Simplifying Progress

Investigating the Use of Automation and Integrated Analytics to Enhance Cell Line and Process Development Capabilities

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Abstract

Automated small-scale bioreactor systems like the Ambr® 15 and Ambr® 250 have been proven to increase the speed and throughput of cell line and process development. Combining the Ambr® systems with integrated analytics like pH modules, cell counters, and metabolite readers not only ensures accuracy and consistency of the measurements but also allows for automatic data transfer and advanced control strategies.

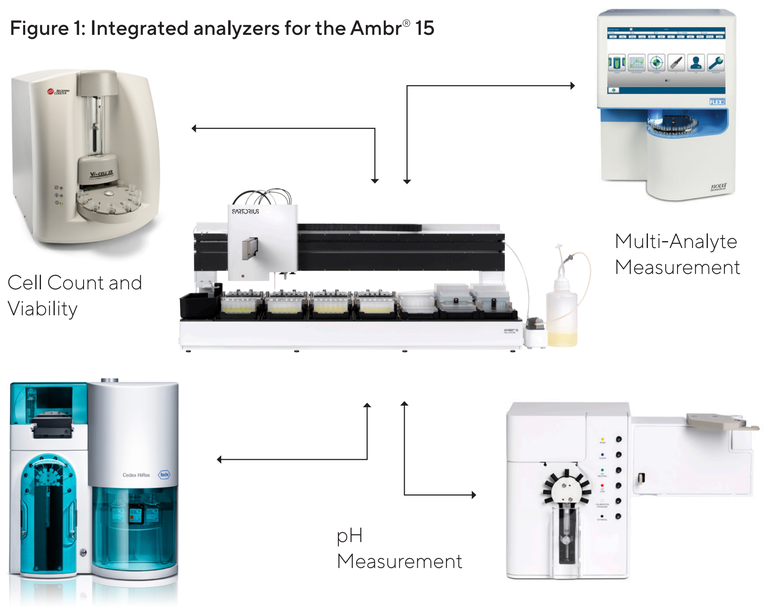
This poster highlights the work carried out to evaluate these integrated systems, which includes comparing manual versus automated samples and implementing automated feedback control using both cell count and glucose measurements for a CHO fed-batch process.

Key takeaways from the case study to be presented include identifying glucose control strategies that led to increased peak cell densities and prolonged culture duration as well as showing that automated sampling and data transfer allows for walk-away glucose control.g in reduced bioprocess complexity and increased process understanding.

Materials and Methods

The Ambr® systems can be used with a suite of integrated analytics (Figure 1) for automated sampling, analysis and data transfer to the Ambr® software. These include the Vi-CELL™ Cell Viability Analyzer (Beckman Coulter Life Sciences) and Cedex HighRes Analyser (Roche Innovartis) for cell count and viability, the Ambr® 15 analysis module (Sartorius Stedim Biotech) for pH and the BioProfile® FLEX2™ Automated Cell Culture Analyzer (Nova Biomedical) for multi-analyte measurement.

Figure 1: Integrated analyzers for the Ambr® 15

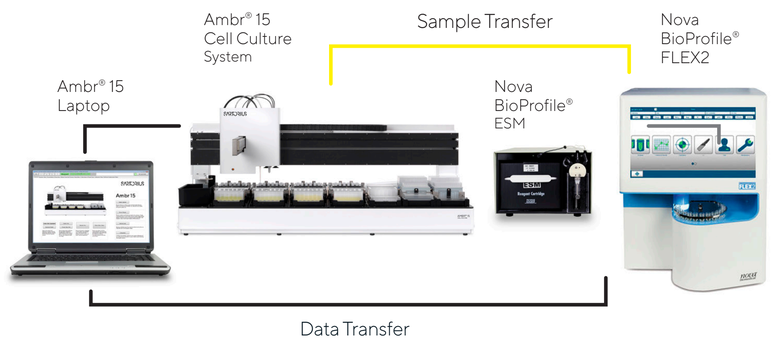


Integrated Metabolite Analytics:

The BioProfile® FLEX2™ directly connects to the Ambr® systems via an External Sampling Module (ESM) (Figure 2). The Ambr® liquid handler withdraws a sample from the bioreactor to the sample cup which is routed to the FLEX2 via the ESM. Once the FLEX2 analysis is done, the data generated is transferred directly back to the Ambr® software. The software then tracks and processes the data. If required, the Ambr® software can be programmed to perform in-run calculations e.g. doubling time, growth rate and feed addition volumes.

The powerful combination of Ambr® with FLEX2 enables fully integrated automatic collection of up to 16 cell culture parameters, including total and viable cell density, cell viability and diameter, pH, pCO_2, pO^2, glucose, lactate, glutamine, glutamate, ammonium, Na+, K+, Ca++ and osmolality, which can be sampled and measured within a cycle time of ~6 minutes.

Figure 2: BioProfile® FLEX2™ integrated with the Ambr® 15 Cell Culture system



Integrated pH Analytics

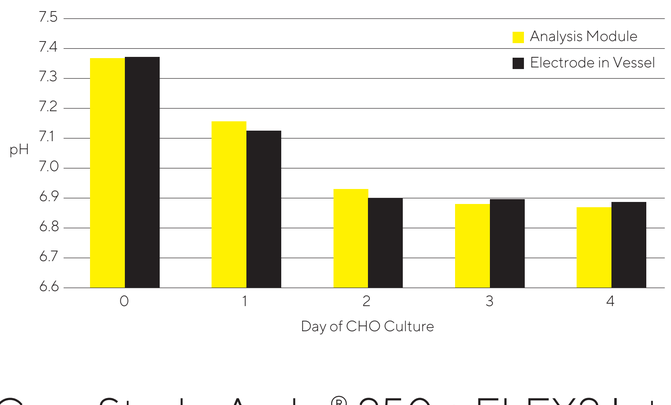
Benefits of the Ambr® Analysis Module:
- Fully integrated at-line pH assay
- Fits into Ambr® 15 and alongside Ambr® 250 biosafety cabinets
- Close coupling and custom liquid handling to prevent sample degassing

Figure 3: Analysis Module specifications

Working range	1.0 - 9.0 pH
Sample volume	60 µL (Ambr® 15)
	250 µL (Ambr® 250)
Cycle time per read	90s (Ambr® 15)
	120s (Ambr® 250)
Resolution	0.01 pH
Calibration buffer accuracy	± 0.01 pH
Minimum sample temperature	ambient +3°C

Analysis Module performance:
- CHO cell culture R&D test in Royston, UK
- N = 24 bioreactors (Ambr® 15)
- ~10^6 cells/mL at Day 6
- Comparable results between analysis module and traditional pH probe over six day culture

Figure 4: Time course comparison of pH measurements taken automatically using the Ambr® Analysis Module (black) vs. manually inserting a Mettler pH electrode into the Ambr® vessel (yellow).



Case Study: Automated Glucose Strategies

- Collaboration between MIT Chemical Engineering Department, NOVA Biomedical, and Sartorius Stedim Biotech
- CHO cell culture with different conditions (Figure 5) and glucose control strategies (Figure 6)
- Automated sampling and data transfer allowed for walk-away glucose control
- Glucose control strategies were identified that led to higher peak cell densities and prolonged culture duration (Figure 7)
- Average cell specific glucose consumption was 9.13 x 10^-12 g/cell which is consistent with values reported in literature

Figure 5: Culture station layout describing conditions for the Ambr® 15 experiment

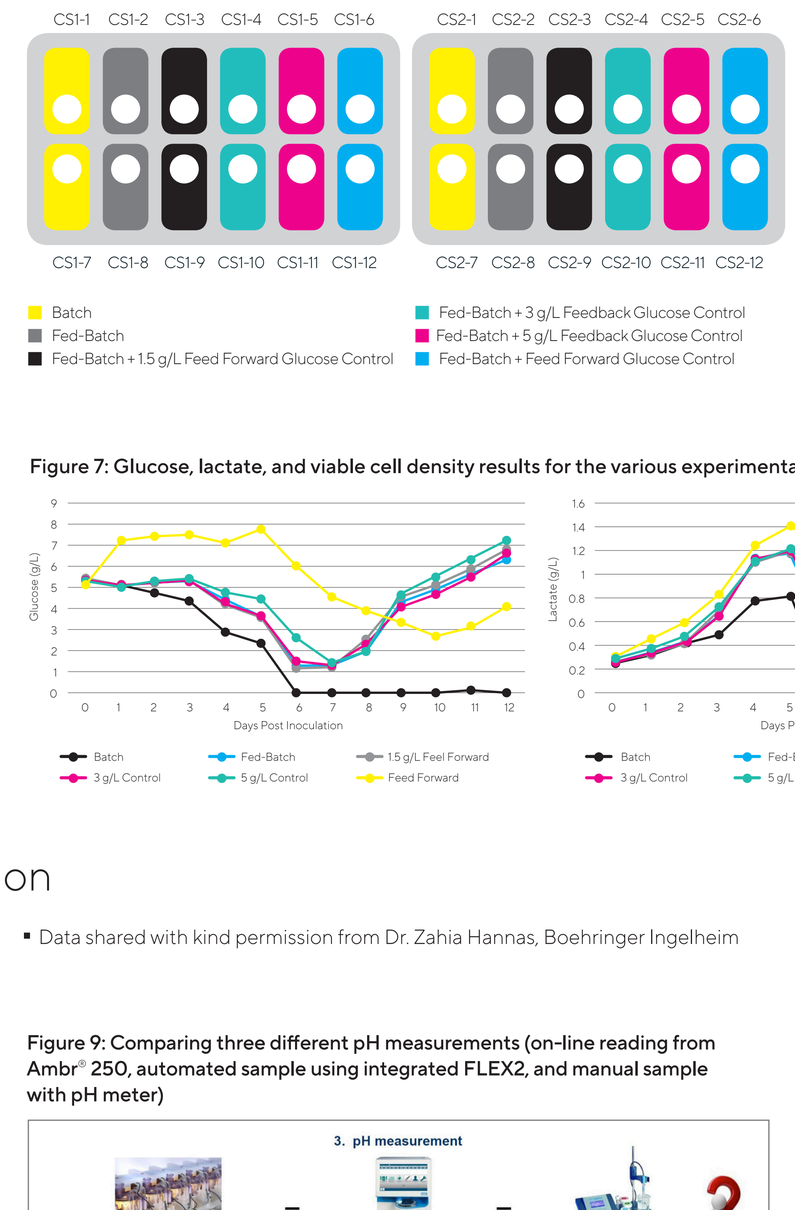


Figure 6: Automated feedback and feed forward glucose control strategies

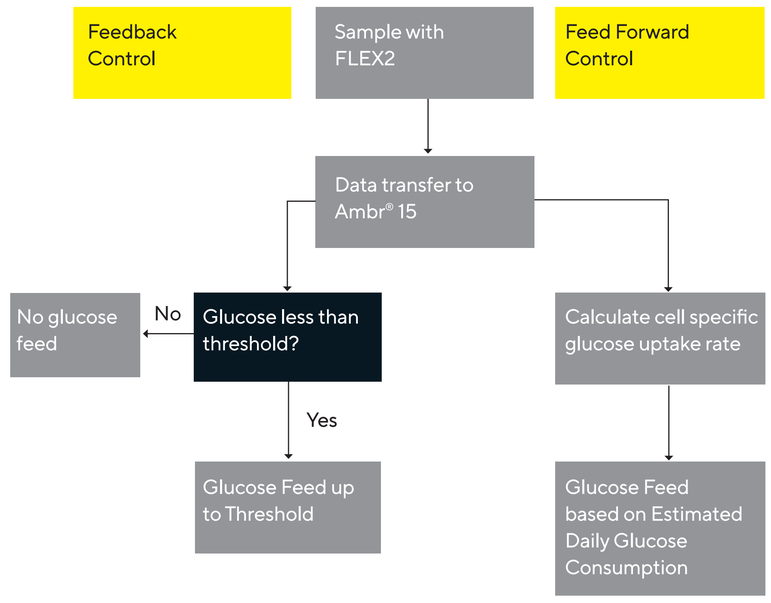


Figure 7: Glucose, lactate, and viable cell density results for the various experimental conditions

Case Study: Ambr® 250 + FLEX2 Integration

- Experiments done by an industry test partner shows comparable results between the automated and manual sampling for viable cell density (VCD), total cell density (TCD), viability, and pH (Figures 8 and 9)
- Data shared with kind permission from Dr. Zahia Hannas, Boehringer Ingelheim

Figure 8: Comparing automated vs. manual sampling for VCD, TCD, and viability at three different cell densities (1, 3, and 7 million cells/mL)

Figure 9: Comparing three different pH measurements (on-line reading from Ambr® 250, automated sample using integrated FLEX2, and manual sample with pH meter)

Summary and Outlook

- Integrating the pH Ambr® Analysis Module and FLEX2 with the Ambr® systems provides accurate and consistent measurements
- Using integrated analytics allows for advanced culture monitoring and control outside of working hours or from different locations
- Automation and integrated analytics enables enhanced, rapid cell line development and process optimization to cost-effectively advance speed-to-clinic for programs in the development pipeline

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