

Monitoring Viable Cell Density for Improved Bioreactor Control



Cell culture and fermentations are used to produce pharmaceutical products or other molecules of interest. To achieve maximum efficiency or quality assessment during the synthesis of targeted products, on-line information about the active metabolic cell concentration is of crucial importance. Although the biomass is the catalyst of the bio-production and its concentration is a key parameter in the reaction, the bioreactors are still commonly monitored with only usual pH, temperature, and dissolved oxygen sensors. In fact the optical techniques that have been proposed to monitor the biomass suffer from a number of disadvantages such as sensitivity to cell debris and other media components or loss of linearity.

From its 20 years of experience in capacitive metrology, FOGALE nanotech has introduced the direct capacitance measurements into a robust, precise and reliable system.

MEASUREMENT PRINCIPLE

The use of capacitance for on-line process monitoring is very attractive and selective by only taking into account the viable cells : under the influence of an electric field between two electrodes, only cells with intact plasma membranes are polarized and behave as tiny capacitors. The resulting capacitance (medium Permittivity) can be measured by a second electrode pair and is accurately and linearly correlated with viable biomass concentration. Dead cells with broken membranes do not contribute to the signal.

ABOUT THE BIOMASS SYSTEM

From the Permittivity measurement technique FOGALE nanotech has recently launched the new generation of on-line biomass monitoring tools. The patented BIOMASS SYSTEM is an affordable sensor for viable cell density monitoring, it is directly

inserted into the bioreactor with in-situ sterilizable standard 12mm and 25 mm diameter probes to fit any bioreactor port. The probes design and materials are ideal for working in a cGMP environment as the wetted material conform to FDA requirements (PEEK, 316L stainless steel).

The BIOMASS SYSTEM is ideal for monitoring animal cell in suspension (including clumping cell lines) or attached to microcarriers, it is also routinely used for yeast or bacteria batch and fed-batch processes with high biomass concentration up to 200g/l dry weight. It includes a unique triple frequency measurement that results in insensitivity to cell size variations and high accuracy in the signal.

CELL CULTURE OPTIMIZATION DURING PROCESS DEVELOPMENT

Figure 1 demonstrates how the on-line Permittivity signal can be used to monitor viable cell density during a 2 L CHO batch culture. The BIOMASS SYSTEM (fitted with a 12 mm diameter sensor) Permittivity is providing a linear correlation with viable cell count over the complete timecourse of the culture. In addition, it is obvious from figure 2 that changes in cell metabolism can be monitored by means of the Permittivity measurement. The first drop in Permittivity corresponds to a metabolic shift where glutamine becomes a limiting nutrient; the oxygen uptake rate also decrease significantly. Alanine is accumulating until glutamine is depleted and is subsequently consumed. The second drop is correlated to the exhaustion of alanine and glucose that marks the beginning of the death phase. The same patterns are obtained from a larger scale culture with a 25 mm diameter sensor.

By providing real time signals, the BIOMASS SYSTEM is a useful tool for cell culture process monitoring and control and process optimization.

Geoffrey Esteban is product manager at FOGALE nanotech, Park Kennedy - Bât A3, 285 rue Gilles Roberval, CS 32028, 30915 Nîmes Cedex 2, France; 33-466-620555, fax 33-466-627160; info@fogale.com; www.fogale.com/Biomass.

Figure 1: Permittivity, viable and total cell counts during a CHO 2 L batch culture (Data courtesy of F. Hoffmann-La Roche Ltd. 2004)

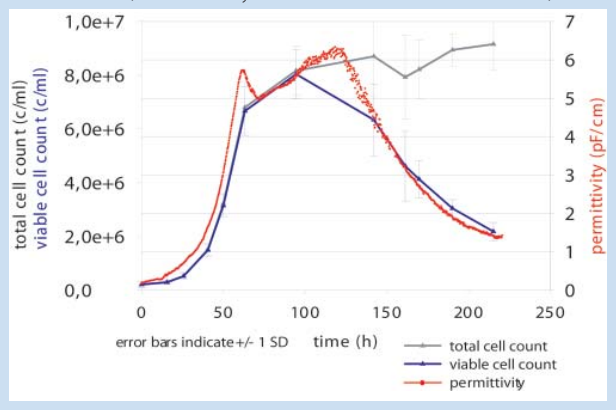


Figure 2: Permittivity, volumetric O₂ uptake rate, and main nutrient concentrations during a CHO 2 L batch culture (Data courtesy of F. Hoffmann-La Roche Ltd. 2004)

