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Fiber optic sensors based on Mach-Zehnder Interferometer for the detection of bacterial biofilms

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Summary:
In this work, a chemical sensor based on an In-fiber optic Mach-Zehnder Interferometer (MZI) is reported. The MZI is fabricated using standard single-mode fiber with a waist-enlarged splicing method, where the spliced point acts as a coupling and re-coupling element for cladding modes and core mode. Unlike conventional transmission based MZIs, a reflective configuration is prepared by coating the fiber tip with silver developing a convenient fiber optic probe. A sensitivity of about 125 nm/RIU to the surrounding medium refractive index is obtained. As an application, the biofilm growth of a Gram-negative aerobic bacterium, i.e. *Pseudomonas alcaligenes*, is observed by monitoring the resonance wavelength shifts in real-time over three days. Biofilm thickness is further analyzed by using Atomic Force Microscopy technique demonstrating the effectiveness of fiber optic technology for real time continuous monitoring of biofilm growth.

Keywords: Mach-Zender interferometer, fiber optic sensors, biofilms, environmental monitoring.

Introduction

Biofilms are microbial communities found on various surfaces, impacting ecosystems, health, and industry. They serve as environmental indicators, capturing data on nutrients, pollutants, and microbial dynamics. Effective monitoring enables early detection of contamination and supports sustainability efforts [1]. Optical fiber sensing technology offers advantages for biofilm monitoring, providing high sensitivity, real-time monitoring, and data deployment in complex environments while minimizing manual sampling [2]. Despite the increased importance of biofilm monitoring, there are only few researches available where it is performed using fiber optic technology [3], [4]. In this work, we develop and test a fiber optic sensing probe based on Mach-Zehnder Interferometer (MZI) achieved by overlap splicing of standard single-mode fibers. After refractometric characterization, the sensor was tested for *Pseudomonas alcaligenes* biofilm detection. This study is a part of the Italian MoBeeFO project, which will further apply the probe for real-time monitoring of honeybee immune responses.

Development of the sensor

The proposed MZI based fiber optic probe is fabricated using a simple and cost-effective approach with commercially available components, as schematically reported in Fig. 1(a).

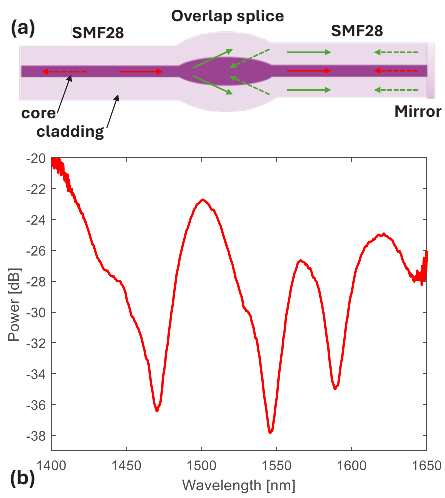

Fig. 1. MZI fiber optic sensing probe: (a) schematic view; (b) reflected optical spectrum.

It consists of two spliced single-mode fibers (Corning SMF28) with an overlap/waist-enlarged region to excite cladding modes. A silver mirror is integrated at the fiber tip to enable reflection-based operation. The interference spectrum is generated as the overlap spliced region re-couples cladding and core modes [5], [6].Fabrication was performed using a Fujikura 62S+ fusion splicer with specific parameters (arc time: 700 ms, overlap: 150 µm), resulting in a 6 mm-long MZI device. The reflected spectrum showed three main interference peaks in the 1400–1650 nm range as shown in Fig. 1(b). Sensitivity testing involved immersing the probe in calibrated refractive index (RI) solutions (within 1.33-1.39), showing a wavelength shift at 1550 nm with a sensitivity of about 125 nm/RIU. This range is relevant for subsequent biofilm experiments.

Results for biofilm monitoring

To monitor biofilm growth experiment, a *P. alcaligenes* biofilm was selected as a case study. A bacterial culture of *P. alcaligenes* was prepared by inoculating a single colony into a growth medium and incubating overnight at 30 °C. The MZI-based fiber optic probe was then immersed in the solution, and resonance wavelength shifts of the device were monitored in real-time for 60 hours using an HBM FiberSensing FS22 interrogator. The solution was continuously stirred at 100 rpm throughout the experiment. Additionally, the Atomic Force Microscopy (AFM) has been used to measure biofilm thickness.

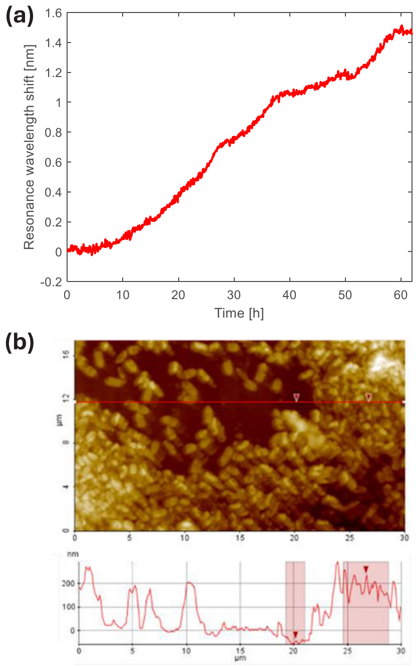

Fig. 2. (a) Sensor response in terms of resonant wavelength shift during bacterial biofilm growth. (b) AFM measurement of biofilm on optical fiber with map view and height profile.

Fig. 2(a) shows the resonance wavelength shift of the MZI peak located at 1550 nm during the biofilm growth experiment, indicating a shift of up to 1.5 nm after 60 hours. The red shifts indicate the increase in surrounding RI and the biofilm formation by a new layer around the fiber. AFM imaging reported in Fig. 2(b) confirmed biofilm formation, revealing typical bacterial morphology and granular structure. A biofilm thickness of about 250 nm and granular surface (avg. roughness: 50 nm) were found, validating the fiber optic probe's capability for real-time detection via wavelength-encoded measurements.

Conclusions

In this work, we designed and fabricated a cost-effective fiber optic probe using an MZI overlap configuration on a single-mode fiber tip for chemical sensing. The probe was successfully tested for detecting *P. alcaligenes* biofilm growth, demonstrating advantages in simplicity, compactness, and robust wavelength-encoded measurements. Future research will focus on comprehensive sensing characterization and biofilm response analysis. Additionally, within the MoBeeFO project, the transducer will be explored for real-time monitoring of honeybee innate immunity dynamics.

Acknowledgements

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