APPLICATION NOTE

Yeast Viability & Cell Count

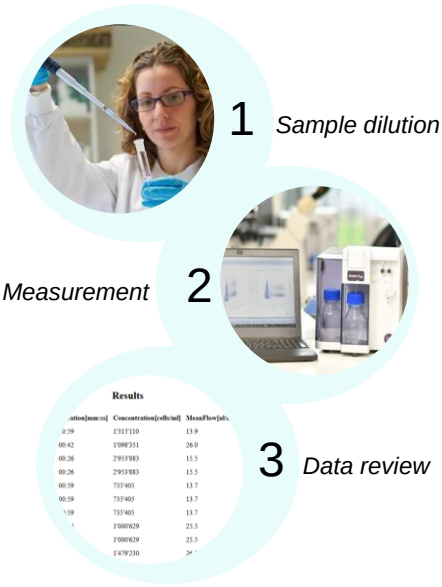
Yeast cultures are at the heart of many production processes for food and alcoholic beverages, the production of pharmaceuticals and even as nutritional supplements.

In brewing, viable yeast concentrations need to be determined to control pitching rates and to avoid production losses due to slow or stalled fermentations.

For the production of biopharmaceuticals, monitoring viability is important to maximize yields by optimizing fermentation times, for example by detecting the onset of apoptosis to stop the process before undesired side products are released.

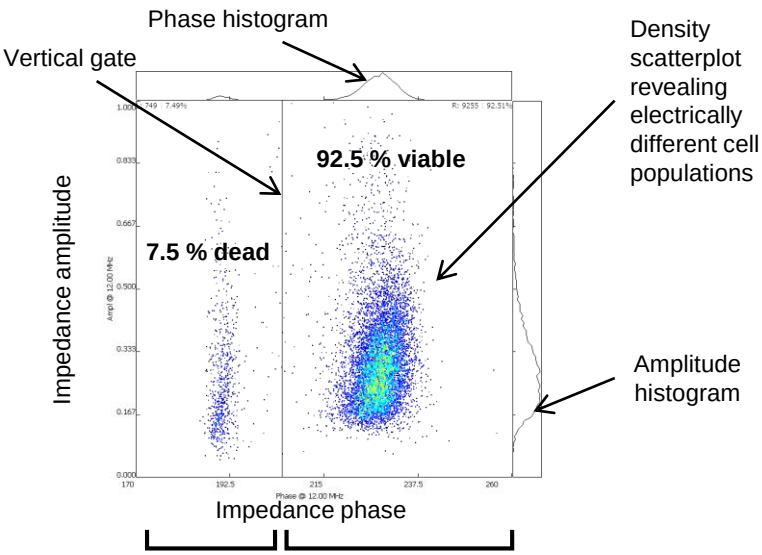
In research, most methods to determine cell viability or toxicity of a compound require labelling, which increases the required time and cost. Measuring viability by counting and distinguishing live, starving and dead cells with the Ampha Z32 does not require any pre-treatment or incubation. It allows to detect changing cell culture states (see page 2) and to pick up certain contaminations (e.g. *Lactobacillus*).

Rapid 3 - Step Workflow



Ampha Z32 – Cell Viability with IFC

Amphasys uses impedance flow cytometry (IFC) to rapidly characterize cells in suspension based on their electric properties. Impedance is measured in a microfluidic chip and results are displayed on a scatterplot. Software supported analysis allows to get precise statistics about individual cell populations.



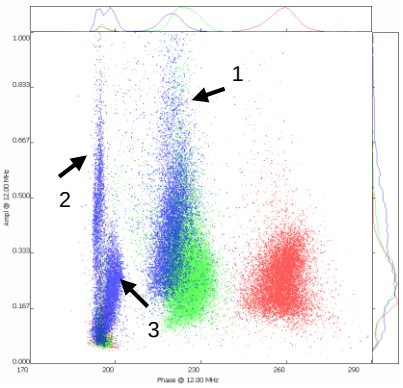
A scatterplot of yeast cells measured with the Ampha Z32 showing two distinct populations of dead and viable cells. Every dot represents the characteristics of one measured cell.

Cell Culture Development Monitoring

Proliferating cells in log phase, sampled after one day (red), are clearly distinguishable from cells in the stationary phase, sampled after two days of fermentation, which is in line with the switch from aerobic to anaerobic fermentation.

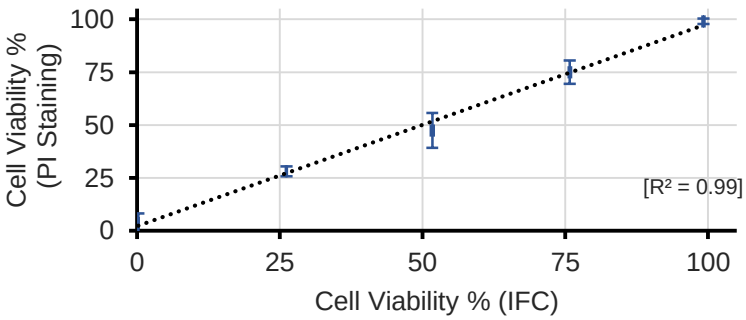
On the eighth day of the fermentation, populations appear higher up on the y-axis (1), indicating larger cell volume. At the same time an increase in dead cells (2) is observed. In addition, a new cell population is appearing (3), which is characteristic for late fermentation stages.

This data shows that high-resolution data obtained with IFC measurements offers an advantage over traditional staining methods, and provides a high level of control to guide process optimization and to make decisions during bioprocessing. This innovative technology provides immediate answers with minimal hands-on time.



An overlay of yeast cells sampled during different stages of beer fermentation shows characteristic impedance patterns. Day one (red), day two (green) and day eight (blue).

Method Comparison to Fluorescence Microscopy



A high correlation (R^2=0.99) between IFC and fluorescence microscopy was found, with higher precision for the IFC analysis due to a robust data analysis and significantly higher number of cells analyzed (average standard deviations of 3 independent viability determinations: IFC = 0.15 %, fluorescence microscopy = 4.4 %).

IFC Analysis offers:

Quality control through fast viability check
- Up to 1'000 cells/sec
- Viability in approx. 1 minute

A simple workflow
- No dilution needed up to 10^7 cells/ml
- No staining required

Reproducible:
- High precision due to large number of cells analyzed

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