Organic and Biological Analytical Chemistry Group

The Century Mix: updates and cool projects

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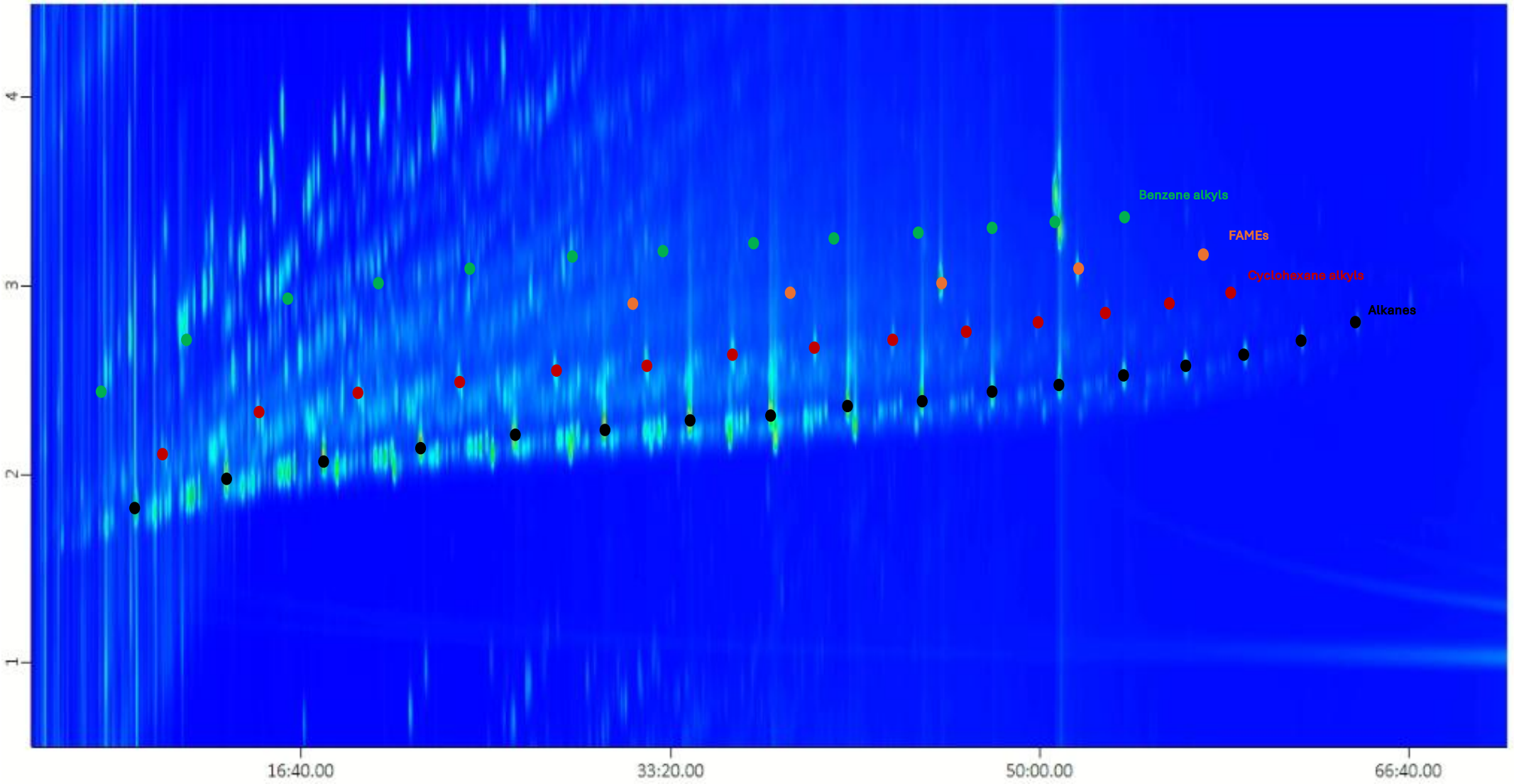
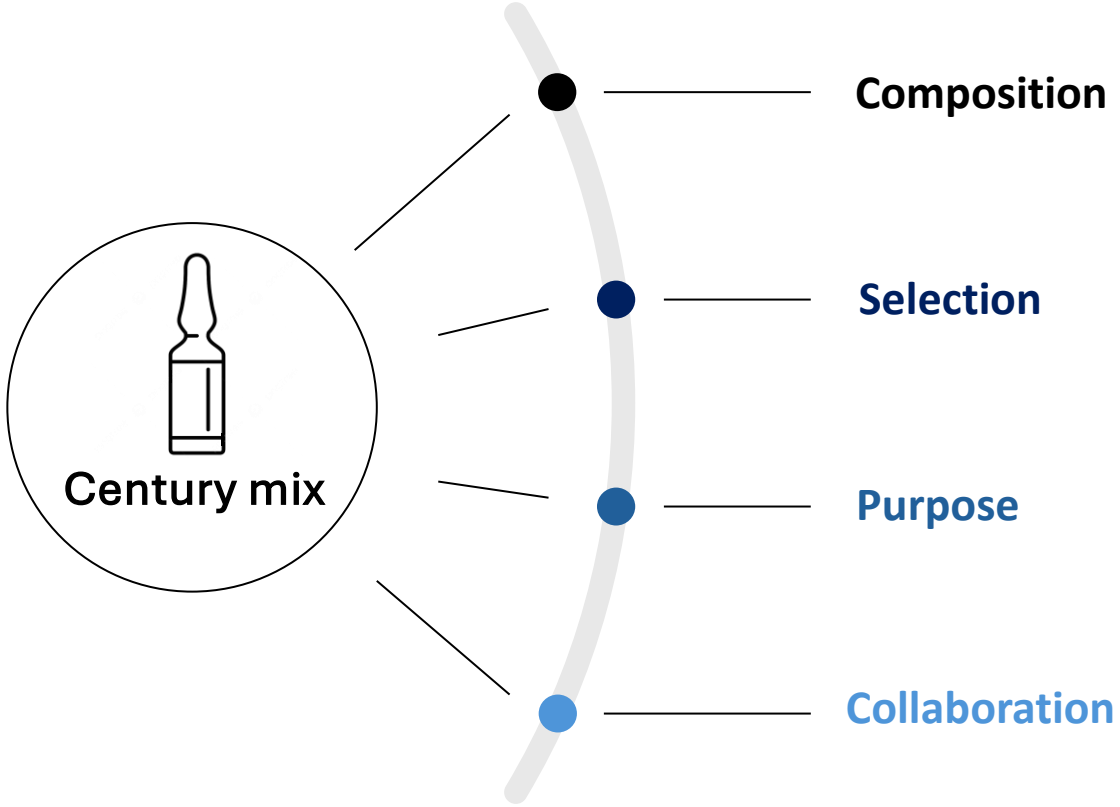
[2]Georgia Institute of Technology, GW Woodruff School of Mechanical Engineering, Atlanta, USA

Many friends from MDCW

Biodiesel structured separation



Century mix



Composition

- **100 compounds :** Alkanes, Alcohols, Ketones, Esters, Lactones, Phthalates, ...

Selection

- Key characteristic: **Stability**
- Wide range of chemical functions to be **representatives** of various samples

Purpose

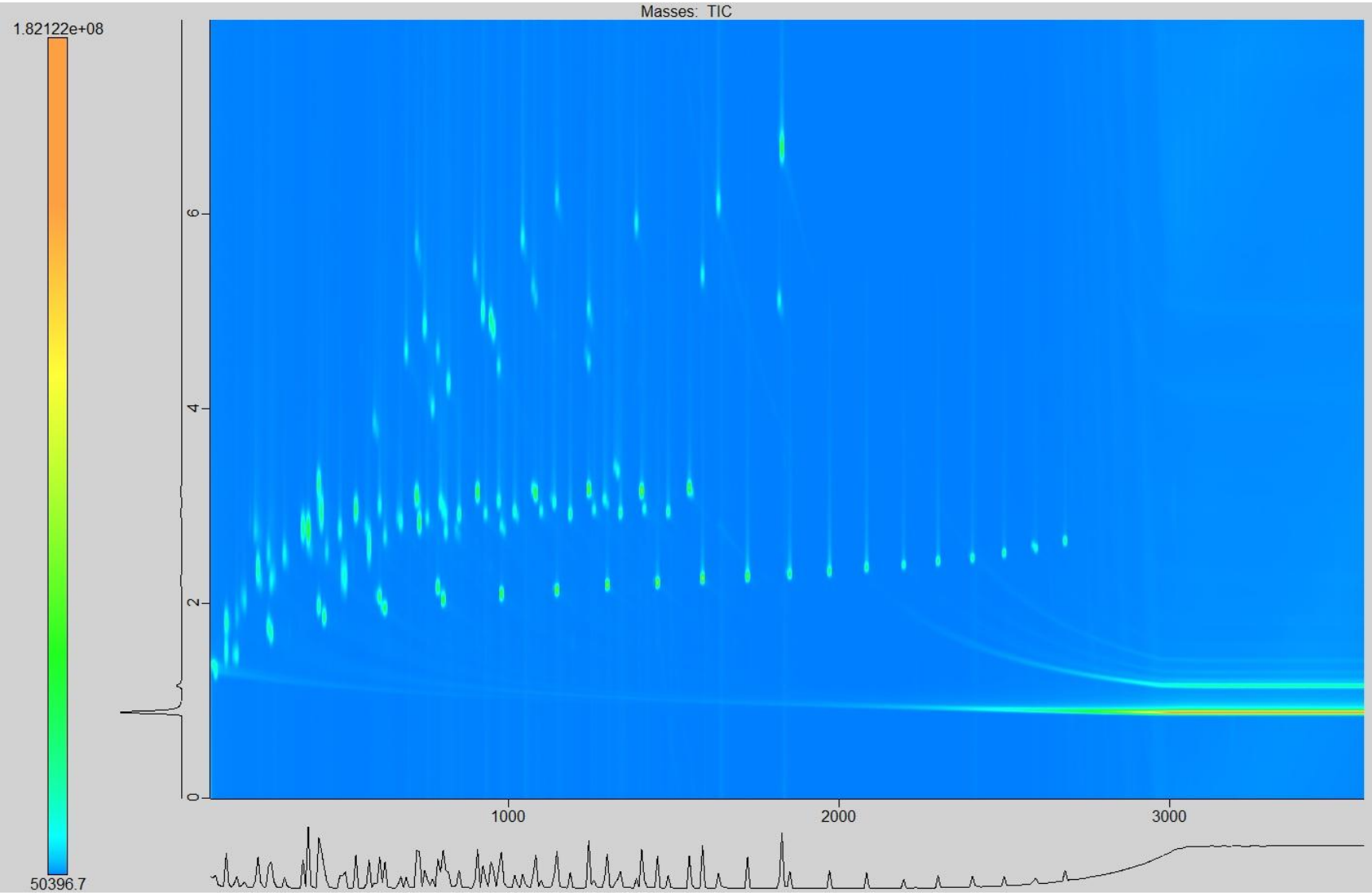
- Column **characterisation**
- Quality control

Collaboration

- Developed with cross-laboratory **input**

Dimandja, J.-M. *et al. LCGC Int.* **2024**, *1*, 12–20.

Century Mix Run



The Task!

2 Runs (at “standard” conditions)

2 Months to provide the following data:

- Stationary phase combination information
- Operating conditions
- Chromatograms
- Peak tables

Factors affecting retention indices

Inter-laboratory study

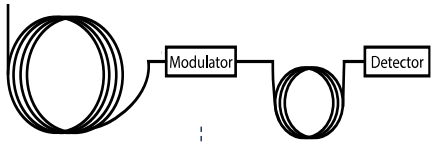
- Standardized conditions : Temperature setting, flow rate, carrier gas, acquisition rate

Cryogenic modulator

Peltier modulator

Flow modulator

11 GC×GC systems



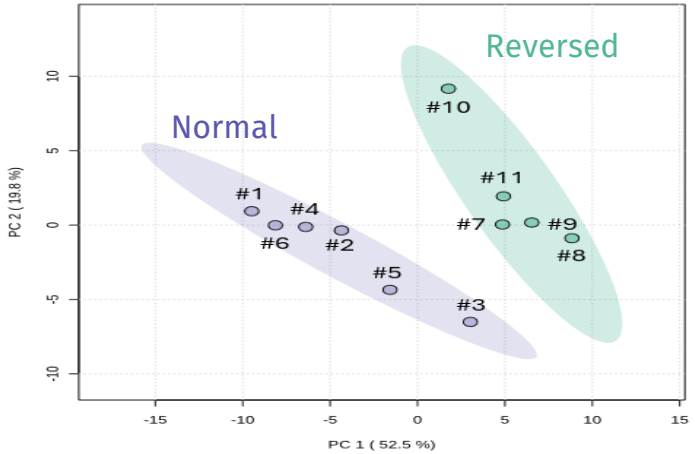
Normal orthogonality

Hybrid orthogonality

Reversed orthogonality

Quadrupole MS, TOF MS, and FID

Principal component analysis



Two clusters according Normal and Reversed conditions

^{1}D Stationary phase impact on retention indices

CM & compounds annotation

Compounds identification
Non-Targeted analysis

Good separation
GC×GC
Deconvolution (HRMS)

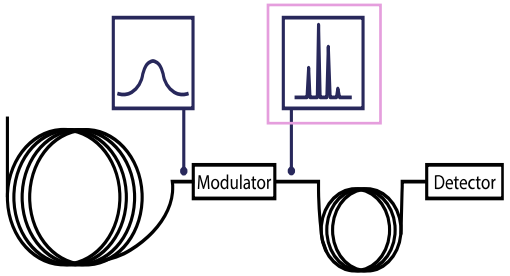
Mass spectra
300,000 EI spectra of unique compounds

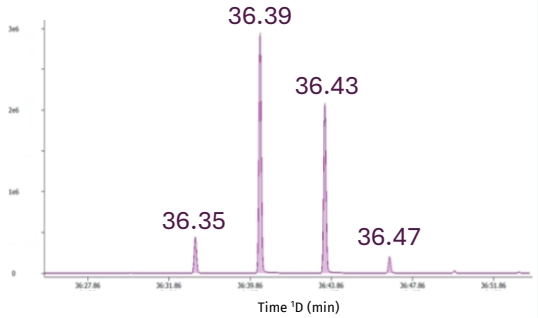
Retention indices
500,000 Retention indices values

Need of predicted data for the **unknown-unknowns**

Retention indices

RI from ^{1}D in GC×GC ⇄ Traditional 1D GC ?





Assess the factors that **affect retention indices** in GC × GC systems

Factors affecting retention indices

Design of experiment (DOE)

- Full factorial design (5×3×3×3)
- All possible combination (140 combinations)
- Reversed orthogonality : Mid-polar × Non-polar (BPX 50 × RXI 5 MS)

Ramp rate

3– 25 °C/min

Flow rate

0.6 – 1.6 mL/min

Modulation time

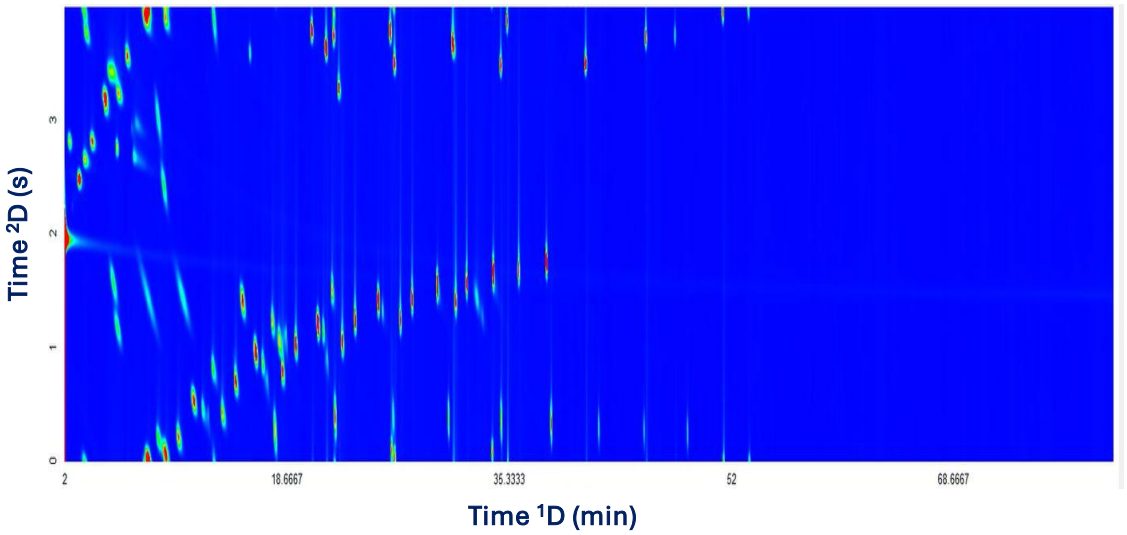
2 - 6 s

Offset oven

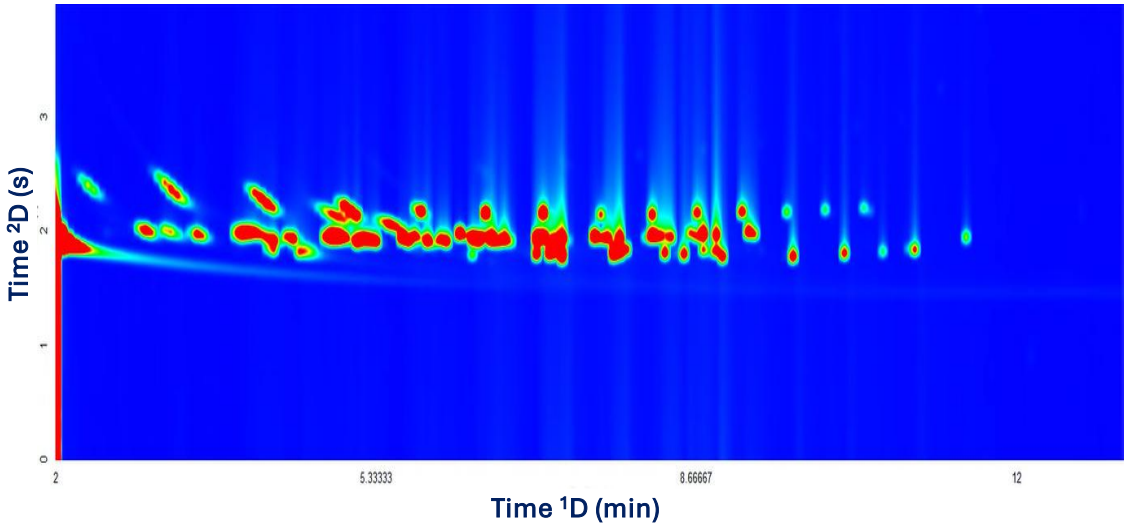
5 – 25 °C

Factors affecting retention indices

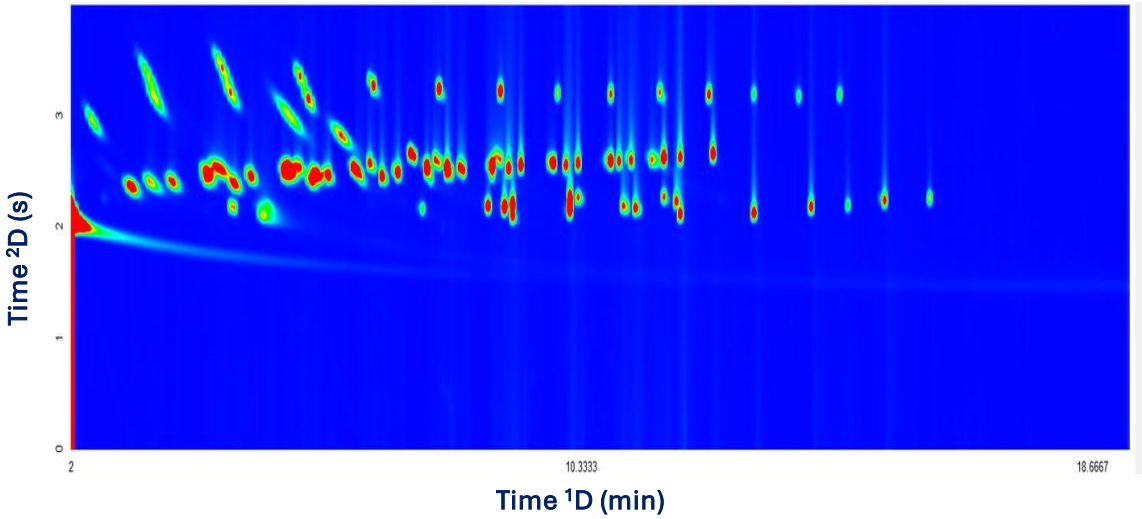
(2221) 1 mL/min - PM 4 s - Offset 15°C - **3°C/min**



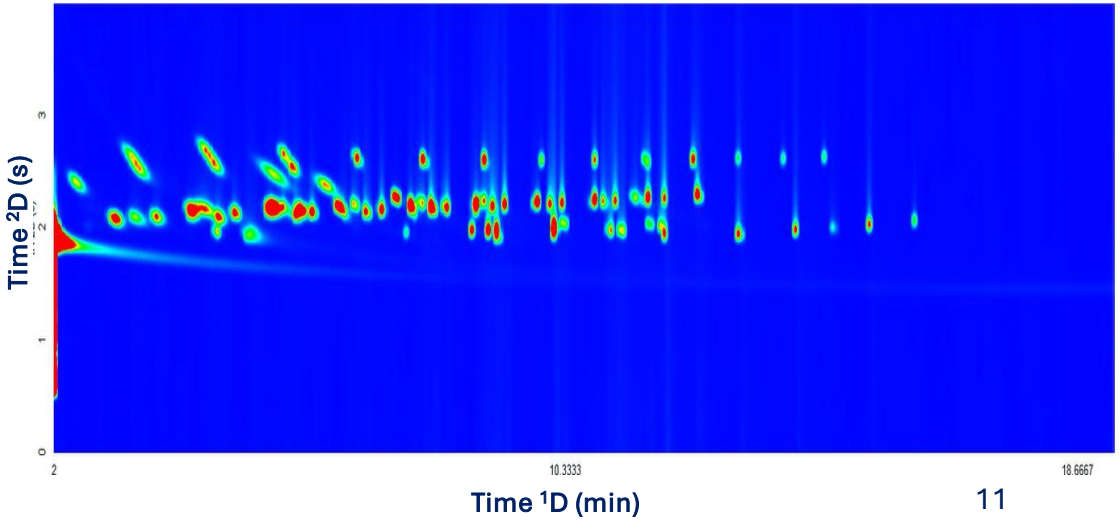
(2225) 1 mL/min - PM 4 s - Offset 15 - **25°C/min**



(2214) 1 mL/min - PM 4 s - **Offset 5°C** - 15°C/min



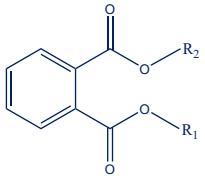
(2234) 1 mL/min - PM 4 s - **Offset 25°C** - 15°C/min



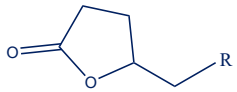
Model fit : Factors affecting retention indices

✓ 33 Compounds fitting the model

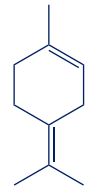
RI variation **linked** to the parameters tested



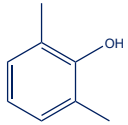
Phtalates



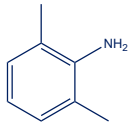
Lactones



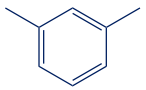
Terpenes



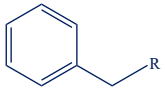
2,6-Xylenol



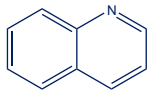
2,6-Xylidine



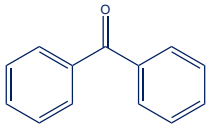
m-cresol



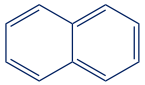
Alkyl Benzene



Quinoline



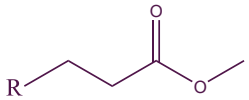
Benzophenone



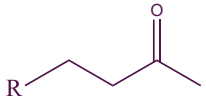
Naphtalene

✗ 43 Compounds not fitting the model

RI variation **not linked** to the parameters tested



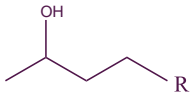
Esters



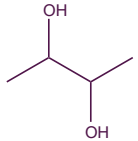
Ketones

Pyridine

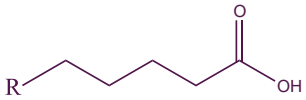
Primary alcohol



Secondary alcohol

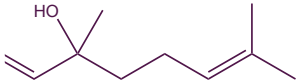


2,3- Butandiol



Carboxylic acids

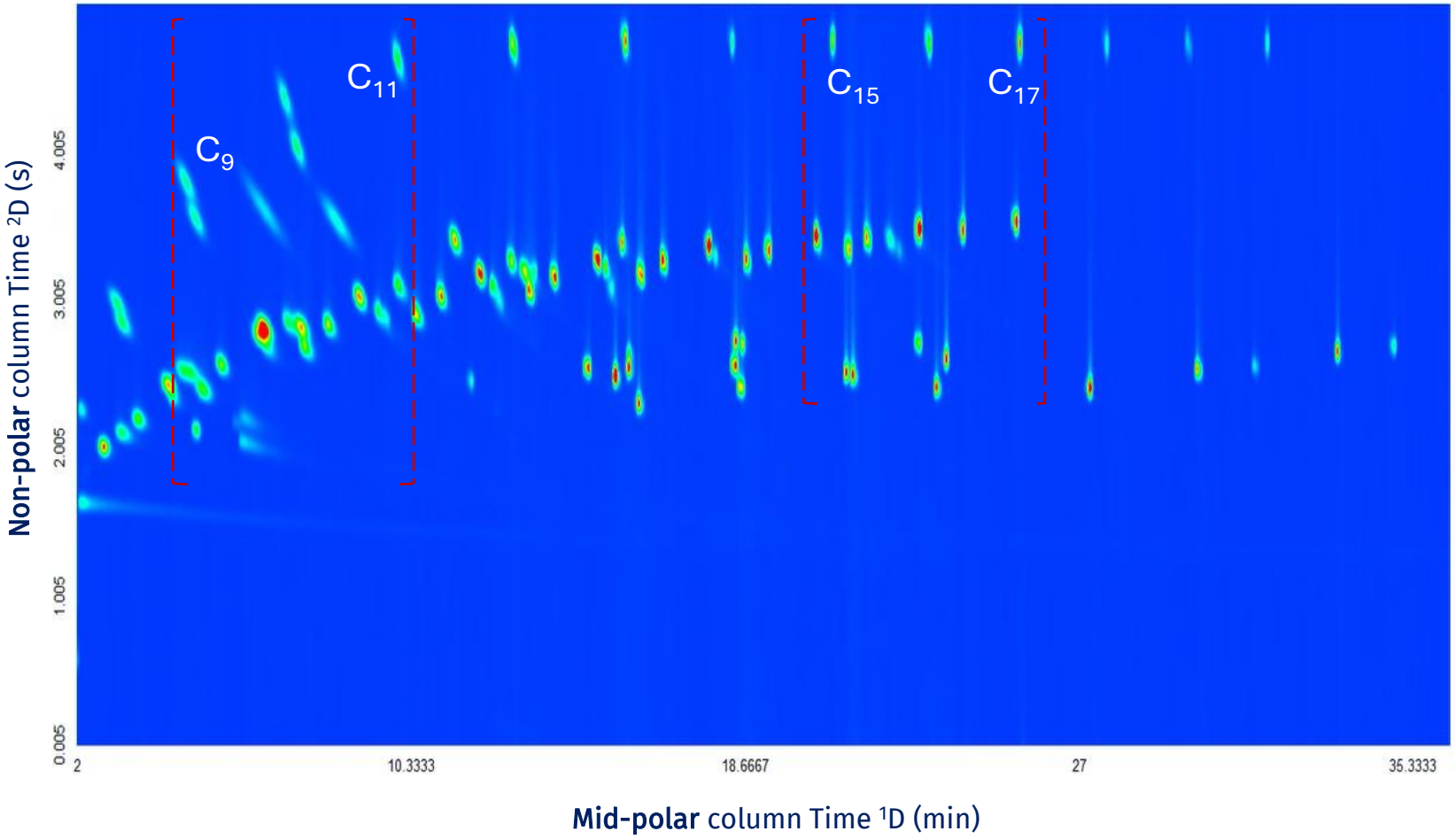
Benzene



Linalool

Model fit : Factors affecting retention indices

Is it related to compounds volatility ?



Nonane C_9 – Undecane C_{11}

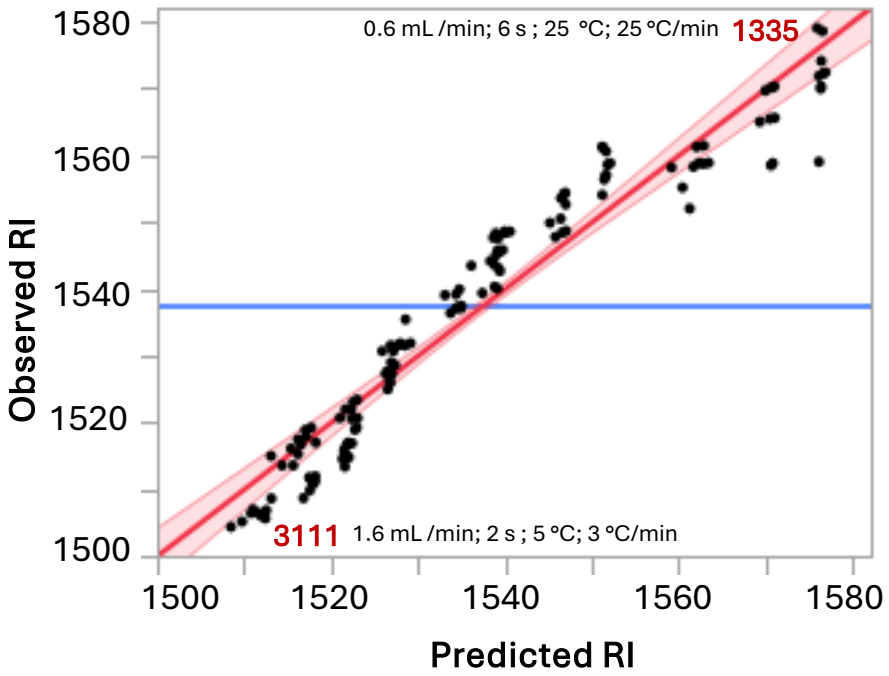
Compounds	Rsquare (R^2)	
Pentanoic acid, methyl ester	0.51	✗
2-Heptanone	0.20	
Benzene propyl	0.74	✓

Pentadecane C_{15} - Heptadecane C_{17}

Compounds	Rsquare (R^2)	
Undecanoic acid, methyl ester	0.16	✗
2-Dodecanone	0.34	
Benzene Nonyl	0.90	✓

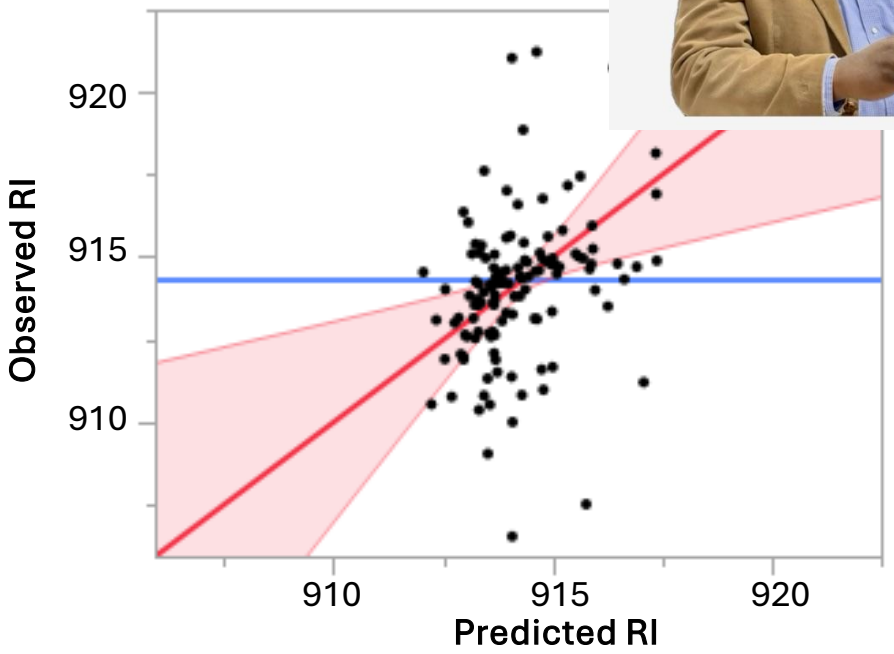
Model fit : Factors affecting retention indices

✓ Quinoline



- **Rsquare (R^2)** : 0.93
- **93%** of response variability **explained** by the model
- ΔRI ≈ 75

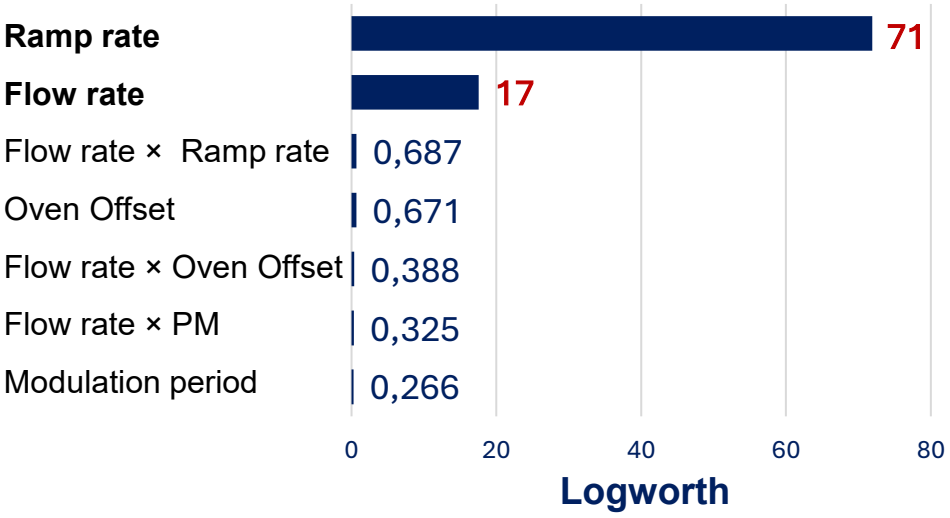
✗ 2- Hexanc



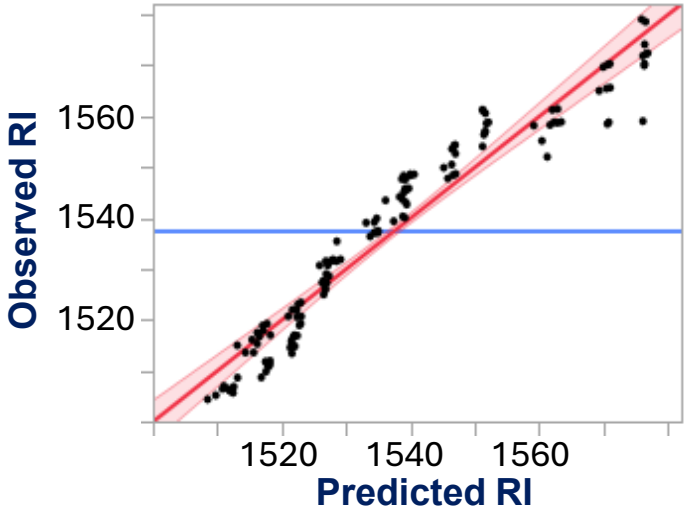
- **Rsquare (R^2)** : 0.24
- Model doesn't explain the variability
- ΔRI ≈ 20

Factors affecting retention indices

Summary of effect

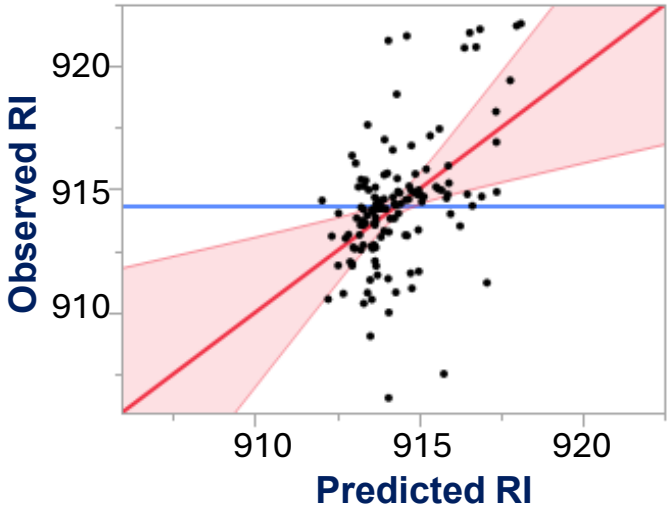


Quinoline



- **Rsquare (R^2)** : 0.93
- **93%** of the variability **explained** by the model
- ΔRI ≈ 75

2- Hexanone



- **Rsquare (R^2)** : 0.24
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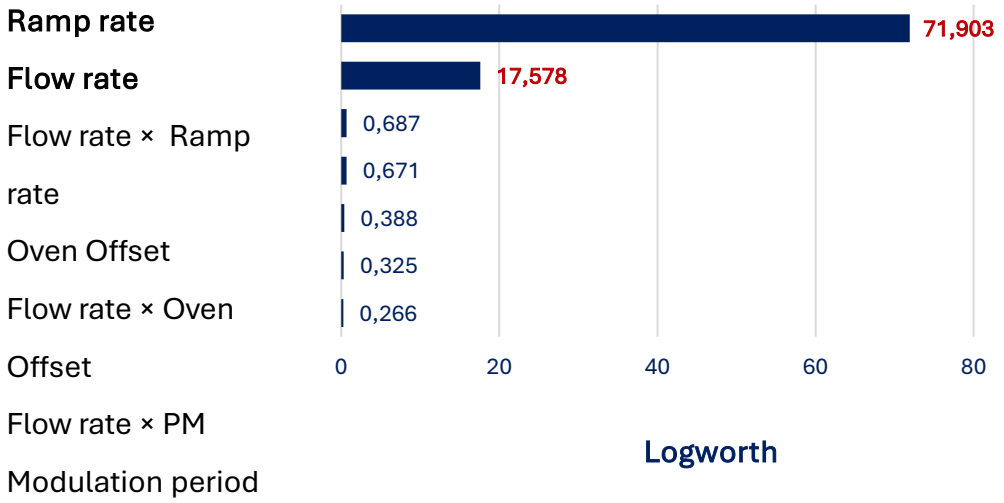
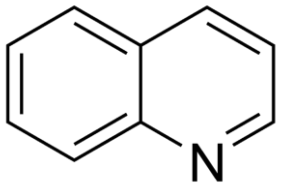
Factors affecting retention indices

Summary of effect





Quinoline

Ramp rate effect on Lactones and phtalates

Logworth

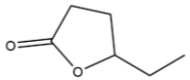
57.26 Dimethyl Phthalate

51.75 Diethyl Phthalate

33.95 Dipropyl Phthalte

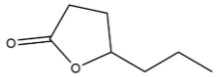
Logworth

49.34



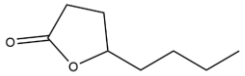
γ-Hexalactone

48.75

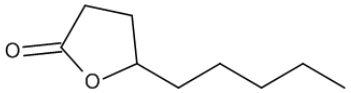


γ-Heptalactone

43.75



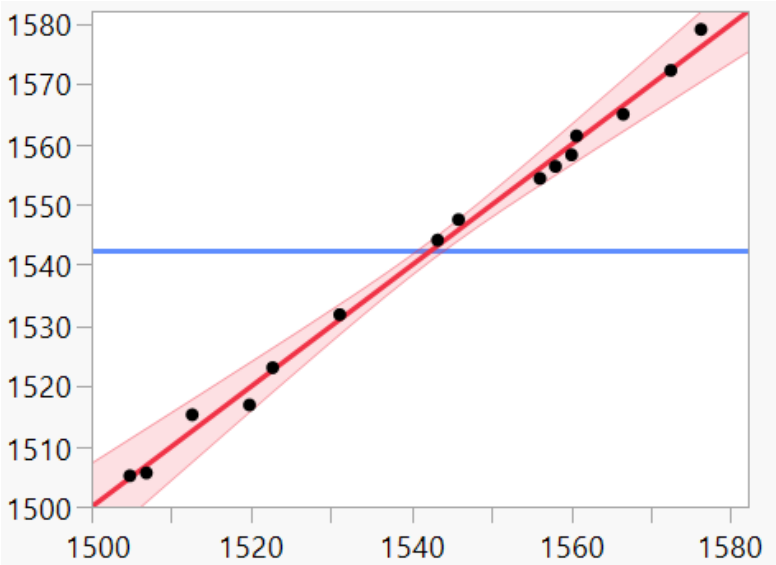
γ-Octalactone



53.62 γ-Nonatalactone

Factors affecting retention indices

Quinoline – Reversed column set

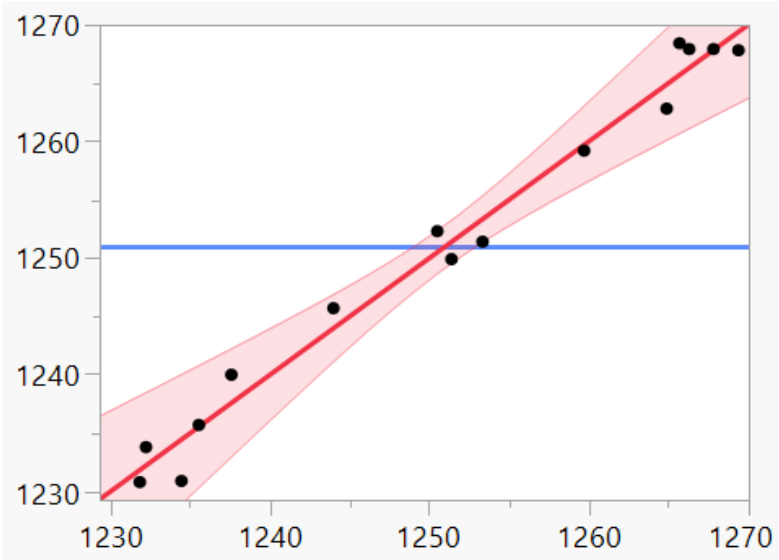


- Rsquare (R^2) : 0.99
- ΔRI ≈ 75

Effect summary

Source	Logworth	PValue
Rampe rate(3,25)	7.151	0.00000
Flow rate(0.6,1.6)	3.923	0.00012
Rampe rate*Rampe rate	3.084	0.00082
Modulation(2,6)	0.784	0.16440
Offset Oven(5,20)	0.563	0.27371
Offset Oven*Offset Oven	0.133	0.73680
Flow rate*Flow rate	0.113	0.77064
Modulation*Modulation	0.010	0.97742

Quinoline – Normal column set



- Rsquare (R^2) : 0.98
- ΔRI ≈ 50

Effect summary

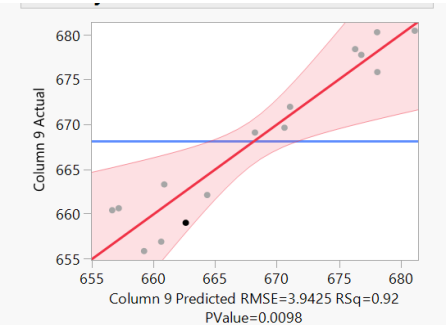
Source	Logworth	PValue
Rampe rate(3,25)	5.570	0.00000
Flow rate(0.6,1.6)	1.327	0.04704
Rampe rate*Rampe rate	0.847	0.14233
Flow rate*Flow rate	0.626	0.23638
Modulation*Modulation	0.532	0.29402
Modulation(2,6)	0.270	0.53678 ^
Offset Oven(5,20)	0.175	0.66855
Offset Oven*Offset Oven	0.138	0.72830

RXI-5MS × RXI-17SiMs

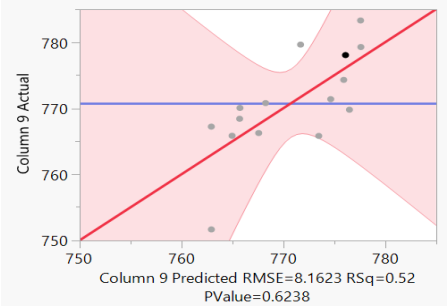
BPX50 × RTX-5MS

Stabilwax × RXI-17 SiMs

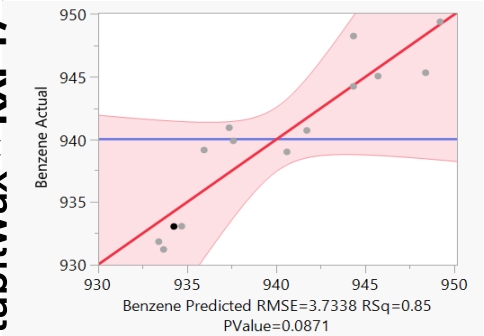
Benzene ΔRI ≈ 25



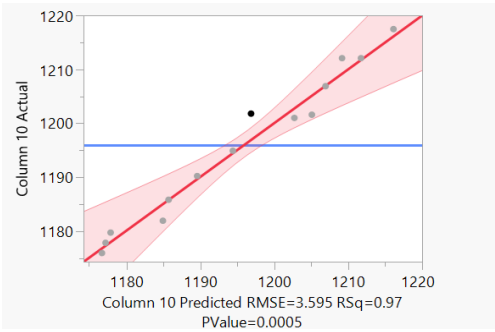
- ΔRI ≈ 30



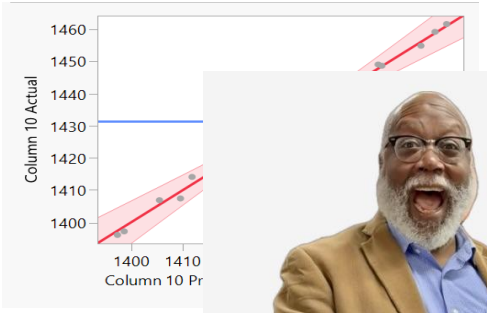
- ΔRI ≈ 18

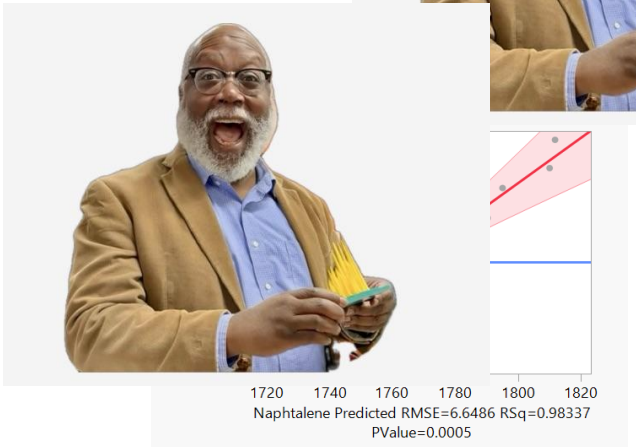


Naphtalene ΔRI ≈ 40

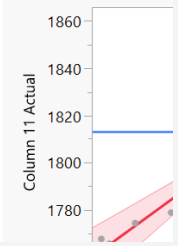


- ΔRI ≈ 60

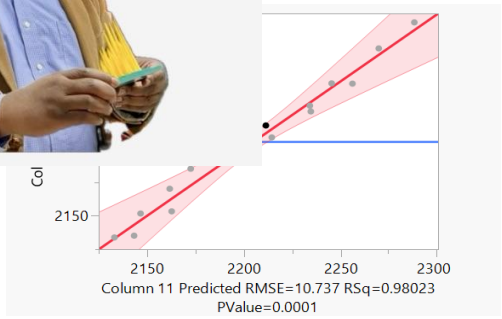




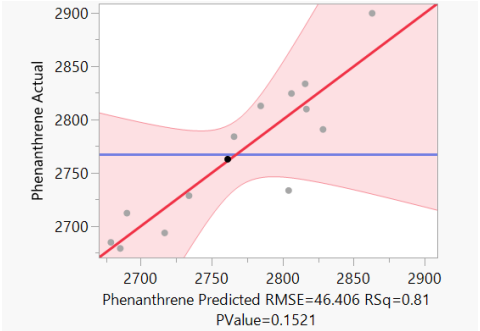
Phenan



l ≈ 150



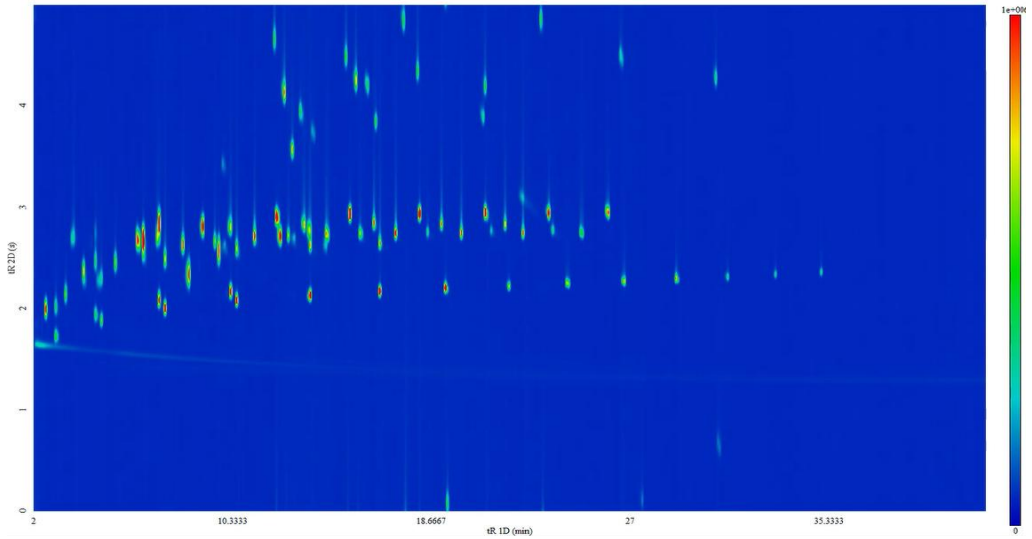
- ΔRI ≈ 200



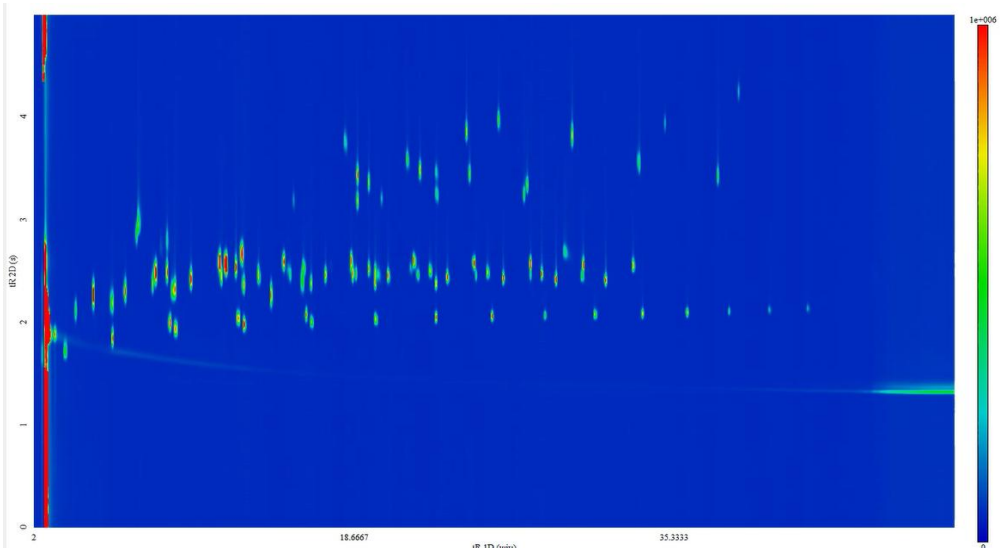
Factors affecting retention indices

Orthogonality	Phases	Length (m)	Int.Diameter (mm)	Film thikness (µm)
Normal	**RXI-5MS** × **RXI-17SilMs**	30 × 2	0.25× **0.25**	0.25× **0.25**
Normal	**RXI-5MS** × **vf 17 Ms**	30 × 2	0.25× **0.25**	1× **0.5**
Reversed	**BPX50** × RTX-5MS	30 × 2	0.25× **0.25**	0.25× **0.25**
Reversed	Stabilwax × **RXI-17 SilMs**	30 × 2	0.25× **0.25**	0.25× **0.25**
Normal	**RXI-624 SilMS** × Stabilwax	30 × 2	0.25× **0.25**	1.4× **0.5**

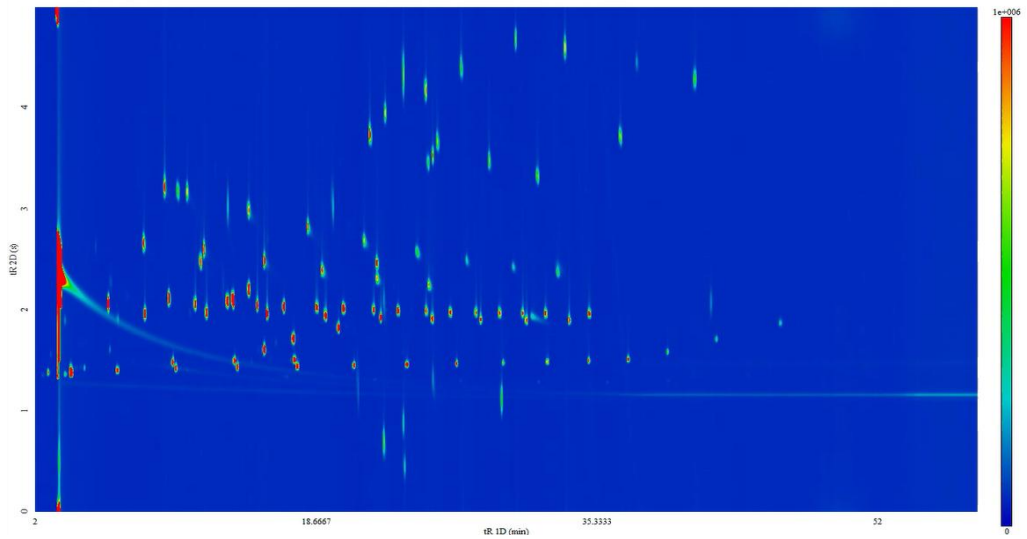
RXI-5MS × RXI-17SilMs



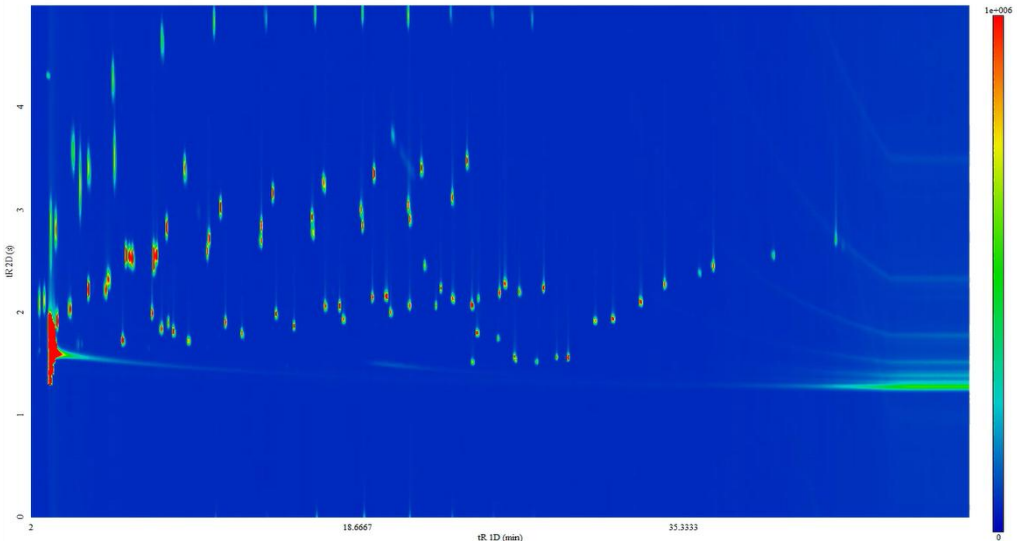
RXI-5MS × VF 17 Ms



RXI-624 SilMS × Stabilwax



Stabilwax × RXI-17 SilMs



Where to go next?

Thank you for your attention !

OBiACHEM group

John Dimandja, GTRI & collaborators

Evolution of gas chromatography

1906 Mikhaïl Tswett
Pioneer of Chromatography

1952 A.J Martin & R.L Synge
Fundamental paper introducing Gas Chromatography

1958 M.J.E. Golay
Open tubular capillary column

Ease hyphenation with mass spectrometry

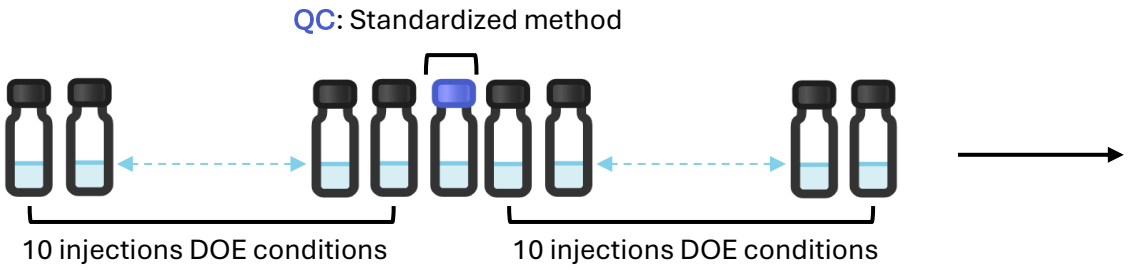
1991 J.B Phillips & Z.Liu
First demonstration of GC×GC

2000s Commercialization of robust GC×GC systems

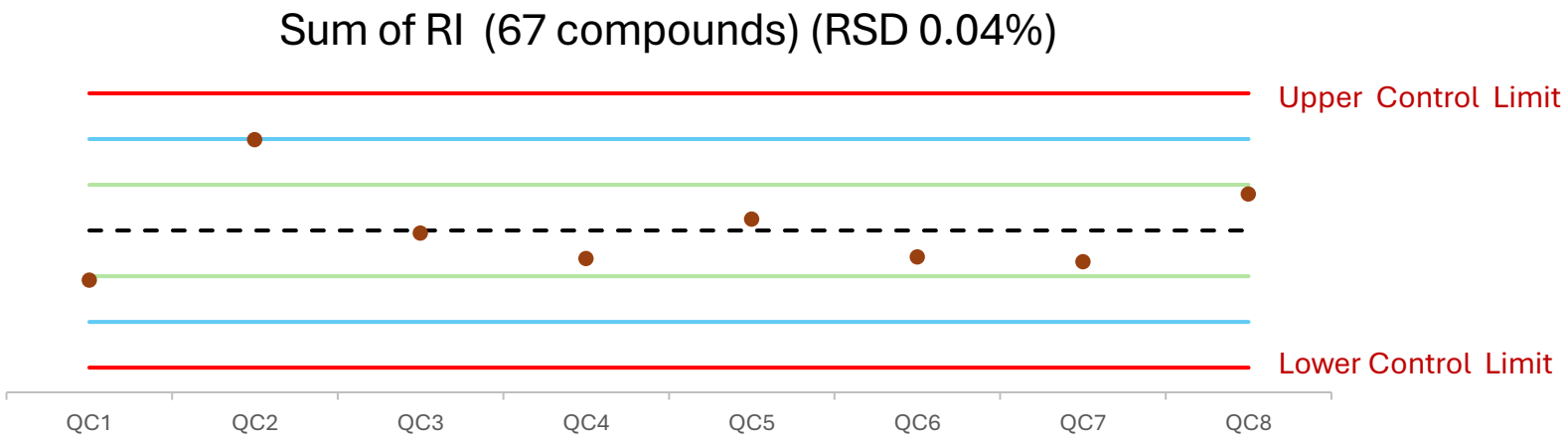
2010s Growth in automated data analysis tools

2020s Integration of machine learning tools

Quality control (CM)



- Monitor the system's stability
- Effects of experimental parameters / potential instrument drift



Essential Insights

Influence

Ramp rate & flow rate are critical parameters

Different impact

Compound-specific effects

Understand

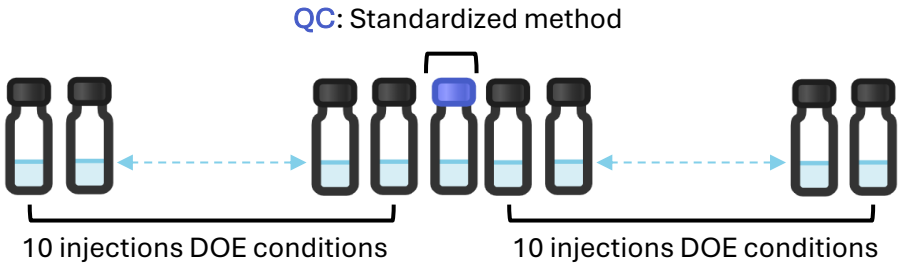
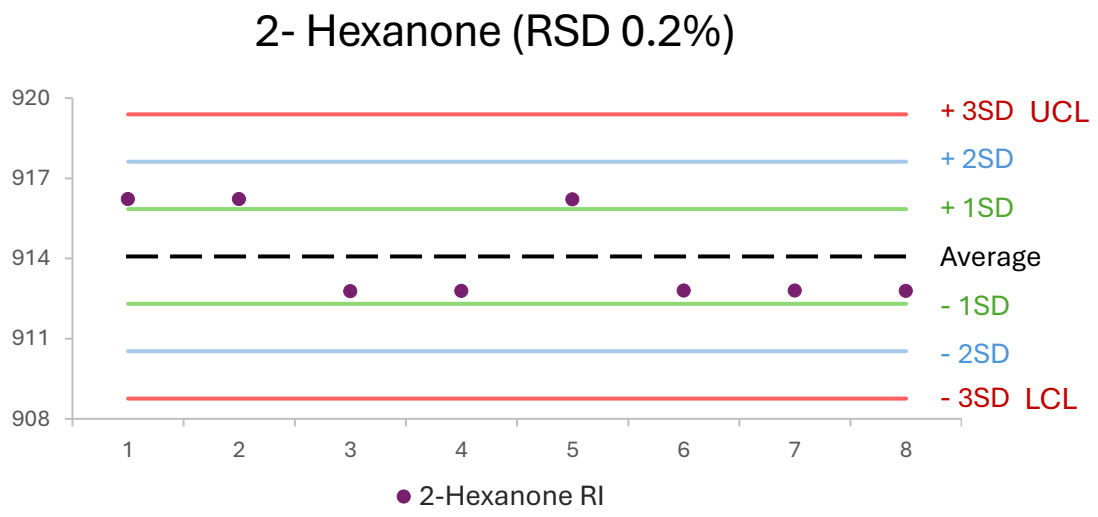
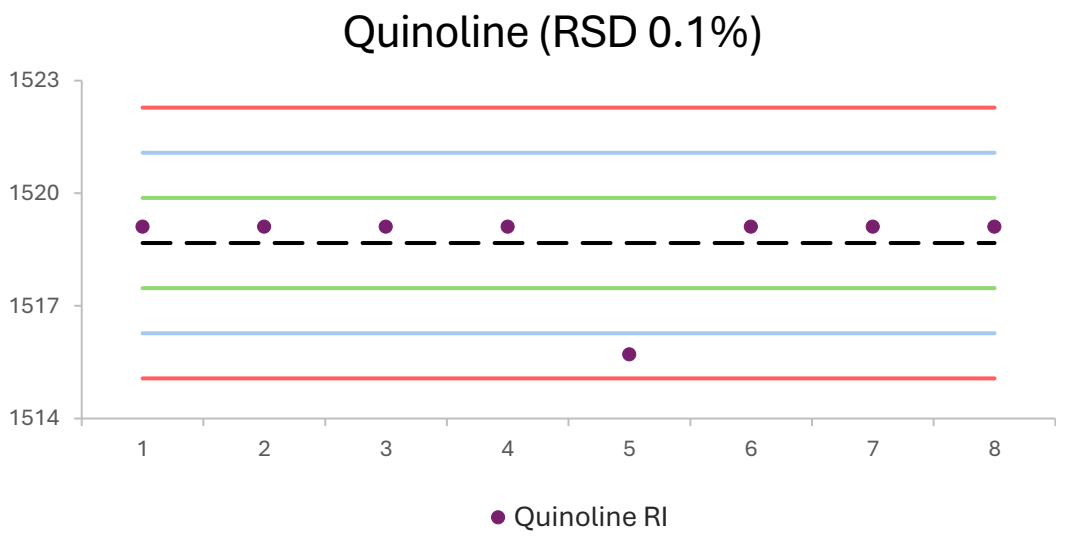
Fundamental chemistry in the GC×GC separation

Test DOE on normal orthogonality

Reliable Prediction

Method development & data processing

Quality control (CM)



- Monitor the system's stability
- Effects of experimental parameters / potential instrument drift