Electronic Supplementary Material (ESI) for Analyst.

ACCURATE QUANTIFICATION OF APOPTOSIS PROGRESSION AND TOXICITY USING A DIELECTROPHORETIC APPROACH

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Supplementary Information

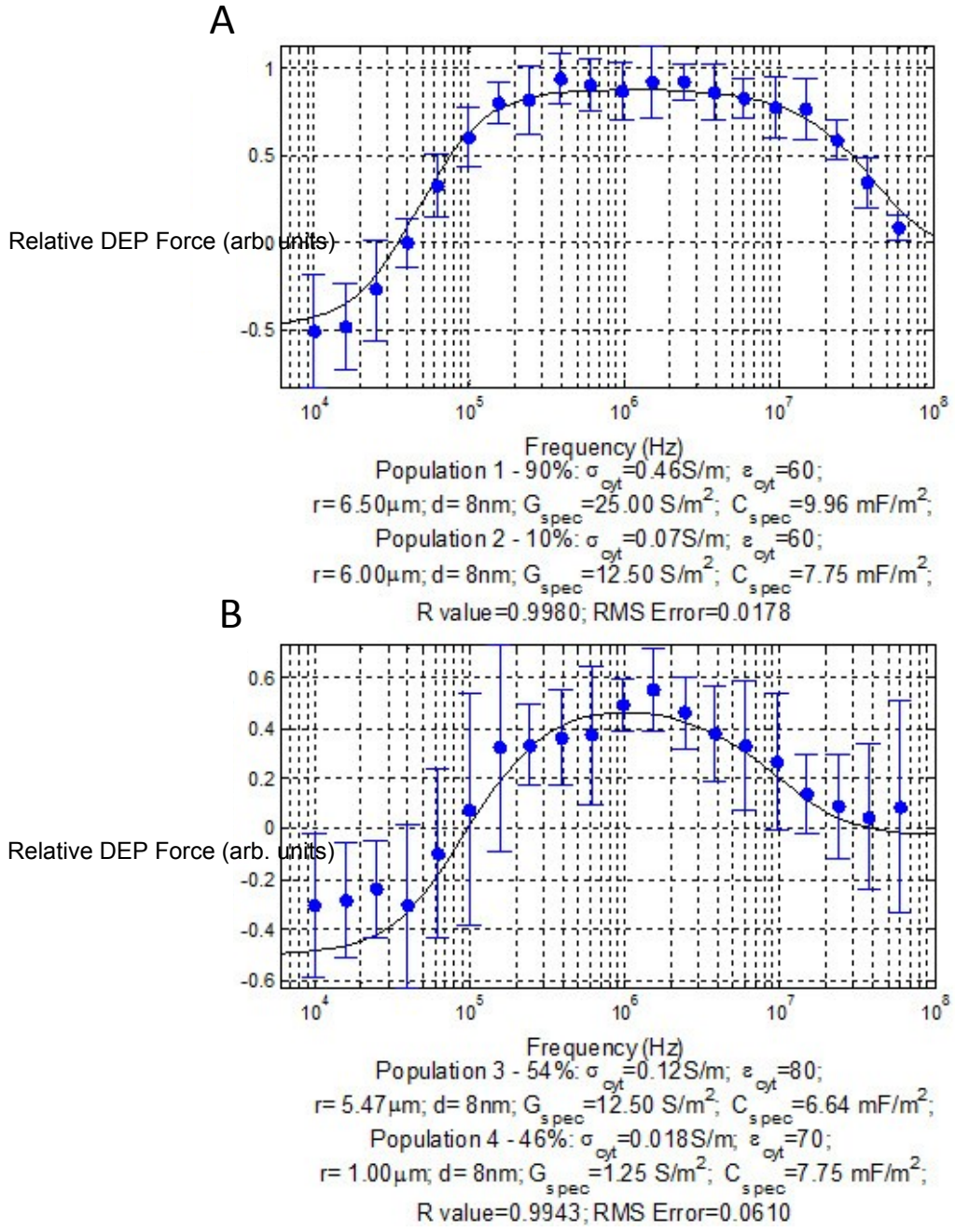


Figure S1. The untreated Jurkat cells (A) fit a two population model and the DOX treated Jurkat cells (B) also fit a two population model. Data points are MEAN±STDEV (n=15). The parameters

determined by the single shell model (solid line) of these two populations were then used to fit the subsequent mixtures. The treated and untreated samples were shown to have statistically ($p<0.1$) varying parameters with cytoplasmic conductivity ($p<0.0001$) and radius ($p<0.0001$) the most effected through 2-way ANOVA and multiple comparison.

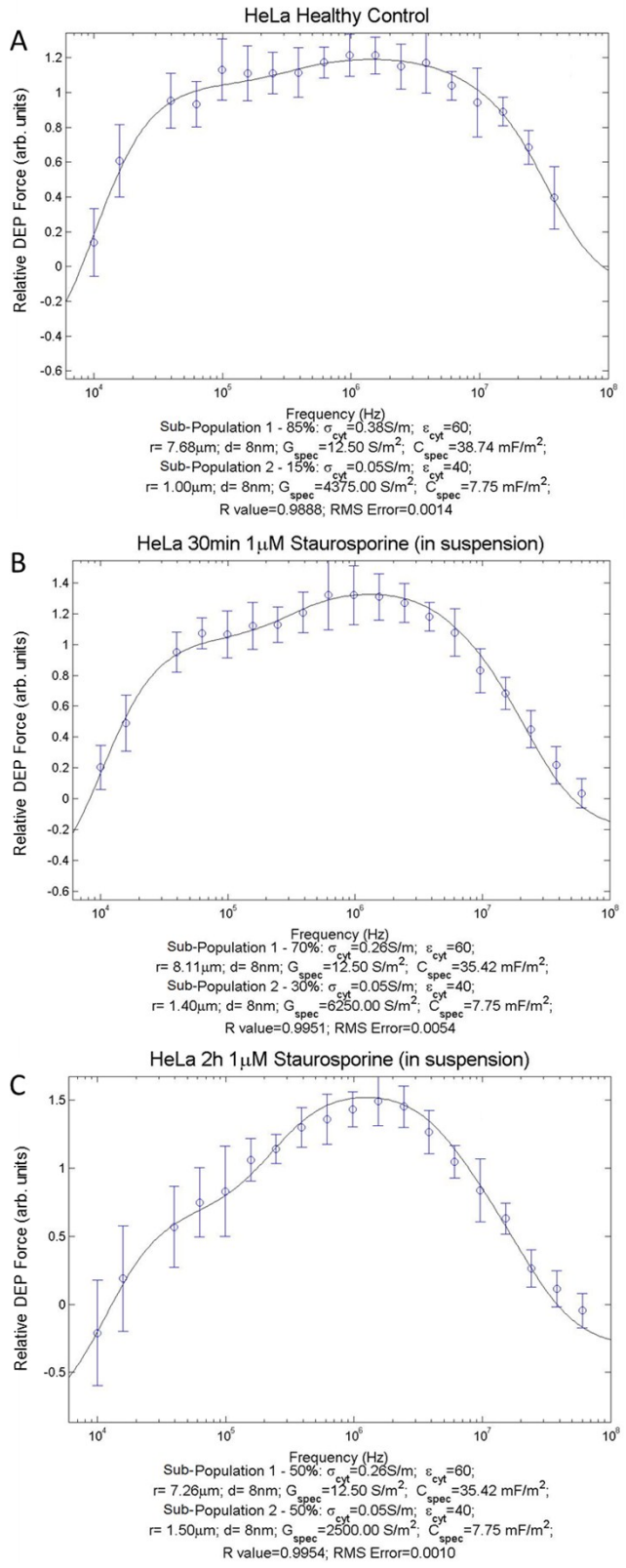
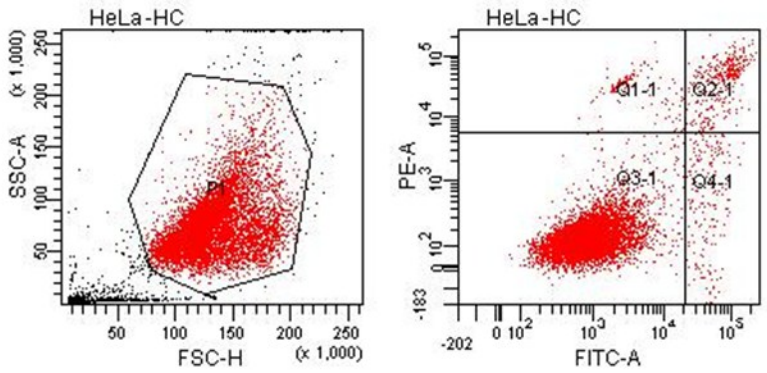


Figure S2. The untreated HeLa cells (A) fit a two population model (indicating healthy and affected populations) and the STS treated HeLa cells also fit a two population model after 30 minute STS incubation (B) and 2 hour incubation (C). Data points are MEAN±STDEV (n=14). The percentages of these two populations were then used to compare DEP with other standard viability assays.

A



Population	#Events	%Parent	FITC-A Mean	PE-A Mean
All Events	10,000	####	5,633	2,739
P1	7,739	77.4	6,280	3,013
Q1-1	124	1.6	5,820	32,558
Q2-1	344	4.4	84,738	51,814
Q3-1	7,066	91.3	1,245	170
Q4-1	205	2.6	48,420	1,235

B

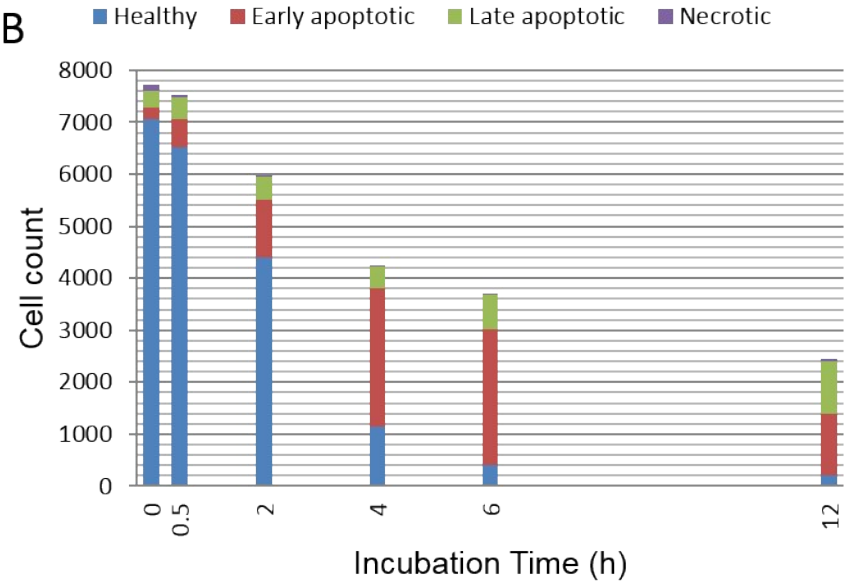


Figure S3. Results of analysis of HeLa cell populations experiencing staurosporine-induced apoptosis at different incubation times, as measured using an Annexin-V assay (n=5). Viable cells have phosphatidylserine (PS) located on the inner membrane. When the cell goes into apoptosis the PS is exposed on the cell surface. When the cell is at a late stage of apoptosis the cell membrane loses its integrity. Annexin V assay consists of adding PI and Annexin V-FITC to the cells. The Annexin V-FITC adheres to the PS. PI leaks into the cell if the membrane is damaged. If a cell is viable, Annexin V-FITC will not bind to PS because it won't be exposed on the cell surface and it will not stain with PI as the membrane will be intact. If the cell is going through early apoptosis, it will be labelled with Annexin V-FITC but not stained with PI as the membrane at this stage remains intact. Once the cell is going through late apoptosis, the membrane loses its integrity and will also stain with PI. This information is represented in a double axis log dot plot of PI vs. FITC, an example of this is on the right hand side of (A). Each quarter represents a different stage of apoptosis. Q3-1 encloses the healthy population, Q4-1 the early apoptotic population, Q2-1 the late apoptotic population and Q1-1 the necrotic population. Electronic compensation was carried out in order to avoid bleed

through of fluorescence. (B) The populations in each quarter were plotted across each incubation time to demonstrate the change in sub-populations of the treated cells with incubation time.

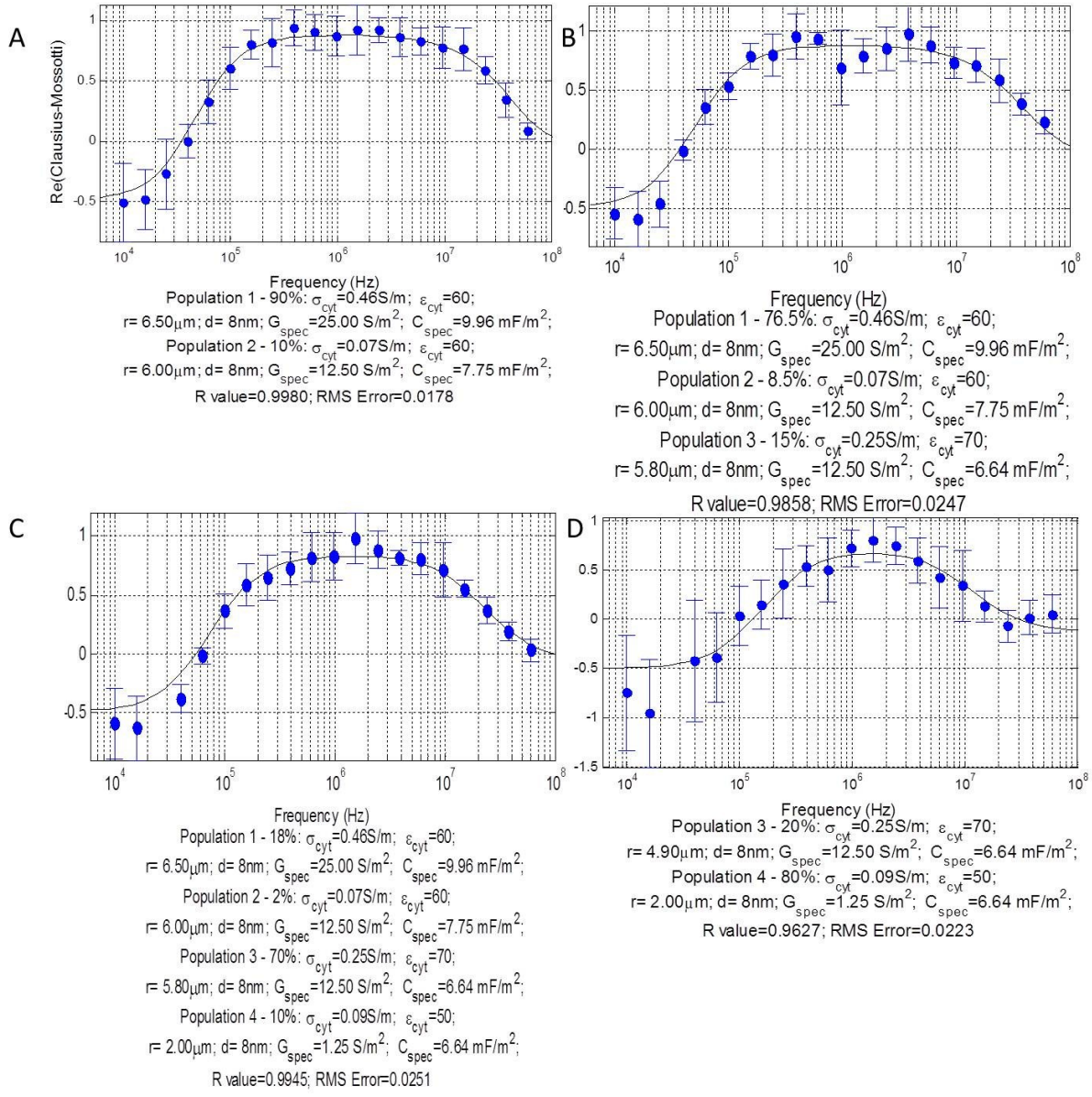


Figure S4: DEP spectra of healthy Jurkat cells (A) and those incubated for 8 h (B), 16 h (C) and 32 h (D) with 1 µM DOX (data points are MEAN ± SEM with n=7). The DEP properties determined by the single shell model (solid line) for each population is given below the spectrum.