



# Effects of the kinematic viscosity and surface tension on the bubble take-off period in a catalase–hydrogen peroxide system

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## ARTICLE INFO

### Article history:

Received 15 October 2008

Received in revised form 14 January 2009

Accepted 22 January 2009

Available online 31 January 2009

### Keywords:

Kinematic viscosity

Surface tension

Catalase

Hydrogen peroxide

Bubble take-off period

## ABSTRACT

The effect of kinematic viscosity and surface tension of the solution was investigated by adding catalase, glucose oxidase, or glucose on the bubble movement in a catalase–hydrogen peroxide system. The kinematic viscosity was measured using a Cannon–Fenske kinematic viscometer. The surface tension of the solution was measured by the Wilhelmy method using a self-made apparatus. The effects of the hole diameter/cell wall thickness, catalase concentration, glucose concentration, and glucose oxidase concentration on the kinematic viscosity, surface tension, and bubble take-off period were investigated. With our system, the effects of the changes in the solution materiality on the bubble take-off period were proven to be very small in comparison to the change in the oxygen-producing rate.

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## 1. Introduction

Measurement of a certain chemical compound using the recognition function of an enzyme is known as biosensing, and this method has been widely applied as enzyme sensors [1–8]. The product amount of the enzyme-catalyzed reaction is converted into an electrical signal, and, therefore, the target (substrate) concentration can be measured as current or voltage. Apart from the pursuit for the high speculation, we proposed a batteryless system that enabled naked-eye measurement [9–12] of glucose [13,14]. Oscillation in the dissolved oxygen concentration is observed in a catalase–dialysis membrane–hydrogen peroxide system [15,16]. When glucose and glucose oxidase coexist in a catalase solution, the oscillation period becomes larger with the glucose concentration. The mechanism of such oscillation is still under investigation; therefore, we focused on the movement of oxygen bubbles produced after the conversion of hydrogen peroxide into oxygen. [17] In this previous study, oxygen bubbles were produced when the oxygen concentration exceeded the maximum dissolved oxygen concentration. Such bubble production occurred at the dialysis membrane surface (Fig. 1(a)). These small bubbles grew into a large bubble through coarsening (Fig. 1(b)). The reaction cell was equipped with a “tunnel ceiling” in front of the membrane. Coarsening continued until the bubble grew larger than the ceiling and, finally, escaped from the tunnel ceiling and floated towards the

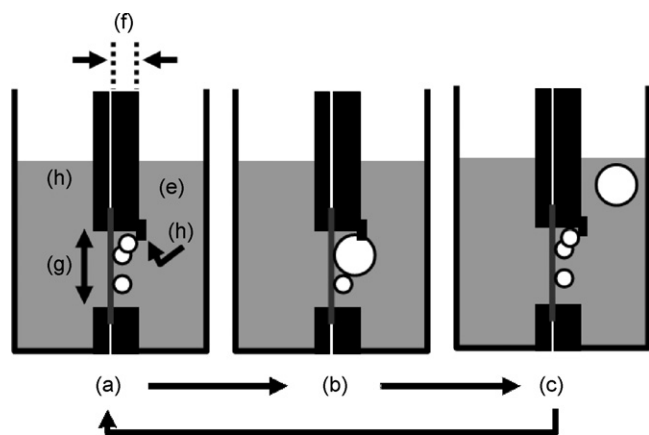
air–liquid interface (Fig. 1(c)). In this previous study, we showed that when hydrogen peroxide diffused into a solution (catalase, glucose oxidase, and glucose) through a dialysis membrane, the bubble take-off period became smaller with a higher glucose concentration. The initial reason might be that the production speed of the oxygen bubble became smaller with the glucose concentration because oxygen was consumed through the glucose oxidation reaction. Experimentally, the bubble take-off period increased with the glucose concentration. This was thought to be due to the decrease in the dissolved oxygen (DO) concentration resulting from glucose oxidation. Although this mechanism was supported by a considerable amount of experimental evidence, we suspected that a change in the solution materiality (viscosity/surface tension) by the addition of glucose might also affect the bubble movement. In this work, we investigated the change in materiality, such as the kinematic viscosity and surface tension, of the solution by adding catalase, glucose oxidase, or glucose and examined their effect on the bubble movement.

## 2. Materials and methods

All the measurements were performed at room temperature adjusted to 25 °C. The effect of the temperature was already known to be negative on the oscillation period. Two solutions contacted via a dialysis membrane, as illustrated in Fig. 1, and 20 ml of a 3% H<sub>2</sub>O<sub>2</sub> (Wako Chemicals) solution (solution A) were used in all the measurements. Eight thousand units of catalase (from bovine liver, Sigma–Aldrich Co.), 2000 units  $\times$   $n$  ( $n = 0, 1$ ) of glucose oxidase (from *Aspergillus niger*, Sigma–Aldrich Co.), and 0.9 mg  $\times$   $n$  ( $n = 0, 1$ ,

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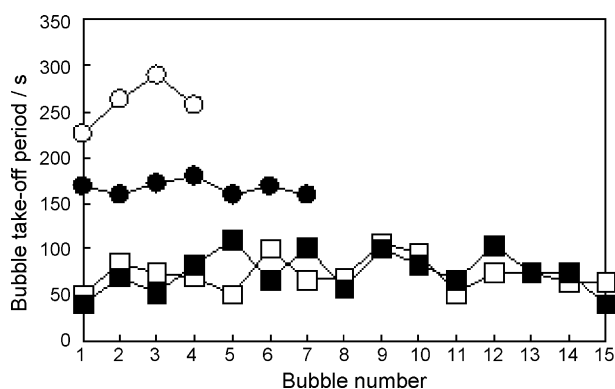
**Fig. 1.** Schematic illustration of bubble production (a), coarsening (b), and take-off (c). Solution A (d) and solution B (e) were placed in each cell. Cell wall thickness (f) and hole diameter (g) were changed to see the reproducibility. A tooth (h) was placed to observe a large bubble when glucose oxidase was used.

2, 3, 4) of glucose (Wako Chemicals Co.) were contained in 20 ml solution (solution B). The cell wall thickness was changed at 1.0, 2.0, and 3.0 mm by changing the acryl plate number. The hole diameter was also changed (2.0, 3.0, 4.0, and 5.0 mm). Bubble movement was recorded with a PC-connected Web camera (Logicool Co.). The kinematic viscosity was measured using a Cannon–Fenske kinematic viscometer (Shibata Scientific Technology Ltd.). The surface tension of the solution was measured with the self-made Du–Noüy ring tensiometer.

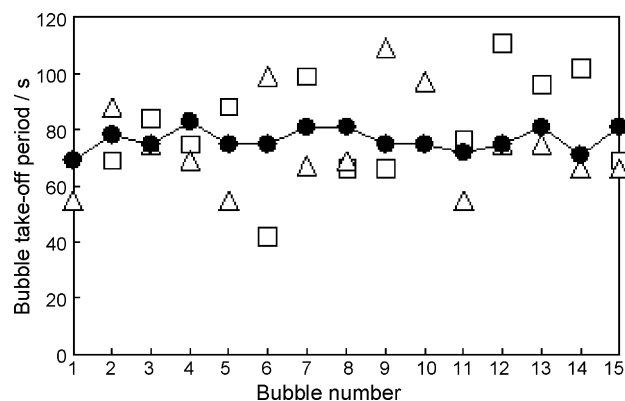
### 3. Results and discussion

#### 3.1. Effects of the hole diameter/cell wall thickness on the bubble take-off period

The reproducibility of the bubble take-off period was initially thought to be an important factor for the discussion. We therefore tried to optimize the cell design by adjusting the hole diameter and cell wall thickness. The bubble take-off period was defined as the time between the  $N$ th and  $(N+1)$ th bubble taking off. First, the cell wall thickness was adjusted to 1.0 mm. The hole diameter was changed to 2.0, 3.0, 4.0, and 5.0 mm, and, among them, the case with a diameter of 3.0 mm showed the best reproducibility (relative standard deviation, R.S.D. = 4.86%,  $n=7$ ) in the bubble take-off period. As shown in Fig. 2, the larger diameter resulted in a smaller take-off period. In the case of a larger diameter, more  $H_2O_2$  molecules were



**Fig. 2.** Effect of hole diameter on the bubble take-off period. Hole diameters of 2.0 mm (○), 3.0 mm (●), 4.0 mm (□) and 5.0 mm (■) were tried. Cell wall thickness was adjusted to 1 mm. Solution A contained 20 ml of 3% (w/w)  $H_2O_2$ . Solution B contained 20 ml of 400 units/ml catalase.

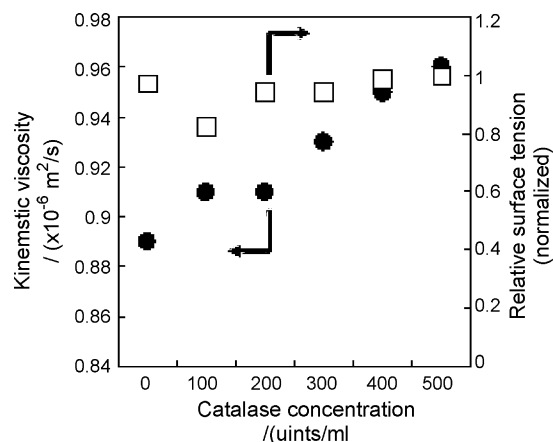


**Fig. 3.** Effect of cell wall thickness on the bubble take-off period. Cell wall thicknesses of 1.0 mm (△), 2.0 mm (□), and 3.0 mm (●) were tried. Hole diameter was adjusted to 4.0 mm. Solution A contained 20 ml of 3% (w/w)  $H_2O_2$ . Solution B contained 20 ml of 400 units/ml catalase.

thought to be supplied to the catalase solution, and more oxygen might thus be produced, resulting in a shorter time to fill the tunnel ceiling of the cell. For the subsequent experiments, we chose a hole diameter of 4.0 mm because of the shorter time required to obtain the results. We then adjusted the cell wall thickness at 1.0, 2.0, and 3.0 mm. Fig. 3 shows the effect of the thickness on the periods. The figure tells us that there is no significant difference in the take-off periods. Among them, the case with 3.0 mm showed the best reproducibility (R.S.D. = 5.48%,  $n=15$ ). At this stage, we could say that the take-off period reproducibility was good when the hole diameter and cell wall thickness were similar. In this case, the growing bubble (almost spherical) can fill most of the “tunnel” space and does not allow tiny bubbles to escape from the tunnel. We were aware that such tiny bubbles often help large bubbles to take-off from the tunnel earlier. The good reproducibility might be due to the reduction of tiny bubbles at the take-off moment.

#### 3.2. Catalase concentration effect

Fig. 4 shows the kinematic viscosity and relative surface tension (normalized) plotted vs. the catalase concentration. In this case, neither glucose oxidase nor glucose was contained. Changes were seen in both values in the range of 0–500 units/ml of catalase. For pure water, the kinematic viscosity and surface tension were reported to be  $0.89 \times 10^{-6} \text{ m}^2/\text{s}$  and 71.96 mN/m at 25 °C. Our result for kinematic viscosity agreed well with the reported value, but not for



**Fig. 4.** Effect of catalase concentration on the kinematic viscosity (●) and on the relative surface tension (□). A Cannon–Fenske kinematic viscometer and a self-made apparatus (a metal ring and a balance) were used for the measurements.

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