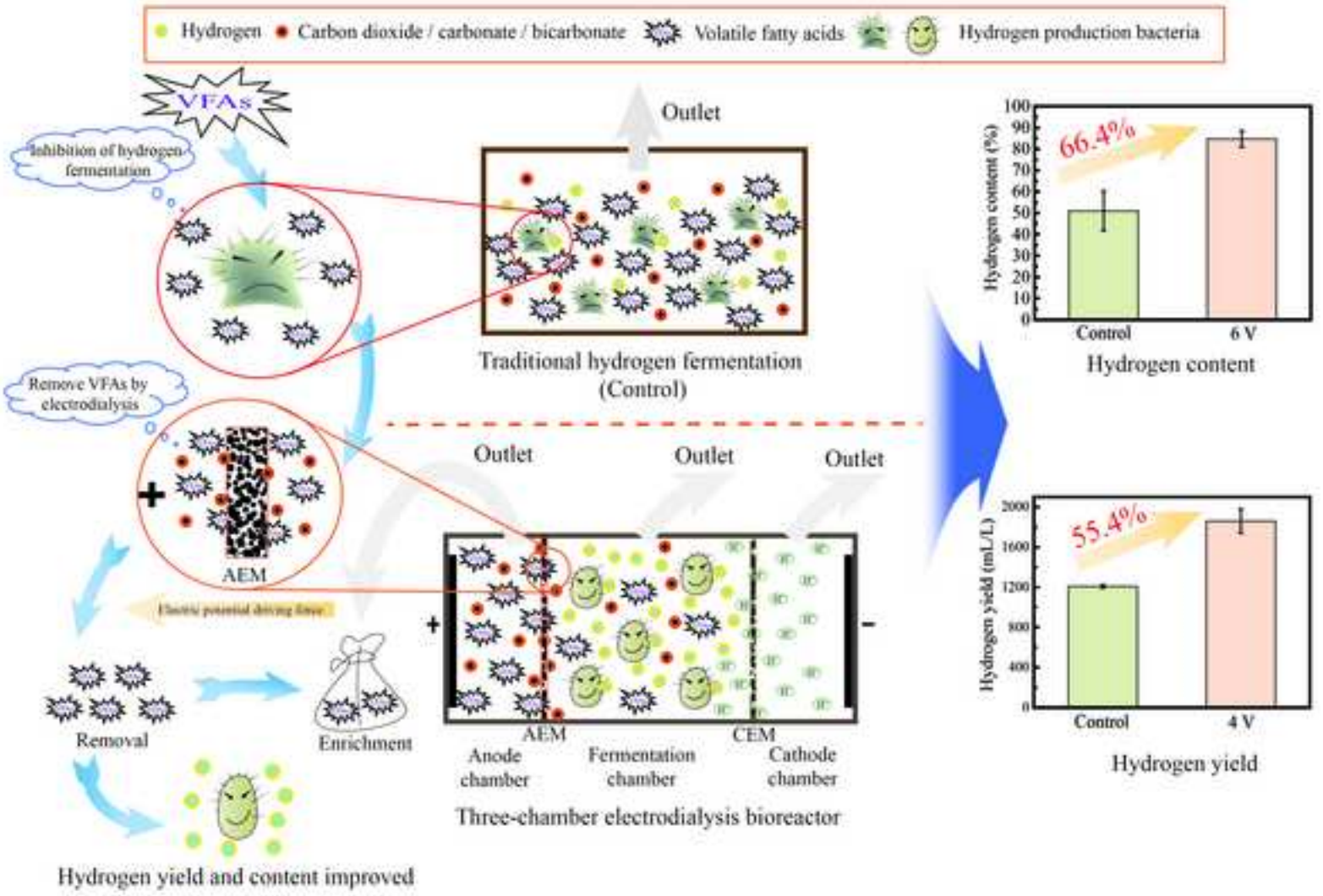
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Graphical Abstract (for review)



***Highlights (for review)**

- A three-chamber electrodialysis reactor was used to enhance hydrogen fermentation.
- VFAs and bicarbonate were effectively removed from fermentation chamber.
- Electrodialysis reactor improved specific H_2 yield up to 55.4% at a voltage of 4 V.
- Electrodialysis reactor enhanced H_2 content up to 66.4% with reduced CO_2 content.

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Enhancing fermentative hydrogen production with the removal of volatile fatty acids by electrodialysis

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Abstract

A three-chamber electrodialysis bioreactor comprising fermentation, cathode and anode chambers was proposed to remove *in situ* volatile fatty acids during hydrogen fermentation. The electrodialysis voltage of 4 V resulted in a volumetric hydrogen productivity of 1878.0 mL/L from the fermentation chamber, which is 55.4% higher than that (1208.5 mL/L) of the control group without voltage applied. Gas production was not observed in the cathode and anode chambers throughout fermentation. By applying different voltages (0-6 V), the hydrogen content accumulated to 54.6%-84.7%, and it exhibited increases of 7.1%-66.4% compared with that of the control. Meanwhile, the maximum concentrations of acetate and butyrate in the fermentation chamber decreased to 10.3 and 13.1 mmol/L at a voltage of 4 V, respectively, which are 68.0% and 62.4% lower than that for the control.

Keywords: Hydrogen production; Fermentation; Volatile fatty acids (VFAs) removal; Electrodialysis; Bioreactor.

1. Introduction

Hydrogen has received increasing attention on account of its clean combustion property and high calorific value by mass (142 MJ/kg) (Noblecourt et al. 2017; Xia et al. 2015). Many conventional methods exist for hydrogen production, such as steam reforming and water electrolysis (Cheng et al. 2012). Nevertheless, such methods may be accompanied by some disadvantages of high temperature, high pressure and high energy consumption (Holladay et al. 2009). In contrast, hydrogen production through dark fermentation of biomass wastes is advantageous due to the low energy demand (Mamimin et al. 2017). Furthermore, a wide range of organic wastes can be degraded by dark fermentation, contributing to significant environmental benefits (Barca et al. 2016). However, the accumulated volatile fatty acids (VFAs), which are generated as a by-product in fermentation, can inhibit the metabolic activity of hydrogen-producing bacteria (HPB) and reduce hydrogen production (Bundhoo & Mohee 2016; Elbeshbishy et al. 2017).

The inhibitory effects of VFAs and some control strategies have been investigated in a number of studies. For example, Zhang et al. studied the inhibitory effects of acetate (0-500 mmol/L) and butyrate (0-250 mmol/L) on dark fermentation using glucose as a substrate and *Clostridium bifermentans 3AT-ma* as the HPB (Zhang et al. 2012). They found that the hydrogen production trended to decrease with increased concentrations of acetate or butyrate. Compared with acetate, butyrate exhibited a more significant inhibition on fermentation. When acetate or butyrate was added to 20 mmol/L, the hydrogen production decreased by 15% and 20%, respectively (Zhang et al. 2012).

Zheng and Yu studied the inhibitory effect of butyrate (4.2-25.1 g/L) on hydrogen production during fermentation (Zheng & Yu 2005). They found that the hydrogen production decreased by 81.7% with 25.1 g/L of butyrate compared with that without the addition of butyrate (Zheng & Yu 2005).

Tang et al. found that the hydrogen production gradually decreased with increasing acetate concentration. When the acetate concentration increased from 0 to 150 mmol/L, the hydrogen production decreased from 2.2 mol H_2/mol glucose to 0.6 mol H_2/mol glucose (Tang et al. 2012). Wang et al. studied the inhibitory effects of ethanol, acetic acid, propionic acid and butyric acid on fermentative hydrogen production at various VFAs concentrations ranging from 0 to 300 mmol/L. They concluded that the hydrogen production and production rate all trended to decrease with increased VFAs concentrations (Wang et al. 2008).

The suitable control of VFAs levels during fermentation can contribute to enhanced hydrogen production. Noblecourt et al. used a submerged membrane anaerobic bioreactor to avoid VFAs accumulation (Noblecourt et al. 2017). The component of VFAs has similar molecular weights as monosaccharides and amino acids, rendering the effective separation of substrates and by-products difficult. As a result, this technology could cause a significant loss of small molecules (such as amino acids and monosaccharides), which are favourable substrates for HPB. There are a few literatures indicate that use of electrodialysis technology can remove VFAs and avoid the loss of small molecules of organic components (Arslan et al. 2017; Jones et al. 2017; Tang et al. 2014). Jones et al. employed conventional electrodialysis to remove and recover VFAs

from model solutions and fermentation broths, resulting in high VFAs removal efficiencies up to 99% at a voltage of 18 V during 60 min of the removal process (Jones et al. 2015). The hydrogen production increased from 0.24 mol H_2/mol hexose to 0.90 mol H_2/mol hexose using conventional electrodialysis (Jones et al. 2017). It should be noted that conventional electrodialysis was used for conducting post-treatment on the fermentation effluent, and the fermentation liquor was subsequently circulated to the fermentation reactor. Such a system includes the fermentation unit and the *ex situ* VFAs removal unit, which cannot directly control the concentration of VFAs in the fermentation reaction zone and may increase the system complexity.

However, previous studies were mainly focused on the batch VFAs removal in a separated electrodialysis reactor. Continuous *in situ* VFAs removal during dark fermentation by electrodialysis has yet been reported. In this paper, a novel three-chamber electrodialysis bioreactor with *in situ* electrodialysis was proposed, for the first time, to simultaneously remove VFAs continuously and to control the concentration of VFAs in fermentation reaction zone directly, thereby enhancing hydrogen fermentation. The aims of this study are to:

- Assess the VFAs removal characteristics using synthetic fermentation liquor.
- Compare the performance of hydrogen fermentation at various voltages.
- Analyse the changes in concentrations of VFAs during hydrogen fermentation.

2. Materials and methods

2.1. Bioreactor

A three-chamber electrodialysis bioreactor was constructed using polymethyl methacrylate. The inner length, width and height of the reactor are 12, 4 and 5 cm, respectively. This bioreactor has a total volume of 240 mL. It comprises an anode chamber (inner length, width and height are 3, 4 and 5 cm; 60 mL), a cathode chamber (inner length, width and height are 3, 4 and 5 cm; 60 mL), and a fermentation chamber (inner length, width and height are 6, 4 and 5 cm; 120 mL) separated by an anion exchange membrane (AEM, 20 cm^2) and a cation exchange membrane (CEM, 20 cm^2). AEM and CEM were purchased from Hangzhou Green Environmental Protection Technology Co. LTD (Hangzhou, China). Graphite electrodes were used as the anode and cathode with a thickness of 2 mm and an area of 20 cm^2 (Beijing Electric Carbon Plant, Beijing, China). A programmable DC power supply (ARRAY 3646A, Bost Electronic Instrument Co. LTD, Shenzhen, China) was used as an external power supply for the electrodes.

2.2. Inoculum and medium

The mixed HPB was isolated and acclimated from the anaerobic digestion sludge derived from a rural digester treating straw and manure in Chongqing, China. The sludge was heated at 100 °C for 30 min to inactivate methanogens and hydrogen consumers, and subsequently enriched three times (3 d each time) to enrich the

spore-forming HPB (Xia et al. 2015). The composition of the acclimation medium was described in a previous study (Cheng et al. 2012).

2.3. Experimental procedures

The three-chamber electrodialysis bioreactor was used to assess the VFAs removal characteristics by using a synthetic VFAs solution with an initial acetic acid concentration of 20 mmol/L or a butyric acid concentration of 20 mmol/L. Eighty millilitres of synthetic VFAs solution was added to the fermentation chamber, and 40 mL of deionized water was added to the anode and cathode chambers to ensure an equal liquid surface level in the fermentation chambers.

For hydrogen fermentation, 8 mL of acclimated HPB and 72 mL of deionized water mixed with 0.8 g of glucose were added to the fermentation chamber. For all reactors, glucose was used as the substrate at a concentration of 10 g/L. Forty millilitres of deionized water was added to the anode and cathode chambers, respectively. The initial pH value of the fermentation chamber was adjusted to 6.5 ± 0.1 using 6 mmol/L HCl or NaOH solution.

The voltage was set at 0-6 V by a programmable DC power supply in the VFAs removal experiments and hydrogen fermentation. A single chamber bioreactor (without voltage and ion-exchange membrane) was used as the control, which was operated with 80 mL of fermentation medium with HPB. A three-chamber electrodialysis bioreactor without substrate addition (no glucose) was used as the hydrogen fermentation blank (as shown in Table 1). All fermentation chambers were purged with nitrogen gas for 5 min

to ensure an anaerobic environment. The headspace of the fermentation chamber was 40 mL. All bioreactors were kept in a thermostat water bath maintained at 35 °C for 96 h. The gas produced was discharged from the headspace of the fermentation chamber and subsequently collected using a graduated container. The gas and liquid samples were collected at a time interval of 12 h for further analysis, and the pH value of the fermentation solution was adjusted to 6.5 ± 0.1 using 6 mmol/L HCl or NaOH at each time interval.

2.4. Analytical methods

The concentrations of acetic acid, propionic acid, butyric acid and hexanoic acid from the fermentation liquor were quantified by a gas chromatograph (Agilent 7890B, USA) equipped with a flame ionization detector (FID) and a polar capillary column (Agilent DB-FFAP Column, 30 m × 0.25 mm × 0.25 μm). The temperatures of the inlet, oven, and FID were 250, 240, and 300 °C, respectively. N_2 was used as a carrier gas at a column flow rate of 1 mL/min. The volume of the liquid samples injected into GC was set as 1 μL with split mode (split ratio 50). The pH value of the fermentation liquid sample was adjusted with HCl to 2 (Cheng et al. 2012). The total inorganic carbon (including bicarbonate, carbonate concentration and dissolved carbon dioxide) in the liquor phase was measured by a Multi N/C 3000 analyser (Analytik Jena AG, Jena Germany). The glucose concentration in the fermentation liquor was determined using the 3,5-dinitrosalicylic acid method (Miller 1959).

The gas composition (H_2 and CO_2) was determined through a gas chromatograph (model Trace 1300; Thermo Scientific) equipped with a micropacked column (ShinCarbon ST Columns, 2 m, OD 1/16", ID 1.0 mm, Mesh 100/120), and N_2 was used as a carrier gas. H_2 and CO_2 concentrations were detected with a thermal conductivity detector (TCD). The temperatures of the inlet, oven, and TCD detector were 120, 110, and 300 °C, respectively. The volume of the gas samples injected into GC was 0.1 mL with split mode (split ratio 29).

The hydrogen production was calculated from the amount and composition of the total volume of hydrogen production in the graduated container at each time interval. Hydrogen content in biogas was expressed as the ratio of the volume of hydrogen to the total volume of hydrogen and carbon dioxide. During fermentation, the gas composition and VFAs were measured every 12 h, and the total inorganic carbon and residual glucose were analysed at the end of the experiment. All of the experimental trials were conducted in triplicate, and the results are expressed as the mean (± standard deviation).

3. Results and discussion

3.1. Removal characteristics using synthetic fermentation liquor

In dark fermentation, hydrogen gas is usually produced through the acetate and butyrate pathways (as shown in Eqs. (1) and (2)) (Barca et al. 2015; Gupta et al. 2014; Xia et al. 2016).

$$C_6H_{12}O_6 + 4H_2O \rightarrow 2CH_3COO^- + 2HCO_3^- + 4H_2 + 4H^+ \quad (1)$$

$$C_6H_{12}O_6 + 2H_2O \rightarrow CH_3CH_2CH_2COO^- + 2HCO_3^- + 2H_2 + 3H^+ \quad (2)$$

Acetate and butyrate have been identified as the major components in fermentation liquor in various dark fermenters. In this study, to assess the VFAs removal characteristics, acetate and butyrate solutions at a typical concentration of 20 mmol/L were used as the synthetic fermentation liquor in the three-chamber electrodialysis bioreactor. Fig. 1a shows the change in acetate concentration during the electrodialysis removal. When the voltage was set as 0 V (without voltage), the concentration of acetate slightly decreased with increasing time. The average removal rate of acetate was 0.09 mol/L/h, and the final concentration was 11.0 mmol/L at 96 h, which corresponds to an overall removal efficiency of 44.8%. In this case, the acetate removal was driven mainly by the diffusion force caused by the different acetate concentrations between the AEM. Such a process is slow, and the concentration diffusion force was reduced by decreasing the acetate concentration gradient.

When the voltage was set as 2 V, the average removal rate of acetate improved to 0.17 mol/L/h. Meanwhile, the final concentration was remarkably reduced to 5.2 mmol/L, corresponding to a removal efficiency of 73.9%. This can be attributed to the enhanced driving force built upon the electric field, in which the process uses an electrical driving force to transfer acetate ions from the fermentation chamber to the anode chamber, thereby improving acetate removal (Mei & Tang 2018; Prochaska et al. 2018).

As the voltage further increased to 4 V and 6 V, the final acetate concentration obtained at 2 V (at 96 h) was achieved at approximately 40 h and 20 h, respectively.

Meanwhile, the average removal rates of acetate were 0.22 and 0.35 mmol/L/h, respectively, at the voltage of 4 V and 6 V. As a result, the acetate removal efficiency increased to 94.7% and 95.6% at 96 h. This was due to the driving forced further enhanced by intensifying the electric field, which significantly improves acetate removal.

The accumulation of acetate in the anode chamber is shown in Fig. 1b. The concentration of acetate gradually increased with time in all groups. The acetate concentrations in the anode chamber achieved with voltage application were even higher than those in the fermentation chamber after removal experiments. This can be explained by the fact that the electrodialysis played a dominant role rather than the concentration diffusion. It should be noted that the volume of fermentation chamber was two times larger than that of the anode chamber, leading to a faster concentration change in the anode chamber compared with the fermentation chamber. Nevertheless, the sum amount of acetate in the fermentation and anode chambers was slightly lower than the initial total amount of acetate (20 mmol/L with 80 mL volume = 1.6 mmol). This can be attributed to the partial adsorption of acetate on the AEM.

The trend of butyrate removal was similar to that of acetate removal (Fig. 2a). The butyrate concentration in the fermentation chamber also decreased with increased time. As the voltage increased from 0 to 6 V, the average removal rate of butyrate gradually increased and achieved 0.10, 0.15, 0.19, 0.26 mmol/L/h, respectively, and the removal efficiency of butyrate was increased from 47.7% to 94.6%. However, the molecular

weight of butyrate is greater than that of acetate, resulting in an increased mass transfer resistance when transferring through the AEM. As a result, the removal rate was lower for butyrate than for acetate. Meanwhile, the butyrate removed from the fermentation chamber was enriched in the anode chamber as shown in Fig. 2b.

3.2. Hydrogen production during fermentation

Dark fermentation in the three-chamber electrodialysis reactor was performed to assess the effects of voltage on hydrogen production performance. When the fermentation was conducted in a single chamber bioreactor without voltage and a membrane (control group), the volumetric hydrogen productivity slowly increased to 61.3 mL/L at the initial fermentation stage (12 h) due to the adaption of HPB (Fig. 3). As the fermentation time increased to 60 h, the volumetric hydrogen productivity rapidly increased to 1116.3 mL/L. This suggests a high activity of HPB metabolism and efficient hydrogen production. When the fermentation time further increased to 96 h, the volumetric hydrogen productivity slowly increased to 1208.5 mL/L (corresponding to a specific hydrogen yield of 1.0 mol H_2/mol glucose). The later stage of hydrogen production was less efficient, which may be explained by the depletion of glucose and the inhibitory effect by the accumulated VFAs (Wang et al. 2008; Zhang et al. 2012; Zheng & Yu 2005).

When the three-chamber reactor was applied without voltage, a slight increase in volumetric hydrogen productivity after 60 h was observed. The accumulation of VFAs in the later stage would be inhibitory for HPB and not advantageous for hydrogen production. This inhibitory effect would be reduced via *in situ* VFAs removal by

diffusion across the AEM, thereby improving the volumetric hydrogen productivity. As a result, the final volumetric hydrogen productivity increased from 1208.5 to 1330.4 mL/L, and the average hydrogen production rate increased from 12.6 to 13.9 mL/L/h (Fig. 3).

When the voltage was set as 2 V, the effect of VFAs removal was enhanced by the electrical driving force. The volumetric hydrogen productivity increased to 1386.4 mL/L, and the hydrogen production rate improved to 14.4 mL/L/h. As the voltage increased to 4 V, the electrical driving force was further enhanced and the VFAs removal performance was accordingly improved. The inhibition of VFAs in the late stage of fermentation was dampened, with a significant hydrogen production improvement after 36 h. Consequently, the volumetric hydrogen productivity further increased to 1878.0 mL/L (corresponding to a specific hydrogen yield of 1.5 mol H_2/mol glucose) with an average hydrogen production rate of 19.6 mL/L/h. As the voltage increased to 6 V, however, the volumetric hydrogen productivity slightly decreased to 1859.3 mL/L with an average hydrogen production rate of 19.4 mL/L/h. This may be attributed to the fact that the ionization of weak acid is enhanced at 6 V, adversely affecting the microbial activity. It could also be explained that the electrical driving force affects the surface charge distribution of the cell and then changes the permeability of the ion to cell. Furthermore, the removal rate of VFAs by electrodialysis was in accordance with the VFAs production rate at 4 V during the fermentation; as a result, the fermentation chamber can be operated stably and efficiently at a low VFAs concentration. The residual glucose concentration in the fermentation effluents for all trials was in the range

of 0.22-0.30 g/L, corresponding to 97.0%-97.9% of the glucose utilization efficiency. This suggests that most of the substrate was consumed by HPB during the dark fermentation.

It should be noted that no gas production was observed in the anode and cathode chambers during fermentation. Furthermore, a blank group (inoculum without substrate added) was tested, and no gas production was observed in any of the three chambers. These results confirm that the hydrogen production was sourced from the fermentation of glucose rather than from the electrolysis of water.

In control group, the hydrogen content was 50.9%. Interestingly, the hydrogen content increased when the voltage was increased from 0 V to 6 V and achieved 54.6%, 65.3%, 69.5%, 84.7%, respectively. Apart from hydrogen, carbon dioxide is a major gaseous product that can be easily dissolved in the liquid phase to form bicarbonate and carbonate (Eqs. (3) and (4)). The electrical driving force affects the transportation of bicarbonate and carbonate across the AEM, thereby reducing the bicarbonate and carbonate concentration in the fermentation liquor. This can be confirmed by the results where the total inorganic carbon (including for bicarbonate, carbonate concentration and dissolved carbon dioxide) in the anode chamber were 214.7, 437.5, 796.7, and 1143.4 mg/L at a voltage of 0 V, 2 V, 4 V, and 6 V at 96 h, respectively. As a result, carbon dioxide dissolution was promoted and the carbon dioxide content decreased, whereas hydrogen content increased. The increased hydrogen content can reduce the gas storage requirement and lower the gas upgrading cost, which is very beneficial for biohydrogen production at an industrial scale.

$$CO_2 + H_2O \rightarrow H^+ + HCO_3^- \quad (3)$$

$$HCO_3^- \rightarrow H^+ + CO_3^{2-} \quad (4)$$

3.3. VFAs removal during fermentation

The concentrations of acetate and butyrate in the fermentation chamber are shown in Figs. 4a and 4b, respectively. When the fermentation was conducted in the single chamber bioreactor (control group), the acetate and butyrate concentrations rapidly increased to 26.9 and 32.3 mmol/L, respectively, with the increased fermentation time of 60 h. This suggests that glucose was quickly metabolized to VFAs during this stage. As the fermentation time further increased to 96 h, the acetate and butyrate gradually increased to 32.2 and 35.0 mmol/L. A number of studies have confirmed that the hydrogen production tends to decrease with increasing concentrations of VFAs (Wang et al. 2008; Zhang et al. 2012; Zheng & Yu 2005). Zhang et al found that with the addition of acetate or butyrate to 20 mmol/L the hydrogen production decreased by more than 15% and 20%, respectively (Zhang et al. 2012). Therefore, high levels of VFAs would be inhibitory for hydrogen production. Consequently, a small amount of hydrogen was produced after 60 h (see Fig. 3).

When the three-chamber electrodialysis reactor was operated without voltage, the concentrations of acetate and butyrate in the fermentation chamber decreased slightly compared with those in the control group (single chamber bioreactor). This can be attributed to the concentration diffusion between the fermentation and anode chambers in which acetate and butyrate can pass through the AEM to the anode chamber. However,

the diffusion by the concentration gradient is not efficient; thus, the concentrations of acetate and butyrate in the fermentation remained at high levels, achieving 26.8 and 32.0 mmol/L at 96 h.

When the voltage was set as 2 V, the concentrations of acetate and butyrate still increased with the fermentation time of 60 h and remained stable with the fermentation time of 96 h. The final concentrations of acetate and butyrate achieved were 18.0 and 21.9 mmol/L, which were 32.8% and 31.6% lower compared with the 0 V group, respectively. This implies that the electrical driving force boosted the VFAs removal. As a result, the hydrogen production in the later stage (after 60 h) was significantly improved (Fig. 3).

When the voltage was increased to 4 V, the VFAs removal was enhanced. No significant increases in the concentrations of acetate and butyrate were observed during 12 h to 60 h. The final concentrations of acetate and butyrate were only 10.3 and 13.1 mmol/L, which were 61.6% and 59.1% lower than in the 0 V group, respectively. Wang et al found that with the addition of acetate or butyrate to 10 mmol/L the inhibitory effect on substrate degradation efficiency and hydrogen production decreased just slightly compared with that without the addition of acetate or butyrate (Wang et al. 2008). The effective removal of VFAs facilitated hydrogen production at the later fermentation stage. Therefore, the hydrogen production rate during 60-84 h could remain at high level (18.2 mL/L/h) compared with the 22.5 mL/L/h obtained during 0-60 h.

As the voltage was further increased to 6 V, the concentrations of acetate and butyrate were similar to those obtained at the voltage of 4 V. This suggests that a further increase of voltage could not improve the VFAs removal when the VFAs concentrations were at extremely low levels in the fermentation chamber. The final concentration of acetate and butyrate were decreased slightly to 7.6 and 8.5 mmol/L, respectively.

Acetate and butyrate were removed from the fermentation chamber but enriched in the anode chamber. Figs. 4c and 4d show the changes in the concentrations of acetate and butyrate in the anode chamber. When the voltage was set as 0 V, the acetate and butyrate concentrations gradually increased with the fermentation time. The final concentrations of acetate and butyrate achieved were 5.3 and 7.1 mmol/L, respectively. As the voltage was increased from 2 V to 4 V, the VFAs enrichment effect was enhanced, the final acetate concentration increased from 6.8 to 21.4 mmol/L, and the final butyrate concentration increased from 7.9 to 14.9 mmol/L. However, the concentration of acetate in the anode chamber increased slightly in the 6 V group after 48 h, and the concentration of acetate at 48 h and 96 h were 17.2 and 17.8 mmol/L, respectively. The concentration of butyrate increased continuously and reached 16.7 mmol/L at 96 h. This may be attributed to the shift in metabolism of HPB from the acetate to butyrate pathway, leading to the reduction in acetate production and increase in butyrate production.

The final concentration of total VFAs (mainly acetate and butyrate) in the fermentation chamber decreased with increasing voltage (see Fig. 5a). In the control group of the single-chamber reactor, the final concentration of total VFAs was 70.3

mmol/L. The experimental results show that the three-chamber electrodialysis bioreactor has obvious effect on control VFAs concentrations (compared with control, total of VFAs decrease by 11.9% to 75.7% when voltage increased from 0 to 6 V). The performance of VFAs removal was greatly improved when a low voltage was applied, as compared with the electrodialysis reactor for post-treatment of fermentation effluent in a recent study (total of VFAs decreased by 26.1% per 24 h at a voltage of 18 V) (Jones et al. 2017). These results indicate that the total VFAs concentrations can be maintained at a desired level by controlling the voltage during the fermentation.

3.4. Utilization of VFAs as substrate

Enhanced hydrogen production can be achieved in the three-chamber electrodialysis bioreactor, in which the accumulated VFAs in the fermentation chamber can be effectively removed. Meanwhile, the VFAs recovered in the anode chamber were considered to be valuable products. As shown in Fig. 5b, the total VFAs concentrations were 13.1, 15.6, 37.5, and 34.5 mmol/L in the anode chamber at voltages of 0 V, 2 V, 4 V, and 6 V, respectively. VFAs can be used as an important raw material in various industrial applications (Jones et al. 2017; Motte et al. 2015). For example, VFAs can be used as precursors for biodiesel production. VFAs can also be used as an external carbon source for the biological denitrification of wastewater rich in nitrogen (Motte et al. 2015) and for electricity generation via microbial fuel cells (Pham et al. 2012; Wang et al. 2014).

4. Conclusion

The three-chamber electrodialysis bioreactor was proposed to effectively remove the VFAs to promote hydrogen fermentation. A volumetric hydrogen productivity of 1878.0 mL/L was achieved in the fermentation chamber at a voltage of 4 V, which is 55.4% higher than that (1208.5 mL/L) of the control group. By applying different voltages (0-6 V), the hydrogen content accumulated to 54.6%-84.7%, exhibiting increases of 7.1%-66.4% compared with the control. Meanwhile, the maximum concentration of acetate and butyrate in the fermentation chamber was maintained at low levels of 10.3 and 13.1 mmol/L, respectively, which were 68.0% and 62.4% lower than the control group.

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Table caption

Table 1. Experimental design for the VFAs removal and hydrogen production trials.

Table 1 Experimental design for the VFAs removal and hydrogen production trials.

	Bioreactor	Voltage	Substrate	Inoculum
VFAs removal experiments	Three-chamber electrodialysis bioreactor	0-6 V	Simulated fermentation broth (acetic or butyric acid)	No
Hydrogen fermentation	Three-chamber electrodialysis bioreactor	0-6 V	Glucose solution	HPB
Hydrogen fermentation blank	Three-chamber electrodialysis bioreactor	0-6 V	No	HPB
Control	Single-chamber	No	Glucose solution	HPB

VFAs: volatile fatty acids; HPB: hydrogen-producing bacteria.

Figure captions

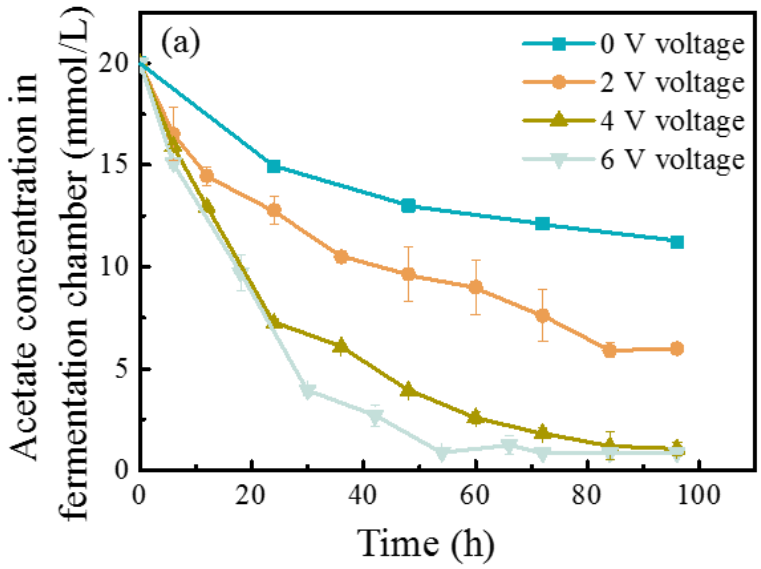
Fig. 1. Removal experiments of simulated fermentation broth. (a) Acetate concentration in the fermentation chamber; (b) Acetate concentration in the anode chamber.

Fig.2. Removal experiments of simulated fermentation broth. (a) Butyrate concentration in the fermentation chamber; (b) Butyrate concentration in the anode chamber.

Fig. 3. Hydrogen fermentation with electrodialysis.

Fig. 4. VFAs removal during fermentation. (a) Acetate concentration in the fermentation chamber; (b) Butyrate concentration in the fermentation chamber; (c) Acetate concentration in the anode chamber; (d) Butyrate concentration in the anode chamber.

Fig. 5. Total VFAs in the fermentation chamber (a); Total VFAs in the anode chamber (b).



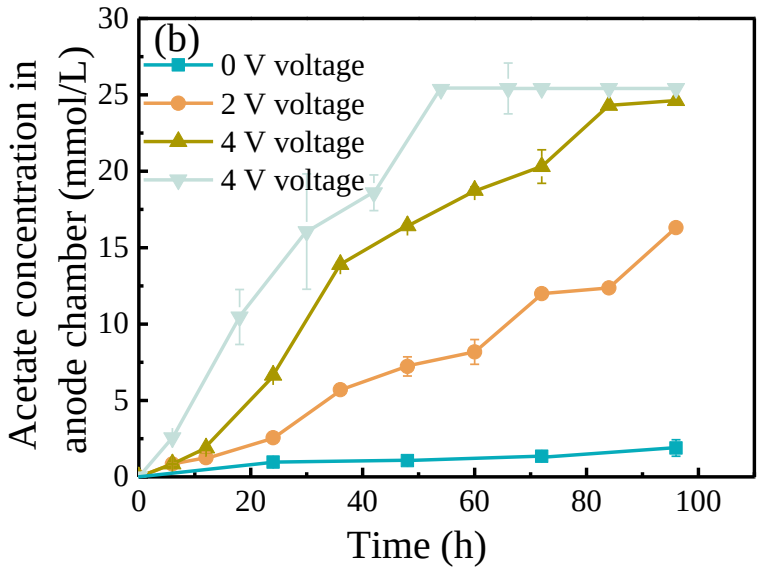
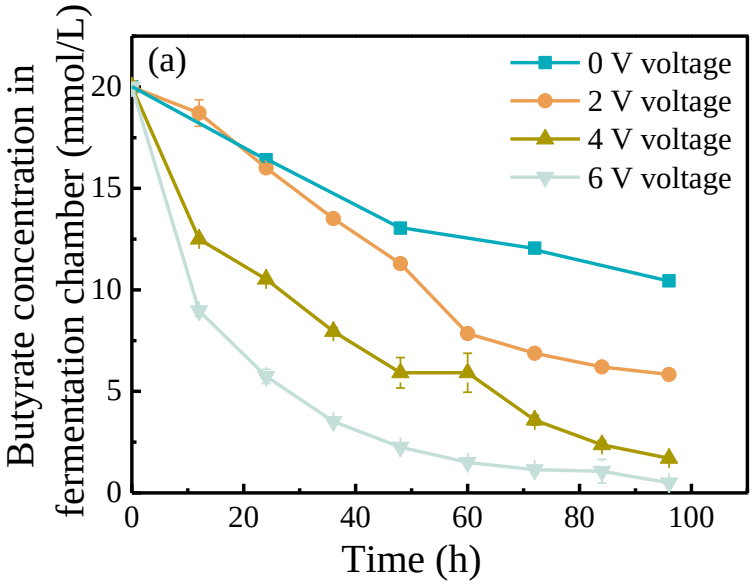


Fig. 1 Removal experiments of simulated fermentation broth. (a) Acetate concentration in the fermentation chamber; (b) Acetate concentration in the anode chamber.



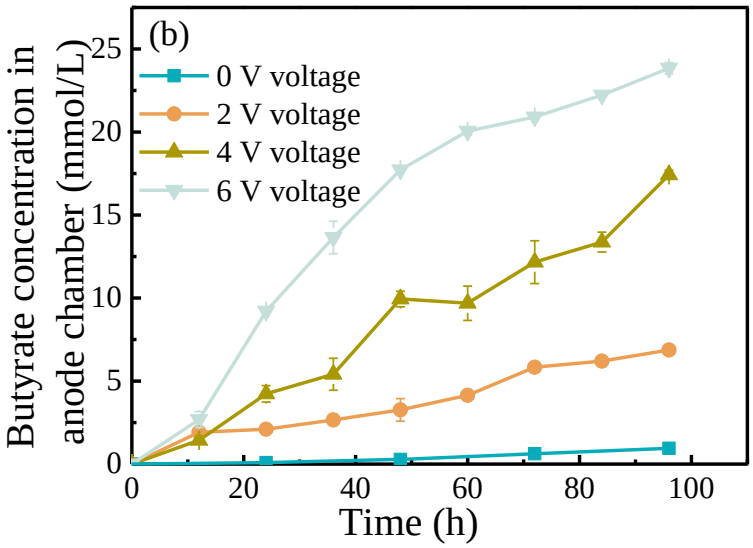


Fig. 2 Removal experiments of simulated fermentation broth. (a) Butyrate concentration in the fermentation chamber; (b) Butyrate concentration in the anode chamber.

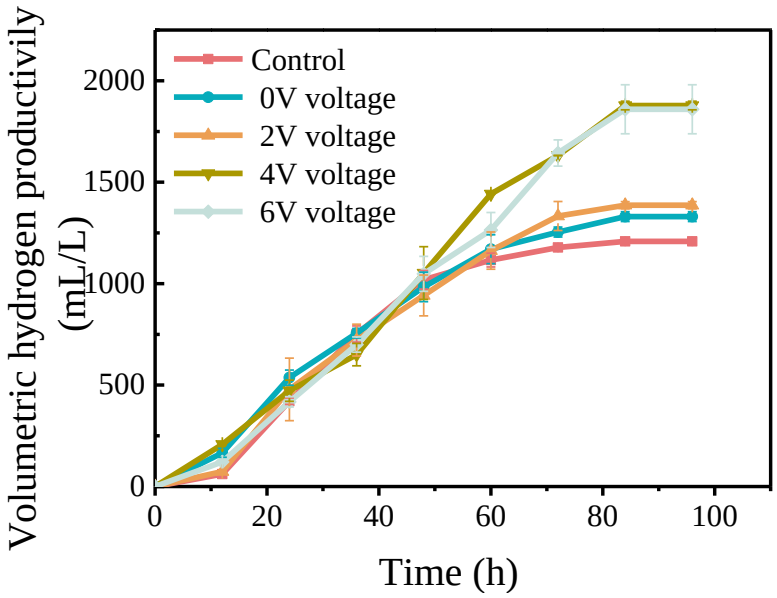


Fig. 3 Hydrogen fermentation with electrodialysis.

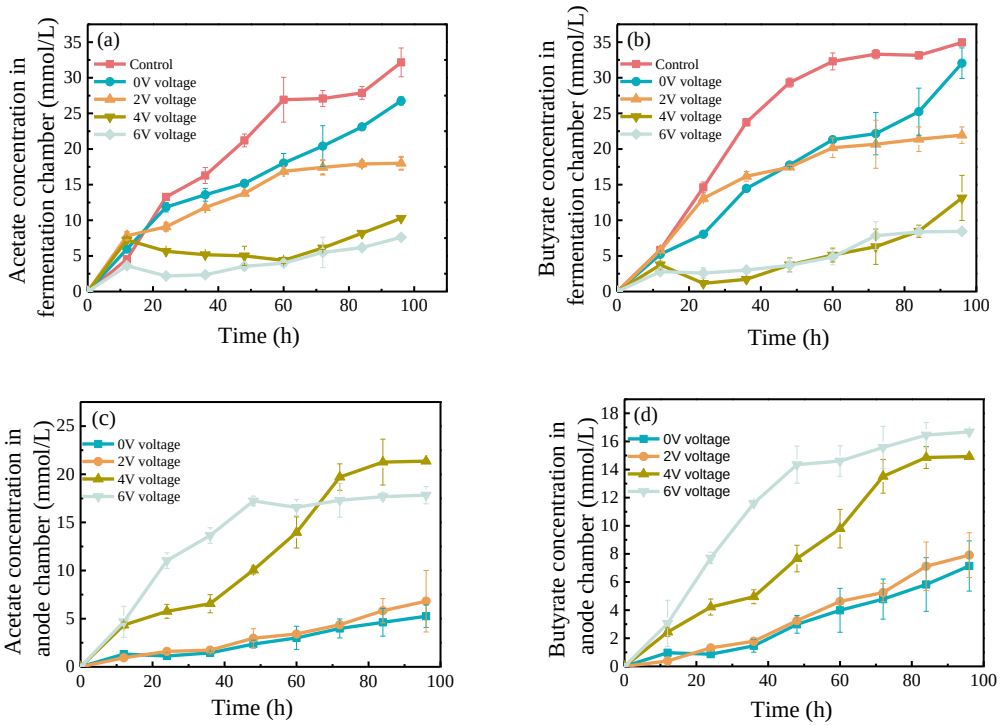
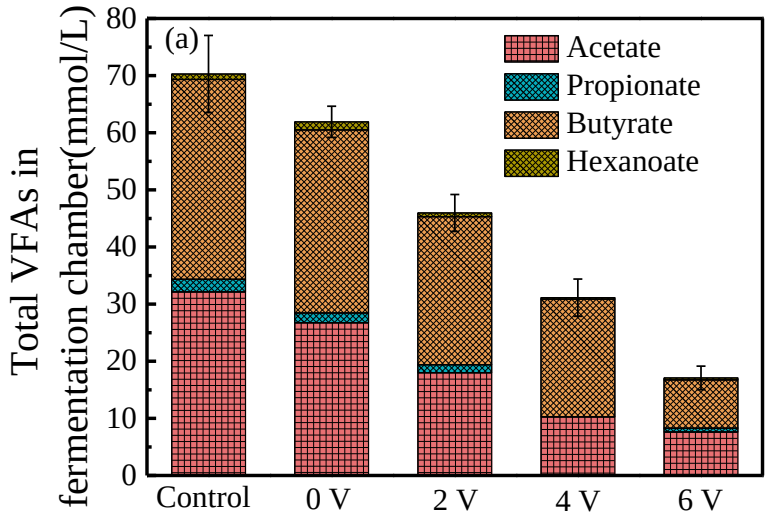


Fig. 4 VFAs removal during fermentation. (a) Acetate concentration in the fermentation chamber; (b) Butyrate concentration in the fermentation chamber; (c) Acetate concentration in the anode chamber; (d) Butyrate concentration in the anode chamber.



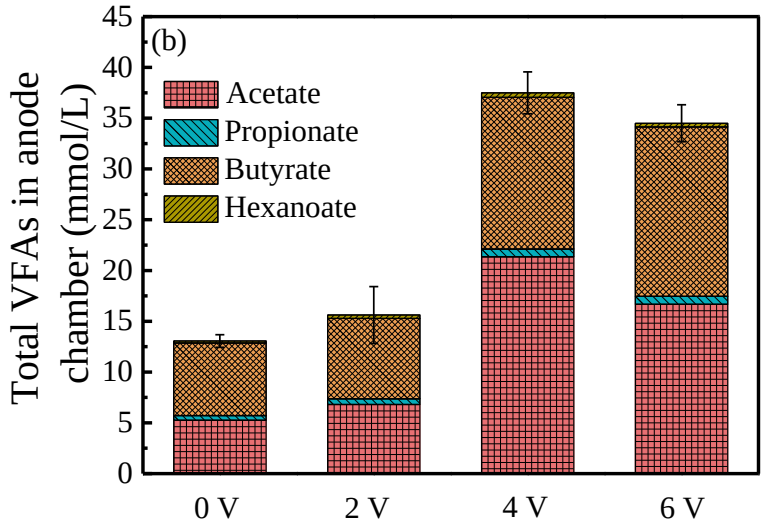


Fig. 5 Total VFAs in the fermentation chamber (a); Total VFAs in the anode chamber (b).