MICROFLUIDIC DEVICES MADE OF UV-CURABLE GLUE (NOA81) FOR FLUORESCENCE DETECTION BASED APPLICATIONS

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ABSTRACT

The UV-curable adhesive NOA81 (Norland Optical Adhesive) is a promising liquid photopolymer for low-cost microfluidic chip production. Different fabrication methods for NOA81 are suggested [1,2]. However, optical characterization for fluorescent based detection systems is missing. We present a comparison of fluorescent emission spectra of various NOA types as well as the time evolution of the spectra of the most promising one - NOA81. We report on a simple rapid prototyping technique to produce chips for fluorescent based capillary electrophoresis (CE). As an application, we show the CE-injection of a fluorescent dye, and its detection by fluorescence microscopy.

KEYWORDS: NOA81, Fluorescence Emission Spectra, Microfluidic Stickers, Microfluidics, Capillary Electrophoresis

INTRODUCTION

Microfluidic channels are often fabricated by silicon or glass bulk micromachining, which is expensive, relatively complex and shows limitations in the channel geometries. In recent years polymers like polydimethylsiloxane (PDMS) or SU-8 photoepoxy have become popular for microfluidic applications due to their high flexibility, ease of fabrication and high reproducibility by rapid prototyping methods [3]. Compared to PDMS, the most widely spread polymeric material for microfluidic channels, NOA81 has better chemical resistance to organic solvents, is impermeable to air and water vapor, is less prone to swelling upon contact with fluids, and surface treatments (for example oxygen plasma) are more stable [1,4,5]. NOA81 is still a new material for microfluidic applications and need to be characterized. To perform fluorescent based detection measurements the auto-fluorescence of the chip material is of crucial importance and induced the comparison of fluorescence spectra of different NOAs.

EXPERIMENTAL

In the visible light range, NOA81 is perfectly transparent. For more specific optical characterization of the material, samples of different NOA types of uniform thickness were analyzed with an "Infinite M200" spectrofluorimeter (Tecan Group Ltd., Switzerland).

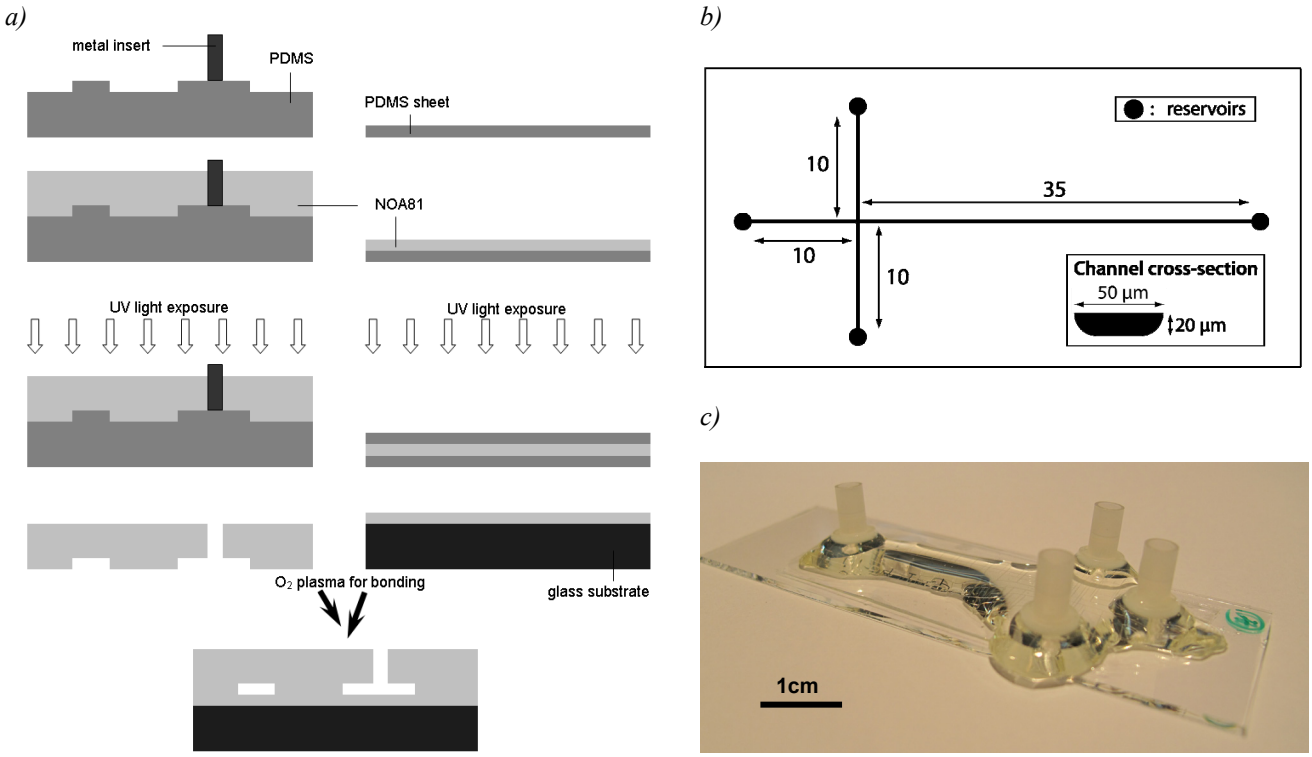

Figure 1: a) The original glass master is fabricated in the cleanroom by isotropic wet etching, on which PDMS is molded. NOA81 is casted and cured on the PDMS. The structured part is pressed on the substrate covered with a cured thin film of NOA81 to form a microfluidic capillary. Permanent bonding can be achieved by an additional O_2-plasma treatment before assembly; b) Design of microfluidic capillary electrophoresis chip. The channel lengths are given in mm; c) Image of the microfluidic chip for capillary electrophoresis experiments made by rapid prototyping of NOA81.

The devices for CE consist of a classical T-channel (50 µm wide, 20 µm deep) and were fabricated by rapid prototyping of NOA81 as explained in Figure 1a. The glass microchannels were etched in a cleanroom, and then PDMS (Sylgard 184, Dow Corning, USA) was molded onto it. Subsequently NOA81 was casted and then cured under the UV-lamp on the structured PDMS master. In parallel also a thin film of NOA81 was cured, sandwiched between two PDMS sheets. After unmolding, the thin film of NOA81 was mounted on a glass substrate and then covered with the structured part of NOA81 to form a microfluidic capillary. Permanent bonding can be achieved by an additional O_2-plasma treatment of the bonding surfaces before assembly.

The CE experiments were performed with a Zeiss Axiovert S100 fluorescence microscope (Carl Zeiss AG, Germany) equipped with a Xenon 100W power light source and a high-sensitivity CCD camera (Kappa AG, Germany). A programmable HVS 448-6000D 8-channels high-voltage supply (LabSmith Inc., CA, USA) allowed to apply three different voltage sequences for the sample loading and injection inside the separation channel of the microchip. We set the injection method and the corresponding voltages according to [6]. The current monitoring method was used to measure the electroosmotic flow (EOF) velocity in the NOA81 channel [7]. Chemical products used for the CE experiments were purchased from Sigma-Aldrich Co. (MO, USA).

RESULTS AND DISCUSSION

At an excitation wavelength of 470 nm, the fluorescence emission of NOA81 is higher than PDMS (see Figure 2a), but it is only half of the intensity of the NOA recommended for fluorescence applications (NOA63), which was found not to be suitable for fluorescence measurements [5]. At the excitation wavelength of 546 nm, NOA81 has comparable fluorescence emission as PDMS (see Figure 2b). The low auto-fluorescence of NOA81 at 546 nm makes it as suitable as PDMS for fluorescent based detection systems. Figure 3a shows the evolution of the fluorescent emission spectrum of NOA81 at the excitation of 470 nm. The intensity is decreasing after fabrication and is stable after 8 days. For the excitation at 546 nm the spectra are stable already after 2 days (see Figure 3b).

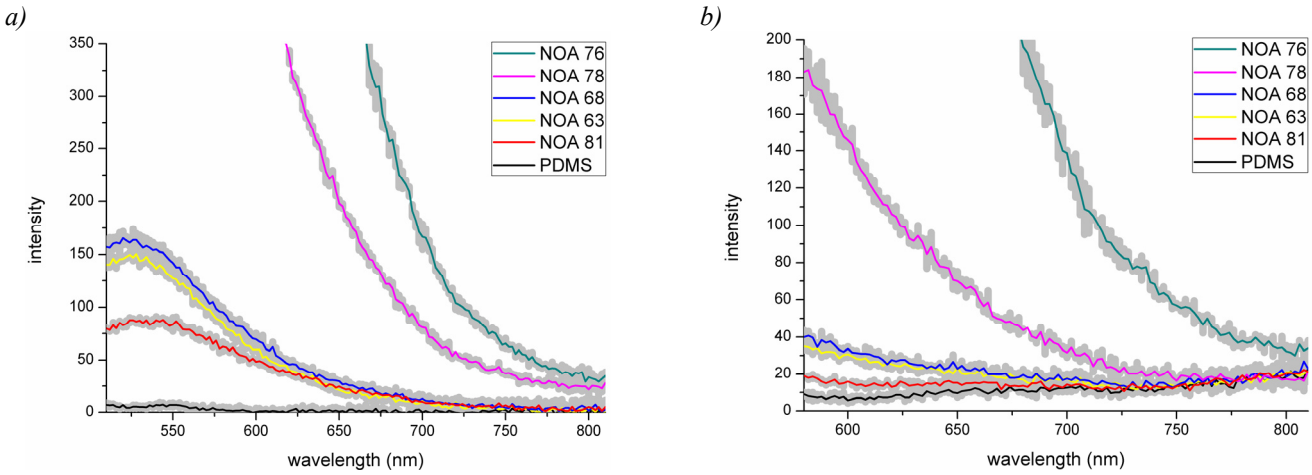

Figure 2: Comparison of fluorescent emission spectra of different NOAs and PDMS as reference value. The spectra are recorded 20 days after chip fabrication. The grey region around each plot represents the standard deviation of the measurement; a) excitation at λ_{ex}=470nm, b) excitation at λ_{ex}=546nm.

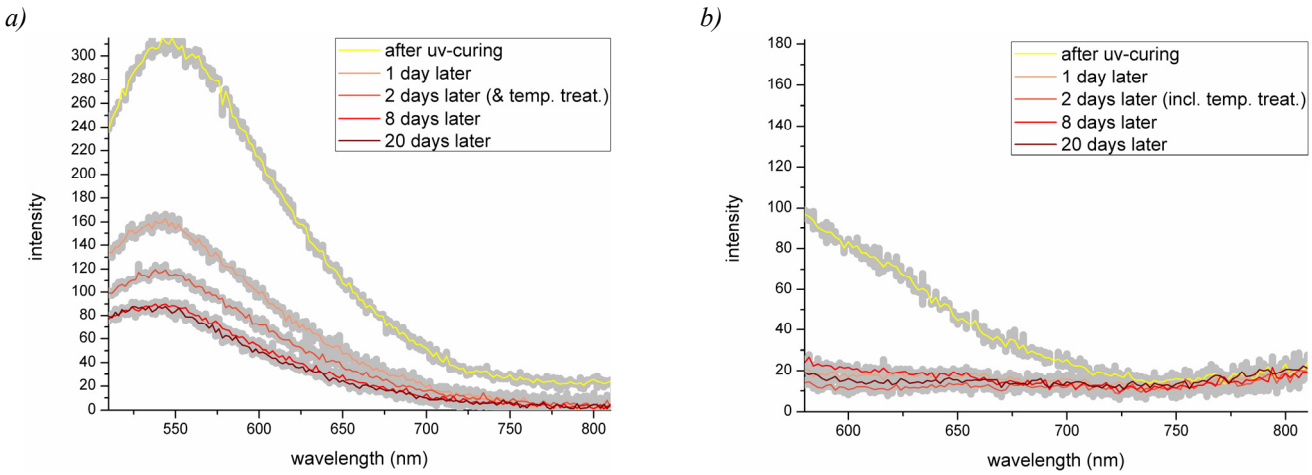

Figure 3: Evolution of fluorescent emission spectrum of NOA81 for excitations at λ_{ex}=470nm (a) and λ_{ex}=546nm (b); directly after the UV-curing, 1 day after the fabrication, 2 days later and after a temperature treatment of 60°C for 2h, 8 and 20 days later. The intensity is decreasing and stable 8 days after the fabrication. The grey region around each plot represents the standard deviation of the measurement.

For the fluorescence detection evaluation, we injected a plug of 200 µM Rhodamine B in the separation channel filled with a 10 mM sodium phosphate buffer at pH 7.0. The fluorescent electropherogram and the corresponding intensity picture of the Rhodamine B plug are presented in Figure 4a. The peak intensity of the dye is well defined and the background level very low.

The electroosmotic flow was found to be very stable during the experiments. This was confirmed by the low-noise current measurement of the EOF velocity (see Figure 4c). From three independent current measurements, we calculated a corresponding electroosmotic velocity of v_{eo} = 1.4 mm·s^{-1} and a corresponding electroosmotic mobility of $\mu_{eo} = 4.15 \cdot 10^{-8}$ m^2(V·s)$^{-1}$ ($\mu_{eo} = L^2 \cdot (V \cdot t)^{-1}$).

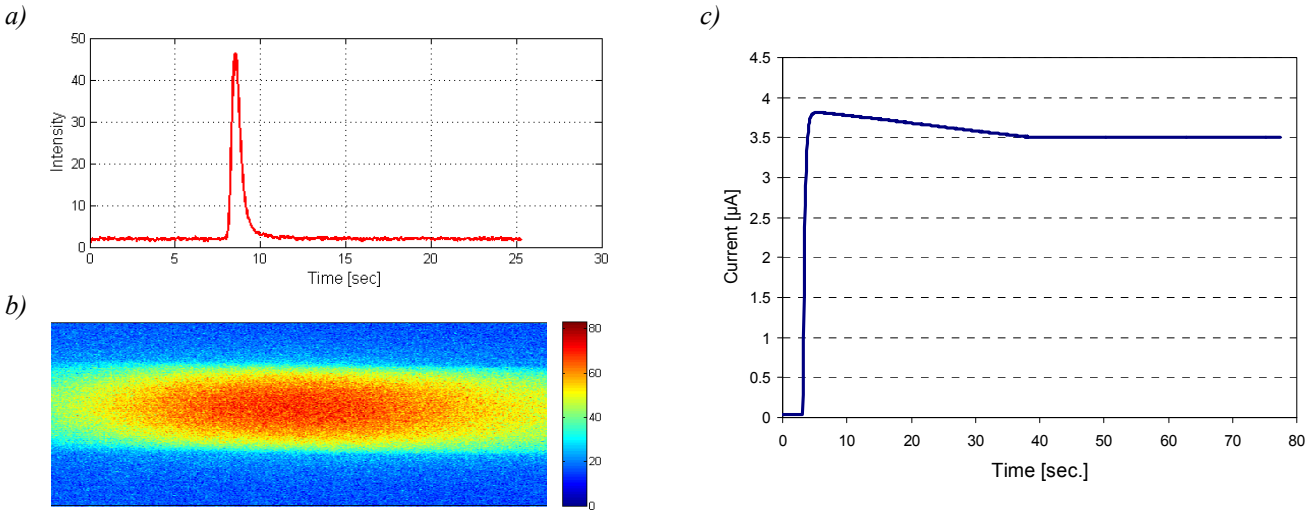

Figure 4: a) CE injection of a sample of Rhodamine B (200 µM, λ_{ex}=540 nm, λ_{em}= 625 nm) in a 10 mM sodium phosphate running buffer at pH 9.0. An electric field of 330 V/cm was applied along the NOA separation channel. The plot shows the mean intensity measurement of a well defined plug over time, 6.2 mm away from the channel intersection; b) Relative pixel intensity picture of the Rhodamine B plug captured by the CCD camera. c) Plot of the EOF measurements using the current monitoring method. A voltage drop of 1500 V (330 V·cm^{-1}) was applied between the reservoirs to load the 10% diluted phosphate buffer at pH 7.0 into the channel. The monitoring of the current gives the time t needed to fill the 45 mm long channel with the diluted buffer and hence the EOF velocity v_{eo}. Here, t = 33 sec corresponds to v_{eo} = 1.4 mm·s^{-1}.

CONCLUSION

The promising result of the fluorescence spectrum measurement and the CE experiments opens the way of using NOA81 as a simple and stable rapid prototyping material for fluorescence-based microfluidic applications. We are currently working on multiple dyes separations**.**

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