Supporting Information for:

Single-Cell Copy Number Analysis of Prostate Cancer Cells Captured with GEDI Microdevices

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1 Supplementary Methods

1.1 Single Nucleus Sequencing (SNS) experimental details

Samples were sorted using the BD Biosystems Aria II flow cytometer, and deposited into a 96-well plate containing 10 µL of lysis solution from the Sigma-Aldrich GenomePlex© WGA 4 kit. Single-read libraries were sequenced using the Illumina GA2 analyzer, and data was processed Illumina GA Pipeline bioinformatics software. Bowtie alignment software was used to align sequence reads to the human genome. S-Plus (Mathsoft, Inc.) software was used to calculate integer differences in copy-number using Gaussian kernel smoothed density plots. Generated copy-number profiles were then used to generate the neighbor-joining tree based on Euclidean distance metrics using Matlab (Mathworks).

1.2 LNCaP preparation via serial dilution

Frozen cell suspensions where thawed and seeded at either 0.5 nuclei per well (limiting dilution) or 1 nuclei per well (2x limiting dilution) in 96-well plates. Plated nuclei then underwent SNS as previously described. Poisson's distribution dictates, at limiting dilution, that the probability of there being no cells per well is 60%, the probability of a well containing a single cell is 30%, and the probability of two or more cells is 10%.

These probabilities can be used to predict the number of wells that should successfully amplify for SNS (Table S1). $50.2\% \pm 0.2\%$ of predicted wells amplified using GEDI-isolated LNCaP nuclei, $44.2\% \pm 0.4\%$ amplified using nuclei from cell culture. 100% of amplified wells generated copy-number profiles. Amplification efficiencies are on the order of, but lower than, conventional FACS (90%). Conversion to copy-number profiles is equivalent.

	Limiting Dilution	Expected # amplified cells	2x Limiting Dilution	Expected # amplified cells
LNCaP (GEDI)	2/12	$4.53\pm1.80/12$	4/12	$7.42 \pm 1.86/12$
LNCaP (Cell Culture)	2/12	$6.05 \pm 2.32 / 12$	n/a	n/a

Table S1. Number of successfully amplified cell nuclei as compared to predicated values using Poisson's distribution.



Figure S1. Simple linear regression for a bulk LNCaP sample from cell culture (150 cells). The averaged genetic profile agrees very well with copy-number profile generated form a single nuclei from a GEDI microdevice (r^2 =0.904).



Figure S2. Pairwise comparisons of various SNS samples.