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## A milliliter-scale yeast-based fuel cell with high performance



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#### ABSTRACT

Microbial fuel cells are attracting attention as one of the systems for producing electrical energy from organic compounds. We used commercial baker's yeast (*Saccharomyces cerevisiae*) for a glucose fuel cell because the yeast is a safe organism and relatively high power can be generated in the system. In the present study, a milliliter (mL)-scale dual-chamber fuel cell was constructed for evaluating the power generated by a variety of yeasts and their mutants, and the optimum conditions for high performance were investigated. When carbon fiber bundles were used as an electrode in the fuel cell, high volumetric power density was obtained. The maximum power produced per volume of anode solution was 850 W/m³ under optimum conditions. Furthermore, the power was examined using seven kinds of yeast. In *Kluyveromyces marxianus*, not only the power but also the power per consumed glucose was high. Moreover, it was suggested that xylose is available as fuel for the fuel cell. The fuel cell powered by *K. marxianus* may prove to be helpful for the effective utilization of woody biomass.

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#### 1. Introduction

Of late, clean energy development is demanded to alleviate global environmental problems and lessen dependence on fossil fuels. Glucose fuel cells have attracted attention as a system producing electric power from renewable fuels. They are divided into two types, an enzyme-based fuel cell and a microbial-based fuel cell (MFC). In the former, enzymes and coenzymes are used directly to extract electrons from glucose, and the power of more than 100 mW is generated in a portable electronic device of 80 cm<sup>3</sup> [1]. In the latter, microorganisms are used, and it is more cost effective than the enzymatic type. As shown in Table 1, the volumetric power densities of MFCs are still low compared with the power of the enzyme-based fuel cell [2,3]. Many researchers have studied MFCs using organic wastes or other organic matter instead of glucose. For example, wastewater [4,5], lactate [6], and acetate [7] have been used as fuels, yielding power densities exceeding 500 W/m<sup>3</sup> [6,7]. To improve the volumetric power, special microminiature fuel cells have been developed [8-10]. Nevin et al. developed a small reactor (anode volume of 0.335 cm<sup>3</sup>) with a large electrode area, in which the power per anode volume reached 2150 W/m<sup>3</sup> [8].

MFCs are in turn divided into two types, a fuel cell depending on a mediator to transfer electrons and a fuel cell with direct electron transfer. For example, yeast (*Saccharomyces cerevisiae*) fuel cells depend upon mediators, such as methylene blue and thionine, to transfer electrons [3,11], and *Shewanella oneidensis* and *Geobacter sulfurreducens* fuel cells generate electricity without a mediator. The extracellular electron transfer systems of *S. oneidensis* [12–14] and *G. sulfurreducens* [15–17] have been studied. Of late, it has been reported that *Candida melibiosica*, a yeast, can be used as a biocatalyst in an MFC without artificial mediators [18].

The development of the fuel cell components such as electrodes is also important for the improvement of power [4,19]. In this study, we investigated a catalytic microorganism. In a previous study [20], relatively high volumetric power was obtained in an MFC using commercial baker's yeast (*S. cerevisiae*) as shown in Table 1. The yeast is a safe microorganism used for food, and its genetic manipulation is also easy. A technique for displaying enzymes on the cell surface was already established, and was utilized in the MFC [21,22], for example, in display of glucose oxidase on a microorganism used as a catalyst [21]; therefore, we utilized yeast as a biocatalyst in the MFC. To select a yeast suitable for a fuel cell from various yeasts and their mutants, a fuel cell capable of stable evaluation in a short period is necessary. Such a fuel cell may also lead to the development of a portable MFC.

The development of a small-sized dual-chamber highperformance fuel cell powered by baker's yeast was carried out, and consequentially, the high maximum power per volume of anode solution was obtained. Furthermore, the biocatalytic activities of various yeasts were also evaluated using the developed fuel

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**Table 1**Summary of performance of MFCs (including reference data of an enzyme based fuel cell).

Catalyst	Substrate	Cell type/cathode/mediator	Cell or Anode volume used for calculation of <i>Pv</i>	Maximum Ps (mW/m²)	Maximum Pv (W/m³)	Ref.
Glucose dehydrogenase	Glucose	Cell composed of two multi-stacked cell units/FeCN/NADH and vitamineK3	80 cm <sup>3</sup> cell	14,500	1250	[1]
Saccharomyces cerevisiae	Glucose	Dual chamber/FeCN/Methylene blue	500 cm <sup>3</sup> anode cell	n.a.	0.147	[3]
Saccharomyces cerevisiae	Glucose	Dual chamber/FeCN/2-hydroxy- 1,4-naphtoquinone	30 cm <sup>3</sup> anode solution	750	300	[20]
Shewanella oneidensis DSP-10	Lactate	Dual chamber stacked cell/FeCN/anthraquinone- 2.6-disulfonate	1.2 cm <sup>3</sup> anode cell	3000	500	[6]
Mixed bacteria	Acetate	Double cloth electrode assembly/Air/None	2.5 cm³ cell	1120	627	[7]
Mixed bacteria	Acetate	Double cloth electrode assembly continuously flow/Air/None	2.5 cm <sup>3</sup> cell	1800	1010	[7]
Saccharomyces cerevisiae	Glucose	Dual chamber/FeCN/2-hydroxy- 1,4-naphtoquinone	7.5 cm <sup>3</sup> anode solution	22	850	This study
Kluyveromyces marxianus	Glucose	Dual chamber/FeCN/2-hydroxy- 1,4-naphtoquinone	7.5 cm <sup>3</sup> anode solution	22	860	This study
Kluyveromyces marxianus	Xylose	Dual chamber/FeCN/2-hydroxy- 1,4-naphtoquinone	7.5 cm <sup>3</sup> anode solution	11.2	443	This study
Mixed bacteria	Xylose	Single chamber/Air/None	12 cm <sup>3</sup> cell	2330	38.8	[25]
Mixed bacteria	Xylose	Single chamber/Air/None	770 cm <sup>3</sup> cell (working)	673	13	[24]
Shewanella oneidensis	Trypticase soy broth	Dual chamber, microfluidic fuel cell/FeCN/None	0.004 cm <sup>3</sup> cell	6.25	62.5	[10]
Geobacteraceae-enriched culture	Acetate	Dual chamber, semi continuous/FeCN/None	0.0045 cm <sup>3</sup> anode cell	47	2300	[9]
Geobacter sulfurreducens	Acetate	Dual chambered stacked cell/FeCN/None	0.335 cm <sup>3</sup> anode cell	1900	2150	[8]

n.a., not applicable.

cell. Of them, *Kluyveromyces marxianus* was found to be useful as a biocatalyst for the MFCs.

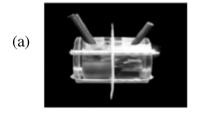
#### 2. Materials and methods

#### 2.1. Manufacture of a small-sized fuel cell

The fuel cell described in this study consists of an anode and a cathode separated by a cation-specific membrane. Electrons generated by glucose oxidation in yeast cells in the anode part are transferred to the electrode by mediators. A fuel cell with anode and cathode tanks of a capacity of 8.3 cm<sup>3</sup> was constructed from an acrylic rod with a diameter of 3.4 cm (Fig. 1a). Commercial carbon rods (diameter 0.5 cm) were used as electrodes. After preliminary testing, three rods were used in the anode and two in the cathode. In addition, carbon fiber bundles (Mitsubishi Rayon Co. Ltd., Tokyo, Japan) were also used as electrodes to obtain high performance. The diameter of the filament was 7 µm and one bundle consisted of 12,000 filaments. For an electrode, 16 bundles of approximately 7 cm length were used to maintain contact with the anode solution. A cation-specific membrane, GORE-SELECT (Japan Gore-Tex Inc., Tokyo, Japan), with a thickness of 30 µm was used to separate the electrode tanks.

### 2.2. Composition of the solution used for the fuel cell

Glucose of 1.5–7% was used as the fuel and baker's dry yeast at 0.1– $0.2 \text{ g/cm}^3$  was used as the microorganism, 2-hydroxy-1,4-naphthoquinone (HNQ) at 14–28 mM was used as the mediator, and  $K_3[\text{Fe}(\text{CN})_6]$  at 160–800 mM was used as the oxidizing agent.



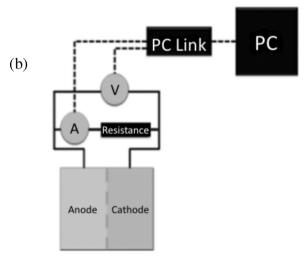


Fig. 1. (a) Small glucose fuel cell and (b) connection of the fuel cell and detectors.

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