Supporting information

Pretreatment with γ-valerolactone/[Mmim]DMP and enzymatic hydrolysis on corncob and its application in immobilized butyric acid fermentation

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Tables

Table S1. Factors and levels of orthogonal experimental design of enzymatic hydrolysis conditions

Level	Enzyme concentration (FPU/g)	Substrate concentration (g/L)	Reaction temperature (℃)	pH	Reaction time (h)
1	45	40	40	4.6	64
2	50	45	45	4.8	68
3	55	50	50	5.0	72
4	60	55	55	5.2	76

21 **Table S2.** Design and results of the orthogonal enzymatic hydrolysis experiments

Sample	Enzyme concentration (FPU/g)	Substrate concentration (g/L)	Reaction temperature (℃)	pH	Reaction time (h)	Glucose yield (%）
1	45	40	40	4.6	64	76.3
2	45	45	45	4.8	68	80.9
3	45	50	50	5.0	72	93.2
4	45	55	55	5.2	76	75.4
5	50	40	45	5.0	76	81.8
6	50	45	40	5.2	72	87.1
7	50	50	55	4.6	68	72.6
8	50	55	50	4.8	64	71.9
9	55	40	50	5.2	68	77.7
10	55	45	55	5.0	64	62.9
11	55	50	40	4.8	76	85.5
12	55	55	45	4.6	72	81.3
13	60	40	55	4.8	72	83.4
14	60	45	50	4.6	76	79.4
15	60	50	45	5.2	64	77.6
16	60	55	40	5.0	68	72.5

K1	81.45	79.80	80.35	77.40	72.18
K2	78.35	77.58	80.40	80.43	75.93
K3	76.85	82.23	80.55	77.60	86.25
K4	78.23	75.28	73.58	79.45	80.53
R	4.60	6.95	7.00	3.03	14.08

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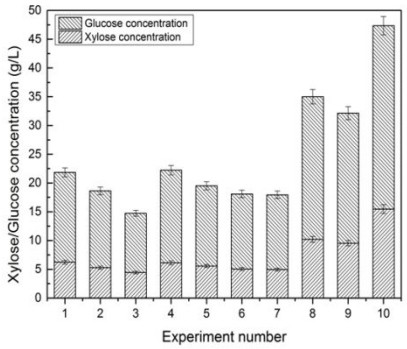
Figures



Number	1	2	3	4	5	6	7	8	9	10
Glucose con. (g/L)	15.6	13.3	10.3	16.1	13.9	13.0	13.0	24.8	22.6	31.9
Glucose Yield (%)	57.8	50.2	39.4	59.5	52.4	49.6	49.4	89.0	82.2	94.9
Xylose con. (g/L)	6.2	5.2	4.4	6.1	5.5	5.0	4.9	10.2	9.5	15.5
Xylose Yield (%)	28.4	24.5	19.3	27.8	25.8	23.6	23.2	44.7	42.7	53.3

Figure S1. Comparison of glucose and xylose yields and concentrations of enzymatic hydrolysis of pretreated substrates under different pretreatment methods. The 1st to 9th experimental conditions were corresponding to the 9 conditions in **Table 2**, and the 10th was the optimal pretreated condition. The yield of glucose and xylose were calculated as Eq. (1) and Eq. (2):

$$Y_1\% = \frac{C \times V \times M_r \times 0.9}{m \times M_0} \times 100 \quad (1)$$

$$Y_2\% = \frac{C \times V \times M_r \times 0.88}{m \times M_0} \times 100 \quad (2)$$

where Y_1 is the yield of glucose; Y_2 is the yield of xylose; C is the obtained glucose/xylose concentration (g/L); V is the volume of enzyme solution (mL); M_r is the total amount of corncob residue recovered after pretreatment; m is the amount of residue used in the enzymatic reaction; M_0 is the total amount of cellulose/xylan in corncob raw material used for the pretreatment; 0.9 is the conversion coefficient between glucose and cellulose; 0.88 is conversion coefficient between xylose and xylan.

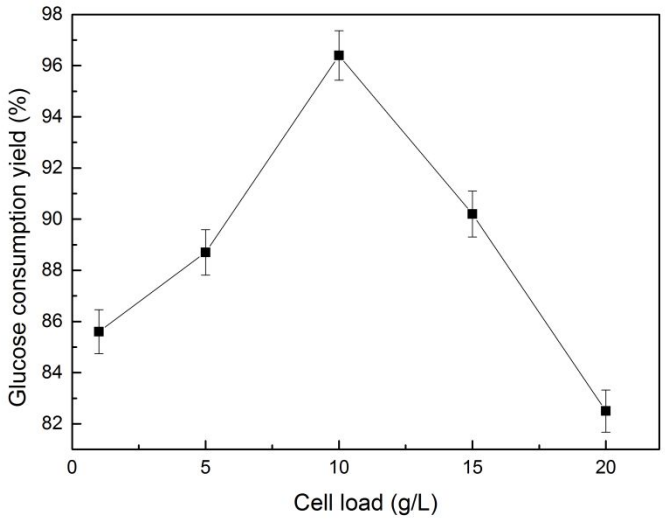


Figure S2. The glucose consumption yield of MCAL beads with five different cell load: 1 g/L, 5 g/L, 10 g/L, 15 g/L and 20 g/L. The glucose consumption yield was calculated as Eq. (3):

$$Y\% = \frac{C_0 - C_{48h}}{C_0} \times 100 \quad (3)$$

where C_0 is the initial glucose concentration in culture; C_{48h} is the glucose concentration in culture after incubation for 48 hours.

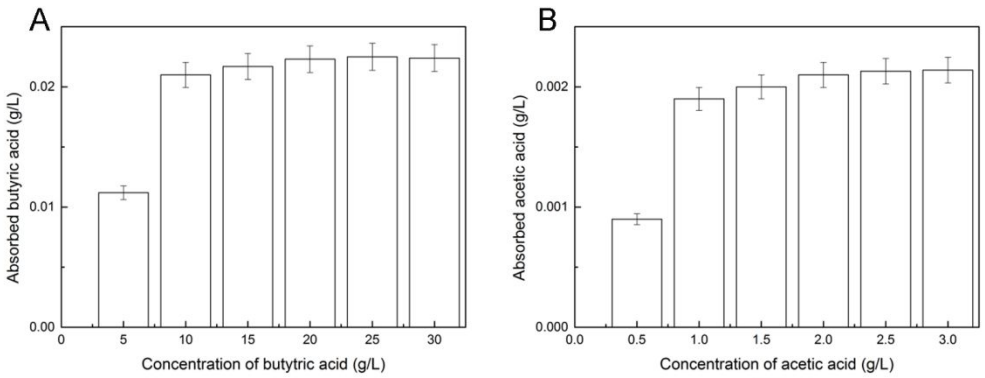


Figure S3. Adsorption isotherms on MCAL beads with different concentration of butyric acid (A) and acetic acid (B). Different amount of butyric acid and acetic acid (e.g., 5, 10, 15, 20, 25, 30 g/L of butyric acid and 0.5, 1, 1.5, 2, 2.5, 3 g/L of acetic acid) were prepared with the addition of 50 g/L MCAL beads in each solution. After the incubation for 48 hours, the final concentration of butyric acid and acetic acid in the supernatant were determined by GC analysis to calculate the amount of butyric acid and acetic acid lost. All assays were done in triplicates and run under parallel conditions.