Use of Fusion QbD for Automated Method Screening for Biotherapeutics

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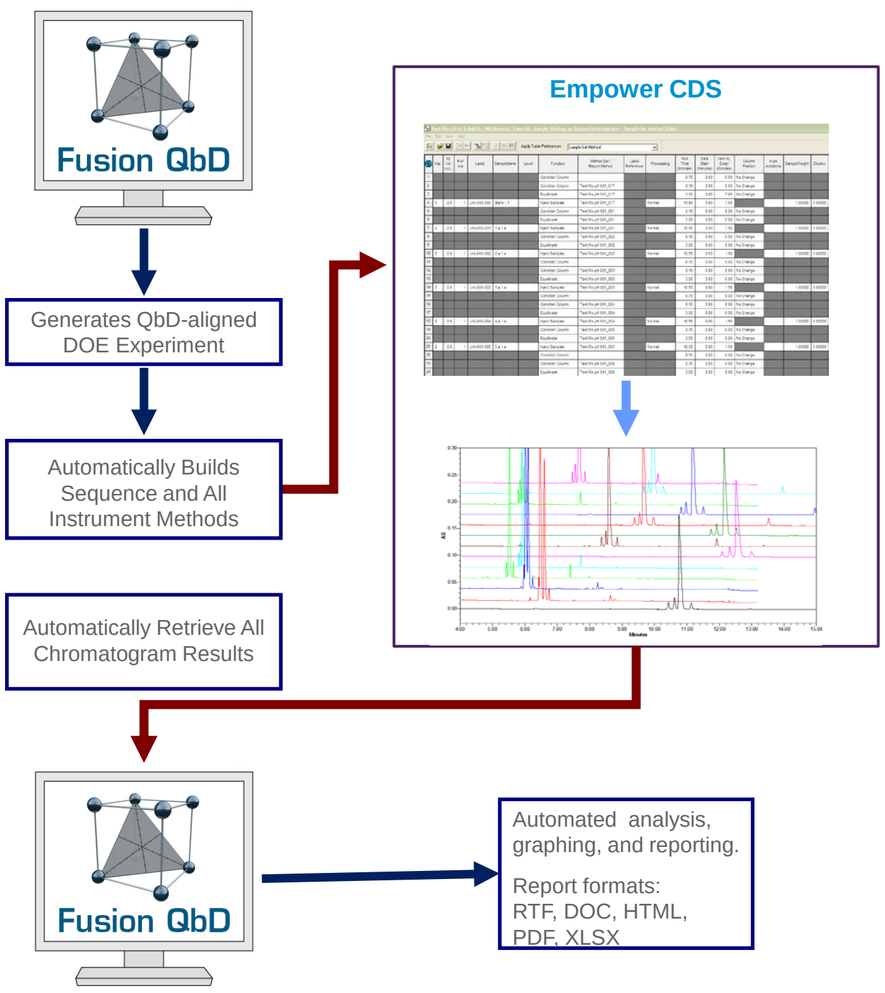
ABSTRACT

Analytical organizations focused on biotherapeutics spend the bulk of their time investment pursuing robust methodologies that ensure drug substances and drug products are pure and stable. In order to achieve faster delivery of therapies to patients, those organizations are continually improving the method development process. One way to improve method development throughput is by moving towards automated method screening and automated method optimization.

Fusion QbD software (S-Matrix Corporation, Eureka CA) was used to automate the screening process for several different modes of chromatography. Fusion allows the user to input relevant chromatographic variables dependent upon which mode of chromatography is being evaluated. Fusion then uses statistical-based experimental design to assess all chosen variables. The design can be exported to Empower to automatically generate method files, which eliminates a large portion of the method development effort. After running the generated methods, results can be imported back into Fusion for modeling and evaluation of each chromatographic variable.

Fusion Workflow

QbD aligned DoE is generated in Fusion with user defined variables. Fusion writes methods from the DoE into Empower. Metrics from processed chromatograms can be brought back to Fusion to determine optimal chromatographic variables.



Case Study 1 - WCX Development

Fusion QbD Screening and Optimization

- 69 Instrument methods generated by Fusion and exported into Empower 3
- 5 Full time employee (FTE) hours, 120 instrument hours
- Variables in DoE: pH, gradient time, mobile phase composition, organic additive, salt concentration, and column temperature
- Resulting method showed no fronting, better resolution of acidic species, and better resolution of basic species
- Resulting method comparable to method developed in 5 months prior to use of Fusion QbD

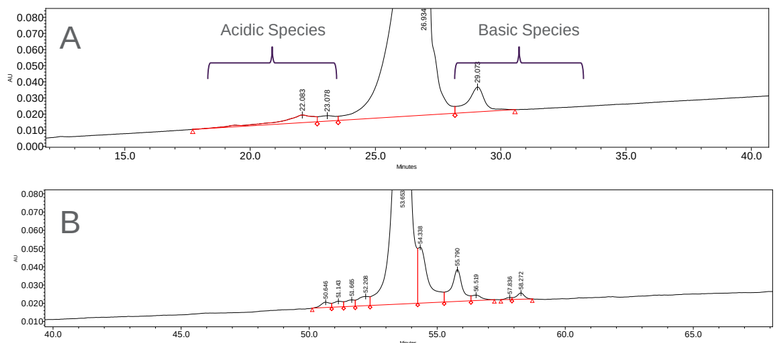


Figure 2. WCX HPLC Zoom of (A) Method prior to development and (B) Fusion QbD generated method showing increased resolution of both acidic and basic species.

Case Study 1 - Overlay Plots

Overlay Plots were generated for WCX HPLC using data imported back into Fusion from Empower. The areas in white highlight acceptable performance regions based upon user defined criteria. Colored regions indicated operating ranges that do not meet the nearest listed requirement.

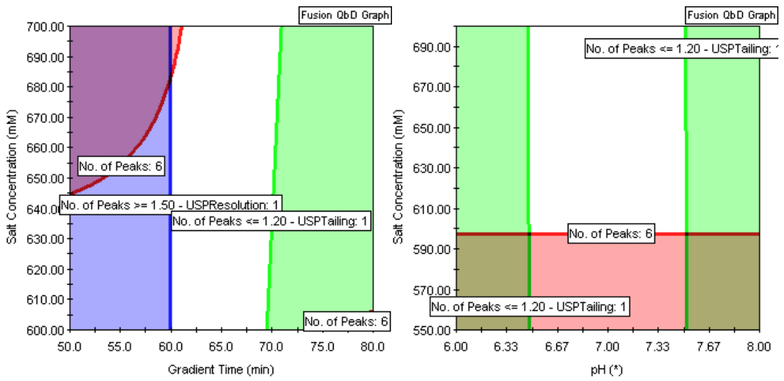


Figure 3. Overlay Graphs showing acceptable performance regions in white. Left: Salt Concentration vs. Gradient Time. Right: Salt Concentration vs. pH.

Case Study 2 - HILIC Development

Fusion QbD Screening

- 38 Instrument methods generated by Fusion QbD and exported into Empower
- 2 Full time employee (FTE) hours and 15 instrument hours
- Variables in DoE: pH, column temperature, gradient time
- Resulting method shows increased resolution between Protein 1 and Protein 2 in addition to less tailing of both protein peaks

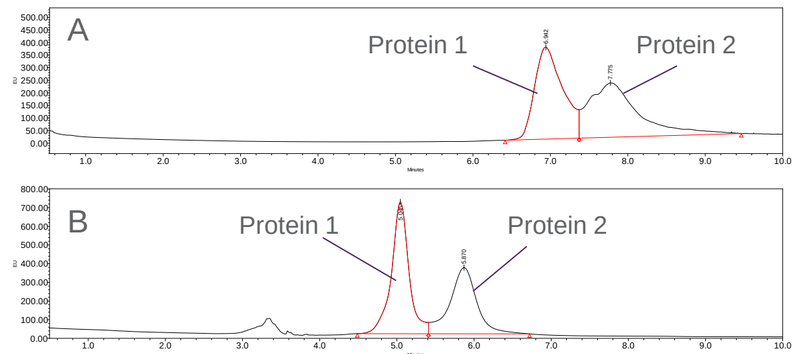


Figure 4. HILIC HPLC Chromatogram of (A) Original method for separation of Protein 1 from Protein 2 and (B) Fusion QbD generated method showing increased resolution and less tailing.

CONCLUSION

Fusion QbD was successfully used to generate design of experiments (DoE) for hydrophobic interaction (HILIC) and cation-exchange chromatographies needed for the analysis of biotherapeutics. Using the method export function in Fusion, methods were automatically created in Empower according to the DoE that was built by Fusion. Results from the automated screening achieved the goals of better resolution in the HILIC method, and increased peak count and resolution in the cation-exchange method. Processed Empower 3 results from the cation exchange method were then imported back into Fusion and modeled to provide optimal operating space for relevant chromatographic variables. The amount of time saved using Fusion QbD is estimated at 2.5 full time employees (FTE's) over the course of a month.

ACKNOWLEDGEMENTS

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