Conference on Advances in Microfluidics and Nanofluidics
The Hong Kong University of Science & Technology
Jan 05-07, 2009, Hong Kong

From Inkjet to Bubble-Actuator Enhanced Micro DNA Biosensors

Introduction

To date, inkjet is one of the most successful microfluidics products. Although early inkjet printheads was made by conventional fabrication methods, the inkjet manufacturing companies, such as HP and Canon, gradually adopt MEMS technology to achieve high-speed high-quality printing. For example, Canon developed her proprietary FINE (Full-photolithography Inkjet Nozzle Engineering) technology. The inkjet microchip for Canon's iP8600 print cartridge can achieve large integration of 6,144 micro nozzles and high printing resolution of 4,800×2,400 dpi [1].

In recent years, micro bubble technology has also been applied to the biomedical field. A micro bubble jet-printing device was used to eject DNA segments (presynthesized oligonucleotides) on DNA micro-array glass chips [2]. To induce cavitation microsteaming, micro bubble actuators have also been applied for micro mixing, micro cell sorting, cell/vesicle lysing, DNA hybridization, micro pumping and so on [3].

Micro Bubble Generation in DNA solution

Generally speaking, vapor bubble generation in polymers is more complicated than the corresponding problems in a pure substance because the former involves phase change in a multi-component fluid. In the study of the cavitation problem in polymer solutions, an initial small non-condensable gas bubble was presumed to exist in the solution, and the adiabatic expansion-compression oscillation of the gas bubble was owing to pressure fluctuations in the fluid. The influence of different polymer properties on the damping effect on the bubble radius versus time has been investigated numerically and experimentally in these studies.

Micro bubble generation has been demonstrated in single-stranded DNA (ssDNA) solutions using a micro bubble actuator under pulse heating. The growth and collapse of a micro vapor bubble was observed using a high-speed CCD camera [4-6].

Micro DNA biosensor enhanced by micro bubble actuation

The proposed micro biosensor consists of an array of micro bubble actuator and a PDMS reaction chamber as shown in Fig. 1. Different types of 2-D micro heater arrays (2×1 and 2×2) were fabricated. The dimension of the micro heater of 2×1 array was 10×3 μm2, and that of the 2×2 array was 3×1 μm2 [7].

Molecular beacons are dual-labeled oligonucleotide probes that have a fluorescent dye molecule at one end and a fluorescence quencher at the opposite end. The probe is designed with a target-specific hybridization domain positioned centrally between short sequences that are self-complementary [8]. The sequence of the loop structure of the molecular beacon was designed according to a portion of the sequence of 16S subunit ribosomal DNA in E.coli (MC41000) and was 17 bases long. The stem part of the molecular beacon was 6 bases in length with a rich G/C percentage of 66%. The 5' end was labeled with Fluorescein (FAM) and the 3' end was labeled with Dabcyl quencher. The molecular beacon was synthesized by IDT (Integrated DNA Technologies, Inc., USA), and the complete molecular beacon sequence was: 5'-/56-FAM/CAGTCGTATTAACTTTACTCCCT CGACTG/3Dabcyl/-3', where the underlined nucleotides are the stem sequences.

Various concentrations of the molecular beacon and beacon complement have been tested to study the concentration effects on the fluorescence intensity change as a function of time. The intensity of the fluorescence emission of the DNA hybridization solution was recorded by a Microplate Fluorescence Reader (CytoFluorTM II, Applied Biosystems, USA), where the solution was excited with a 485±20 nm laser and the emission filter was set at 530±20 nm. For hybridization assay I and III (CR = 1:6), it required about 2 hours to reach the fluorescence saturate value. For hybridization assay II and IV (CR =1:1), it required almost 48 hours to reach the fluorescence saturate value. Apparently, excessive existence of the beacon complements in the hybridization solution increased the accessibility of the molecular beacon with the beacon complement and therefore dramatically shortened the hybridization time.

Experimental Summary

A novel bubble actuator enhanced DNA micro biosensor was proposed for E. Coli bacteria detection. The DNA biosensing was implemented by the fluorescence light emission during the hybridization between molecular beacon and its complement on conventional assay and fabricated micro biosensors. Compared to the hybridization time for the hybridization without bubble agitation, it was distinctively shorten by 18% with 10 cycles of micro bubble agitation from a 2×1 heater array, and by 43% with 10 cycles of micro bubble agitation from a 2×2 heater array as shown in Fig. 2.

香港科技大學
THE HONG KONG UNIVERSITY OF SCIENCE AND TECHNOLOGY

Prof. Yi-Kuen Lee

The Hong Kong University of Science and Technology