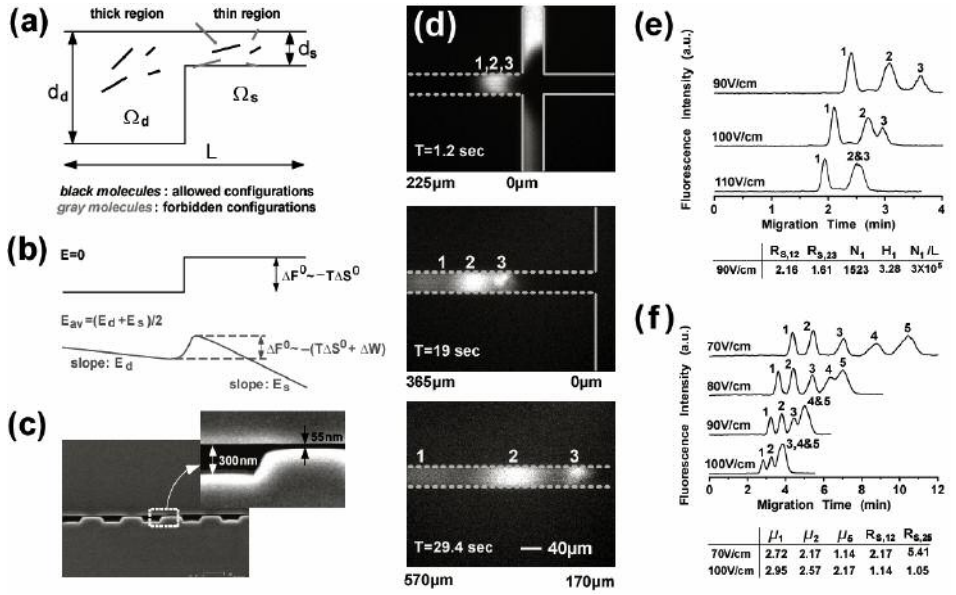

Figure 1. (a) Partitioning of rigid, rod-like molecules in a slit-like nanofilter. (b) Free energy landscape of a nanofilter. (c) SEM images of a periodic array of nanofilters with alternating thin and thick regions. (d-f) Separation of SDS-protein complexes (d&e) and dsDNA molecules (f) in a one-dimensional nanofilter array chip (d_s: 55nm, d_d: 300nm, L: 1μm). Band assignment for SDS-protein complexes: (1) cholera toxin subunit B (MW: 11.4kDa); (2) lectin phytohemagglutinin-L (MW: 120kDa); (3) low density human lipoprotein (MW: 179kDa), for DNA: (1) 50bp; (2) 150bp; (3) 300bp; (4) 500bp; (5) 766bp.

Since the attempted transition rate and the free energy barrier ΔF^0 both favor the successful passage rate of smaller molecule over the barrier, smaller molecule migrates faster though the nanofilter array with a less characteristic trapping life time [5]. Figure 1(d-f) summarizes the separation results of SDS-protein complexes and dsDNA molecules in a one-dimensional nanofilter array chip (d_s=55nm, d_d=300nm, L=1μm). The speed and resolution obtained by the nanofilter array chip is comparable to current state of the art systems (i.e. capillary gel electrophoresis).

3. CONTINUOUS-FLOW SEPARATION IN 2-D NANOFILTER ARRAY CHIP

The continuous-flow biomolecule separation in 2D nanofilter array chip takes advantage of differential bidirectional transport of biomolecules of different sizes through two-dimensional periodic arrays of nanofilters (Fig. 2a). The successful jump passage rate though the nanofilter is size-dependent, so after some traveling period, larger molecules stay close to the original thick channel with less deviations, while smaller molecules execute more successful passages of the nanofilters and the trajectory of them will deflect more from their original injection direction. Two different operation modes can be applied for the two-dimensional nanofilter array chip: continuous field separation mode and pulse-field separation mode. In the continuous field separation mode, constant field is applied across the nanofilter arrays during the separation process. The field distribution and strength can be modulated by alternating the voltages applied at different fluidic reservoirs.

The continuous-flow separation can also be achieved using alternating electric fields of different strengths or durations in the pulse-field separation mode (Fig. 2b-2d).

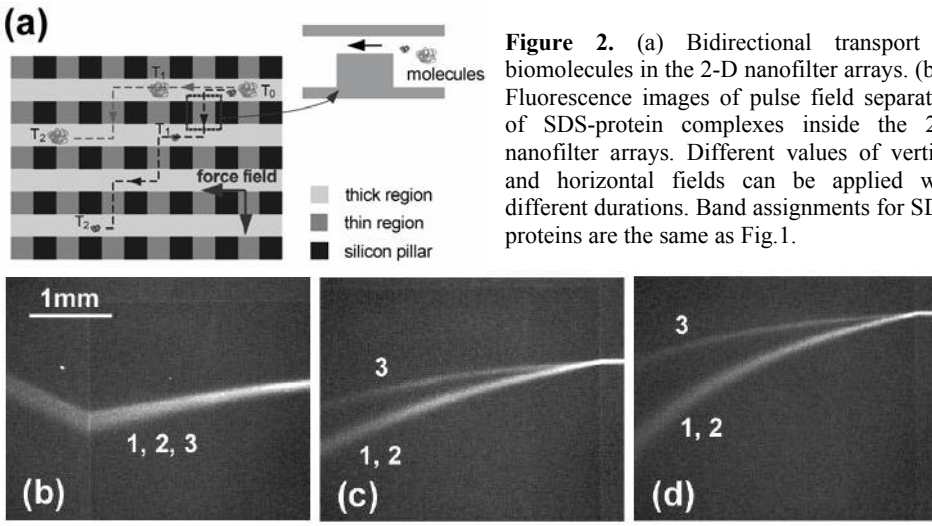

Figure 2. (a) Bidirectional transport of biomolecules in the 2-D nanofilter arrays. (b-d) Fluorescence images of pulse field separation of SDS-protein complexes inside the 2-D nanofilter arrays. Different values of vertical and horizontal fields can be applied with different durations. Band assignments for SDS-proteins are the same as Fig.1.

4. CONCLUSIONS

In conclusion, we have successfully size-separated SDS-protein complexes and small dsDNA molecules in nanofilter array based chips. The speed and resolution obtained in the one-dimensional nanofilter array chip is comparable to current state of the art systems without using any sieving gel. This opens up possibilities for integrating many different biomolecule sensors, and separation and reaction chambers in a single chip, without the concern of sieving matrix crosstalk and contamination. The separation efficiency could be further improved by scaling down the nanofilter period by advanced lithography techniques. The continuous-flow operation of the two-dimensional nanofilter array chip is ideal for preparatory sample fractionation with increased sample throughput. The retrieval of separated samples is straightforward and this greatly facilitates further downstream analysis.

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REFERENCES

[1] J. Han and H. G. Craighead, *Science* **2000**, *288*, 1026-1029.
[2] L. R. Huang, J. O. Tegenfeldt, J. J. Kraeft, J. C. Sturm, R. H. Austin, and E. C. Cox, *Nat. Biotechnol.* **2002**, *20*, 1048-1051.
[3] A. G. Ogston, *Trans. Faraday Soc.* **54**, 1754 (1958).
[4] J. C. Giddings, E. Kucera, C. P. Russell and M. N. Myers, *J. Phys. Chem.* **72**, 4397 (1968).
[5] J. Fu and J. Han, in preparation.