Passive Lab-in-a-Foil Devices for Phaseguiding and Multiple Displacement Amplification of DNA

Eriksen, Johan; Marie, Rodolphe ; Schira, Julien; Vincent, Nadine; Kristensen, Anders

Published in:
Proceedings of NanoBioTech - Montreux 2013

Publication date:
2013

Passive Lab-in-a-Foil Devices for Phaseguiding and Multiple Displacement Amplification of DNA

Johan Eriksen[a], *Rodolphe Marie*[a], *Julien Schira*[b], *Nadine Vincent*[b], *Anders Kristensen*[a]
[a]*DTU Nanotech, Technical University of Denmark, DK-2800 Kongens Lyngby, Denmark*
[b]*FASTERIS SA, CH-1228 Plan-les-Ouates, Switzerland*
E-mail: joher@nanotech.dtu.dk

The amplification of DNA via multiple displacement amplification (MDA) requires a minimal sample volume to enable further analysis of the sample. It can be a challenge to realize a microfluidic chip handling microliter volumes of fluids. In this paper, we present the fabrication and operation of a passive device for MDA of DNA. Furthermore, an on-chip amplification of λ phage DNA is presented.

In previous work, MDA of DNA has been performed on a microfluidic device containing active push-up valves [1]. We present a fast and cheap fabrication protocol of passive Lab-in-a-Foil devices by using hot embossing. The presented polymer device consists of a micro fluidic channel, the bus, connecting three inlets to a waste reservoir and four reaction chambers parallel connected to the bus by nanoslits with the height of 500 nm, see Figure 1. Embossing large protrusions in a polymer for fabricating large microfluidic chambers is not trivial. Furthermore, the MDA protocol is a stoichiometric reaction where monitoring and measuring volumes of different reagents is crucial. We want to start off with empty chambers that are gradually filled with reagents in a serial manner. Filling such a microfluidic chamber without encapsulating air bubbles isn't trivial either [2]. We enable the controlled filling of the chambers and ease the fabrication of the device by using an array of pillars as showed in Figure 1. Each pillar has a radius and height of 10 μm and facilitates a preferred flow-direction due to minimizing of surface tension and thereby guide the filling of the fluids involved in the MDA protocol – phaseguiding.

The lab-on-a-Foil devices were fabricated by using the Compact Nanoimprint Tool, CNI Tool, from NIL Technology, by thermal imprinting a 4" silicon stamp with 10 μm and 500 nm protrusions into a TOPAS® 6013 Cyclic Olefin Copolymer (COC) foil. Afterwards, a flattened lid of the COC foil was thermal bonded to the structured foil. The total fabrication time (device imprint, lid flattening and wafer bonding) of a wafer containing four devices was 90 minutes and the final device is showed in Figure 1.

The pillars are arranged in a rectangular array. The fluid is pinched on the pillars and the front of the fluid will be guided transverse in the reaction chamber. The flow in the wetted reaction chamber is not guided by the pillars and thereby allows a two-dimensional flow and diffusion. Figure 2 shows a sequence of images where MilliQ water is filled into the reaction chamber and guided by the pillars. Furthermore, the pillars also prevent collapse of the lid and adding another functionality compared to the phaseguiding presented in [2]. As a proof-of-concept, 28 pg of λ phage DNA was amplified > 500 times by sequentially exchanging the reagent in the bus by a pressure-driven flow and thereby adding the solution containing the DNA, the denaturation solution, the neutralization solution and polymerase solution to the reaction chamber from the three inlets. In all solutions 0.01 % Triton-X 100 was added in order to lower the hydrophobic behavior of the COC polymer foil and thereby lowering the adsorption of DNA [3]. Sequencing data showed a low contamination with 93 % of the sequenced DNA mapping to the genome of λ phage DNA. This work was supported by the EC-FP7 HEALTH IP "CELL-O-MATIC" (No. 278204)

1. H. C. Fan, J. Wang, A. Potanina, S. R. Quake, Nature Biotechnology, **29** (2011) 51-57
2. P. Vulto, S. Podszun, P. Meyer, C. Hermann, A. Manz, G. A. Urban, Lab on a Chip, **11** (2011) 1596-1602
3. E.K. U. Larsen, N. B. Larsen, Lab on a Chip, **13** (2013) 669-675

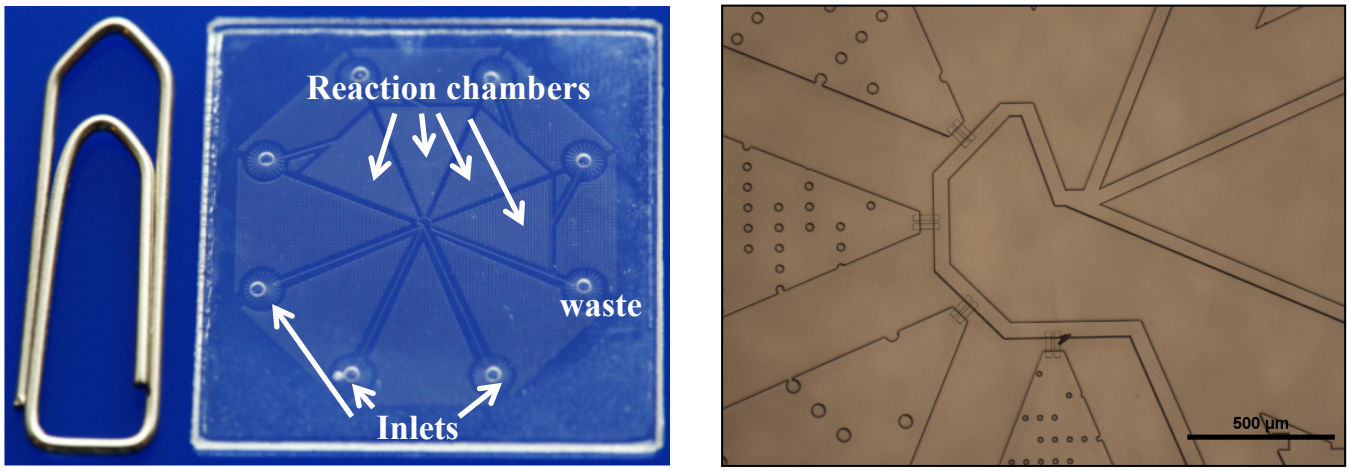

Figure 1. Left: *Image showing the final chip with the dimension of 2.5 x 2.5 cm. The chip contains of a micro fluidic channel connecting three inlets and a waste reservoir and four separated reaction chambers are connected to the microchannel by nanoslits of the height of 500 nm where the microstructures are at high of 10 µm.* ***Right:*** *Microscope image showing a zoom of the center of the polymer chip.*

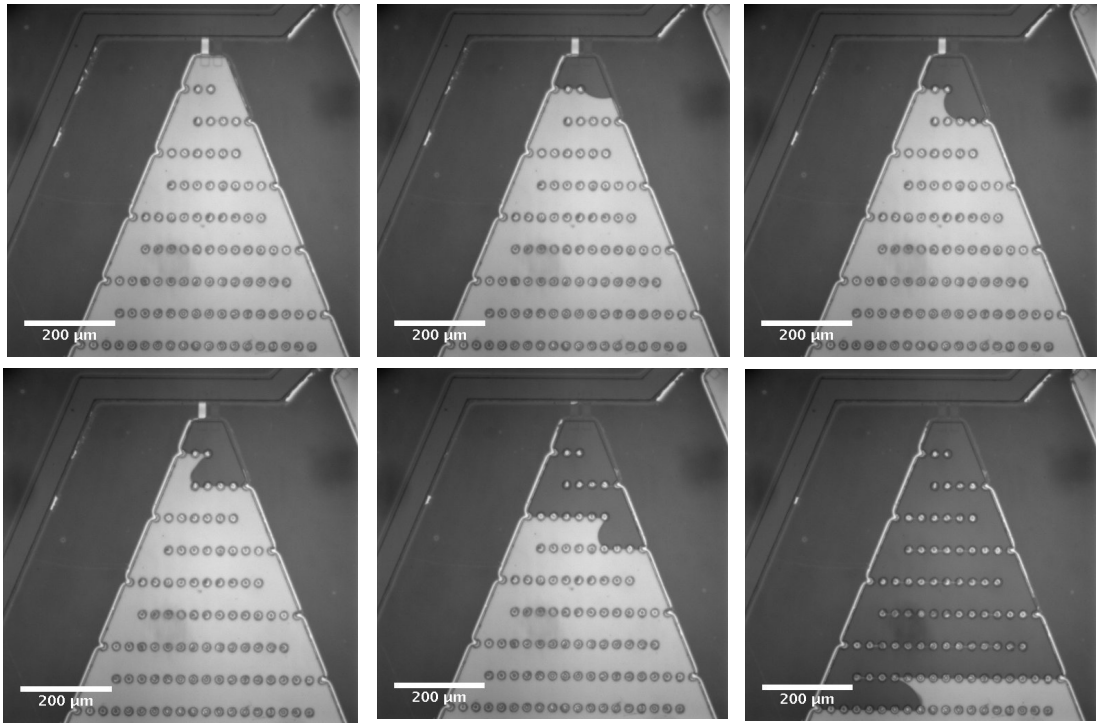

Figure 3. *Sequence of microscope images showing the principle of phaseguiding. The array of pillars facilitates a preferred flow-direction. By removing pillars on each row of pillars bursting from one row to the next is specified in the design. The filling showed in the images is performed within 30 s corresponding to a volume of ~16 nL.*