Modelling of analytical (U)HPLC: An important element in the QbD toolbox

Pharma Lab Congress 2013, Düsseldorf, Germany, November 13-14th 2013
Petersson, Patrik, Novo Nordisk A/S, +45 3079 2146, ppso@novonordisk.com

Outline

- QbD in general
- Analytical QbD at Novo Nordisk
- Modelling of analytical (U)HPLC
Quality by Design (QbD)

“A systematic approach to development that begins with predefined objectives and emphasizes product and process understanding and process control, based on sound science and quality risk management”

*from ICH Q8 pharm. dev.*
Quality by Design for analytical methods

- The pharma industry presented their vision for analytical QbD 2009*
- Expectation of regulatory relief

*EFPIA/PhRMA white paper to FDA and EMA 2009

Currently no guidance
EMA/FDA - “There is currently no international consensus ... until this is achieved, any application that includes such proposals will be evaluated on a case-by-case basis.”*

- No regulatory relief at this stage
- A way to increase method understanding and thereby improve performance and robustness

*EMA-FDA pilot program for parallel assessment of Quality-by-Design applications: lessons learnt and Q&A resulting from the first parallel assessment EMA/430501/2013
Analytical Quality by Design at NN

Analytical Target Profile
- "User requirement specification"
- Giving clear directions for analytical development
- Ensures agreement between stakeholders
- Internal targets – no regulatory relevance

- Method specific targets
  - Performance aspects typically included in validations (accuracy ...)
  - User aspects (handling, safety ...)
  - Customer aspects (re-runs, cycle time ...)
  - Sample aspects (matrix, ranges ... )
  - ...

Risk assessment
Method understanding
Control strategy
Knowledge management
Analytical Target Profile

- Evaluation against
- Validation data
- Customer/user experiences

14 targets for content
Analytical Target Profile

• Special focus on intermediate precision and specifications/OOS
• Validation
• Control sample included in routine analysis sample sets
• Stability study residual variation

Risk assessment

• Method specific risk assessment and knowledge sharing
  • Brainstorm to share knowledge, spot problems/possibilities and anchor actions
  • At transfer to/from discovery, external vendors or QC
  • Different focus (development, re-runs, robustness ... )

<table>
<thead>
<tr>
<th>Failure Mode</th>
<th>Severity (A)</th>
<th>Probability (B)</th>
<th>Detection (C)</th>
<th>Risk</th>
<th>RP, RPMP, EPM</th>
<th>Action Taken</th>
<th>Responsible and Target Completion Date</th>
<th>Action Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Systematic error in column temperature due to difference in HPLC system design effect resolution between API and adjacent impurities</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>15</td>
<td>SST for resolution between temp</td>
<td>SST section of method updated</td>
<td>EPM N</td>
<td>FR, NO</td>
</tr>
<tr>
<td>Very broad or even split peaks obtained when injecting more than 10 µL. This is probably related to the injection of a solvent containing impurities</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>15</td>
<td>No change to the method</td>
<td>EPM N</td>
<td>FR, NO</td>
<td></td>
</tr>
<tr>
<td>Void volume or response anomaly</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>15</td>
<td>No change to the method</td>
<td>EPM N</td>
<td>FR, NO</td>
<td></td>
</tr>
</tbody>
</table>
Method understanding (by modelling)

- Some techniques generic and not optimized or typically involve one factor at the time optimization (e.g. spectroscopic)

- Most techniques are well suited for optimization and/or robustness testing based on statistical models (DoE)

- Chromatography also suitable for models based on theory (later ...)

\[ t_R = \left[ \frac{t_g F}{m \Delta \Phi} \log(2.3 V_M \frac{\Delta \Phi}{t_g F k_0 + 1}) + V_M + V_D \right] \frac{1}{F} \]

Control strategy

- To ensure that the analysis is fit purpose each time
- Involves
  - System suitability tests
  - Selection of parameters
  - Definition of data based acceptance criteria
  - Adjustments ... later
- Control charts
- Instructions for data evaluation
- Preventive maintenance parameters
Method operational design ranges (MODR)

- MODR = design space for analytical method
- MODR with acceptable results typically very small for (U)HPLC

*EMA-FDA pilot program for parallel assessment of Quality-by-Design applications: lessons learnt and Q&A resulting from the first parallel assessment EMA/430501/2013*
Method operational design ranges (MODR)

- We do not define any MODR for chromatographic methods
- Instead SST to justify changes in %ACN and temperature to compensate batch to batch and instrument to instrument differences
- Described in EP, USP and JP ±5°C
- Graphical guidance e.g.

Knowledge management

- Concise method specific development report
  - A table with conclusions and references
  - Purpose - easy documentation/retrieval of method development and validation information
  - Continuously updated

- “Follow the molecule”
  - Hands on experience and personal contacts
Analytical Quality by Design at NN

- Analytical Target Profile
- Risk assessment
- Method understanding
- Control strategy
- Knowledge management

Method understanding by modelling

- Generic screen of columns and mobile phases
- DoE: Optimisation of mobile phase composition
- Semi-theoretical models: Optimisation of gradient and temperature
- DoE: Robustness testing
Semi-theoretical models

Linear gradient ⇒ analytical solution

\[ L = \int_{0}^{t_{R}} u_{0} \frac{1}{k + 1} dt \]

Non-linear gradient ⇒
Numerical solution
Non-linear least squares fitting

\[ t_{R} = \left[ \frac{t_{G}F}{b\Delta\Phi} \log(2.3V_{M} \frac{\Delta\Phi}{t_{G}F} k_{0} + 1) + V_{M} + V_{D} \right] \frac{1}{F} \]

Solvent strength models

- The retention models that can be found in most commercial software have been developed for RPC and small molecules
- Novo Nordisk is a company developing products based on proteins
- In order to model proteins other chromatographic techniques and retention models are needed
- Collaboration between and
Solvent strength models

- RPC Reversed phase chromatography
  $$\ln k = a + b\Phi$$
- IEC Ion exchange chromatography
  $$\ln k = a + b\ln(\Phi)$$
- HILIC hydrophilic interaction chromatography
  $$\ln k = a + b\ln(\Phi)$$
- HIC hydrophobic interaction chromatography
  $$\ln k = a + b\Phi$$
- NPC Normal phase chromatography
  $$\ln k = a + b\ln(\Phi)$$


Proteins respond much more strongly to changes in %ACN

$$\ln k = a + b\Phi \approx a - 1.1MW^{0.44}\Phi$$

<table>
<thead>
<tr>
<th>MW</th>
<th>b</th>
<th>b(MW)/b(100)</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>8</td>
<td>1</td>
</tr>
<tr>
<td>1000</td>
<td>23</td>
<td>3</td>
</tr>
<tr>
<td>10000</td>
<td>63</td>
<td>8</td>
</tr>
<tr>
<td>100000</td>
<td>174</td>
<td>21</td>
</tr>
</tbody>
</table>

Temperature models

\[ \ln k = a + \frac{b}{T} \]

\[ \ln k = a + \frac{b}{T} + \frac{c}{T^2} \]

Small molecules - M. Euerby et al., Hichrom Ltd
Proteins – P. Petersson et al., Novo Nordisk A/S
Input

- From 2, 6 or 9 experiments we need:
  - Retention time
  - Peak width (main + 1 to 2 imps.)
  - Peak area
  - Peak asymmetry for main peak
  - Dead volume
  - Dwell volume
  - Flow

A 1st example: Protein >100 kDa

- Demonstration
- [LINK]
- Improved resolution
- Improved robustness
- Isolation/preparative
A 2nd example: Peptide map ~0.5-3.5 kDa

- In general:
- 60 to 80% reduction in cycle time
- Maintained or better resolution
- Improved robustness
- Identification of critical peak pairs

A 3rd example: Excipient <0.5 kDa

- 2 methods → 1
Accuracy

- $\Delta t_R < 2\%$ and systematic
- $\Delta w < 20\%$
- $w \times 1.2 \Rightarrow R_s \times 0.83$

Peak tracking

- Peak areas
- Retention change linear or somewhat curved
- Guess and confirm
- MS!!!
Peak tracking overwhelming?

- Model the first and the last peak
- Model the API and critical peak(s) which are easy to follow
- Ignore the rest

Peak tracking overwhelming?

- Predict e.g. 6 conditions which gives good retention and resolution for the peaks which have been modelled

R_s-map based on modelled peaks
Peak tracking overwhelming?

- Test these conditions experimentally
- Pick the condition that gave the best results

What the R²-map would look like if all peaks had been modelled

Conclusions

- Analytical QbD
  - NN interpretation of analytical QbD
  - A way to obtain better methods
    - Understanding
    - Performance
    - Robustness
Conclusion

• Semi-theoretical modelling
  • Works very well also for large proteins after some adaption
  • Not all softwares provide the necessary models
  • Application areas:
    • Optimisation
    • Robustness evaluation (DoE still needed)
    • Fraction collection
    • Troubleshooting

Acknowledgements

• Anders Dybdal Nielsen
• Anette Skammelsen Schmidt
• Babak Jamali
• Birgit Gaarde-Nielsen
• Bonnie Schmidt
• Helle Holton
• Jytte Pedersen
• Karin Juul Jensen
• Mads Wichmann Matthiessen
• Marika Ejby Reinau
• Steffen Frandsen
• Thorbjørn Strøm-Hansen
• Tine Sloth Høg