Electronic Supplementary Information (ESI)

Table S1 Yields of xylitol production in function of different parameters tested for the optimization of Kraft pulp fermentation by cell surface-engineered strain YPH499-XR-XYL-XYN of *S. cerevisiae*. The influence of parameters such as the amount of commercial hemicellulases cocktail ([CHC]; 0 to 0.1 g/g-DW) used during pre-treatment, the amount of substrate (LBKP; 25 to 125 g-DW/L) or the amount of yeast cells (25 to 100 g-WW/L) added for the fermentation, on the yields of xylitol production has been evaluated. Values represent averages ± standard deviation of the results from three independent experiments.

Parameters	Yields	
Amount of CHC (g/g-DW)	Yxylitol/xylose (g/g)	Yxylitol/CHC (g/mg)
0	0.02 ± 0.01	ND
0.01	0.05 ± 0.01	0.04 ± 0.01
0.02	0.27 ± 0.03	0.11 ± 0.02
0.05	0.42 ± 0.04	0.07 ± 0.01
0.1	0.64 ± 0.05	0.05 ± 0.01
Amount of LBKP (g-DW/L)	Yxylitol/xylose (g/g)	Yxylitol/LBKP (g/g-DW/L)
25	0.26 ± 0.04	0.04 ± 0.01
50	0.27 ± 0.03	0.04 ± 0.01
75	0.13 ± 0.03	0.02 ± 0.01
100	0.02 ± 0.01	0.004 ± 0.001
125	0.01 ± 0.002	0.001 ± 0.0005
Amount of yeast cells (g-WW/L)	Yxylitol/xylose (g/g)	Yxylitol/yeast cells (g/g-WW/L)
25	0.09 ± 0.01	0.03 ± 0.01
50	0.19 ± 0.02	0.03 ± 0.01

75	0.24 ± 0.03	0.02 ± 0.005
100	0.27 ± 0.03	0.02 ± 0.01

Table S2 Yields of xylitol production from Kraft pulp by different cell surface-engineered strains YPH499-XR (control), YPH499-XR-XYL-XYN and YPH499-XR-XYLsss-XYNsss of *S. cerevisiae*. Values represent averages ± standard deviation of the results from three independent experiments.

Strains	Yields	
	Yxylitol/xylose (g/g)	Yxylitol/CHC (g/mg)
YPH499-XR	0.05 ± 0.01	0.02 ± 0.01
YPH499-XR-BGL	0.10 ± 0.01	0.04 ± 0.01
YPH499-XR-BGL-XYL-XYN	0.28 ± 0.03	0.11 ± 0.02
YPH499-XR-BGL-XYLsss-XYN	0.30 ± 0.02	0.13 ± 0.01
YPH499-XR-BGL-XYLsss-XYNsss	0.44 ± 0.02	0.18 ± 0.01
YPH499-XR-BGL-XYLsss-XYNsss (scale up)	0.5 ± 0.01	0.2 ± 0.06

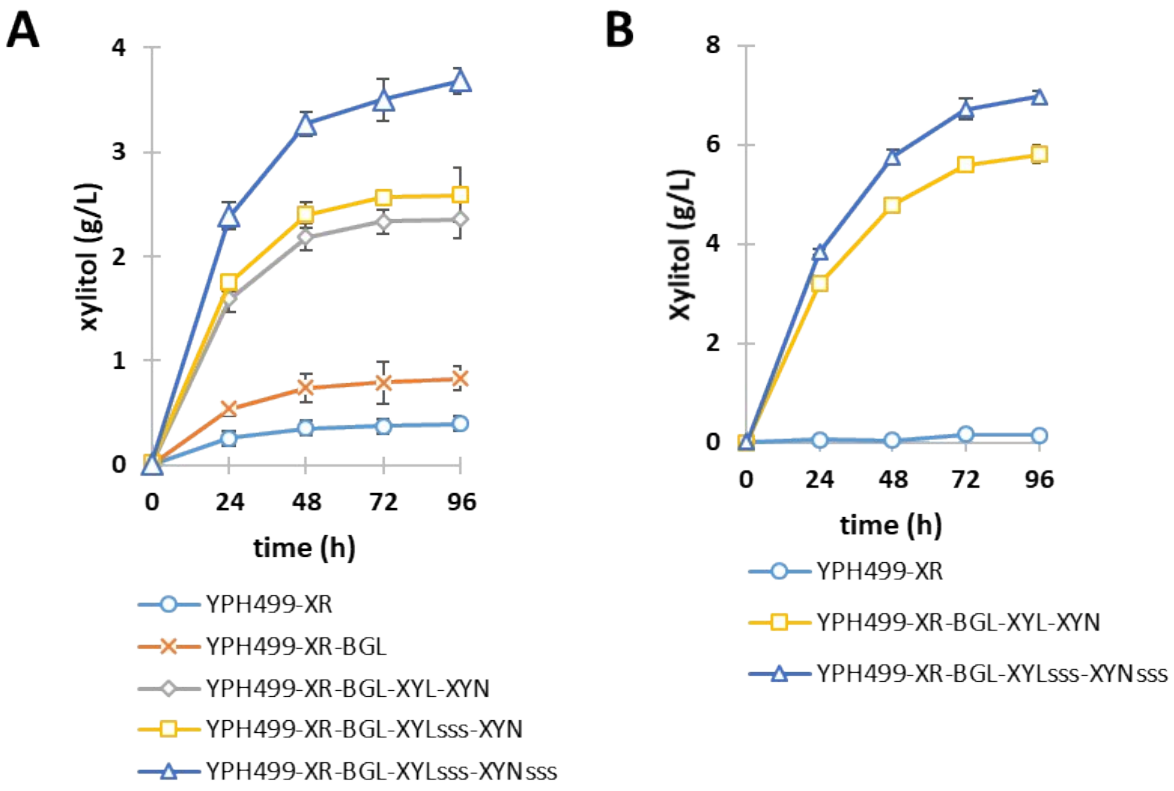

Figure S1 Time course monitoring of xylitol production by different strains and from different substrate. **A**: Fermentation of pretreated LBKP by different cell surface engineered strains of *S. cerevisiae* such as YPH499-XR (negative control), YPH499-XR-BGL, YPH499-XR-BGL-XYL-XYN, YPH499-XR-BGL-XYLsss-XYN and YPH499-XR-BGL-XYLsss-XYNsss. **B:** Fermentation of 50 % rice straw hydrolysate by YPH499-XR (negative control), YPH499-XR-BGL-XYL-XYN, and YPH499-XR-BGL-XYLsss-XYNsss, in absence of CHC in the same conditions than previously described [34].

Table S3 GenBank and/or UniProt accession numbers of the genes/proteins/promoters/signal sequences/anchors that were used for metabolic engineering in this study

Components	Accession No.
Trichoderma reesei β-xylanase (XYN2)	U24191.1
Aspergillus oryzae β-xylosidase A (XylA)	AB013851.1
Aspergillus aculeatus β-glucosidase (BGL1)	D64088.1
Saccharomyces cerevisiae TDH3 promoter	NC_001139.9 (883834..884477, complement)
Saccharomyces cerevisiae SED1 promoter	NC_001136.10 (599993..600792)
Rhizopus oryzae glucoamylase secretion signal sequence	D00049.1 (115..188)
Saccharomyces cerevisiae SED1 secretion signal sequence	NC_001136.10 (600793..600849)
Saccharomyces cerevisiae SAG1 anchor	NC_001142.9 (442909..443871, complement)
Saccharomyces cerevisiae SED1 anchor	NC_001136.10 (600796..601809)
Saccharomyces cerevisiae SAG1 terminator	NC_001142.9 (442464..442899, complement)