

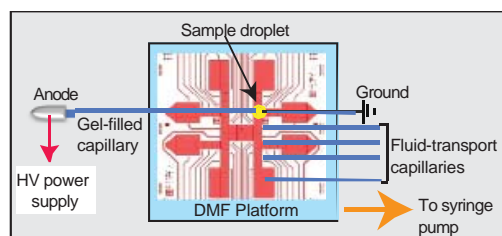
INTEGRATED DIGITAL MICROFLUIDIC PLATFORM FOR QUALITY CONTROL IN NEXT-GENERATION SEQUENCING LIBRARY PREPARATION

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Abstract

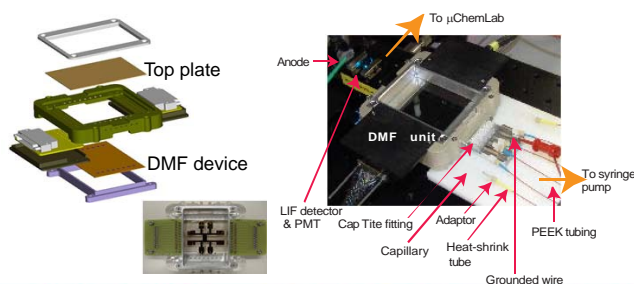
We have developed, characterized and demonstrated a fully automated, integrative digital microfluidic (DMF)-capillary electrophoresis (CE) platform for evaluation of transpose-fragmented peripheral blood mononuclear cells (PBMC) DNA library for Illumina-compatible next generation sequencing.



Integrated Platform

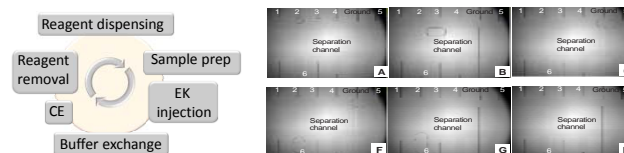
The platform consists of three functional units:

- I. Fluidic transport module
 - Capillary interface through via-holes of manifold
 - Reservoirs for reagent storage, mitigate evaporation
 - Precise volume control via an 8-port syringe pump
- II. Two-plate DMF sample processing module
 - Transparent indium-tin oxide (ITO) electrodes
 - Highly hydrophobic Teflon AF surface
 - Discrete μL -scale droplet manipulation via manual keystrokes or script sequences
- III. In-plane CE module
 - Qualitative size-based separation
 - Sensitive, tunable miniaturized LIF detector
 - Minimal sample consumption via droplet-channel interface



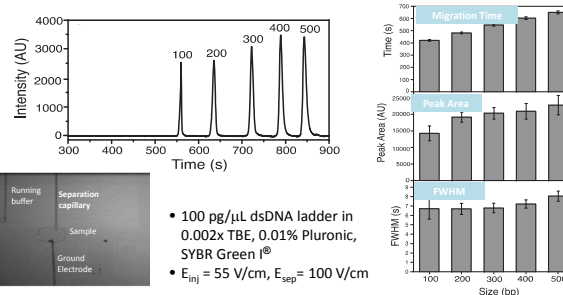
Automated Operation

The entire operation procedure was fully computer controlled. Multiple runs can be performed continuously without manual reagent replacement.



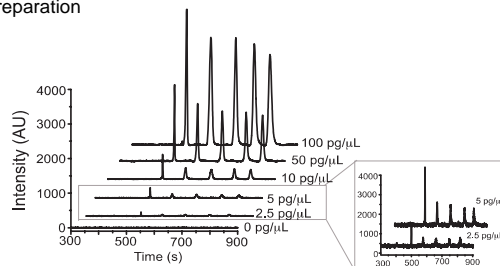
Repeatability Test

Robust DMF-Capillary interface allows sample droplets to be reproducibly prepared and analyzed.



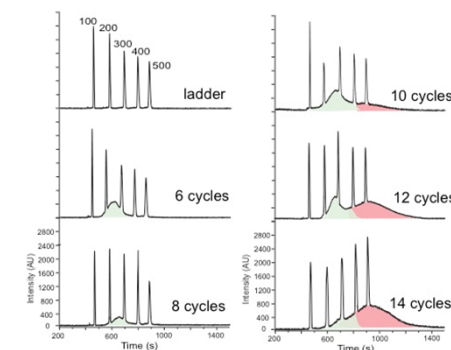
Serial dilution and Sensitivity

- Require $<1 \mu\text{L}$ of sample
- Sensitivity level (5-100 $\text{pg}/\mu\text{L}$) appropriate for NGS sequencers
- Low volume & high sensitivity analytical tool towards small scale library preparation



NGS Library Characterization

QC: sizing & quantification of NGS library prepared from transpose-fragmented PBMC gDNA using PicoGreen® on a single DMF device



Quantitation results using internal standard ladder are in agreement with Agilent BioAnalyzer® (High Sensitivity)

	DMF-CE	BioAnalyzer
6 cycles	380 $\text{pg}/\mu\text{L}$	380 $\text{pg}/\mu\text{L}$
8 cycles	220 $\text{pg}/\mu\text{L}$	280 $\text{pg}/\mu\text{L}$

Future Work

The integrated platform will interface to upstream next generation sequencing systems with a goal of detecting unknown pathogens by enriching informative nucleic acids sequences (from the pathogen) and suppressing background DNA (from the host).

Acknowledgements

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