Single-Use shake flasks for online monitoring of process parameters

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Introduction
Shake flasks are still a preferred system for cultivation of cells or small scale production of cell-related products. They are easy to use, affordable and the manufacturing process is simple. Our goal was to establish an analytical system in order to monitor dissolved oxygen, pH and cell density online. Especially single-use flasks and sensors are suitable to sustain GMP/GLP standards during growth and production process.

Principle of measurement
The non-invasive measurement of biomass is performed by detection of 180° scattering light at a dominant wavelength of 625 nm. The included acceleration sensor triggers each measuring at the moment of maximum liquid level (synchronized) (Fig. 1/3). The measuring interval can be reduced to 7s. pH value and O₂ saturation are measured optically by specific sensor spots (Fig. 2). In order to prevent the effect of boundary layer reflections a simple bezel was attached to the flask bottom (Fig. 4). In addition it provides a measuring zone with constant width.

Non-invasive online monitoring
Since suspension-cell cultures of mammalian strains, such as CHO-K1, are quite sensitive to contamination, online monitoring via light is an appropriate alternative to classical offline sampling. Cell proliferation can be detected from 6·10⁴ - 15·10⁸ cells·mL⁻¹ (and more). In the case of low density cultivations, calibrations with the cell count exhibit a linear correlation (Fig. 5/6). The pH measurement is limited by the given range of 5.5-8.0, which is compensated by a high resolution of 0.01 pH units. In respect to the resolution of DO measurement is high with 0.1 % absolute saturation (Fig. 7).

Conclusion
The combination of single-use shake flasks offers a complete non-invasive monitoring of the most critical process parameters in mammalian cell culture. Calibration with cell count is possible in the common growth range. pH and pO₂ measurements provide additional informations about the growth phase and oxygen supply in the individually used cultivation system.

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