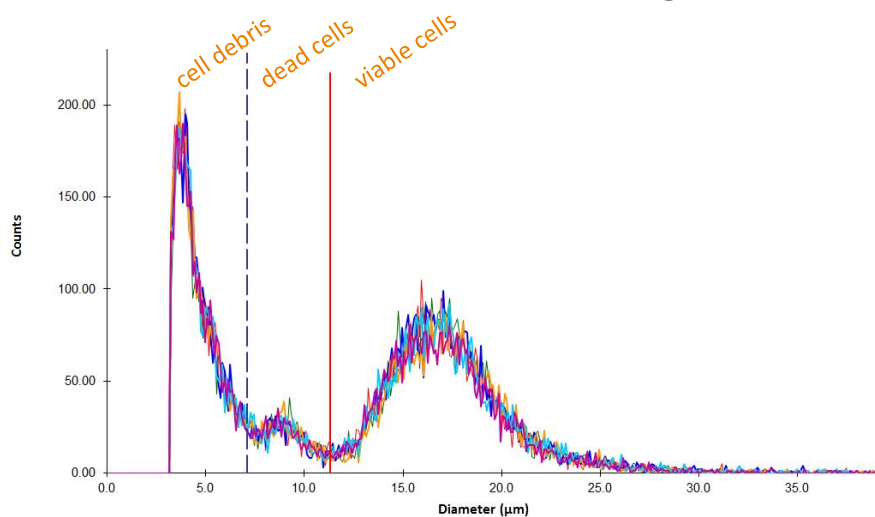


Cell Counting, Viability Testing and Cell Aggregation Control with High Fidelity Analysis of Keratinocytes using CASY



Introduction

Here we present original data generated during a live on-site CASY TT instrument demonstration. Aim was to verify CASY's capabilities regarding high reproducibility, fast and easy sample preparation and straightforward live / dead discrimination. After a brief introduction, assays were performed by Kerstin Schenk and Eliane Wandeler, DSM Nutritional Products, R&D Personal Care, Kaiseraugst, Switzerland.



Sample

Cell Type

Keratinocytes

CASY Analysis

All samples were analyzed with CASY (150µm Capillary; 3x400µl sample volume; 0-40µm size scale, dilution factor 102)

Graphs of the CASY measurements were created with CASYworX software

Fig. 1: Overlay of all Keratinocyte technical replicates measurements

Overlay was plotted from 6 measurements of Keratinocytes using CASY. Single cell peak at 16.59µm, average single cell volume $2.4 \cdot 10^3$ fl.

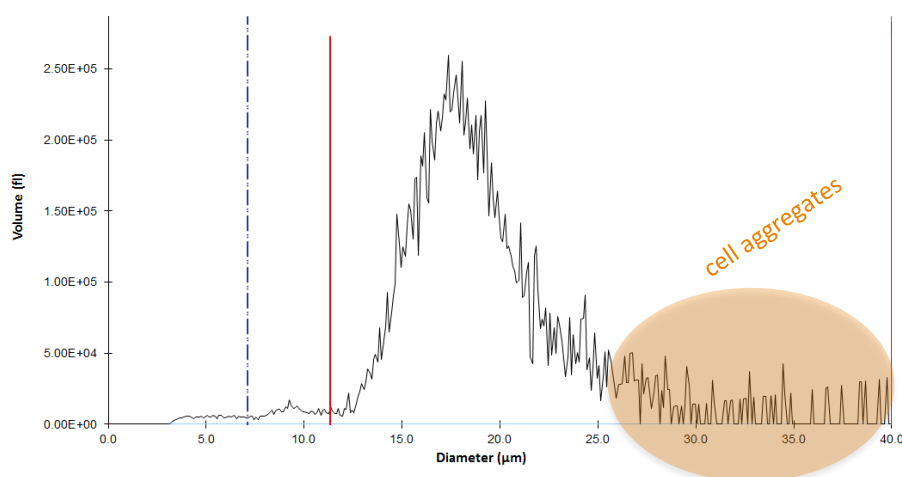


Fig. 2: Visualization of Biomass

Plotting particle volume against particle diameter displays the biomass of the sample present in cell aggregates (orange area).

Results

Standard deviations of repeated measurements

Sample	Agg.Factor	Viable Cells / mL	Total Cells / mL	% Viability	Peak Dia / μm
1 st Measurement	n.a.	477500	556000	85.88	16.68
2 nd Measurement	n.a.	454200	525500	86.44	16.38
3 rd Measurement	n.a.	478500	550300	86.94	16.52
4 th Measurement	n.a.	446800	519900	85.9	16.81
5 th Measurement	n.a.	460000	532300	86.4	16.63
6 th Measurement	n.a.	440400	513100	85.8	16.49
Measurement 1-6	Agg.Factor	Viable Cells / mL	Total Cells / mL	% Viability	Peak Dia / μm
Average	n.a.	459567	532850	86.23	16.59
Standard deviation absolute	n.a.	14373	15559	0.41	0.1
Standard deviation %	n.a.	3.13%	2.92%	0.47%	0.84%

Standard deviations taking aggregation control into consideration

Sample	Agg.Factor	Viable Cells / mL	Total Cells / mL	% Viability
1 st Measurement	1.273	607900	686400	88.6
2 nd Measurement	1.266	575000	646300	86.4
3 rd Measurement	1.315	629200	701100	89.7
4 th Measurement	1.335	596500	669600	89.1
5 th Measurement	1.314	604400	676700	89.3
6 th Measurement	1.269	558800	631600	88.5
Measurement 1-6	Agg. Factor	Viable Cells / mL	Total Cells / mL	% Viability
Average	1.30	595300	668617	88.60
Standard deviation absolute	0.03	22864	23473	1.06
Standard deviation %	2.08%	3.84%	3.51%	1.20%

Conclusion

High reproducibility of CASY analysis:

+ Cell count StDv < 3.9%

+ Cell Viability < 1.2% StDv

+ Average viable cell diameter < 0.84% StDv

It was demonstrated that CASY allows **fast & simple sample preparation and measurement**. **Cell debris, live and dead cells** were discriminated without the need of dyes instantly during the measurement in size distribution plots (Fig. 1) and statistically in the results section. **Aggregation control** can easily be visualized in biomass plots (Fig. 2) and allows for accurate cell number determination in the statistical evaluation.

Contact

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