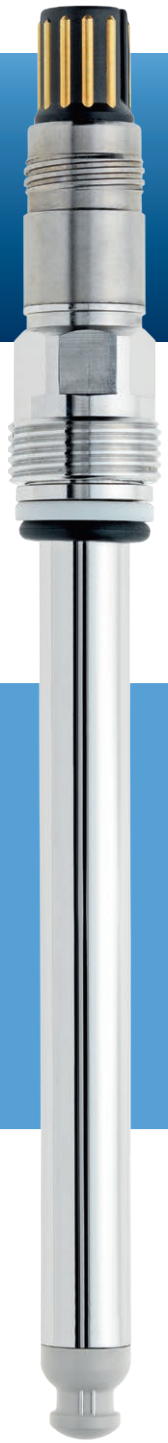
Online monitoring of CHO cells



Comparison of cell density measurement technologies in R&D

Industry: University
Application: Monoclonal Antibody production with CHO cells
Hamilton products: Incyte, PC Box

Chinese Hamster Ovary (CHO) cell lines are common host cells for the industrial production of monoclonal antibodies for therapeutic and diagnostic purposes. As the costs of growing mammalian cells are high any negative influence on the cells has to be avoided. Studies estimate that up to 15 % of cell culture experiments are misidentified or contaminated. Other issues that might be generated are nutrient depletion, accumulation of apoptotic cells, changes in the media such as pH value, or genetic alterations. Therefore sterile conditions of CHO cell cultures in bioreactors ensure economical and scalable production processes. Most of the information about the growth conditions can be gained by sampling and offline monitoring of the most relevant growth parameters as well as the cell viability. Various drawbacks come along with sampling and offline measurements such as risk of contamination, qualified personnel has to spend a lot of time onsite performing the analyses, and data are only generated at certain points in time, usually during working hours. Luckily, at least the cell growth can be monitored online and thus reduce the risk of missing events. The Technical University of Budapest, Hungary, is experienced in cell culture processes and has used the viable cell density technology provided by Hamilton to compare it to established offline methods.

Benefits Incyte

- Dilution in fed-batch easy recognizable
- Constant information about cell growth
- Reliable
- Convenient
- Increased yield
- Optimized feeding strategy

Determination of the cell factor

First, for each type of cells the cell factor has to be determined prior to the online measurement. During this procedure the correlation between permittivity and cell concentration is evaluated. Figure 1 shows the linear correlation. In order to determine the number of living cells present the cells need to be stained and counted once using an offline method. With the correlation factor the permittivity signal can be directly converted in a more convenient and meaningful dimension, i.e. cells/mL, or similar.

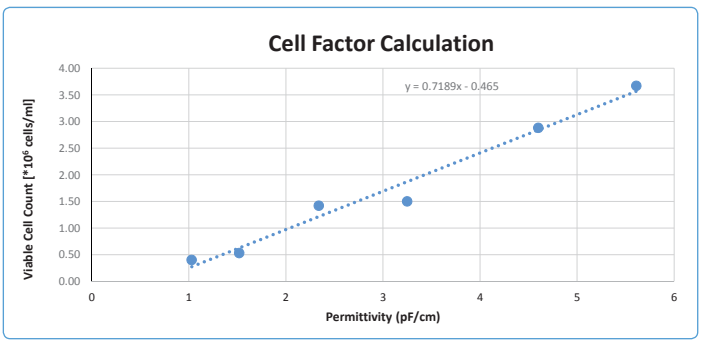


Figure 1: Calculation of factor between capacity and concentration of living cells.

Batch process

The first comparison was done in a batch process. Figure 2 shows that during the growth phase the different methods provide consistent cell density values. At a later stage of the process when cell densities are higher and nutrients are lacking, offline methods get less accurate and show different values than the inline sensor Incyte.

Fed-batch process

The three methods were also compared in a fed-batch process. At every feed 15% of the volume was added and carried out every 48 hours from the 72th hours after the start of the culture. The growth curve presented in Figure 3 shows that every feed was detected by Incyte immediately as the permittivity drops due to a dilution of the cells. Overall online and offline methods correlate very well during the growth phase of cells. On the other hand side figure 3 also demonstrates that the results of the offline methods depend very much on the time of sampling, resp. cell count.

Conclusion

All methods shown correlate well during the growth phase irrespective the type of process. As culture time increases the amount of cell debris increases accordingly. Cell debris might affect offline methods in a way that it'll be more difficult to use them properly. However, the Incyte sensor does not get noisy or somehow affected by a relatively high amount of dead cells or cell debris. The longer the process the more results deviate. This is because offline methods require more attention and labor so that it's hard to monitor cell culture processes very closely. Incyte sensors monitor online and permanently so that deviations of the ideal process can be detected instantly and corrective actions can be taken in real time to expand the growth phase leading to higher yields and more efficiency.

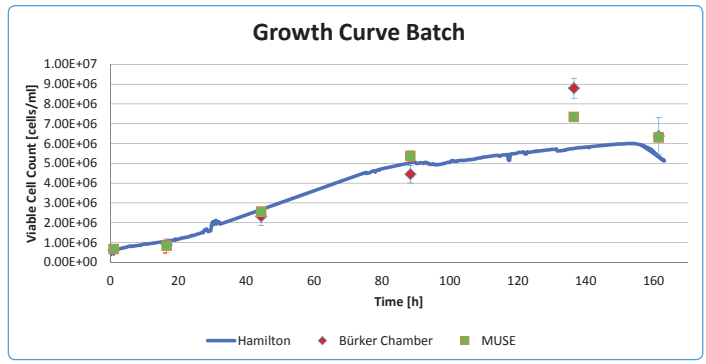


Figure 2: Growth curve of CHO batch culture by three different methods: Hamilton's real-time, in-situ capacitance sensor, and off-line measurement by Bürker-chamber and MUSE (Millipore) cell counter.

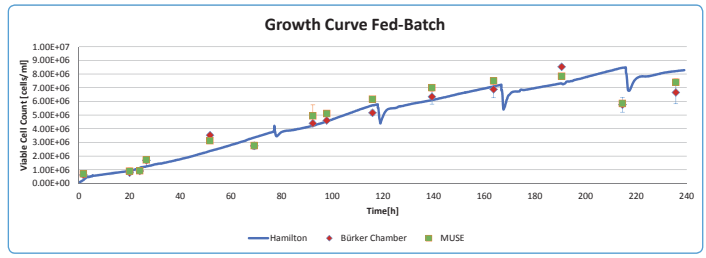


Figure 3: The growth curve of CHO fed-batch culture generated by three different methods: Hamilton's real-time, in-situ capacitance sensor, and off-line measurement by Bürker-chamber and MUSE (Millipore) cell counter.

Author

Eszter Szabó, Annamária Fricska, Edit Hirsch, András Ballagi*

Budapest University of Technology and Economics, Biotechnology unit at Pharmatech Model Laboratory, Hungary, H-1111 Budapest, Muegyetem rkp. 3

**present address*
Diagon Ltd.
Hungary, H-1047 Budapest, Baross u. 48-52

REF 695225/00 — 09/2017

HAMILTON®

Web: www.hamiltoncompany.com
USA: 800-648-5950
Europe: +41-58-610-10-10

Hamilton Americas & Pacific Rim
4970 Energy Way
Reno, Nevada 89502 USA
Tel: +1-775-858-3000
Fax: +1-775-856-7259
sales@hamiltoncompany.com

Hamilton Europe, Asia & Africa
Via Crusch 8
CH-7402 Bonaduz, Switzerland
Tel: +41-58-610-10-10
Fax: +41-58-610-00-10
contact.pa.ch@hamilton.ch

To find a representative in your area, please visit www.hamiltoncompany.com.