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Towards a femtosecond laser micro-machined optofluidic device for distinguishing algae species

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ABSTRACT

We demonstrate a small device with a microfluidic channel and an integrated waveguide as a compact rudimentary tool for the detection, real-time monitoring, and potentially classification of algae. In order to reduce parasitic noise the micro-device used a curved subsurface optical waveguide to illuminate particles transiting through a microfluidic channel. The changes in the transmitted signal are monitored using a quadrant-cell photo-detector. The signals wavelets from the different quadrants are used to qualitatively distinguish different families of algae. Additional information, such as flow direction, is also provided. The channel and waveguide are fabricated out of a monolithic fused-silica substrate using a femtosecond laser-writing process combined with chemical etching. This proof-of-concept device paves the way for more elaborate femtosecond laser-based optofluidic micro-instruments incorporating waveguide network designed for the real-time analysis of cells and microorganisms in the field.

Keywords: algae detection, femtosecond laser, fused-silica, optofluidic device, waveguide

1. INTRODUCTION

Early detection of algae proliferation is an important environmental issue as well as a public health issue since some algae can be particularly virulent and toxic. The human health risk not only relates to drinking water but also bathing water: for instance, as a result of intensive human activities, eutrophication of freshwater systems has favored the presence of cyanobacteria or "blue-green algae" that can produce potent toxins. Exposure to these compounds can result in acute poisonings that may even be fatal [1]. At a lower dosage, some cyanobacterial toxins are tumor promoters, if they are repeatedly taken in with contaminated water [2]. Cyanobacterial toxins have been implicated in countless fatalities among domestic animals and adverse ecological effect [3]. Today, the identification of microbiological load generally requires that the specimen be collected and transported to a laboratory. Individual tests for each micro-organism are cumbersome and must be performed by trained personnel. The associated delay reduces our ability to react effectively to an outbreak.

There is strong need for monitoring methods that can identify threats posed by waterborne pathogens in fresh and marine waters within a much shorter time period than current practice and that can be widely spread. Further these devices should be operable by personnel with limited training. In this context, portable microfluidic–based compact instruments that can detect the presence of potential pathogens in various water bodies are particularly attractive.

The recent development of femtosecond processing of fused silica ([4][5][6]) providing for the multifunctional integration of waveguides, fluidic channels and micromechanical features [7] have opened new exciting avenues for the fabrication of compact and robust microorganism monitoring instruments . Although Foturan® glass has been proposed as a platform for Lab-on-a-Chip by others [8], we prefer fused-silica for this type of application due to its outstanding optical properties (the material is transparent to a broad range of wavelength) and its exceptional chemical stability.

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In a previous work [9], we demonstrated a microdevice which counted particles flowing through a micro-fluidic channel. This device relied on a conceptually very simple shadowgraphy implementation, where the flowing particles obstruct a light source. A salient feature of the device was the integration of a subsurface optical waveguide with a microfluidic channel in a monolithic fused-silica substrate.

This paper introduces an improved version of our original device and shows its t application to the monitoring and classification of algae. In this new implementation, we use a more powerful light detection technique based on four-quadrant photodetector.
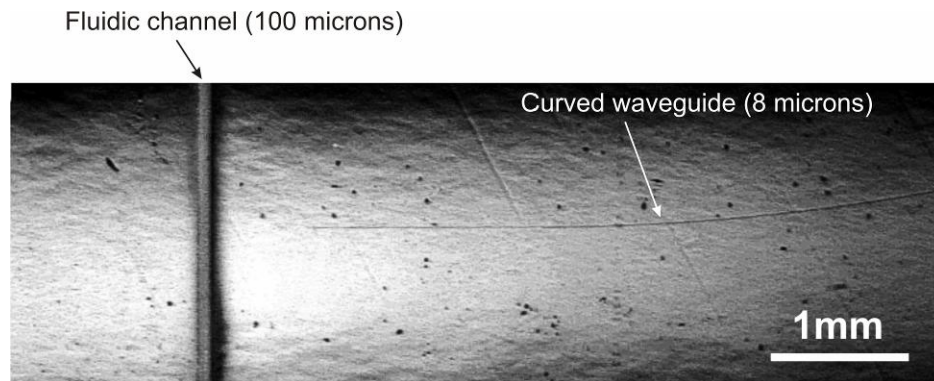

Fig. 1. The optofluidic device has a curved subsurface waveguide which is femtosecond laser-formed in the fused silica substrate. The waveguide starts near the top edge (not shown here) of the device and ends near a 100 μm wide fluidic channel.

2. METHOD

2.1 Fabrication

The bulk fused silica device comprises a curved waveguide with a 8 x 8 μm^2 cross section. To prevent uncoupled light from reaching the detector, the waveguide is 90-degree curved. The 18-mm radius of curvature is dictated by the delta n (68 10-3) of the waveguide. The waveguide ends perpendicular to a micro-channel with a 100 x 100 μm^2 cross section (see Fig. 1). A laser emitting at 1550 nm is used to probe the fluidic channel. At this wavelength the waveguide is single mode. The waveguide is fabricated in the bulk of the substrate (buried at a depth of 50 μm from the surface) using femtosecond laser pulses that locally increases the refractive index of fused-silica [4]. In the same laser-writing step, a volume region defining the micro-channel is structurally modified. This region is later preferentially etched away in an HF bath [5] to leave behind a surface with a trench dug out (more detail of the process can be found in refs [6][7]). A thick-film for covering the trench and fluidic interconnects is made from PDMS. It is treated with oxygen plasma in order to form a robust semi-permanent bond with the glass optofluidic device.

2.2 Setup and measurement

The packaged optofluidic device is interfaced with a syringe pump and a reservoir. To illuminate the fluidic channel locally, a fiber-coupled infrared laser source (Thorlabs S1FC1550) is aligned at the beginning of the waveguide. In order to detect and identify components as they flow past the waveguide, a quadrant-cell photoreceiver (New Focus 2903) is placed on the optical axis where the curved waveguide ends to measure the transmitted light (see Fig. 2). In the absence of micro-particles, the beam diameter emerging from the waveguide and falling on the active area of the sensor (3 x 3 mm^2) ranges from 300 to 500 μm (depending on whether the channel is filled with fluid or air).

Apart from the sum signal from all four quadrants of the photoreceiver, a signal measures the relative difference in the light distribution across the quadrant cells in the direction of the channel. The signals are collected at 1 kHz.

To evaluate the detection and identification performance of the optofluidic device, a high speed camera (AVT PIKE F-032B) is used to image micro-particles as they flow past the waveguide. This camera is to be used only during the development phase to assess false positives for instance or to calibrate the device.
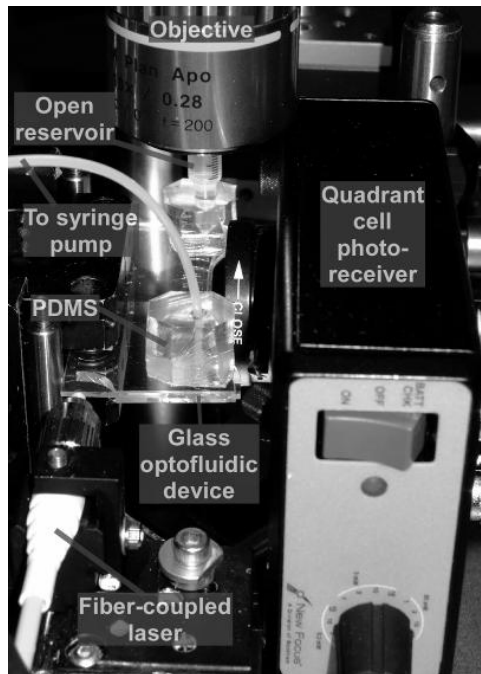
Fig. 2. The packaged optofluidic device has a fiber-coupled laser source and a quadrant cell photoreceiver aligned with the optical axes of the integrated waveguide.

A real-time Simulink model is used to control data acquisition and simultaneously trigger high-speed image acquisition (120fps) when a micro-particle is detected.

3. RESULTS & DISCUSSION

3.1 Detection of a single glass bead

Fig. 3 shows experimental results associated with a 50-micron glass bead as it transit through the channel Fig. 3a shows a collection of images as the glass bead transits by the waveguide (faint horizontal line on the right in each frame). Since the micro-particles under consideration are smaller than the beam diameter, we expect deviations in the light signal to arise from shadow effects (for the opposite regime where refraction dominates, see [9]). The collective signal in Fig. 3b shows a dip as the moving bead casts a shadow on the receiver.

Observing the difference signal reveals a wavelet with a peak and then a trough (Fig. 3c). When the bead traverses in the opposite direction, we detect a time-reserved wavelet as shown in Figure. 4. Hence by looking at the difference signal and determining the peak-trough sequence we can determine the direction of flow of the micro-particle. Due to axial symmetry of the bead, the wavelet is negatively symmetric about its mid-point.

Finally, from the extent of the wavelet we can estimate the size of the micro-particle provided its speed is known or vice-verse. By fabricating another waveguide further down the channel, we can correlate signals to determine the speed in our case from the image sequence we determine the bead diameter to be 61 +/- 4μm, and the speed to be 7.3+/- 0.2mm. Using this speed we estimate, from the extent of the difference signal wavelet, the size of the bead to be 66 +/- 8μm which is in agreement with the image- based value.
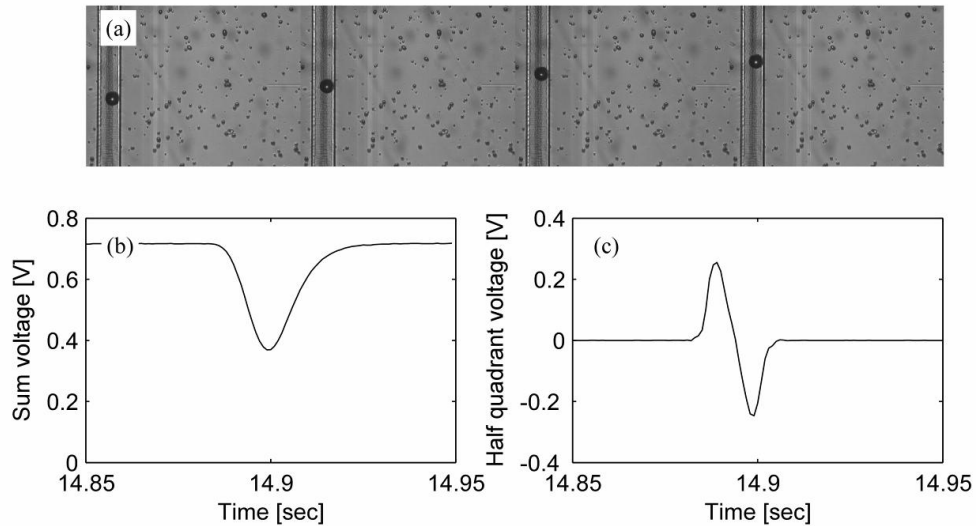
Fig. 3. Detection of a flowing bead. (a) Sequence of captured images as a bead flows past the waveguide (visible on the right of every frame). (b) Total signal from all four quadrants of the photoreceiver. (c) Difference signal from two halves of the detector along the direction of the channel.
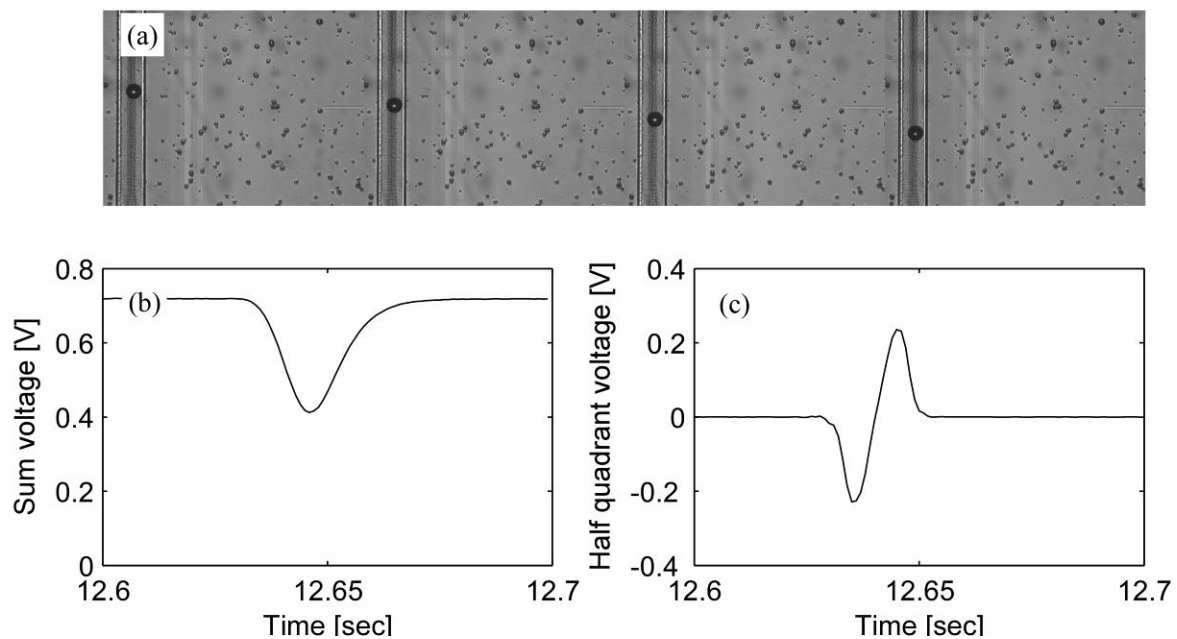

Fig. 4. Detection of a flowing bead. (a) Sequence of captured images as a bead flows past the waveguide. (b) Total signal from all four quadrants of the photoreceiver. (c) Difference signal from two halves of the detector along the direction of the channel.

3.2 Distinguishing various algae cells

Next, a set of five different species of algae with different geometries but similar sizes were made to flow through the optofluidic device. Their geometrical properties are illustrated in the left column of Fig.5. The other columns show representative images and measurements obtained for each of these families of algae. Note that the flow velocity was not constant during these various experiments. Also note that in the measurement from Fig. 4a, the Cyanothece is traveling down the channel, whereas measurements in Fig. 4b-Fig. 4e the algae travel in the opposite direction.
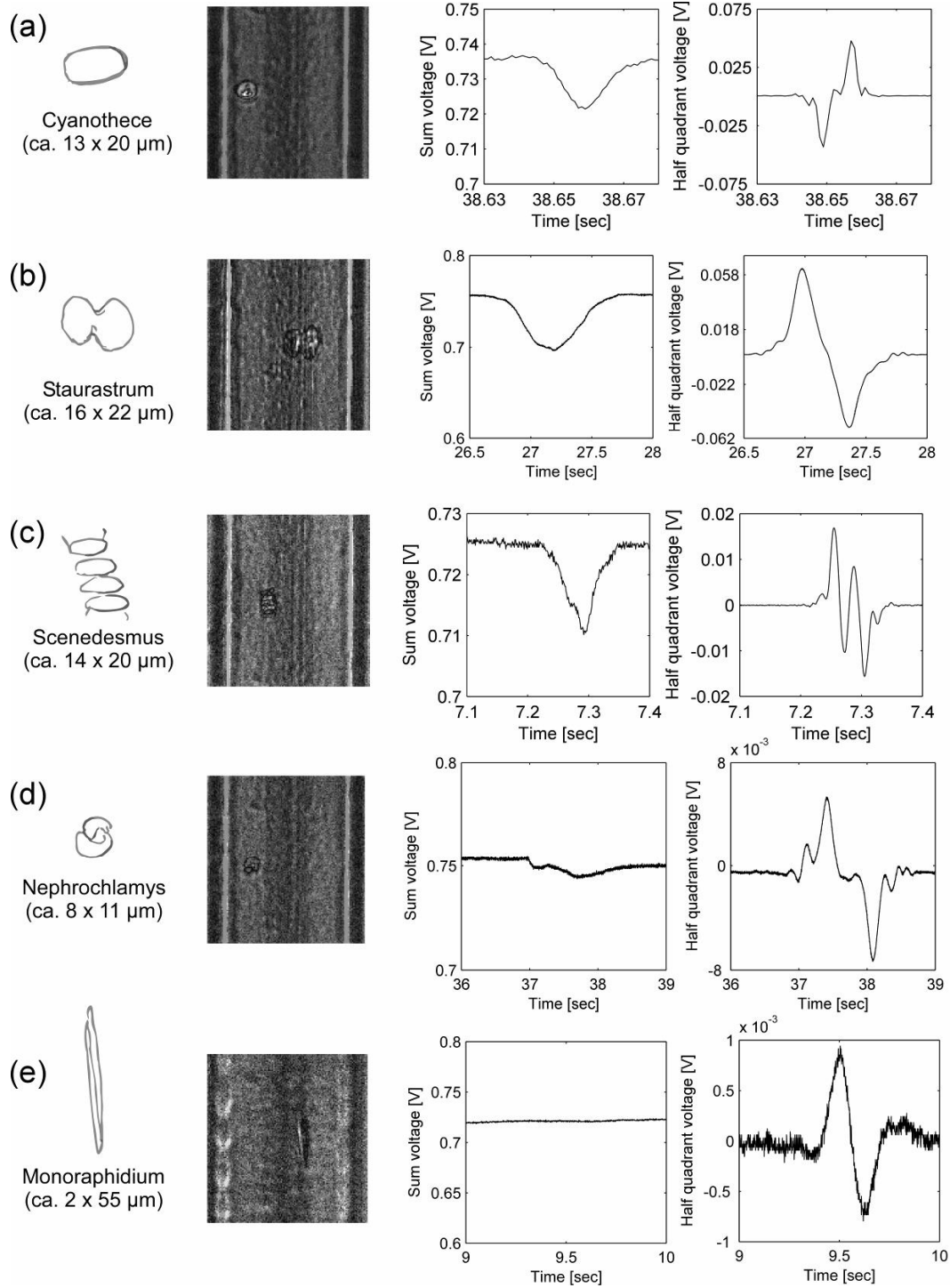
Fig. 5. Algae cells of various geometries but similar sizes are detected with the optofluidic device. For different types of cells, the difference signals give rise to distinct wavelets.

The sum signals show some attenuation which mark the presence of algae, The difference signals give rise to distinct wavelets which a qualitative mean of distinguishing the various type of algae. The distinct nature of the wavelets is related to the particular geometry, and associated optical properties, of the algae. These distinguishing wavelets are centrosymmetric (with their respective mid-points).

If the cell is not axially symmetric and/or happens to be rotating as it flows past the waveguide the wavelet centro-symmetry is broken. Qualitatively, we can easily distinguish Scenedesmus and Nephrochlamys A thorough quantitative analysis is underway to determine to what extent each of these algae type can be correctly identified.

4. CONCLUSIONS & PROSPECTS

We have fabricated a functioning compact femtosecond laser-machined optofluidic device that can detect, monitor, and qualitatively distinguish algae species. Using a quadrant-cell photoreceiver, we have shown that algae generate distinctive wavelet that can be associated with the particular algae geometry. These results demonstrate that even a single waveguide instrument can perform rudimentary algae identification function Adding another waveguide, we expect to extract more quantitative information, calibrated algae size , flow rate, etc. Our field-compatible compact optofluidic device will provide most of the information provided by more complex image-based systems. Furthermore, the approach based on integrated optics has the potential to provide additional information including specific optical properties such as fluorescent response.

The optofluidic device has important characteristics which are made possible when fused-silica is micro-machined with femtosecond laser. The single-mode curved waveguide yields robust detection results particularly when difference signals are used. Since the waveguide is only 8 x 8 μm^2, the micro-channel device could be further miniaturized by a factor of ten. Furthermore, the vicinity of the waveguide to the channel can be tuned to control the portion of the channel height that is illuminated to optimize detection or study refractive properties. The exclusive use of free-space optics involved in the setup, and considering that laser diode source and quad-cells can be packaged with the device open interesting prospects for field-based pathogens detection devices.

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