2	Response 3	Response 4	Response 5	Response 6
A:Acid	B:Solid	C:Time	Xylose	Arabinose	Mannose	Glucose	Galactose	TSR
%	g	min	%	%	%	%	%	%
72	88	10	85.884	81.9984	60.6902	59.6278	68.2019	66.5
72	88	20	87.0735	83.441	56.7288	59.225	70.6048	66.8
72	88	30	86.4911	82.5009	60.8039	62.2934	71.8252	69.1
72	88	45	70.0256	74.7077	55.0523	63.5906	71.3724	67.7
72	88	60	69.6242	70.5006	55.6386	65.4579	70.6235	68.6
72	146	10	75.4458	65.2008	48.718	58.3799	53.4173	61
72	146	20	77.7617	68.3713	47.6461	59.6632	56.5751	62.8
72	146	30	78.1586	73.6046	55.9853	61.1811	59.0493	64.6
72	146	45	77.8036	76.5299	53.3468	62.4374	57.6254	65.3
72	146	60	75.6279	74.1501	45.9938	62.4786	56.2926	64.7
75	88	10	96.5034	88.732	96.75	64.738	84.2735	74.4381
75	88	20	93.098	89.6	67.3911	62.6105	82.6347	72.3235
75	88	30	90.5149	93.1807	72.7463	67.5097	85.2091	76.0518
75	88	45	73.2768	71.1189	49.9432	71.7211	73.7217	73.9168
75	88	60	69.9611	71.3789	52.1395	74.7427	76.0392	76.0791
75	146	10	78.7904	69.3231	23.5408	74.9463	59.069	73.8568
75	146	20	81.5814	69.502	33.5582	76.0427	61.079	75.2972
75	146	30	82.5658	91.1132	48.6011	76.4729	71.8114	78.7132
75	146	45	81.3606	78.389	46.6263	76.7677	74.3835	78.6793
75	146	60	74.2808	78.3503	41.282	75.5563	70.0463	76.2824
75	234	10	89.4317	92.628	66.0454	54.283	63.0235	62.7141
75	234	20	71.1307	74.2994	28.0973	54.8666	49.6821	57.7945
75	234	30	78.265	77.21	31.4212	51.4776	61.4067	58.5614
75	234	45	73.9466	73.4777	42.5779	48.1394	51.1465	53.7445
75	234	60	65.0747	68.4976	29.2905	50.9797	49.5084	54.1962
80	88	10	93.3538	93.2515	84.2486	80.6987	96.1017	87.4229
80	88	20	91.0399	87.3033	80.0573	80.2229	86.8725	84.8493
80	88	30	79.6808	75.2729	72.9254	85.2301	85.6977	86.2878
80	88	45	70.8597	67.7026	73.2639	77.0323	79.2572	78.1954
80	88	60	49.7646	57.0079	38.3233	75.9678	67.7603	72.5521
80	146	10	78.9481	82.3753	49.3044	76.1613	68.5758	77.1105
80	146	20	73.5335	75.4874	55.4834	76.143	70.2756	76.5312
80	146	30	74.8775	70.6823	35.727	76.7008	69.6922	76.7028
80	146	45	48.5896	48.0591	19.571	69.9469	56.8263	65.8342
80	146	60	45.8856	49.8942	28.8059	69.3566	60.7182	65.966
80	234	10	92.8574	90.1176	55.7727	75.9209	71.9542	79.4153
80	234	20	63.9632	62.1653	27.9216	70.1997	61.0673	69.088
80	234	30	73.295	68.2389	37.5031	71.7769	60.7528	71.3872
80	234	45	53.0129	56.0728	25.1143	60.8079	56.0266	60.2795
80	234	60	46.3612	50.1719	36.6855	63.8501	56.7875	61.538

Table S2 ANOVA for xylose, arabinose, glucose, and galactose

ANOVA for Xylose						ANOVA for Arabinose				
Source	Sum of Squares	df	Mean Square	F value	P value	Sum of Squares	df	Mean Square	F value	P value
Model	5474.43	9	608.27	16.29	< 0.0001*	4022.82	9	446.98	9.13	< 0.0001*
A-Acid	245.86	1	245.86	6.58	0.0155	321.56	1	321.56	6.57	0.0157
B-Solid	0.2614	1	0.2614	0.0070	0.9339	0.3454	1	0.3454	0.0071	0.9336
C-Time	745.79	1	745.79	19.97	0.0001	900.96	1	900.96	18.39	0.0002
AB	19.69	1	19.69	0.5271	0.4734	29.01	1	29.01	0.5923	0.4476
AC	882.38	1	882.38	23.63	< 0.0001	1032.62	1	1032.62	21.08	< 0.0001
BC	61.38	1	61.38	1.64	0.2097	26.94	1	26.94	0.5499	0.4641
A ²	221.81	1	221.81	5.94	0.0210	287.17	1	287.17	5.86	0.0217
B ²	109.76	1	109.76	2.94	0.0968	220.97	1	220.97	4.51	0.0420
C ²	15.56	1	15.56	0.4165	0.5236	0.8471	1	0.8471	0.0173	0.8962
Residual	1120.42	30	37.35			1469.36	30	48.98		
Cor Total	6594.86	39				5492.19	39			

ANOVA for Glucose						ANOVA for Galactose				
Source	Sum of Squares	df	Mean Square	F value	P value	Sum of Squares	df	Mean Square	F value	P value
Model	2755.66	9	306.18	16.18	< 0.0001*	4201.73	9	466.86	18.94	< 0.0001*
A-Acid	93.37	1	93.37	4.93	0.0340	230.87	1	230.87	9.37	0.0046
B-Solid	17.47	1	17.47	0.9229	0.3444	101.84	1	101.84	4.13	0.0510
C-Time	152.25	1	152.25	8.05	0.0081	321.58	1	321.58	13.04	0.0011
AB	69.30	1	69.30	3.66	0.0653	12.71	1	12.71	0.5155	0.4783
AC	131.31	1	131.31	6.94	0.0132	340.12	1	340.12	13.80	0.0008
BC	78.28	1	78.28	4.14	0.0509	13.04	1	13.04	0.5289	0.4727
A ²	81.66	1	81.66	4.32	0.0464	206.18	1	206.18	8.36	0.0071
B ²	357.91	1	357.91	18.91	0.0001	244.97	1	244.97	9.94	0.0037
C ²	0.0735	1	0.0735	0.0039	0.9507	8.09	1	8.09	0.3282	0.5710
Residual	567.74	30	18.92			739.57	30	24.65		
Cor Total	3323.41	39				4941.29	39			

* refers to statistical “significant” results.

Table S3 ANOVA for total sugar recovery

Source	Sum of Squares	df	Mean Square	F value	P value	
Model	2273.76	9	252.64	16.78	< 0.0001	significant
A-Acid	142.33	1	142.33	9.45	0.0045	
B-Solid	21.64	1	21.64	1.44	0.2400	
C-Time	257.47	1	257.47	17.10	0.0003	
AB	31.53	1	31.53	2.09	0.1582	
AC	252.79	1	252.79	16.79	0.0003	
BC	18.29	1	18.29	1.22	0.2791	
A ²	125.82	1	125.82	8.36	0.0071	
B ²	66.87	1	66.87	4.44	0.0435	
C ²	1.36	1	1.36	0.0907	0.7654	
Residual	451.67	30	15.06			
Cor Total	2725.44	39				

Standard Xylose

R² = 0.99944

Standard Arabinose

R² = 0.99888

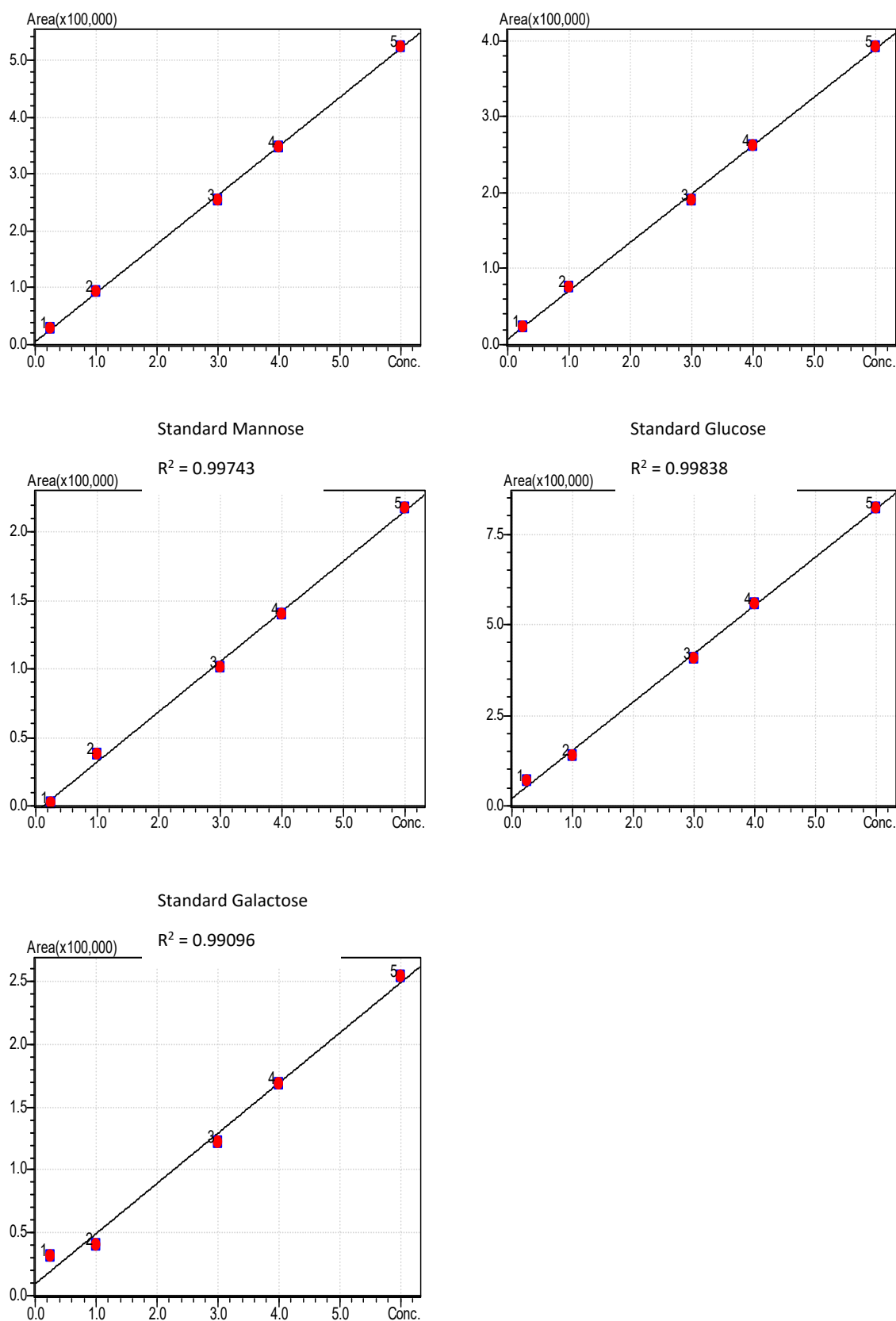


Figure S1 Five-point calibration curve of standard sugars in HPLC by 0.25, 1, 3,4,6 mg/mL

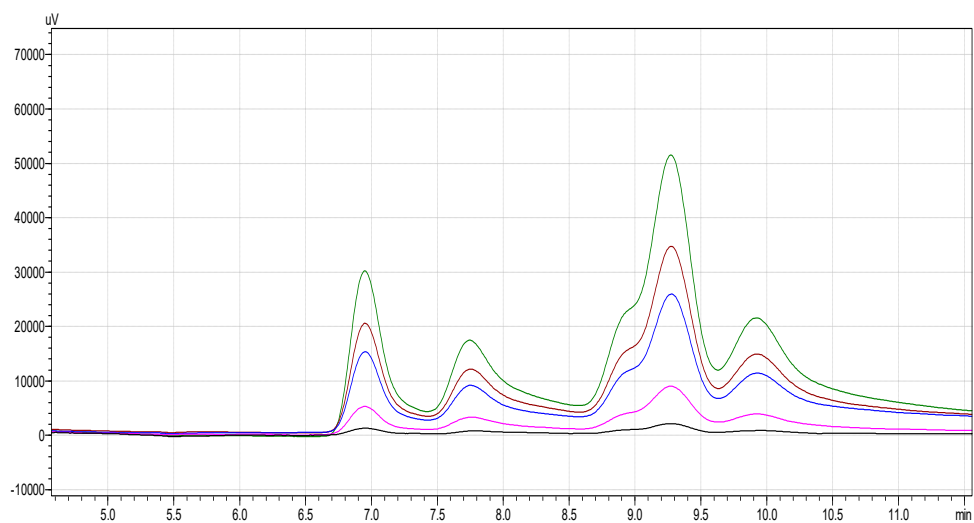


Figure S2 HPLC spectra of the mixed standard sugars solution for calibration curve. Points are 0.25, 1, 3,4,6 mg/mL. Peaks from left: xylose, arabinose, mannose, glucose, galactose

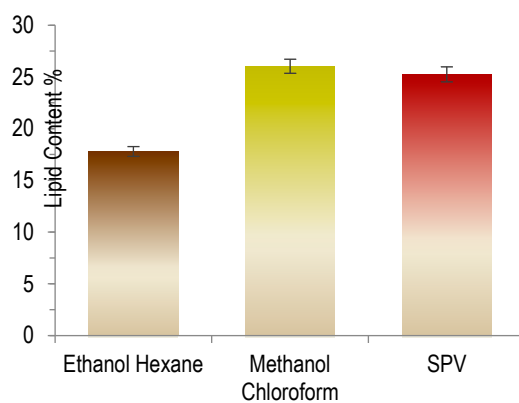


Figure S3 Lipid extraction efficiency from *Y. lipolytica* using different methods: hexane, methanol chloroform, and Sulpho-phospho-vanillin

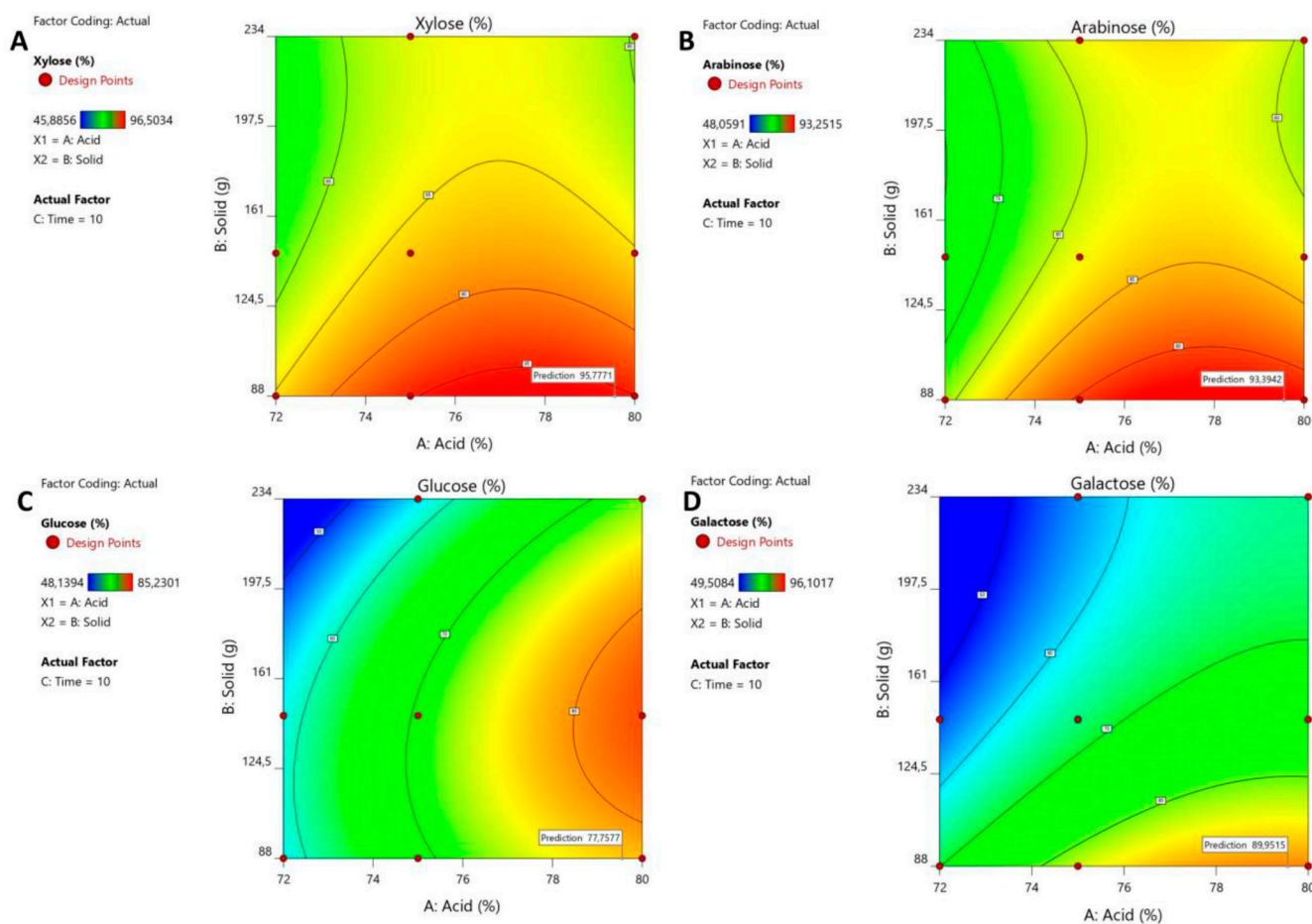


Figure S4 Contour plots indicating the simultaneous maximum recovery of a) xylose b) arabinose c) glucose and d) galactose with respect to acid vs. solid