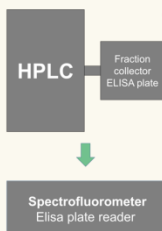


## Third-order data modelling – HPLC-EEM data analysis

**Methodology 1**  
**Collection of fractions**Flow rate: 2 mL min<sup>-1</sup>

- For each sample:
  - 1 Chromatographic run collecting 25 fractions, every 2 s = 2 min
  - 25 EEMs = 40 min.

**Methodology 2**  
**Multi-chromatographic run / Multi-excitation**Flow rate: 2 mL min<sup>-1</sup>

- For each sample:
  - 10 Chromatographic runs recording time-emission matrix at fixed excitation wavelength (different excitation wavelength for each run) = 40 min

**Methodology 3**  
**On-line EEM detection**Flow rate: 0.5 mL min<sup>-1</sup>

- For each sample:
  - 1 Chromatographic run recording 25 sequential EEMs = 7 min

Analysis time per sample

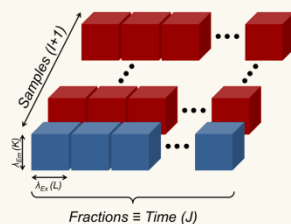
(HPLC + FD) = 45 min

Matrix size

(J×K×L): 25×17×25

Data pre-processing:

EEM\* smoothing

Data processing:PARAFAC, APARAFAC, MCR-ALS,  
U-PLS/RTLAnalysis time per sample

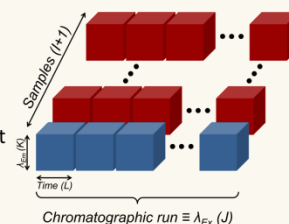
40 min

Matrix size

(J×K×L): 10×121×25

Data pre-processing:

TEM smoothing, Peak alignment

Data processing:PARAFAC, APARAFAC, MCR-ALS,  
U-PLS/RTLAnalysis time per sample

(HPLC + FD) = 7 min

Matrix size

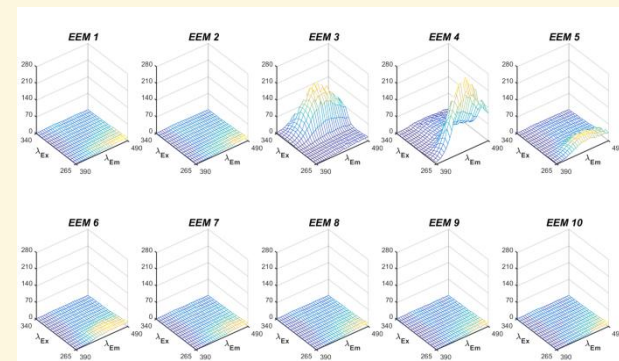
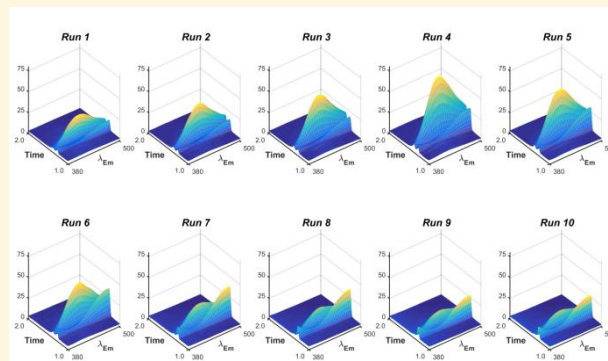
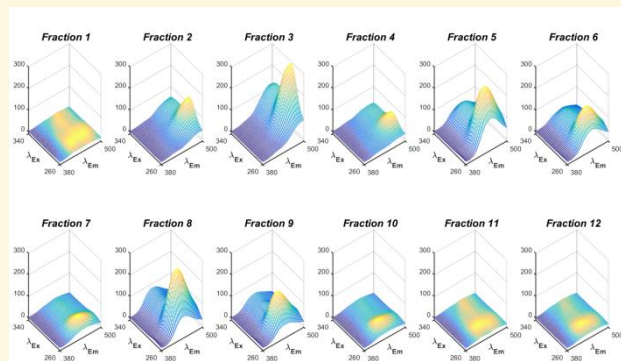
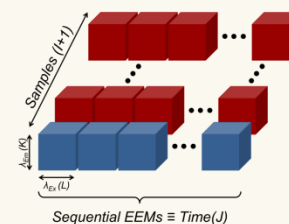
(J×K×L): 15×28×15

Data pre-processing:

TEM smoothing

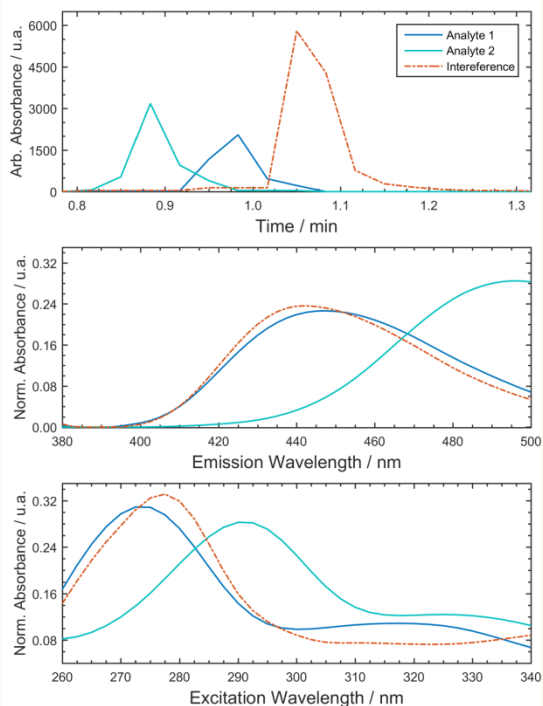
Data processing:

Work in Process



## Third-order data modelling – HPLC-EEM data analysis

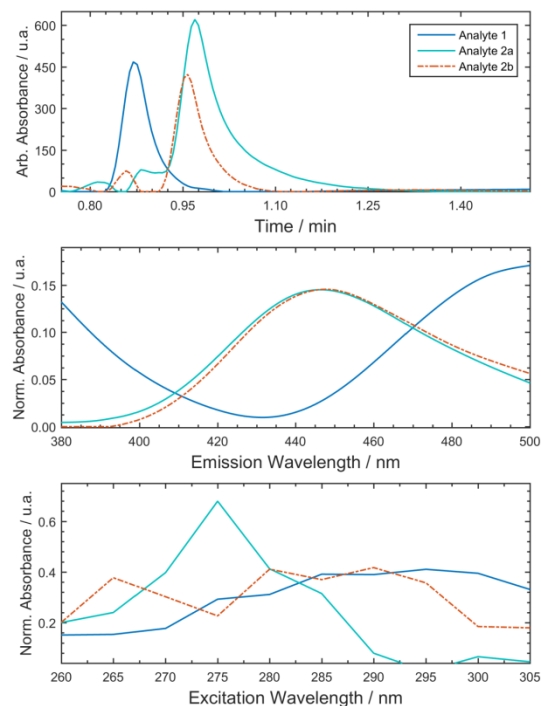
## Methodology 1

3<sup>rd</sup> order data (TEEC\*)

Trilinear data

- General features**
- Minimal data pre-processing
  - Simple data processing
  - Semi-automated data generation
  - Three instruments are required
  - Time consuming
- 4-way data array (Conc-TEEC)**
- Non-quadrilinear
  - Breaking mode=Elution time (Peak shifting between samples)

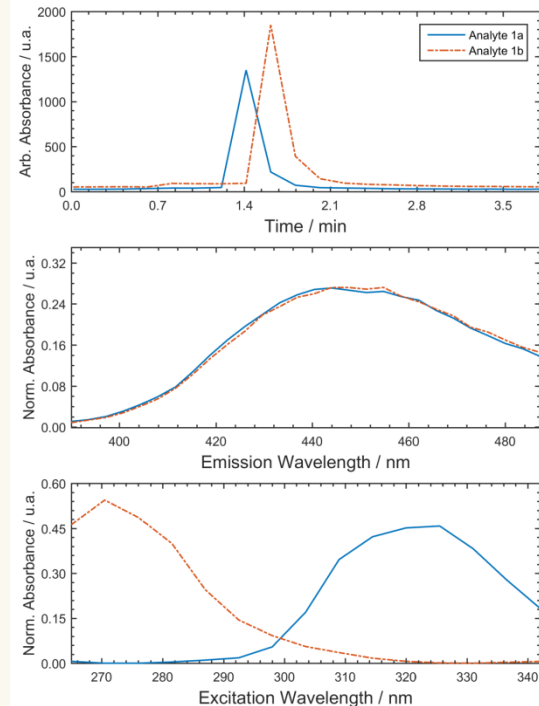
## Methodology 2

3<sup>rd</sup> order data (TEEC\*)

Non-trilinear data

- General features**
- Complex data pre-processing
  - Simple data processing
  - Automated data generation
  - One instrument is required
  - Time consuming
- 3<sup>rd</sup> order data (TEEC\*)**
- Breaking mode=Elution time (Peak shifting within sample, leading to a trilinear break in excitation mode)
  - High selectivity in two modes
  - Low selectivity in one mode

## Methodology 3

General features

- Minimal data pre-processing
- Very complex data processing
- Automated data generation
- Two instruments are required
- Non-time consuming

3<sup>rd</sup> order data (TEEC\*)

Non-trilinear data

- Breaking mode=Excitation (Excitation is strongly affected by concentration/elution time)
- High selectivity in the three modes