



# 2021「中技社科技獎學金」

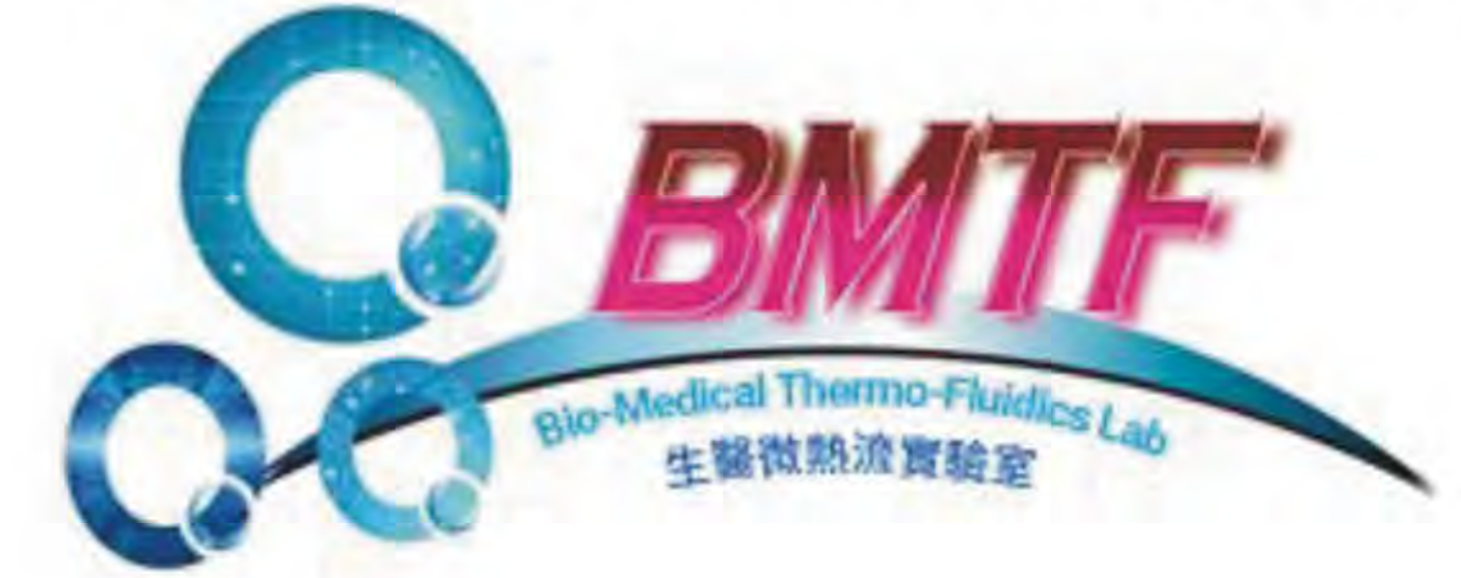
## 2021 CTCI Foundation Science and Technology Scholarship

### 境外生研究獎學金

Research Scholarship for International Graduate Students

## ELECTRO-WETTING-ON-DIELECTRIC PLATFORM FOR CELL FREE DNA (cf-DNA) EXTRACTION FROM MOUSE EMBRYO CULTURE MEDIUM

Anand Baby Alias, Da-Jeng Yao  
4<sup>th</sup> Ph.D. Scholar, Advisor



Institute of NanoEngineering & Microsystems, National Tsing Hua University, Hsinchu, Taiwan  
alias.anand@gmail.com, djyao@mx.nthu.edu.tw

### Abstract

In Digital microfluidic (DMF) system specialized for an electro-wetting-on-dielectric (EWOD) mechanism, we extracted cell-free DNA (cf-DNA) at a small concentration from a mouse embryo culture medium (2.5 days & 3.5 days) using bio-reagents of micro-scale quantity. By EWOD, a mean extraction of 23% of DNA has been achieved. In 2.5 days' sample (E 2.5), the extraction of cf-DNA ranges from 7 femtograms to 243 femtograms, which gives an average weight of 91.47 femtograms. For 3.5 days' sample (E 3.5), the extraction of cf-DNA in samples ranges from 0.14 femtogram to 5.71 femtogram, which gives an average weight of 3.28 femtogram. EWOD enables to extract DNA at ultra-small concentrations (fg/ $\mu$ L to pg/ $\mu$ L) from an embryo culture medium. These results show that DNA extraction with EWOD appears promising and paves a new path towards a renowned lab-on-a-chip concept. The extraction protocol is now adapting for gender determination of mice embryo culture medium has been put forward.

### Research Focus

#### Procedure

The typical way of cf-DNA extraction from an embryo culture medium is explained in figure 1. The placement and adaption steps of various bio-buffers on a EWOD chip with respect to figure 1 is shown in figure 2.

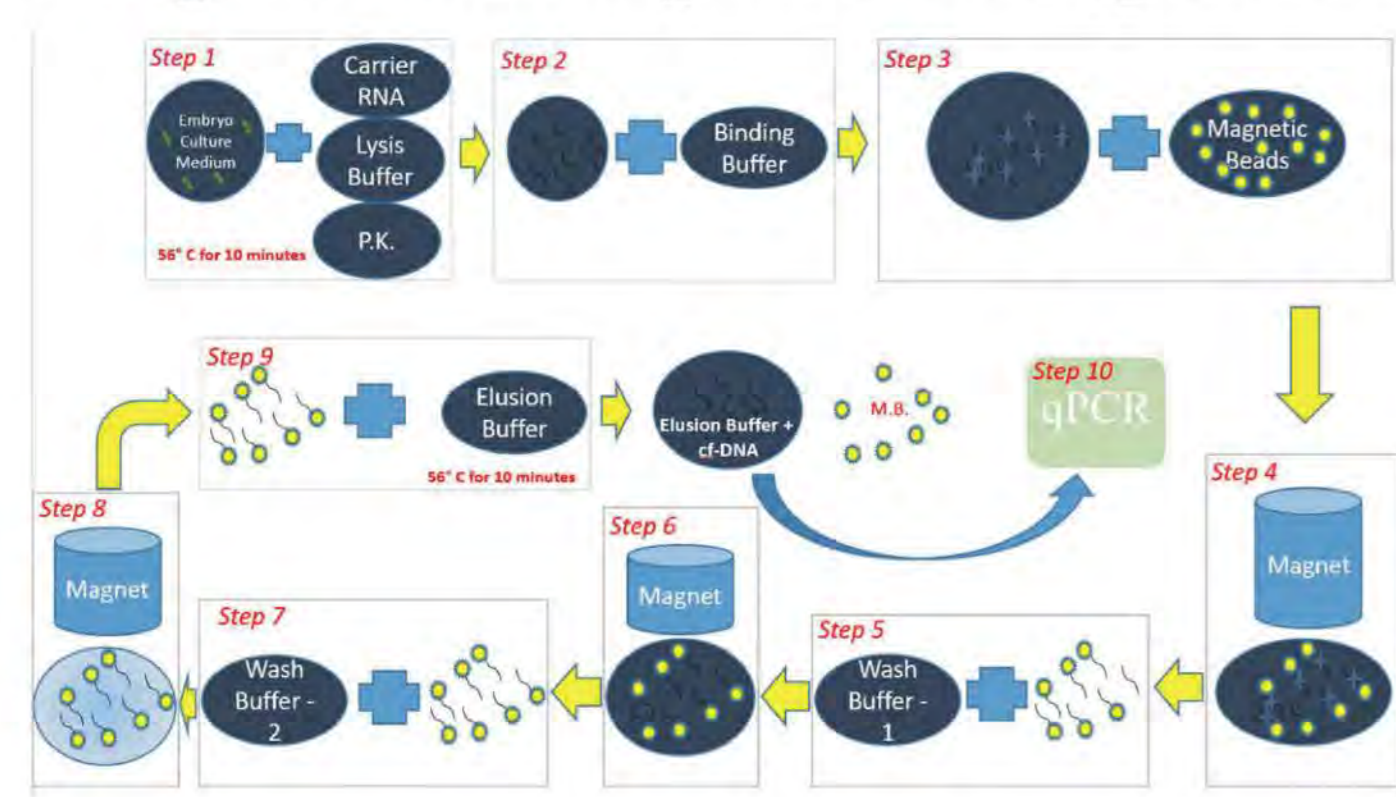


Figure 1: Typical way of cf-DNA extraction

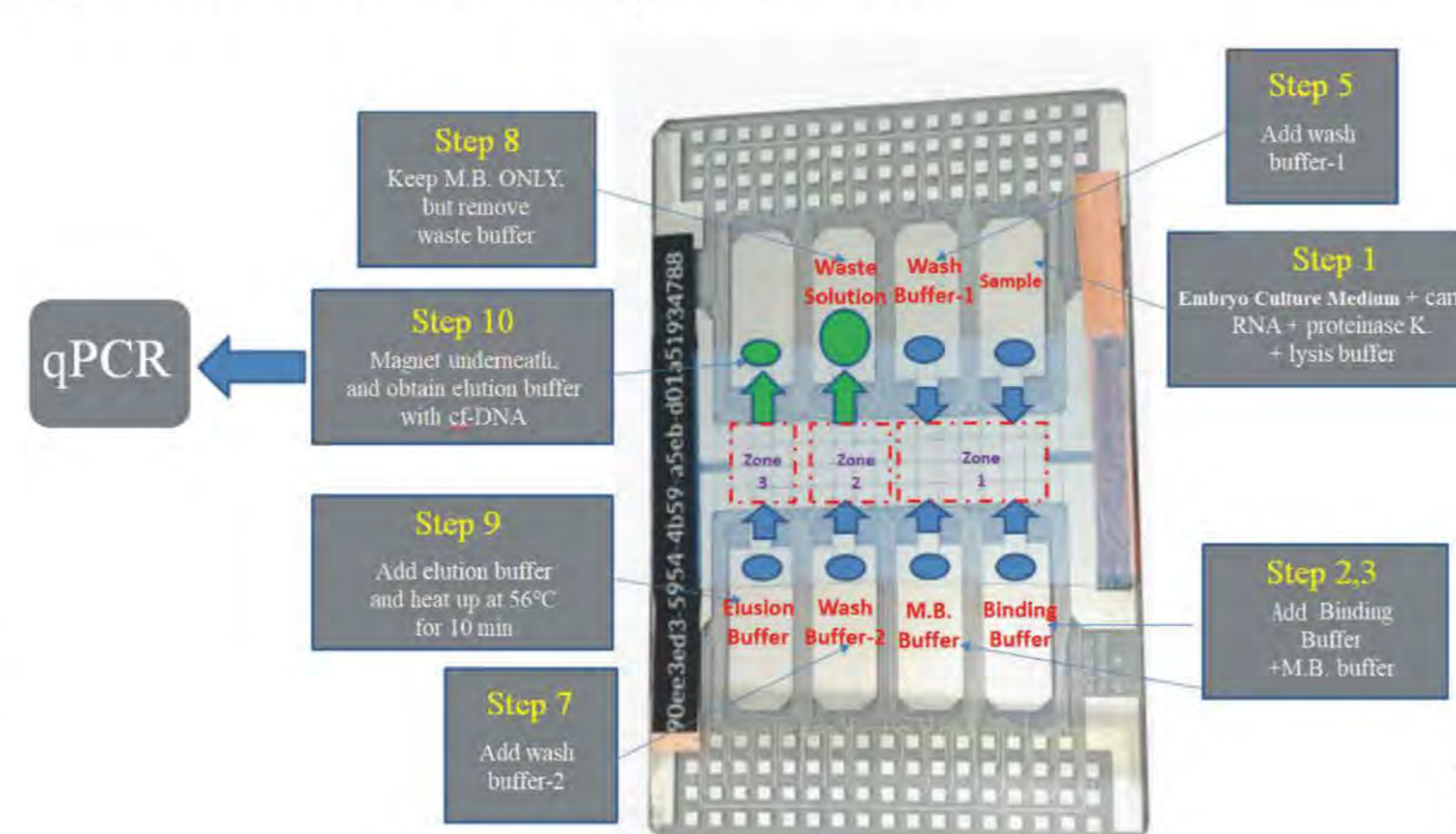


Figure 2: EWOD adaptation for cf-DNA extraction

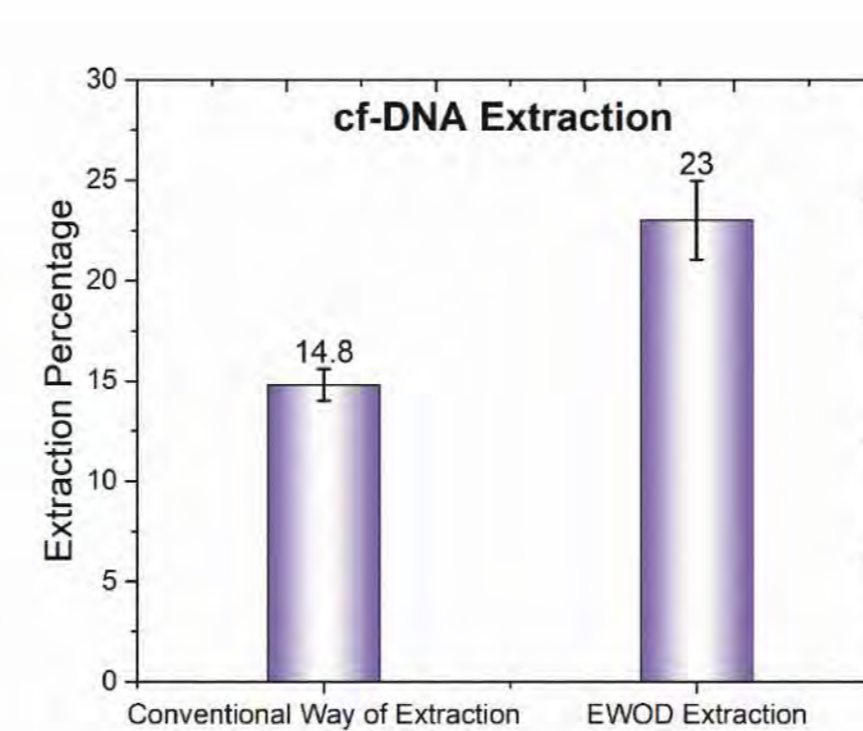


Figure 3: Comparison of EWOD and conventional way of cf-DNA extraction

The extraction of designed-DNA was performed with both conventional and EWOD methods; the mean extraction percentage was calculated for comparison. Cycle threshold (Ct) results were obtained with a quantitative polymerase chain reaction (q-PCR); the mean extraction percentages obtained were 14.8 percentages for the conventional method and 23 percentages for the EWOD at 100 V, 2 kHz is given in figure 3.

#### Summary

We have developed the first magnetically based cf-DNA extraction with an EWOD system in IVF. The new EWOD format allowed the realization of avoiding harm to an embryo and there was no effect on embryo growth before the implantation. Compared to conventional techniques, this method decreased the reagent volumes and duration of analysis. We have successfully extracted the cf-DNA from mouse embryo culture medium. In future, we are planning to implement our EWOD methodology for cf-DNA extraction from human sample.

#### Results & Discussion

The quantitative result analysis is shown in figure 4. For 2.5 days' sample (E 2.5), the weight of cf-DNA in samples ranges from 7 femtograms (fg) to 243 femtograms (fg) which in turn gives an average weight of 91.47 femtograms (fg). For a 3.5 days' sample (E 3.5), the weight of cf-DNA in samples ranges from 0.14 femtograms (fg) to 5.71 femtograms (fg) which in turn gives an average weight of 3.28 femtograms (fg).

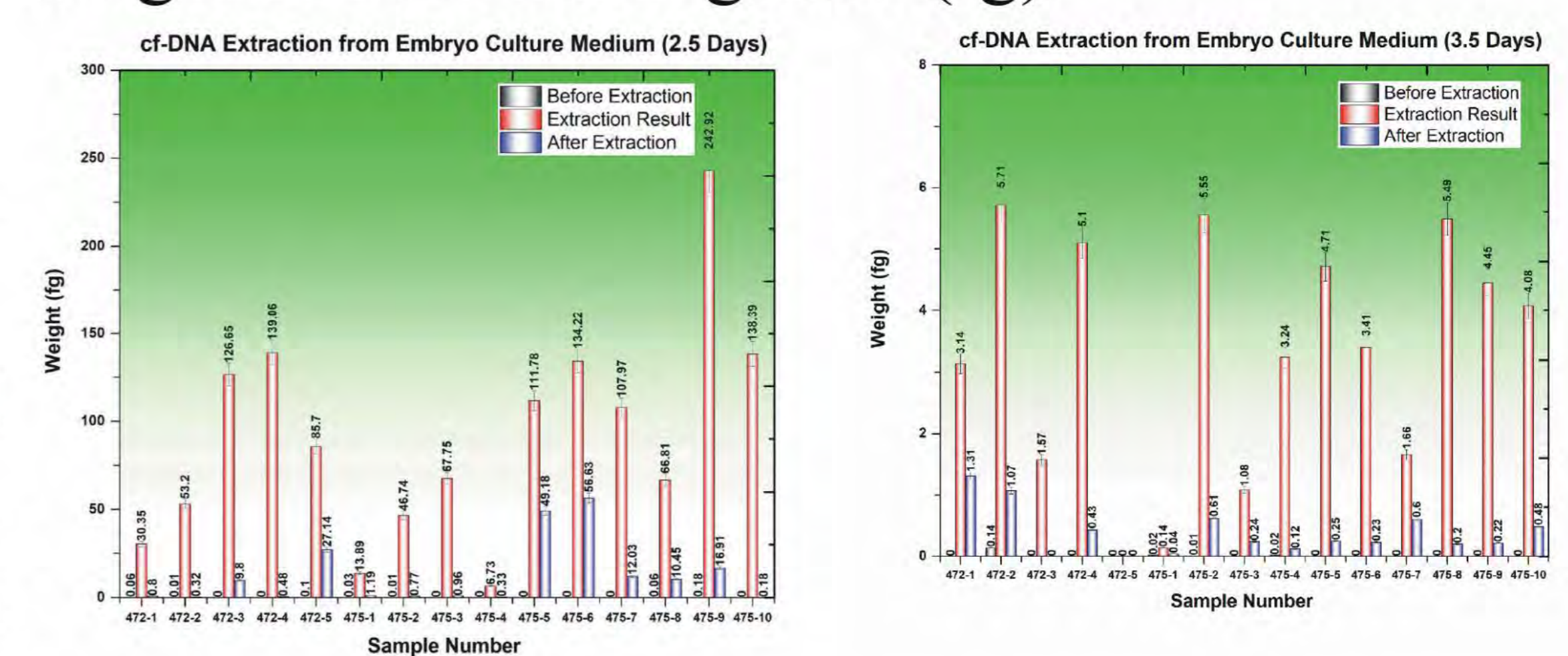


Figure 4: cf-DNA extraction from 2.5 days and 3.5 days of mouse embryo culture medium

#### Selected Journal for Publication

1. Anand BabyAlias, Cheng-EnChiang, Hong-Yuan Huang, Kai-Ti Lin, Pei-Jhen Lu, Yi-WenWang, Tzu-HuiWu, Pei-Shin Jiang, Chien-AnChen& Da-JengYao; Extraction of Cell-free Dna from An Embryo-culture Medium Using Micro-scale Bio-reagents on Ewod
2. Anand Baby Alias, Hong-Yuan Huang and Da-Jeng Yao; A Review on Microfluidics: An Aid to Assisted Reproductive Technology
3. Cheng-En Chiang, Hong-Yuan Huang, Kai-Ti Lin, Anand Baby Alias, Pei-Jhen Lu, Yi-Wen Wang, Tzu-Hui Wu, Pei-Shin Jiang, Chien-An Chen, Da-Jeng Yao; A medical innovation: a new and improved method of DNA extraction with electrowetting-on-dielectric of genetic testing in-vitro fertilization (IVF)

