Real-Time Concentration Prediction of Proteins, LNPs, and Cells Using Viscosity-Based Regression Modelling

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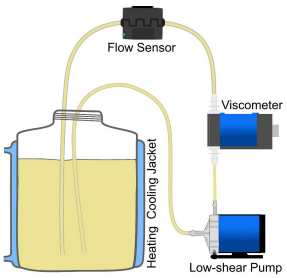
Introduction

Accurate concentration measurements of bio-pharmaceutical modalities are crucial for ensuring robust manufacturing processes and high-quality products. However, the availability of accurate real-time measurement methods and sensors remains limited, requiring at-line or off-line analysis. Viscosity in particular remains an underestimated process parameter that can be leveraged across various modalities to enable real-time concentration prediction, allowing for much tighter process control.

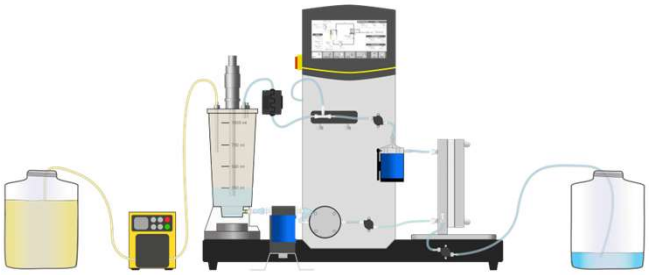
Experimental Approach

A single-use inline viscometer was developed to measure both fluid viscosity and temperature. Regression modeling was applied based on the viscosity-temperature dependence of biopharmaceutical solutions. By calibrating the inline viscometer at various concentrations and varying temperatures, concentration prediction models were established for proteins (mAbs), LNP-like test materials (Lecithin), and cell cultures.

Calibration Setup



During process operation, the viscometer can be readily integrated into the process stream to measure real-time concentration.

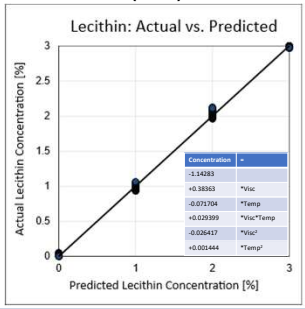


Results and Discussion

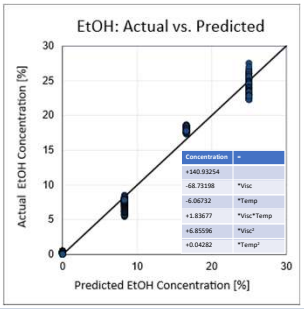
Calibration Models

- A reliable correlation between Lecithin (LNP) concentration and viscosity could be established by linear regression, resulting in a polynomial quantification formula.
- Similar prediction models were generated for EtOH concentration and for protein concentration.

Lecithin (LNP) Model

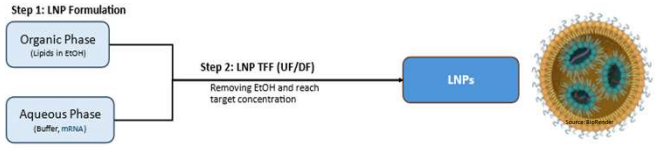


EtOH Model

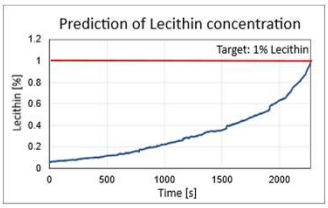


Process Monitoring of Lecithin (LNPs) in TFF UF/DF:

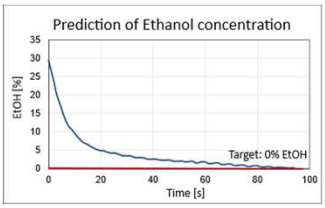
- UF: Real-time monitoring of Lecithin (LNP) concentration during TFF operation was possible due to real-time temperature and viscosity measurements by the inline viscometer. Lecithin concentration can be monitored based on the established polynomial quantification formula.
- DF: EtOH removal could similarly be monitored based on the previously established prediction formula.



UF: Lecithin (LNP) Concentration

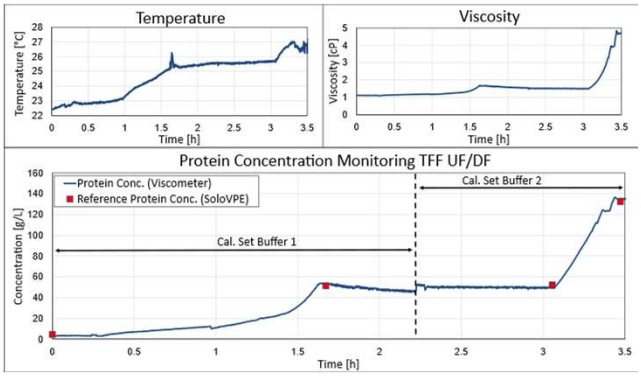


DF: EtOH Removal



Process Monitoring of Proteins (mAb in TFF UF/DF):

- Real-time monitoring of protein concentration during TFF operation was possible due to real-time temperature and viscosity measurements by the inline viscometer. Concentration could be monitored based on the established polynomial quantification formulas.
- Viscometer based protein quantification aligned well with offline reference analytics (SoloVPE).



Source: Zsofia Bencze, Ferring Pharmaceuticals, Levitronix Bioprocessing Conference 2024, Boston, MA, USA

Conclusion and Outlook

The calibration approach enabled the development of accurate and reliable real-time concentration prediction models for various biopharmaceutical modalities. These models facilitate tighter process control in key unit operations, such as TFF UF/DF steps, LNP formulation, and protein compounding, but also with cells. Beyond concentration measurements, viscosity readings themselves provide valuable insights, particularly for optimizing filtration processes, where viscosity significantly impacts filtration performance.