Continuous microfluidic DNA and protein trapping and concentration by balancing transverse electrokinetic forces

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Electronic Supplementary Information (ESI): pUC19 under alternative square wave excitation (Figure S1), flow rate dependence (Figure S2), pulsed field application (Figure S3) and vortex visualization using micro beads (video 1).

1. pUC19 under square wave excitation

Aside from testing the DNA samples using DC and pulsed electric fields, experiments were also conducted using AC fields or applying a square wave. The following image (Figure S1) shows the DNA sample diluted in $1 \times TE$ buffer and infused at 0.5 µl/min during the application of an square wave ($3V_{peak}$ at 1 MHz). As a result, the DNA did not migrate transversely across the channel as seen when using DC or pulsed electric fields. When using AC or square wave fields, the time averaged applied voltage is zero so analytes move in one direction during the first half cycle of the wave and in the opposite direction during the other half cycle, resulting no noticeable net migration. It is noted that the trapping observed using DC fields is the result of the field polarity. In addition, this demonstrates that DNA dielectrophoresis, which is polarity independent, is not the trapping mechanism seen in this work.



Figure S1. DNA sample under alternative square wave.

2. Flow rate dependence

To evaluate the impact of the axial parabolic pressure driven flow on the DNA migration, different flow rates were used to infuse pUC19 in $1 \times$ TE buffer. In Figure S2, images were acquired at the entrance of the channel, where three flow rates were compared: 0.5 µl/min (S2b), 0.3 µl/min (S2c) and 0.1 µl/min (S2d). The flow rate affects the position along the length of the channel where the DNA concentrates but not the transverse location and width of the concentrated DNA plug. A decrease in the axial velocity allows the DNA to concentrate closer to the entrance of the channel, since the DNA is convected forward at a slower velocity, At 0.5 µl/min, the initial DNA migration can be seen but DNA trapping occurs further down the channel and was not observed at entrance region. At 0.3 µl/min and 0.1 µl/min, the DNA begins to concentrate towards the center of the channel and is captured in the images at the channel entrance.



Figure S2. Images at the entrance of the main channel using DNA samples in $1 \times TE$ buffer at different flow rates. (a) No field applied. The field was applied (3 V_{DC}) while infusing at (b) 0.5 µl/min, (c) 0.3 µl/min and (d) 0.1 µl/min. The DNA can be seen migrating towards the center of the channel in images (c and d).

3. Pulsed field application

Another approach which has proved useful for avoiding electrolysis and decreasing transport time has been through the use of a pulsed electric field using a signal generator (Agilent Technologies, model 33220A) and high-voltage amplifier (Tegam model 2350, Tegam Inc., Madison, OH). The pulsed electric field consists of a pulse signal of 15 V_{DC} at a 1 MHz frequency and 20% duty cycle (so the pulse is only applied for 200 ns followed by a 800 ns rest period). In this manner the DC field is not applied long enough at each pulse for electrolysis bubbles to nucleate. This allows a higher total electric field to be applied without solution electrolysis. The same DNA concentration behaviour is seen as when using a constant electric field, but the DNA accumulates at its equilibrium position faster.



Figure S3. Images of pUC19 sample in TE buffer at the device outlet as time increases when a pulsed electric field is applied (15 V_{DC} , 1 MHz, 20% duty cycle).

4. Vortex visualization using micro beads

The following video was acquired at the entrance of the device showing EO recirculation vortex formation. Using both uncharged melamine and negatively charged polystyrene beads in $1 \times$ TE buffer the flow profiles can be visualized. The charged particles move quickly to the positive electrodes (thought to be due to the large particle EP mobility). However, the melamine particles recirculate with the EO flow profile moving down the channel in a helical fashion showing the recirculation vortex formed near the center of the channel.