Development of a microfluidic System for DNA Extraction and Analysis

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Abstract

The analysis of DNA molecules holds significant importance in various fields, such as forensics, genetics, medicine, and evolutionary biology. The quality of DNA analysis is substantially contingent upon the purity, integrity, and quantity of the sample material, which poses a challenge when dealing with limited sample volumes. To simplify the handling of small sample quantities and ensure higher quality, the current trend is towards integrating extraction from the collected sample and DNA analysis within a single device. Lab-on-a-Chip (LOC) systems exhibit significant potential in this regard, as they perform complex processes on compact miniaturized systems, operating at the same scale as DNA molecules themselves. [1,2]

This study presents an integrated micro- and nanofluidic device for on-chip DNA extraction and analysis. It is based on a LOC system for on-chip sample preparation and a laser-based analytical method for in-line DNA analysis.
The chips are mainly manufactured in a clean room using direct nanoimprinting, allowing the patterning of large (hundreds of microns) and small (tens of nanometers) 2D and 3D structures within one device in just one fabrication step. For this, a silicon stamp is first fabricated using different micro and nanolithography methods. Depending on the lateral resolution (micro or nano), photolithography or electron beam lithography is used for the 2D structures. Dose-modulated electron-beam lithography allows for additional complex 3D structuring. [3]
After the chips have been manufactured, external heating and magnetic field are connected adding extra functionality to the polymer-based chip, enabling single-use chips for various applications while keeping the fabrication simple and low-cost. In this way we can fabricate microfluidic devices down to nanometer resolution in only two minutes. [4]

The developed LOC plastic device comprises two integrated sections as illustrated in Figure 1: the DNA extraction part and the DNA analysis part.
In the extraction section, DNA molecules selectively bind to magnetic particles, which can be efficiently transferred into a clean buffer solution using external magnets, thereby purifying the samples. In the analysis section, labeled DNA molecules are stretched within nanochannels and read out using a laser system. The system has been applied for different research purposes. We investigated the dynamic behavior of DNA molecules confined in nanochannels and studied the influence of two factors, ion concentration and channel diameter, on DNA stretching and velocity. In addition, we showed the proof of concept of using the system for various biomedical applications, including virus identification and the study of genome intercalation in DNA in tumor cells, showcasing the versatility and capabilities of this system. [5,6]

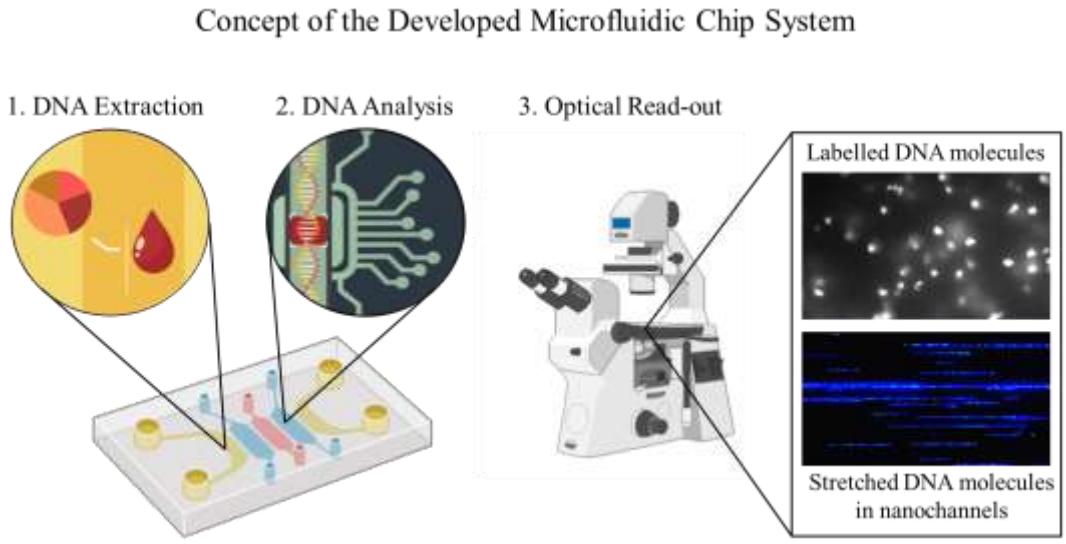

Figure 1: Concept of the microfluidic chip for DNA extraction and analysis. First, the DNA sample is applied to the microfluidic chip and the DNA molecules are extracted on the chip using a magnetic bead system. After extraction, the molecules are stained with intercalating dyes and a fluorescence pattern of the molecules is generated in the DNA analysis area. Here, the DNA molecules are stretched in nanochannels in order to read out the entire molecule. For the optical readout, a laser system is coupled with a fluorescence microscope to detect a fluorescence barcode for each molecule using a single photon counter.

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