ARTICLE

Technical concept of 3D printed fluidic biosensor with polydimethylsiloxane chip based on fluorescence detection system

Jan Zitka [1,2], Zbynek Heger [1,2], Ondrej Zitka [1,2], Vojtech Adam [1,2] and Rene Kizek [1,2,*]

[1] Department of Chemistry and Biochemistry, Laboratory of Metallomics and Nanotechnologies, Mendel University in Brno, Zemedelska 1, CZ-613 00 Brno, Czech Republic - European Union;

[2] Central European Institute of Technology, Brno University of Technology, Technicka 3058/10, CZ-616 00 Brno, Czech Republic - European Union;

* Author to whom correspondence should be addressed; E-Mail: kizek@sci.muni.cz;

Received: 2.2.2015 / Accepted: 9.3.2015 / Published: 1.4.2015

There is a rapidly growing interest in low-cost, fast and sensitive biosensors. In particular, direct determination of important metabolites, serving as biomarkers of various pathological states can significantly enhance the treatment successes. In our study, we introduce a technical concept of a 3D printed biosensor, which employs polydimethylsiloxane chip with volume of 50 μL as an inert and optically clear reservoir for recognition element and fluorescence detection. By using a 3D printing technology, low production cost and high crafting reproducibility were achieved. Due to a presence of controlled electromagnet, the biosensor can be utilized for a broad spectrum of applications, based on paramagnetic nano- or microscaled materials.

Keywords: 3D printing; acrylonitrile butadiene styrene; biomarkers; fluorescence

1. Introduction

A biosensor is an analytical device, which can be easily employed for detection of broad spectra of analytes, including molecules, bacteria, viruses, etc. [1]. The development of biosensors provides an accurate, sensitive and specific detection, together with portability and the ability to furnish continuous real time signals of analytes levels in various matrixes. According to the definition, firmly established in the analytical field "biosensor" is a detection system that relies on a biomolecule for molecular recognition and a transducer to produce an observable output [2]. Molecular recognition element (e. g. antibodies, nucleic acids, receptor proteins, whole cells or bacteria) is fundamental component that interacts with the studied analyte [3]. Currently, very promising recognition elements are produced as a result of driven recombination in biological engineering. The transducers can work in different ways, such as physicochemical, optical, piezoelectric, electrochemical, etc.). Their role is the transformation of obtained signal, which goes from the analyte-recognition element interaction [4]. The transformation leads to more easily measured and quantified signals. Nowadays, few commonly used biosensors such as glucose biosensor are in every-day use all over the world. The development of novel biosensors for easy, fast and cheap detection of various types of biomarkers for cancer, oxidative stress or metabolical disorders, can significantly enhance the treatment successes and the survival rates. From these reasons, we suggested a technical concept of 3D printed biosensor with polydimethylsiloxane (PDMS) chip, utilizing the fluorescence detection, which is one of the most sensitive, to serve as a universal platform for detection of various types of biomarkers. Proposed biosensor was particularly designed to work in coupling with quantum dots-based labeling, which offers exceptional quantum yields. Various recognition biomolecules can be easily labeled by quantum dots, thus high specifity and boosted sensitivity can be achieved.

2. Results and Discussion

2.1. PDMS chip

PDMS belongs to a group of polymeric organosilicon compounds, commonly referred to as silicones. PDMS is optically clear, inert, non-toxic and non-inflammable and these attributes makes PDMS ideal material for large scale of applications, such as production of contact lenses and other medical devices, heat-resistant tiles and it is also used as antifoaming agent in food [5]. In our case, beneficial attributes of PDMS made us to decide to utilize this material for preparation of chip with reservoir where the interactions between analyte and recognition element are carried out. In particular, inertness and optical clearness are fundamental for fluorescence-based biosensors. PDMS chip was produced in 3D printed pattern (Fig. 1a, b), which was smoothed for creating of chips without surface imperfections (Fig. 1c, d). 3D printed patterns are cheap and their manufacturing is highly reproducible, thus their utilization offers few advantages, such as possibility of low-cost, large-scale production of chips with identical attributes.

Resulting volume of reservoir in chip was 50 µL and comprised an input connected with reservoir and an output for waste (Fig. 2). Both, the input and the output were tightly connected with syringe needles, to avoid an unwanted leakage of sample.

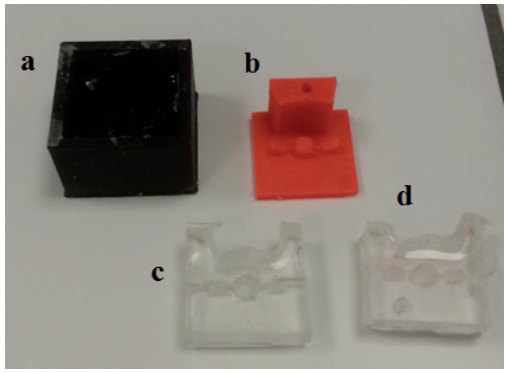

Figure 1: Photography of the patterns used for chip fabrication. The patterns (a, b) were manufactured by using 3-D printing, using acrylonitrile butadiene styrene (ABS) and (c, d) PDMS chips.

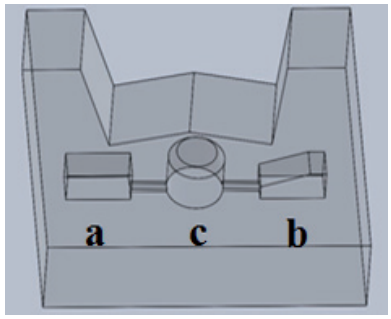

Figure 2: Schematic illustration of polydimethylsiloxane chip with (a) input, (b) output and (c) reservoir with volume of 50 µL.

2.2. Construction of fluidic device

As it is shown in a schematic drawing in Fig. 3A, the entire biosensor contained following 3D printed parts: holder for PDMS chip (3), emission (9) and excitation (10) filters, input (4) and output syringes (5) and part for pressing the PDMS in the holder (1). Other parts were purchased and were as follows: emission bandpass filter (λ = 550(55) nm) (8) and shortpass excitation filter (λ = 425 nm) (7), UV-Vis optical fibers with diameter of 3 mm (6) and electromagnet (11). In the case of excitation part, there is a slot for high-power LED (λ = 400 nm) as an excitation source. In emission part, the biosensor was connected with a control unit Arduino, which was linked to a LED driver and UV-Vis photodiode with photomultiplier. As system was designed to be flow-through, the leakage of analyzed liquid forms a large problem. Hence the PDMS chip with syringe needles was pressed between the holder and the pressing part by using four M3 screws. Such seal allowed for washing of the chip during the load tests without observable leakage. The electromagnet can be employed for both - immobilization of paramagnetic particles during magnetic separation or stirring of samples with paramagnetic materials, and is controllable through the control unit.

As is mentioned above, biosensor is designed particularly for manipulation with paramagnetic nano-, microscaled materials. Due to

communication between electromagnet and control unit, paramagnets can be easily immobilized or stirred, in dependence on application. Thus, the device can be employed for a broad spectrum of applications, such as particle-based immunoassays [6], magnetic separation of low--mass compounds [7], direct isolation of viral particles [8] or study of complex paramagnetic nanostructures, comprising fluorescence labels [9].

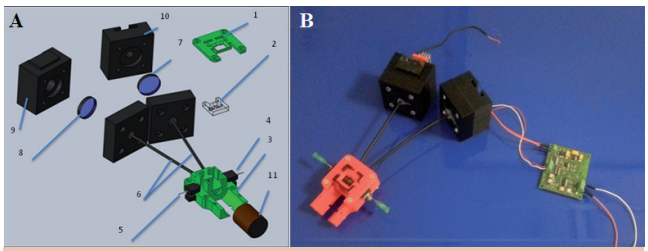

Figure 3: (A) The schematic drawing of individual parts employed for biosensor, where 1 stays for pressing part, 2 for PDMS chip, 3 for PDMS holder, 4 for input, 5 for output, 6 for optical fibers, 7 for shortpass excitation filter, 8 for bandpass emission filter, 9 for emission filter platform and 10 for excitation filter platform, (B) the assembled biosensor ready-to-use

3. Experimental Section

3.1. Fabrication of 3D-printed biosensor device

The platforms for PDMS chip, excitation, emission filters and optical fibres were manufactured by using 3D-printing technology (acrylonitrile butadiene styrene - ABS as the production material) by using 3D printer PROFI3DMAKER (Aroya, Straznice, Czech Republic).

3.2. Preparation PDMS chip

PDMS chip was fabricated in the pattern, made from ABS to produce the resulting structure with reservoir with the volume of 50 µL. Prior to use in biosensor, PDMS (Sylgard 184, Dow Corning, Midland, MI, USA) was evacuated to remove the unwanted air bubbles, which can interfere with optical light paths. For one chip 2 mL of PDMS and 100 uL of curing agent were mixed. Resulting mixture was further evacuated, filled into 3D printed pattern and left at 25 °C.

3.3. Detection system parts

As excitation source was employed power LED (λ = 400 nm, optical power 200 mW) (Roithner LaserTechnik, Vienna, Austria). Emission bandpass filter (λ = 550 ± 55 nm) and excitation shortpass filter (λ = 425 nm) were purchased from Semrock (Rochester, NY, USA). Optical fibers (UV-Vis transparent, core fiber diameter 1960 µm, numerical apparatus 0.5) were obtained from Edmund optics (Barrington, NJ, USA). The system was driven by the control unit Arduino linked to the LED driver. Evaluation of signal from UV-Vis photodiode S3991-01 (Hamamatsu Photonics, Hamamatsu, Japan) was done by multifunction two channels amplifier board (Digi board, Sglux, Berlin, Germany).

4. Conclusions

Biosensors can serve as promising tools for fast, cheap and sensitive analyses of broad spectrum of analytes. There exists many technical concepts and ways how to design and fabricate diverse platforms. 3D printing in combination with non-toxic, inert materials offers a lot of advantages, such as low production costs, biocompatibility, significant reproducibility and fast crafting time. Moreover, in conjunction with fluorescence detection, high sensitivity can be achieved.

Acknowledgments

This work was financially supported by project SPINCANCER NT/14337.

Conflicts of Interest

The authors declare they have no potential conflicts of interests concerning drugs, products, services or another research outputs in this study. The Editorial Board declares that the manuscript met the ICMJE „uniform requirements“ for biomedical papers.

References

1. Banica, F.-G., What are Chemical Sensors? In Chemical Sensors and Biosensors, John Wiley & Sons, Ltd: 2012; pp 1-20.
2. Heger, Z.; Zitka, O.; Fohlerova, Z.; Rodrigo, M.A.M.; Hubalek, J.; Kizek, R.; Adam, V., Use of green fluorescent proteins for in vitro biosensing. Chemical Papers 2014.
3. Oliveira, O.N.; Iost, R.M.; Siqueira, J.R.; Crespilho, F.N.; Caseli, L., Nanomaterials for Diagnosis: Challenges and Applications in Smart Devices Based on Molecular Recognition. Acs Applied Materials & Interfaces 2014, 6, 14745-14766.
4. Wu, C.S.; Du, L.P.; Zou, L.; Zhao, L.H.; Huang, L.Q.; Wang, P., Recent advances in taste cell- and receptor-based biosensors. Sensors and Actuators B-Chemical 2014, 201, 75-85.
5. Lotters, J.C.; Olthuis, W.; Veltink, P.H.; Bergveld, P., The mechanical properties of the rubber elastic polymer polydimethylsiloxane for sensor applications. Journal of Micromechanics and Microengineering 1997, 7, 145-147.
6. Ng, A.H.C.; Choi, K.; Luoma, R.P.; Robinson, J.M.; Wheeler, A.R., Digital Microfluidic Magnetic Separation for Particle-Based Immunoassays. Analytical Chemistry 2012, 84, 8805-8812.
7. Cernei, N.; Heger, Z.; Kopel, P.; Skladanka, J.; Zitka, O.; Adam, V.; Kizek, R., Isolation of Biogenic Amines Using Paramagnetic Microparticles Off-Line Coupled with Ion Exchange Liquid Chromatography. Chromatographia 2014, 77, 1451-1459.
8. Heger, Z.; Cernei, N.; Guran, R.; Michalek, P.; Milosavljevic, V.; Kopel, P.; Zitka, O.; Kynicky, J.; Lany, P.; Adam, V., et al., gamma-Fe_2O_3 Magnetic Core Functionalized with Tetraethyl Orthosilicate and 3-Aminopropyl Triethoxysilane for an Isolation of H7N7 Influenza Serotype Virions. International Journal of Electrochemical Science 2014, 9, 3374-3385.
9. Heger, Z.; Kominkova, M.; Cernei, N.; Krejcova, L.; Kopel, P.; Zitka, O.; Adam, V.; Kizek, R., Fluorescence resonance energy transfer between green fluorescent protein and doxorubicin enabled by DNA nanotechnology. Electrophoresis 2014, 35, 3290-3301.

The article is freely distributed under license Creative Commons (BY-NC-ND). But you must include the author and the document can not be modified and used for commercial purposes.