AN AUTOMATED MICROFLUIDIC SYSTEM FOR OPTIMIZATION OF APTAMER SELECTION BY USING CANCER TISSUE SAMPLES

Cheng-Sheng Lin[1], Yi-Cheng Tsai[1], and Gwo-Bin Lee[1, 2, 3*]

[1] Department of Power Mechanical Engineering, National Tsing Hua University, Hsinchu, Taiwan

[2]Institute of NanoEngineering and Microsystems, National Tsing Hua University, Hsinchu, Taiwan

[3]Institute of Biomedical Engineering, National Tsing Hua University, Hsinchu, Taiwan

Email: *gwobin@pme.nthu.edu.tw

THE 33RD IEEE INTERNATIONAL CONFERENCE ON MICRO ELECTRO MECHANICAL SYSTEMS
MEMS 2020
VANCOUVER CANADA
SPONSORED BY IEEE MEMS
JANUARY 18-22, 2020

Abstract

It is a challenging task to optimize screened aptamers after tedious tissue-based systematic evolution of ligands by exponential enrichment (tissue-SELEX) process. In this study, we used an integrated microfluidic system composed of a formulation chip and an optimization SELEX chip to automatically perform the entire tissue-SELEX process under different screening conditions simulating physiological environments such as human whole blood. The structures and functionalities of screened aptamers during the screening process could be therefore readily applied on clinical samples.

Experiment Process

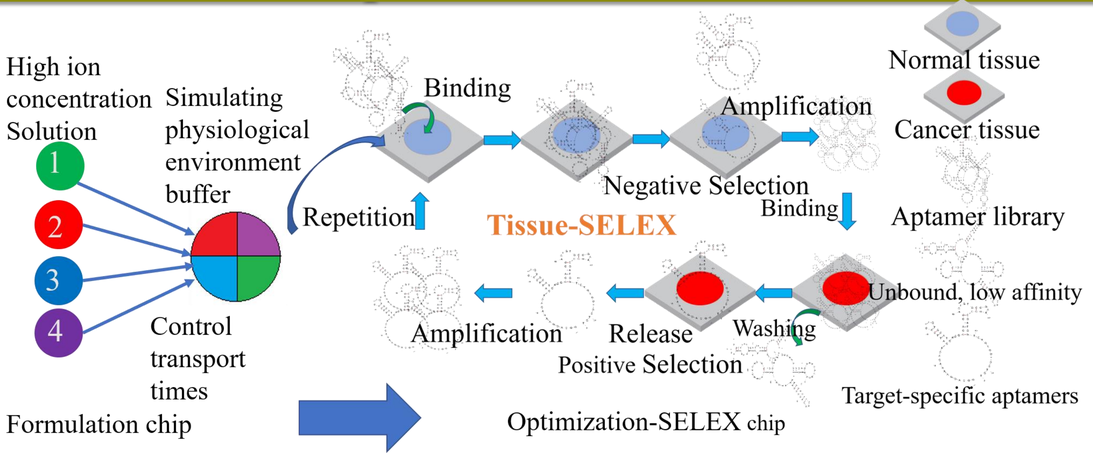


Figure 1: Illustration of optimization tissue-SELEX process. Four kinds of binding buffers with high ion concentrations were formulated by controlling the number of transport times of each buffer on the formulation chip. They were transported to the optimization-SELEX chip for the following SELEX process.

Experiment Setup

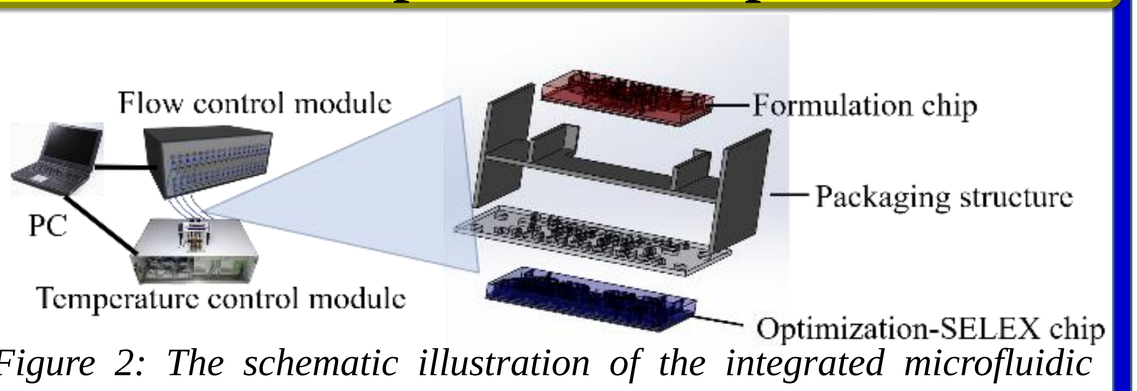


Figure 2: The schematic illustration of the integrated microfluidic system, which was mainly composed of a flow control module, a temperature control module, and two microfluidic chips.

Chip Design

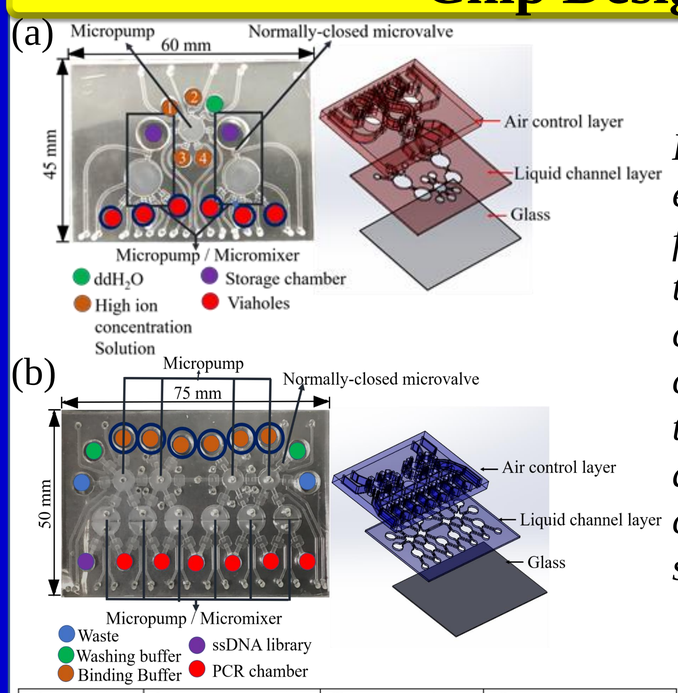


Figure 3: A top view and an exploded view of (a) the formulation chip and (b) the optimization-SELEX chip. Both of microfluidic chips were consisted of three layers, including an air control layer, a liquid channel layer, and a glass substrate.

Ion species	Input concentration	Transport volume	Output concentration
1(Mg^{2+})	a mM	w μL	s mM
2(Ca^{2+})	b mM	x μL	t mM
3(K^{+})	c mM	y μL	u mM
4(Na^{+})	d mM	z μL	v mM

$$s = \frac{a \cdot w}{w+x+y+z}$$

$$t = \frac{b \cdot x}{w+x+y+z}$$

$$u = \frac{c \cdot y}{w+x+y+z}$$

$$v = \frac{d \cdot z}{w+x+y+z}$$

$$w : x : y : z = \frac{s}{a} : \frac{t}{b} : \frac{u}{c} : \frac{v}{d}$$

Figure 4: Operating principle of the formulation chip.

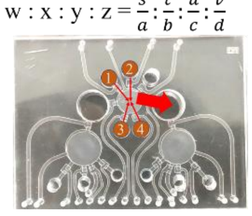
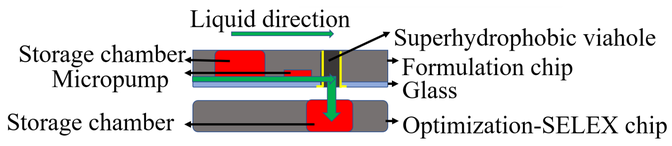
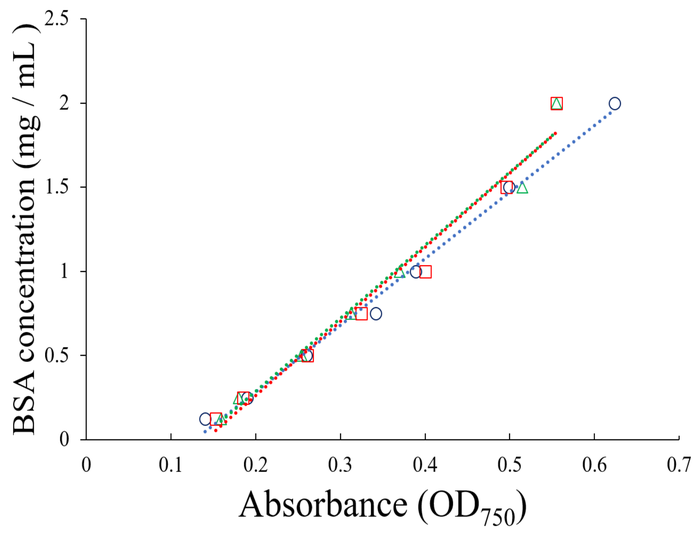


Figure 5: A side view of two microfluidic chips, which showed the transportation between two microfluidic chips.



Results



BSA: ddH_2O	Inaccuracy
10 : 0	0.0%
9 : 1	2.9%
8 : 2	7.7%
7 : 3	8.3%
6 : 4	2.4%
5 : 5	2.3%
4 : 6	7.6%

Figure 6: Bovine serum albumin (BSA) and ddH2O were mixed to measure the concentrations by using a protein assay. Compared with the standard BSA samples, inaccuracy of the on-chip results was measured to be less than 10%, which was similar to the on-bench results..

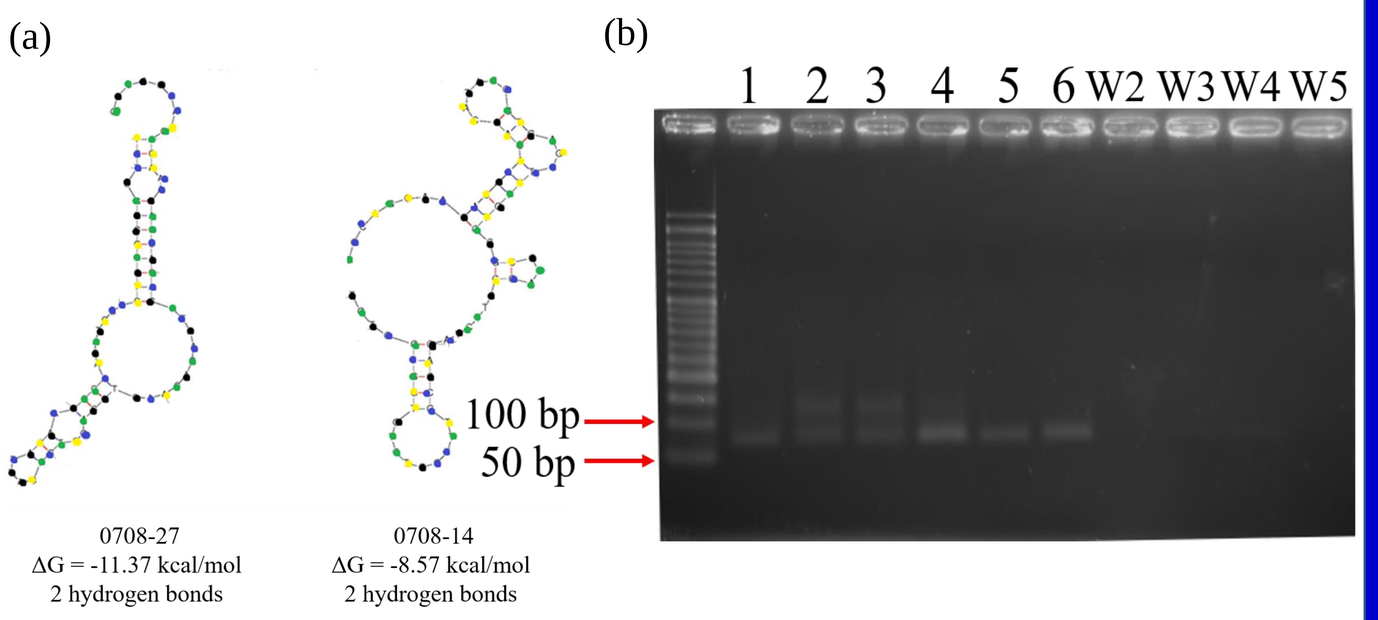


Figure 7: (a)The secondary structures, sequences and ΔG values of aptamer candidates (0708-27, and 0708-14) predicted. (b) Electropherograms of the 10 samples (72 base pairs) collected from 6-round optimization tissue-SELEX, including 6 PCR products and 4 wastes. L: 50-bp ladders. 1~6: PCR products from 1st to 6th round. 7~10: The washed-away ssDNA from 2nd to 5th round.

Conclusions

A new integrated microfluidic system for optimization of aptamer selection has been demonstrated in this work. The entire process including formulation of the buffer and tissue-SELEX could be performed on the developed system automatically. Thus it might be a promising platform for optimization of aptamers screening process and the structures and functionalities of aptamers could be applied for biomedical applications.

Acknowledgements

The authors would like to thank 1) Ministry of Science and Technology (MOST) of Taiwan for funding this work (MOST 107-2314-B-007-005 and 2) National Health Research Institutes (NHRI) (NHRI-EX107-10728EI).

Microfluidic Biochips Lab

國立清華大學 NATIONAL TSING HUA UNIVERSITY