

# Supplementary Data

Characterization of a novel high-throughput, high-speed and high-precision plate-based image cytometric cell counting method

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Celigo image analysis parameters for AO-stained Jurkat and CHO-S cells.			
Image Analysis Parameters	AO-Jurkat	AO-CHO	BF
Algorithm	Fluorescence	Fluorescence	Fluorescence
Intensity Threshold	8	8	255
Precision	High	Normal	High
Cell Diameter	10	12	10
Dilation Radius	0	0	0
Background Correction	Uncheck	Uncheck	Uncheck
Separate Touching Objects	Uncheck	Uncheck	Uncheck
Minimum Cell Area	20	20	
Maximum Cell Area	1200	500	

Average live cell concentration and CV for each sample measured on each instrument.												
Live Cell (cells/mL)	Cellaca MX 1		Cellaca MX 2		Vision 1		Vision 2		Auto2000 1		Auto 2000 2	
	Average	CV										
Sample 1	1.2 x 10 <sup>6</sup>	4.7%	1.2 x 10 <sup>6</sup>	6.0%	1.1 x 10 <sup>6</sup>	6.9%	1.1 x 10 <sup>6</sup>	6.6%	1.1 x 10 <sup>6</sup>	7.6%	1.1 x 10 <sup>6</sup>	6.5%
Sample 2	0.6 x 10 <sup>6</sup>	7.1%	0.6 x 10 <sup>6</sup>	7.2%	0.5 x 10 <sup>6</sup>	5.4%	0.5 x 10 <sup>6</sup>	7.2%	0.5 x 10 <sup>6</sup>	8.0%	0.5 x 10 <sup>6</sup>	7.1%
Sample 3	2.0 x 10 <sup>6</sup>	5.3%	2.0 x 10 <sup>6</sup>	5.7%	1.9 x 10 <sup>6</sup>	2.9%	1.9 x 10 <sup>6</sup>	4.3%	1.9 x 10 <sup>6</sup>	3.9%	1.9 x 10 <sup>6</sup>	4.3%
Viability %	Average	CV										
Sample 1	86.8%	3.4%	85.8%	3.0%	83.3%	4.4%	84.3%	3.9%	83.9%	3.5%	83.5%	3.9%
Sample 2	83.6%	2.3%	83.1%	5.0%	79.1%	4.0%	81.3%	4.1%	80.8%	4.1%	78.4%	5.6%
Sample 3	87.3%	1.5%	86.8%	1.4%	84.5%	1.6%	86.0%	1.9%	84.0%	2.2%	84.5%	1.4%