MONITORING OF YEAST CELL CONCENTRATION USING A COUPLED MICROFIBER BASED OPTICAL SENSOR

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A simple optical sensor is proposed using a coupled microfiber probe to monitor yeast live cell concentration. In this method, intensity and spectrum of the reflected light from the coupled tip is measured for the determination of biomass. It is obtained that the increase of number of cells in the detection medium leads to the enhancement of light scattering and increases the output light intensity. Based on this technique, the proposed sensor is able to detect the concentration of yeast with sensitivity of 0.2121dB/%, slope linearity of more than 98.37 % and a resolution of 1.17%. The spectral analysis can also be used to detect changes in the live yeast concentration whereby the wavelength is shifted from 1530.92 nm to 530.78 nm as the concentration varies from 10% to 40%. These results show that the proposed sensor is suitable for use in industrial fermentation systems.

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1. Introduction

Optical microfibers have recently attracted considerable interest as promising building blocks for a wide variety of photonic applications [1-2]. They possess large refractive index (RI) contrast which is able to provide tight field confinement that makes microfiber particularly suitable for nonlinear optical applications [3]. Microfibers also offer an advantage of the ease of integration with conventional single mode fiber (SMF) as well as the access to the evanescent field provided by tapering since the light is guided by the boundary between the taper and the external environment. The external environment may be chosen to determine the number of modes supported by the waist, the bend tolerance and may provide a means of tuning through index of refraction or absorption control [4]. Up to date, microfibers have been used for variety of applications such as optical probes for biomedical applications [5], add–drop multiplexers [6], optical encoder systems [7], and optical resonators [8]. Microsphere-taper coupling systems were also developed for various applications [9-11]. For instance, optical fiber with micro-lens was utilized to produce large field-of-view video-rate optical beam scanning for micro-endoscopy applications [9]. The optical detection nature of microsphere-taper structure can also be utilized for robust thermal sensing application [11].

On the other hand, there is a growing interest in the miniaturization of cell cultivation systems especially for fermentation studies. For the studies, monitoring of biomass with time is

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important for the assessment of the influence of ferment or conditions. Previously, live cell index sensing based on the reflection mode of tilted fiber tip with gold nanoparticles was demonstrated [12]. In this paper, live yeast cell concentration monitoring is demonstrated using a coupled microfiber sensor probe. In this method, the reflected light from the coupled fiber tip is measured for the determination of biomass. The sensor probe is coated with a polymer to reduce the scattering loss and increases the sensitivity of the sensor.

2. Experimental arrangement

At first a coupled microfiber structure is fabricated using an experimental setup as described in Fig. 1. We fabricate the microfiber coupler by twisting two different location of single mode fiber (Corning SMF 28) to make an overlapping contact. Then, the twisted fibers were tapered using a flame brushing technique to form a microfiber coupler [13]. The coupling region is then perpendicularly cleaved so that the fiber tip can be used as a sensor probe. The coupled microfiber tip is then covered by poly-(methyl methacrylate) (PMMA) provided by Organic ACROS with mole weight 35.000 using dip coating method. In the experiment, the fiber tip of the microfiber coupler structure is immersed in solution containing 1.30 wt % PMMA. After immersion about 25 seconds, the tip is pulled out with a fast speed. Figs. 2 (a) and (b) show the microscope images of the coupled microfiber tip without and with the polymer coating, respectively. The size of the microfiber tip is slightly increased with the coating.

The coupled microfiber sensor probe is then used to monitor the live cell concentration. In the experiment, we use live yeast cell, which was cultured in sterile distilled water in proportions of 10%, 20%, 30%, 40% and 50%. The yeast sample was incubated at 30°C for growth. Figs. 2 (c) shows the fiber tip image as it is immersed into the yeast solution while Fig. 2(d) shows the image of the yeast solution with 10% concentration.

**Fig. 1:** Schematic illustration of a coupled microfiber fabrication setup which uses an oxy-butane burner as a torch
Fig. 2: The microscope images of the coupled microfiber tip a) bare b) microfiber tip with the polymer coating c) microfiber tip immersed into the yeast solution d) yeast image in the solution

Fig. 3 shows the experimental setup for the proposed coupled microfiber based optical sensor to monitor yeast cell concentration. The sensor probe is immersed into yeast solution with various concentration, which is put into a petri dish as shown in the figure. To monitor the transmission spectrum of the sensor during the experiment, amplified spontaneous emission (ASE) source from an Erbium-doped fiber amplifier (EDFA) operating in 1550 nm region is injected into one end of the SMF while the other end is connected to the optical spectrum analyzer (OSA). The sensor probe was first immersed in 10% yeast concentration to measure the change in output spectrum and output power, followed by other yeast concentrations of 20%, 30%, 40%, and 50%. Throughout the experiment, the corresponding output spectra were measured by an OSA which provides accurate measurements. Furthermore, a well-regulated ASE is used to minimize the fluctuation of source intensity.

Fig. 3: Schematic diagram of the experimental setup to monitor yeast cell concentration using a coupled microfiber based optical sensor with tip diameter of 7µm.

3. Results and Discussion

The refractive index of various yeast concentrations was measured using a refractometer and the result is shown in Fig. 4. It is obtained that refractive index reduces from 1.347 to 1.3438 as the concentration of yeast increases from 10% to 50%. The output power from the coupled microfiber probe is then investigated at various yeast concentrations and the result is shown in Fig. 5. It is observed that an increase in yeast concentration can be detected by an increase in the output
power. As the concentration of yeast increases from 10% to 50%, the output power increases linearly.

A higher concentration of yeast solution corresponds to a lower refractive and thus less light can be reflected back into the coupled microfiber due to Fresnel reflection. The lower index contrast reduces the total internal reflection effect at the fiber tip area and this also contributes to the mode confinement ability becomes weaker for the microfiber region. This causes more fiber loss especially at longer wavelength. However, the increase of number of cells in the detection medium leads to the enhancement of light scattering. The scattering causes more light to be reflected back into the coupled microfiber tip in various angles. Since the scattering effect is more significant compared to the index contrast effect, the collected output light increases with the yeast concentration in the solution as shown in Fig. 5. It is obtained that the coupled microfiber with PMMA coating is able to detect the concentration of yeast with sensitivity of 0.2121dB/%, slope linearity of more than 98.37% and a resolution of 1.17%.

![Fig. 4. Concentration of yeast against value of refractive index.](image)

![Fig. 5: Linear relationship between the output power and yeast concentration.](image)

Fig. 6 shows the stability of the sensor at 5 different concentrations against time. As the concentration increases from 10% to 50%, the output power increases. An increase of output power can be observe due to increase in the scattering effect. The output power fluctuations against time are 0.2586 dB, 0.1347 dB, 0.1335 dB, 0.096 dB, 0.1665 dB for concentrations 10, 20, 30, 40 and 50%, respectively. These results show that the proposed sensor is able to detect the difference in the concentration yeast which varies from 10% to 50% using the intensity modulation approach.
Besides the intensity modulation, the spectral shift approach can also be used to monitor the concentration. As the light is transformed to multimode at the coupled microfiber tip area, cladding modes interference is also obtained as the interference pattern is observed at the output spectrum. Fig. 7 shows the output spectrum, which is obtained as the microfiber tip is immersed into various yeast concentrations. It is observed that, as the concentration varies from 10% to 30% and 40%, the wavelength is shifted from 1530.92 nm to 1530.85 nm and 1530.78 nm. The shifting of wavelength shows that the spectral analysis of the sensor can also be used to detect changes in the concentration. These results show that the proposed sensor is also able to sense live cell in real time. The sensor is also suitable for use in larger (up to industrial scale) fermentation systems.

4. Conclusion

A simple optical sensor is demonstrated using a coupled microfiber tipcoated with PMMA to monitor yeast live cell concentration. The sensor can operate based on intensity modulation and spectral analysis. Due to the enhanced scattering effect with the increase of number of live cells in the detection medium, the output light intensity is observed to increase with the concentration. It is obtained that the sensor is able to detect the concentration of yeast with sensitivity of 0.2121dB/%, slope linearity of more than 98.37 % and a resolution of 1.17%. In the spectral analysis, the transmission spectrum is shifted from 1530.92 nm to 530.78 nm as the concentration varies from 10% to 40%.
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