

**Electronic Supplementary Information**

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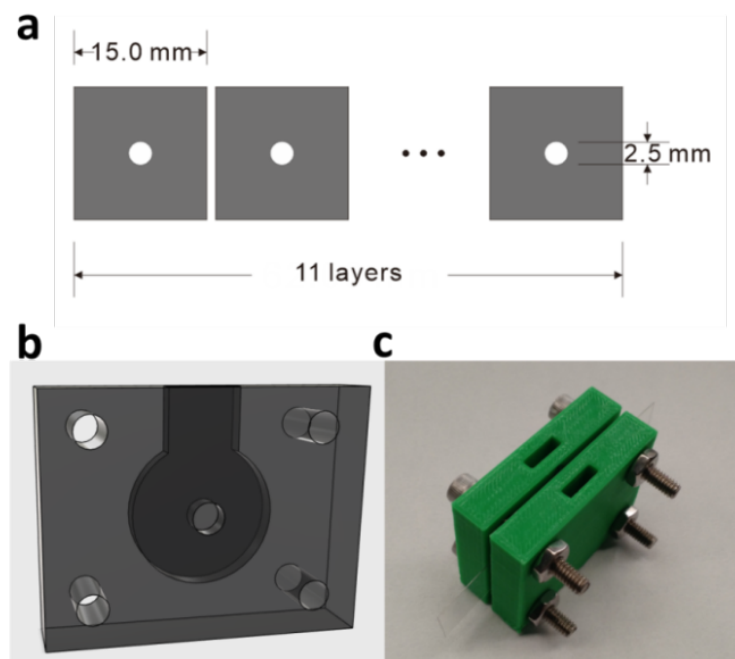
**Low-Voltage Paper Isotachopheresis Device for DNA Focusing**

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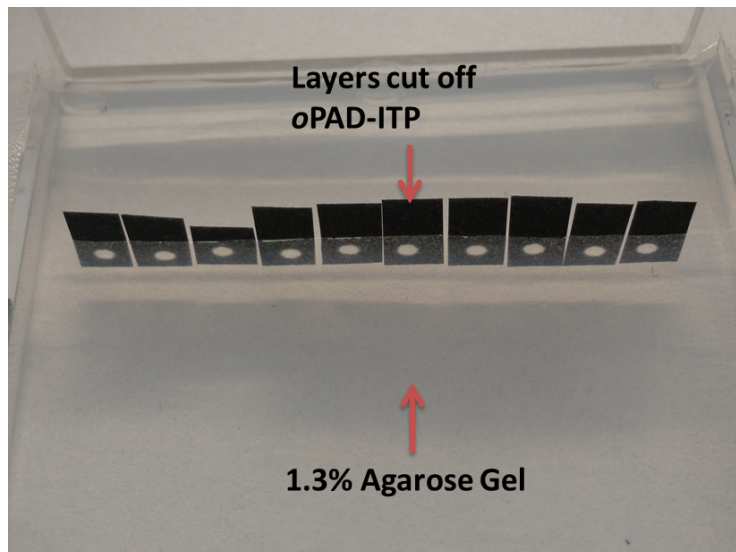
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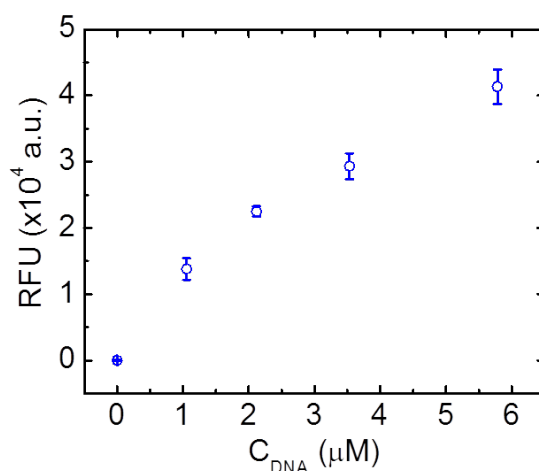
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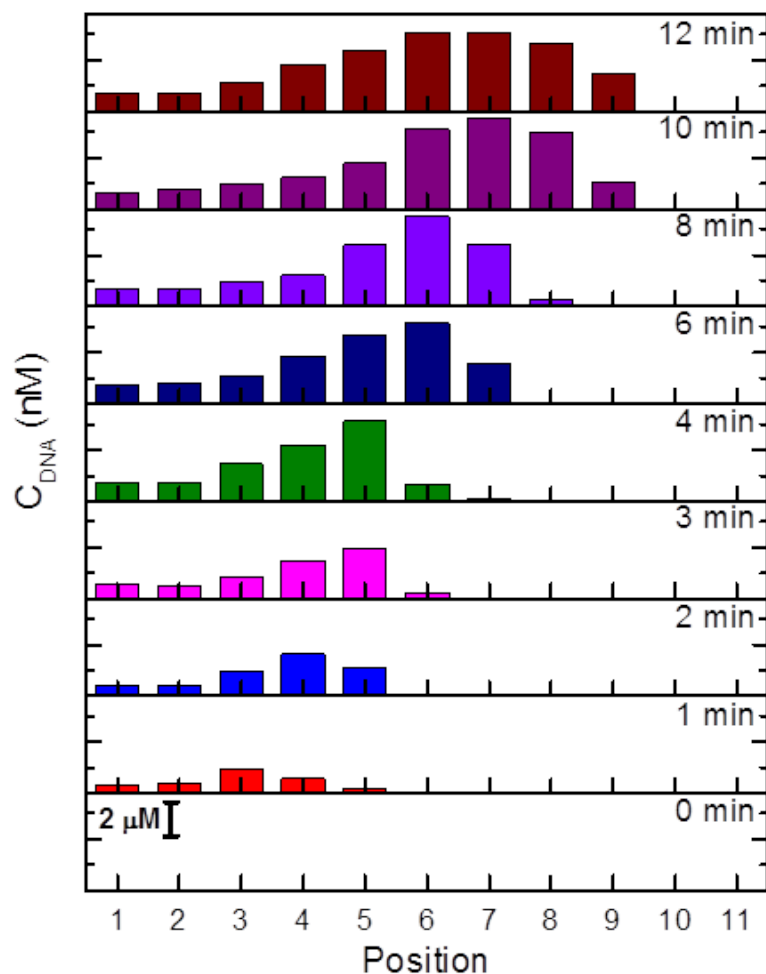
**Figure S1.** Design and photographs of the oPAD-ITP. (a) A CorelDRAW drawing of the origami paper device. The white parts represent unmodified paper and the gray areas are impregnated with wax. (b) A drawing (Autodesk 123D Design) of the 3D-printed reservoirs. (c) A photograph of an actual device. The origami paper is sandwiched between the two green reservoirs using four screws at the corners. Electrodes are inserted into the top holes on the reservoirs.



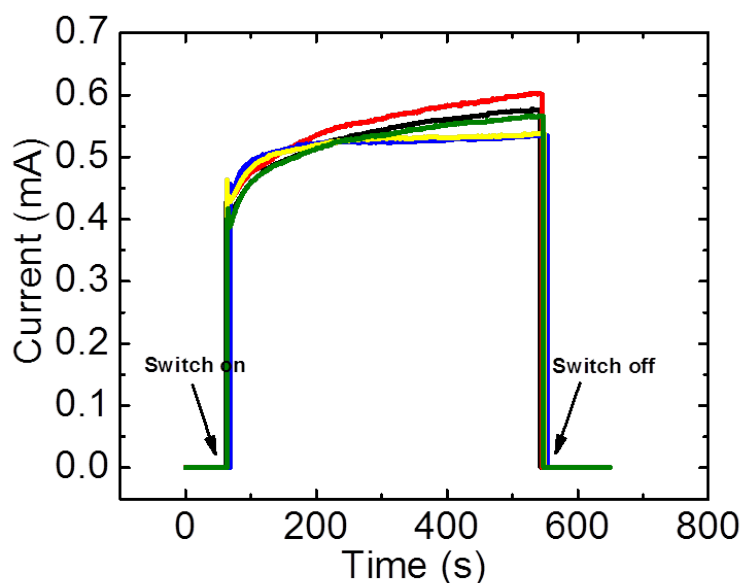
**Figure S2.** Photograph of the gel electrophoresis arrangement. After ITP, each paper layer of the oPAD-ITP was cut off, dried, and inserted into a 1.3% agarose gel containing 10  $\mu\text{g}/\text{mL}$  EtBr for analysis by gel electrophoresis. This photograph shows the individual folds inserted into the gel. A fluorescence scanner was used to image the gel after gel electrophoresis.



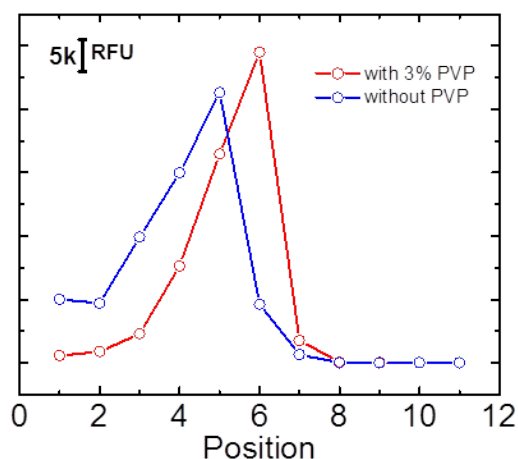
**Figure S3.** ssDNA calibration curve. For each data point, an 11-layer origami paper (same as the oPAD-ITP) was prepared, and 15.0  $\mu\text{L}$  ssDNA solution having the indicated concentrations was added to the inlet and allowed to wet all 11 layers. Excess liquid was removed, the paper was dried in the dark, and then the fluorescence image of each layer was obtained. The average RFU intensity of all layers is plotted as a function of the ssDNA concentration.



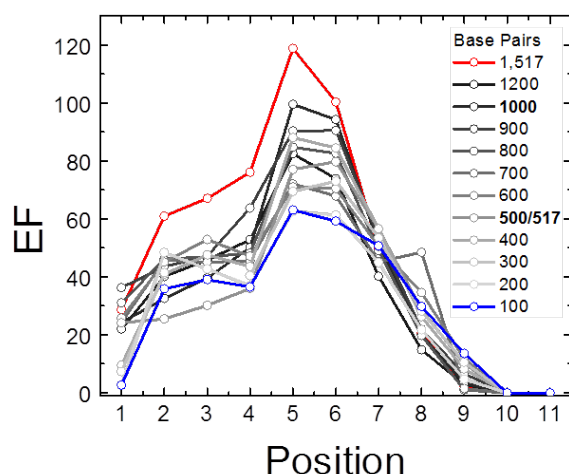
**Figure S4.** Distributions of ssDNA in the oPAD-ITP as a function of time. A 1.0 mL solution containing 40.0 nM ssDNA and TE buffer was added to the TE reservoir, and 1.0 mL of the LE buffer was added to the LE reservoir. After applying a voltage of 18 V for different lengths of time, the oPAD-ITP was unassembled, the individual paper layers were cut out, dried in the dark, and then imaged using fluorescence microscopy. The integrated fluorescence intensities are plotted here.



**Figure S5.** Current vs time curve for a typical ITP experiment. In this case, the ITP voltage (18 V) was applied using a CHI 650C potentiostat (CH Instrument, Austin, TX) so that the current could be recorded. Five replicates are shown in the figure.



**Figure S6.** Electroosmotic flow control experiment. The red trace presents the distribution of ssDNA when there was 3.0% polyvinylpyrrolidone (PVP) initially present in the LE to suppress EOF. The blue trace is same experiment without PVP. Both ITP experiments were run at 18 V for 4.0 min. The results show that EOF did not significantly affect the value of EF and C% in the oPAD-ITP.



**Figure S7.** EF analysis of the ITP of a dsDNA ladder. Same experiment setup and buffer conditions were used as Figure 1. The dsDNA ladder was loaded into TE buffer. After ITP at 18 V for 10 min, the oPAD-ITP was unfolded and each paper layer was cut off and inserted in gel as Figure S2 for gel electrophoresis. The gel electrophoresis image of each DNA band in each lane was analyzed using ImageJ software. The EF plotted here was calculated as the area of filled blue bars in Figure 5b divided by the area of corresponding hollow blue bars in Figure 5c.