

Advanced characterization and extraction of complex bio-based matrices using GC×GC-TOFMS

Kinjal Bhatt*, Marco Beccaria, Yun Zou, Pierre-Hugues Stefanuto, and Jean- François Focant

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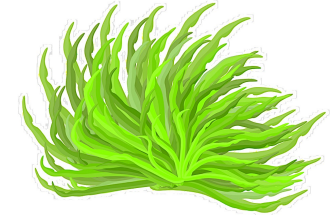
Feedstocks for biodiesel synthesis



Food crops

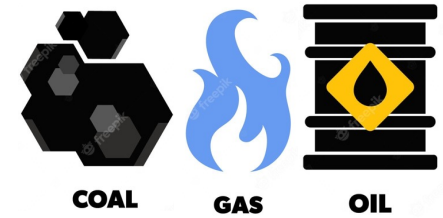


Agricultural waste



Algae or other aquatic plants

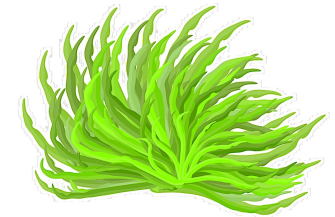
Feedstocks for biodiesel synthesis



Food crops



Agricultural waste

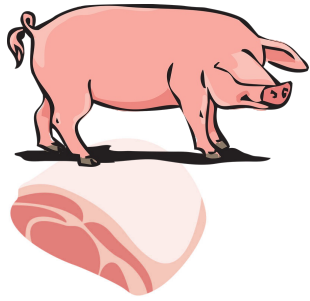


Algae or other aquatic plants

Constant change in impurities → depending on the type of feedstock used in the production process

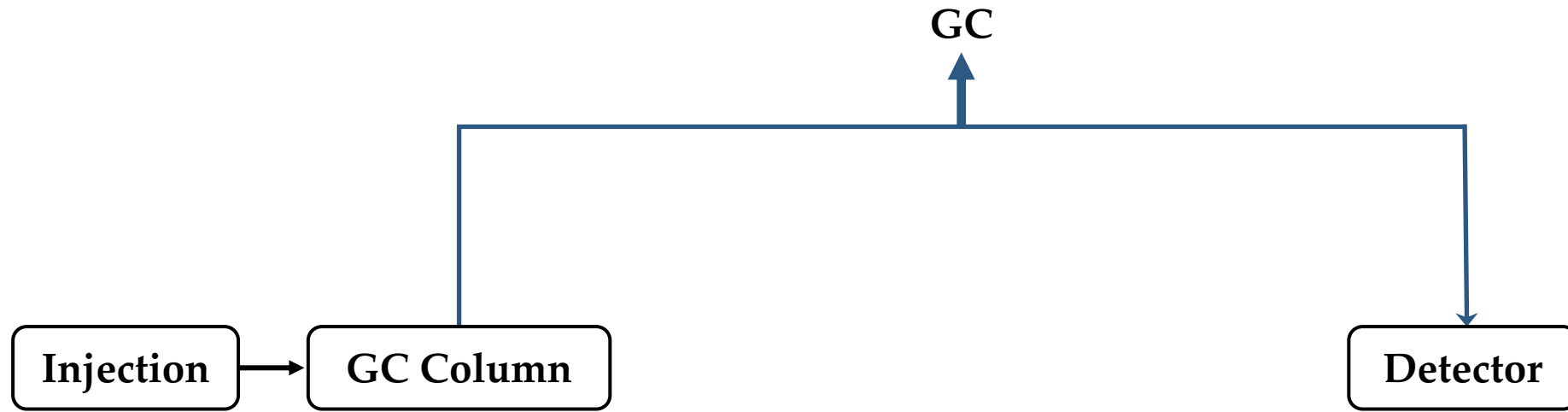
Demands More Advanced Characterization Techniques than Ever Before

Animal fat by-products

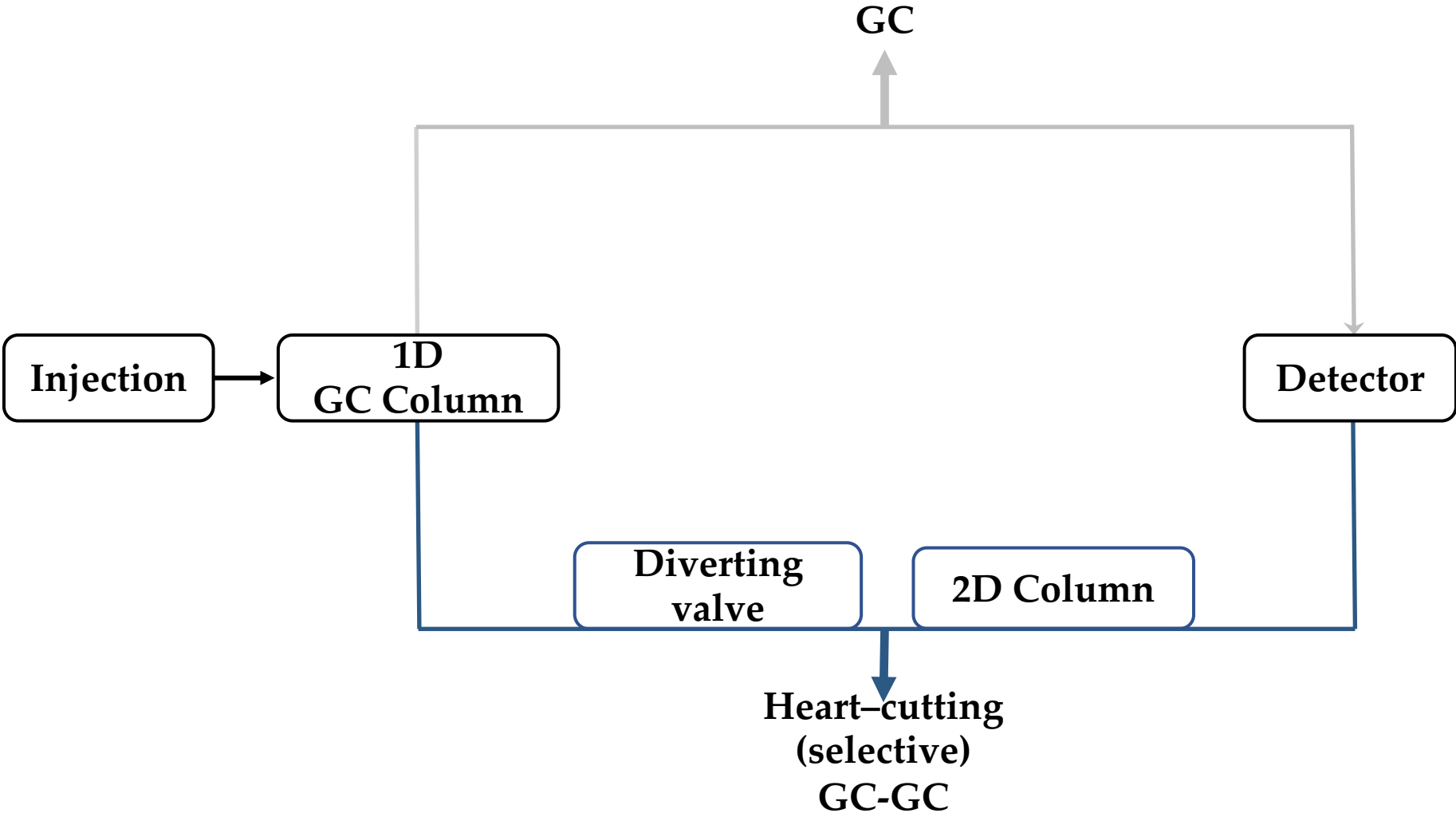


- In the EU, over **20 million tonnes** of animal fat by-products are produced every year
 - Compared to vegetable oil: low raw material cost and higher stability

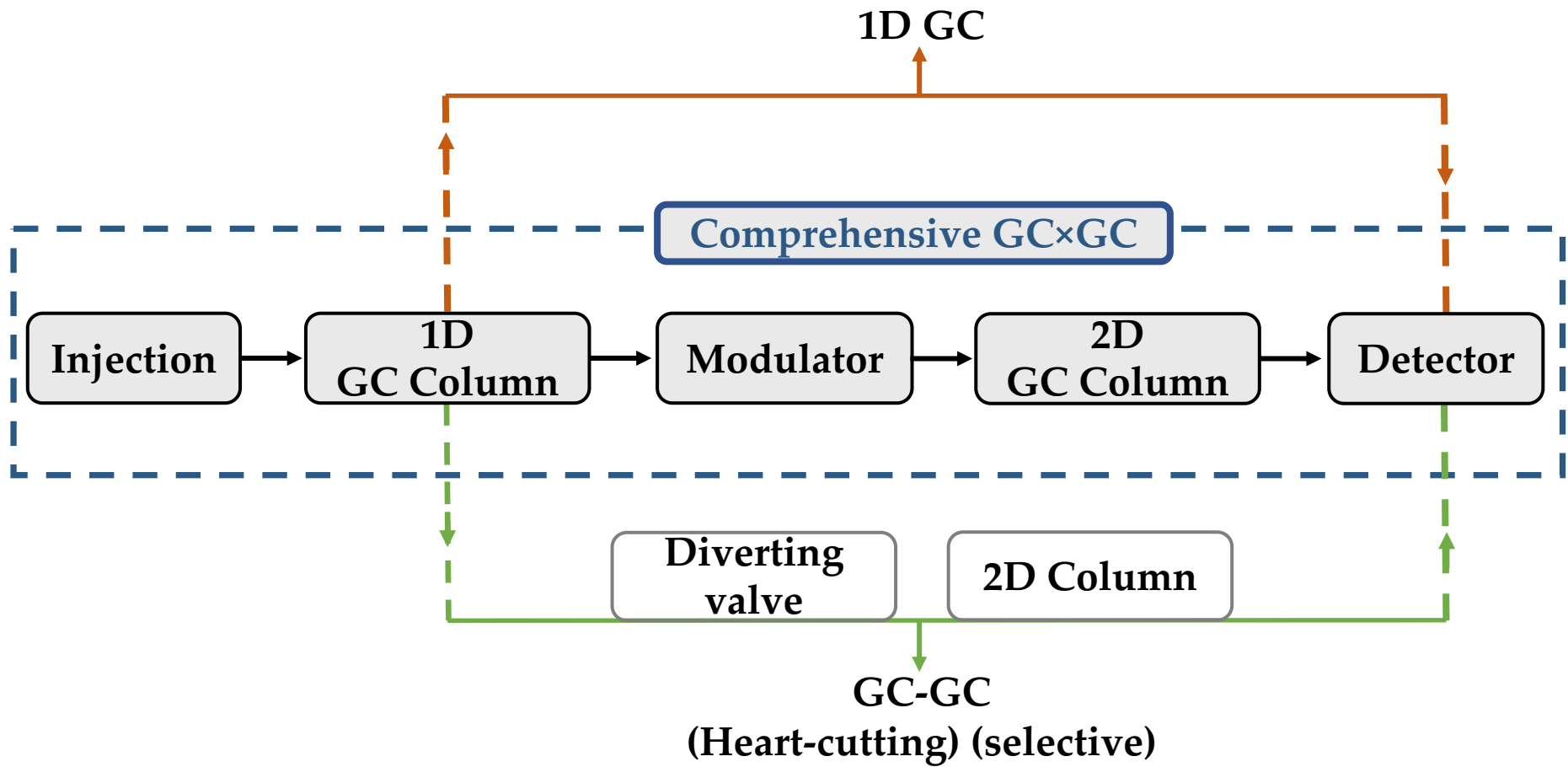
GC×GC-TOFMS



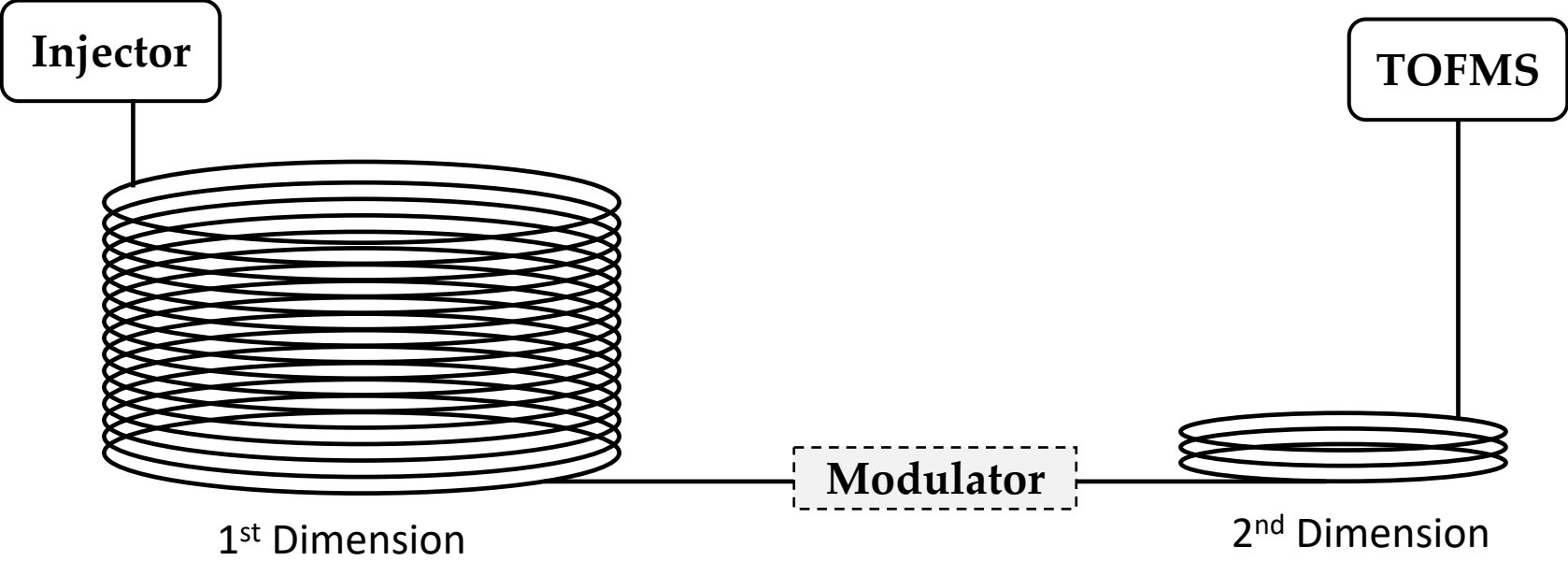
GC×GC-TOFMS Facts



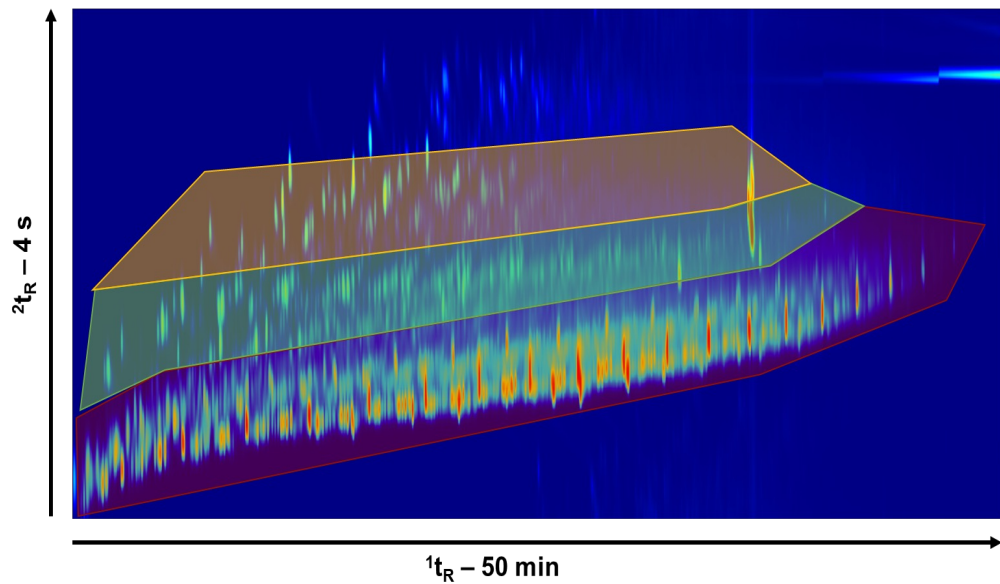
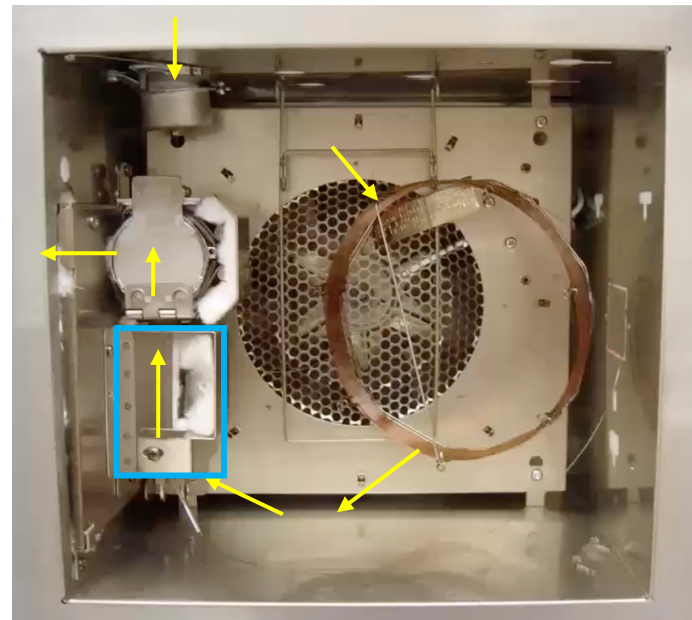
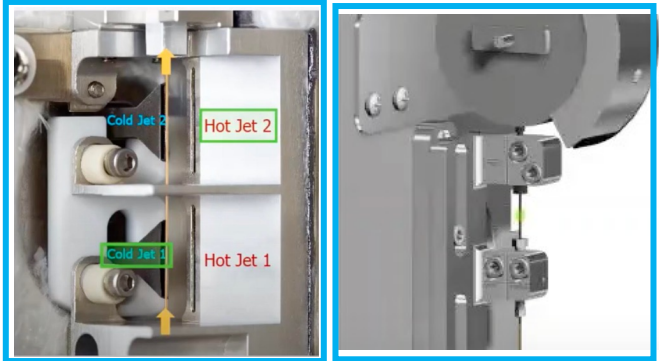
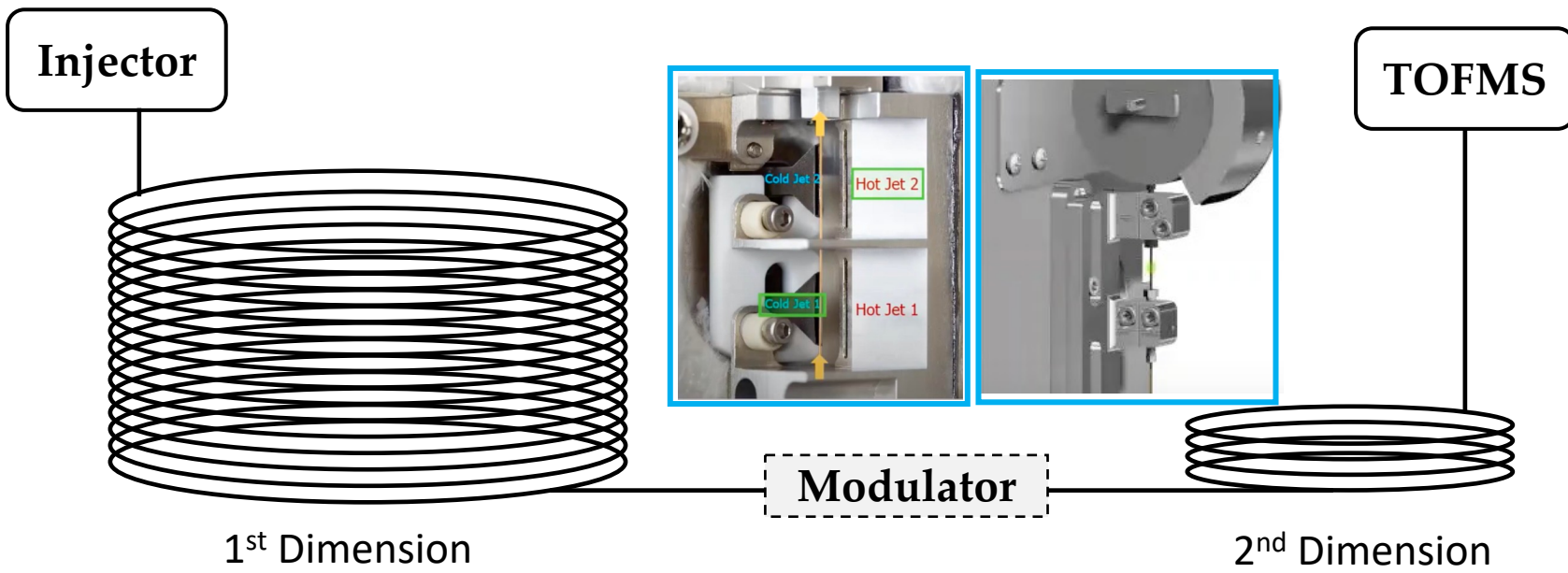
GC×GC-TOFMS Facts

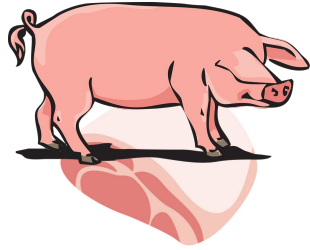


GC×GC-TOFMS Principle



GC×GC-TOFMS Principle



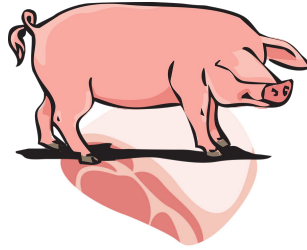


Pig Fat

**Major component
(95-99 %)**

Triglycerides

**Minor component
(1-5%)**



Pig Fat

**Major component
(95-99 %)**

Triglycerides

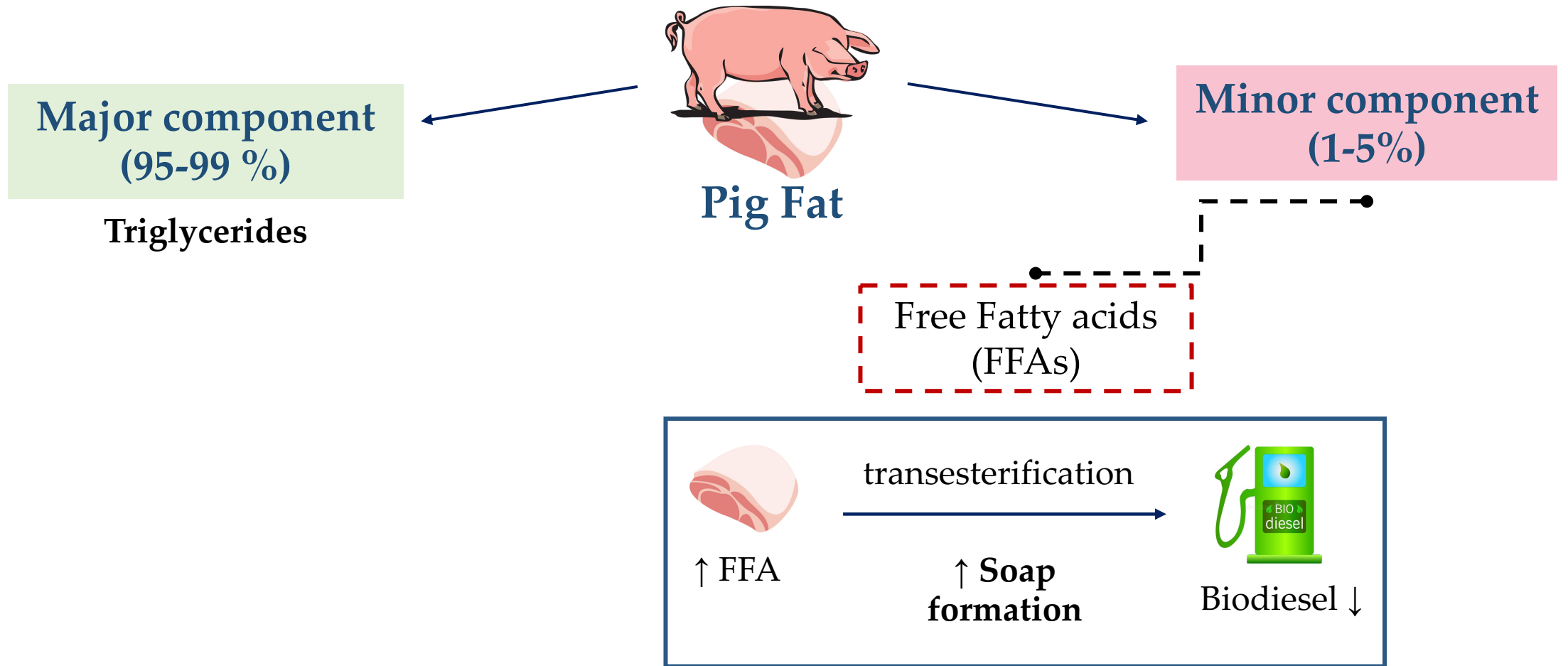
**Minor component
(1-5%)**

Triglycerides Derivatives

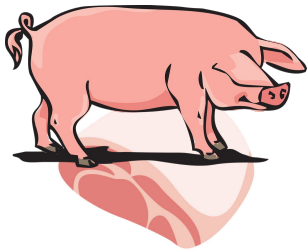
Glycerol
Free Fatty Acids
Mono- and Diglycerides

Non- Triglycerides
Derivatives

Phospholipids
Sterols
Pigments
Vitamin
Antioxidants
Alcohols
Waxes
Hydrocarbons



Quality of the animal fat used and the FFA content in it are important considerations for biodiesel production



Pig Fat

**Major component
(95-99 %)**

Triglycerides

**Minor component
(1-5%)**

Fatty acids
(FAs)

Saturated FAs
(SFAs)

Monounsaturated FAs
(MUFAs)

Polyunsaturated FAs
(PUFAs)

- more prone to oxidation
- can lead to instability in the final product

Analysis of Fatty acids in animal fat feedstock

GC×GC-EI-LR-TOFMS

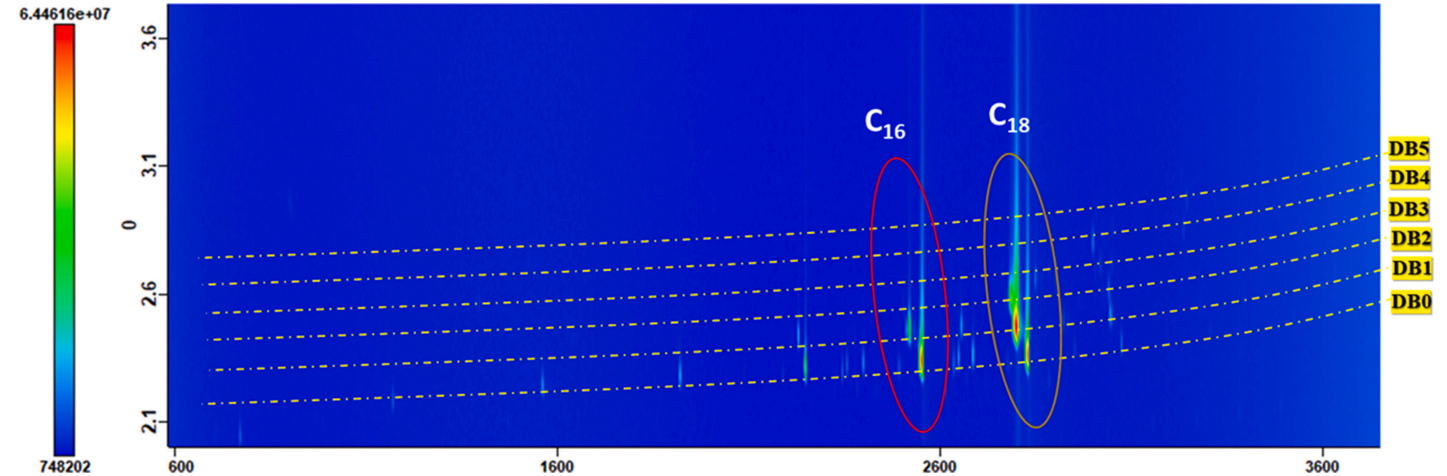
Sample preparation:

10mg of fat + n-hexane, $\text{CH}_3\text{ONa} + \text{BF}_3$

1.5 hrs
70°C

.....
N₂ drying (20-30 minutes)

↓
Analysis



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Deeper investigation of oxygen-containing compounds in oleaginous feedstock (animal fat) by preparative column chromatography and comprehensive two-dimensional gas chromatography coupled with high-resolution time-of-flight mass spectrometry

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^a University of Ferrara, Department of Chemical, Pharmaceutical, and Agricultural Sciences, via L. Borsari 46, Ferrara, Italy

^b Organic and Biological Analytical Chemistry Group, Madsy Research Unit, University of Liège, Liège, Belgium

^c TotalEnergies Marketing Services, Research Center, Solaise, France

^d TotalEnergies Refining and Chemicals, Total Research and Technologies Gonfreville, Harfleur, France

^e International Aolne Laboratory - IZMOC, Complex Materials Molecular Characterization, TRIG, Harfleur, France

^f AgroBioChem Department, Laboratory of Analytical Chemistry, University of Liège, Gembloux Agro-Bio Tech, Passage des Deportes 2, 5030 Gembloux, Belgium

Analysis of Fatty acids in animal fat

Sample preparation

10mg of fat + n-hexane, $\text{CH}_3\text{ONa} + \text{BF}_3$

↓ 1.5 hrs
70°C

.....

N_2 drying (20-30 minutes)

↓
Analysis

Sample preparation

Tedious
Time consuming



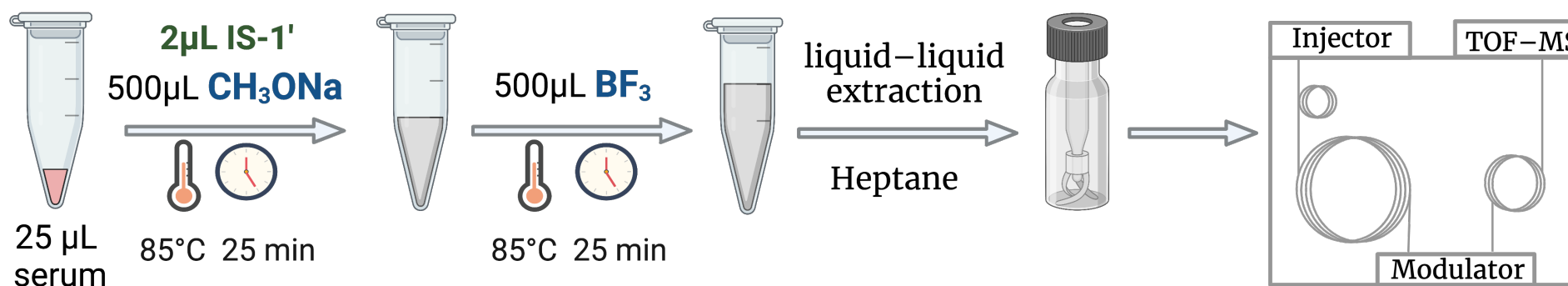
Sample preparation

**Moving towards
automization**



- ✓ Easy transfer to industry
- ✓ Increased efficiency
- ✓ Cost-effective

Specifically tailored and optimized approach for fatty acid analysis

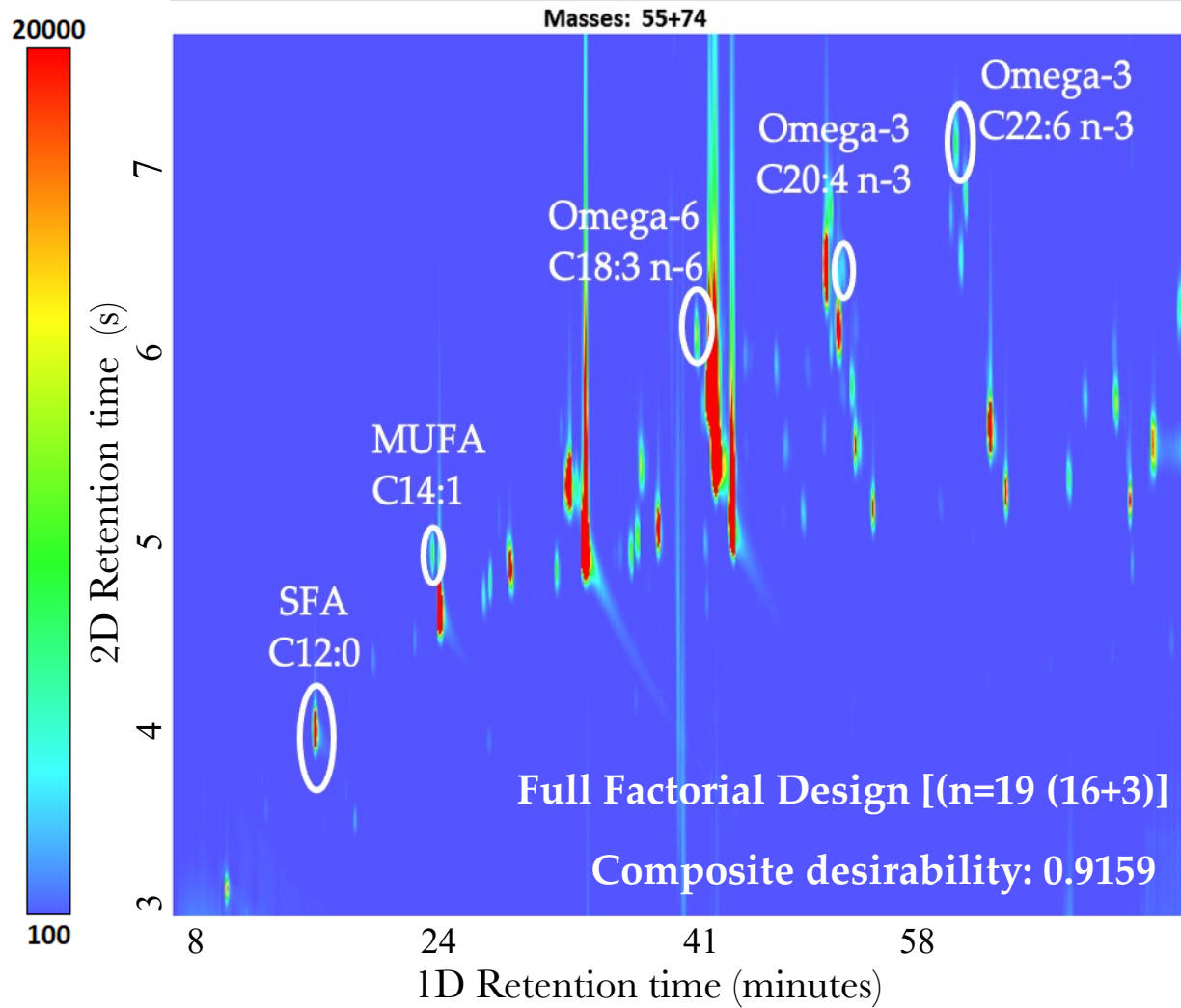


CH₃ONa → base-catalyzed transesterification

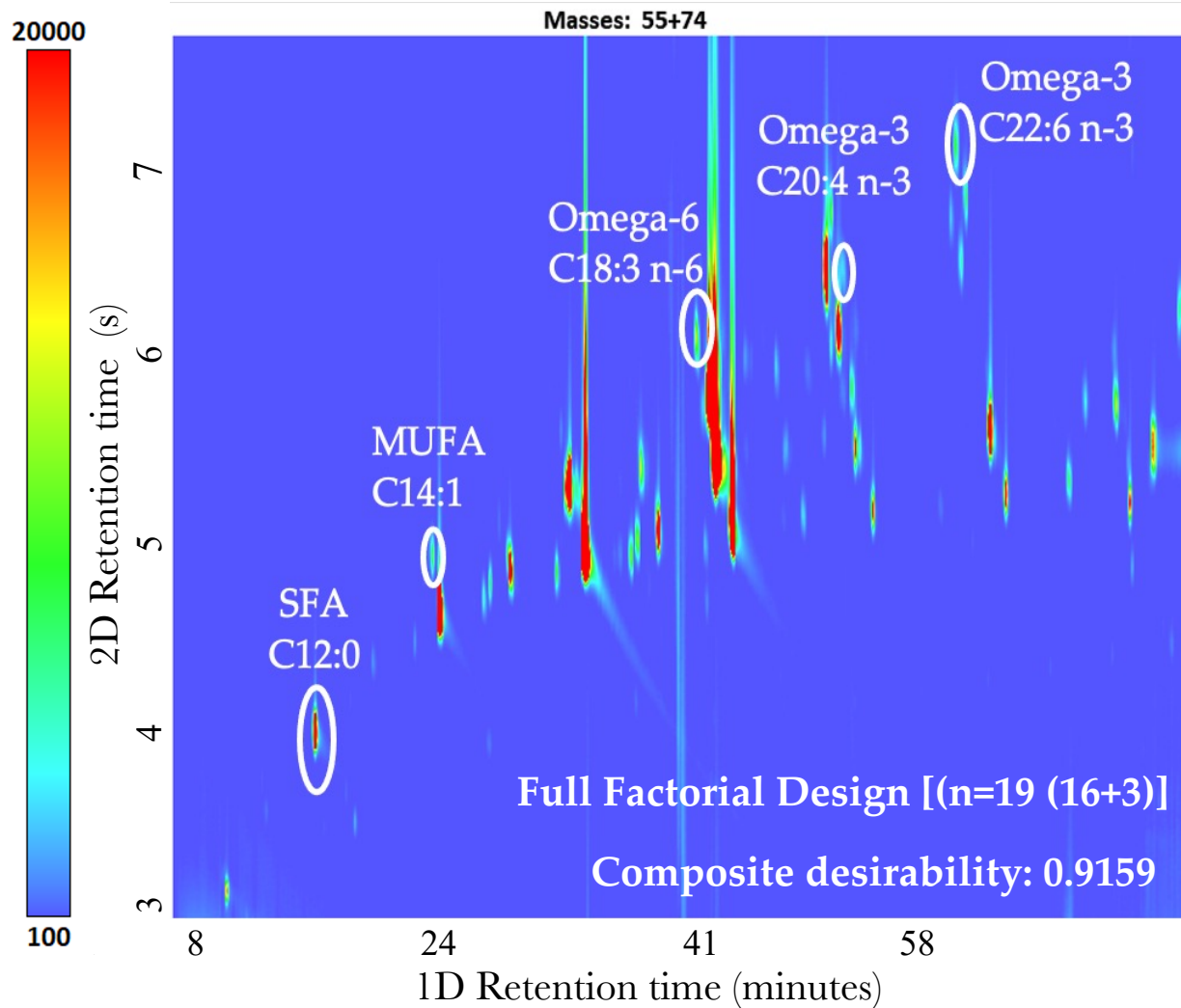
BF₃ → acid-catalyzed esterification



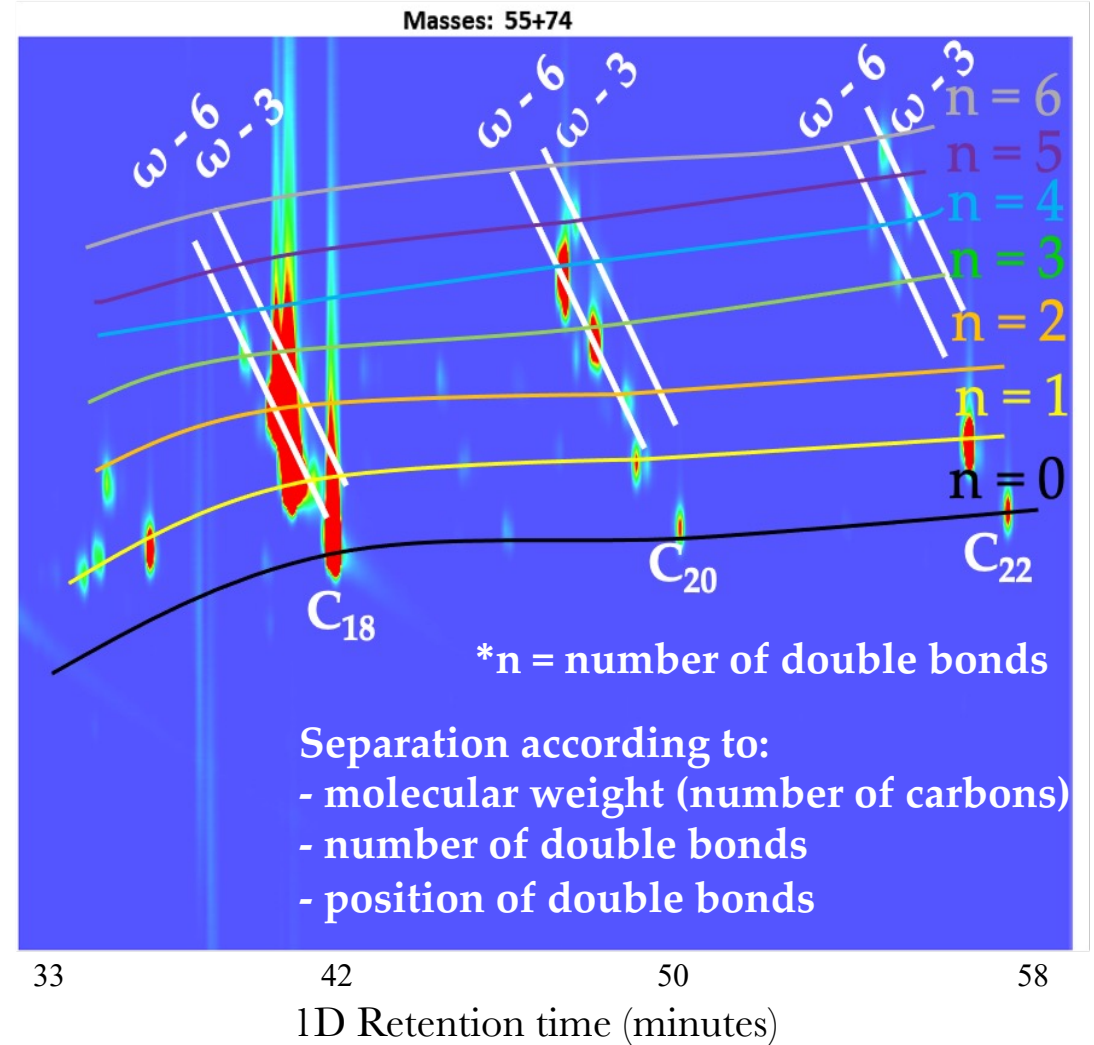
Response Optimization



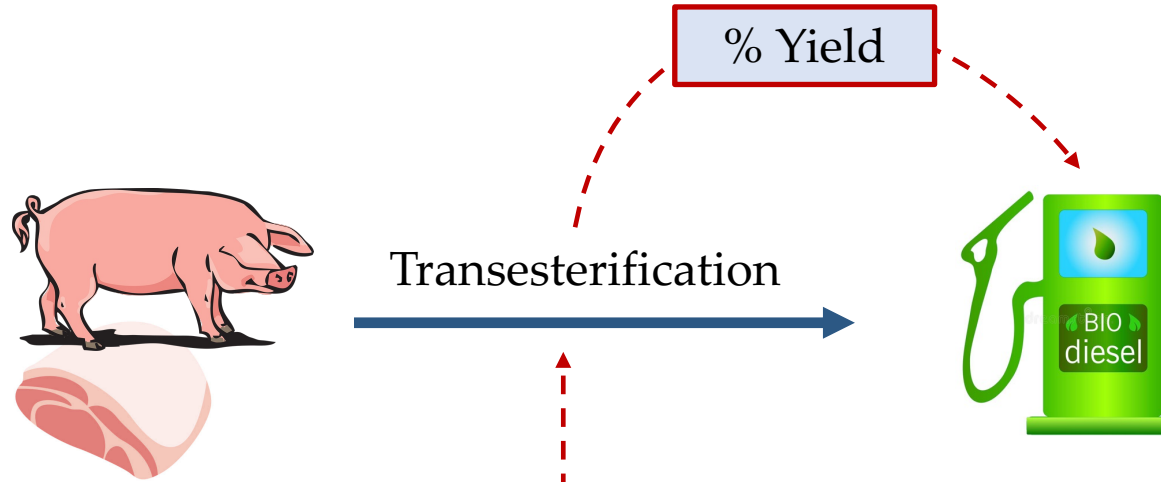
Response Optimization



Chromatographic separation



Production Process:



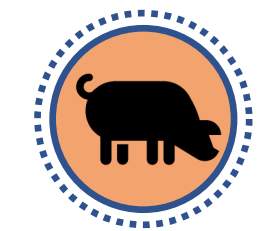
Impurities:

Oxygenated compounds
- Aldehydes
- Ketones
...
non-esterified fatty acids (NEFA)

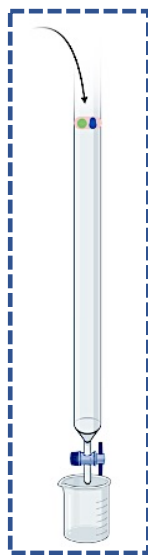
- Corrosion in the fuel
- Formation of gums...



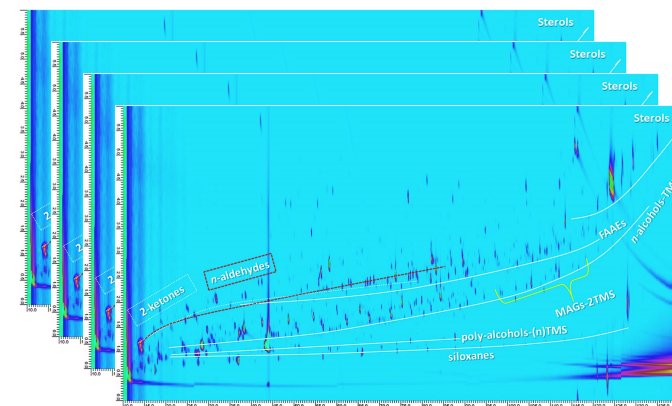
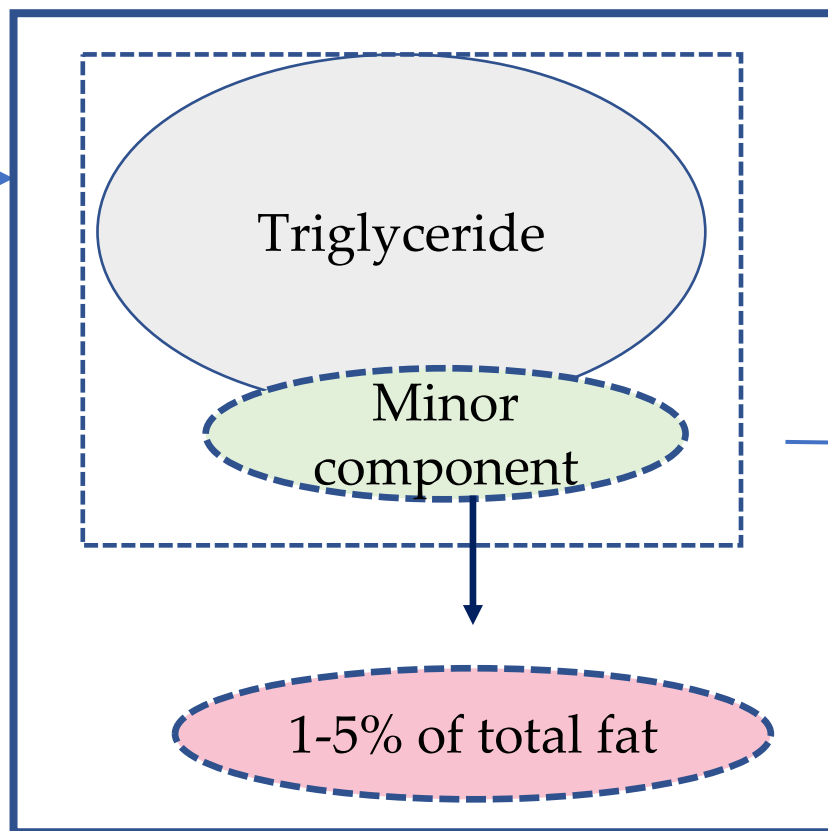
In-Depth investigation of oxygen-containing compounds in oleaginous feedstock (animal fat)



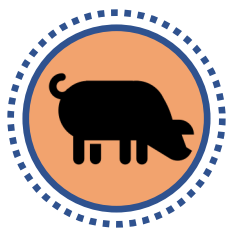
Animal fat feedstock



Extraction

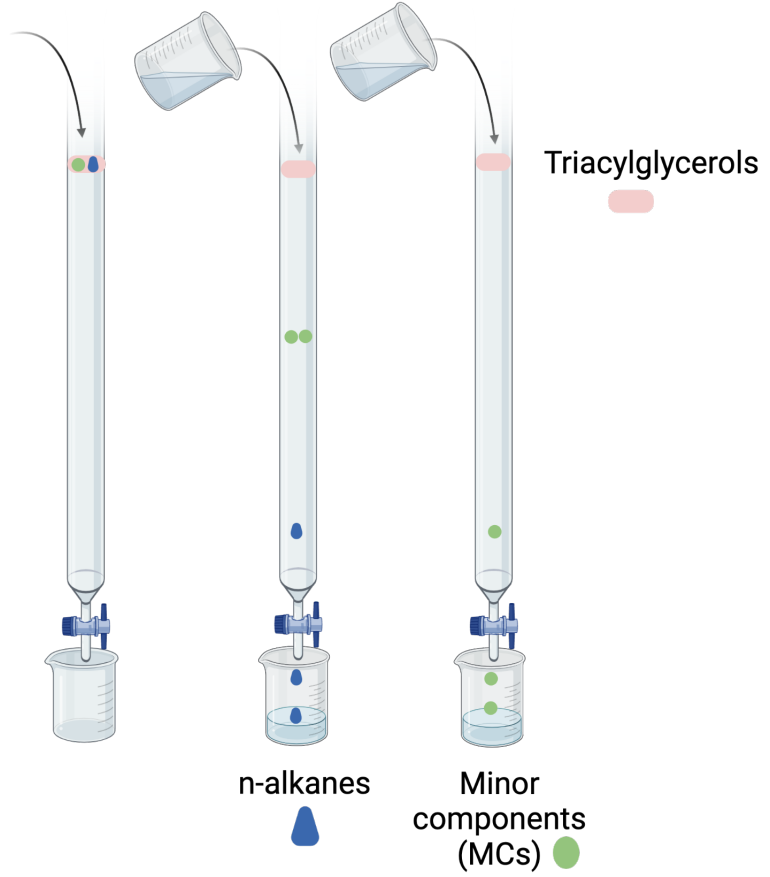


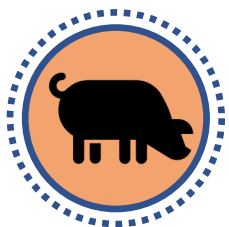
GCxGC-TOFMS



Animal fat
feedstock

TMS-lipid
sample
(100mg/1ml)



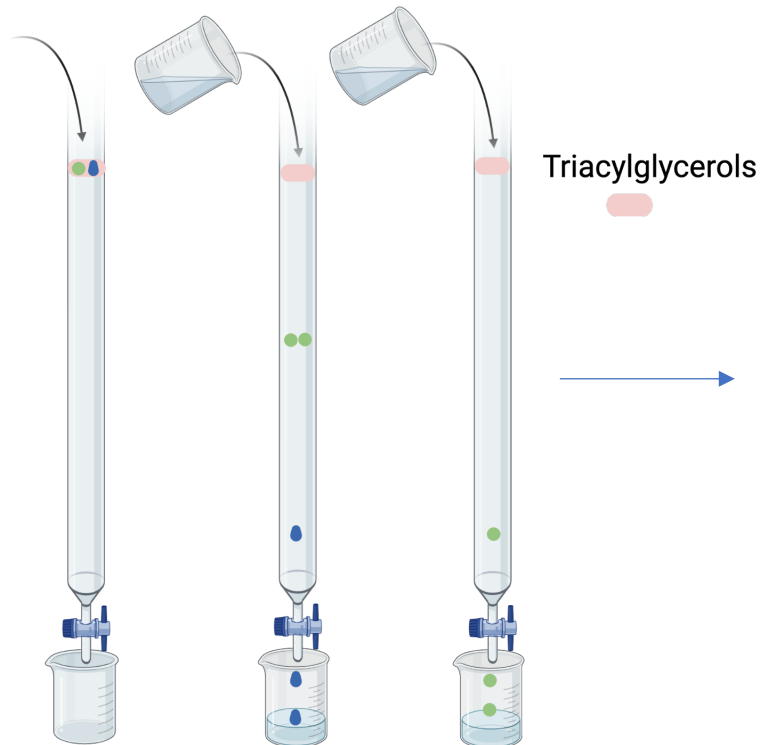


Animal fat
feedstock

TMS-lipid
sample
(100mg/1ml)

50 ml
hexane

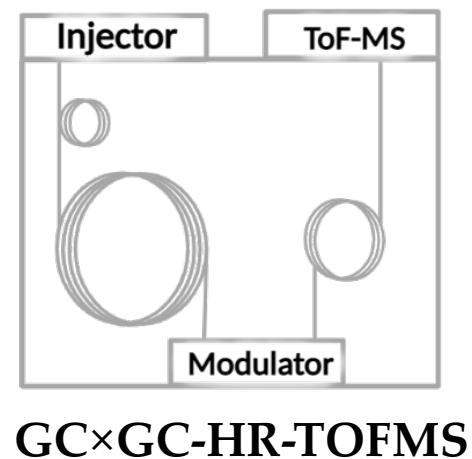
45 ml hex/
ethyl ether
(99/1)



Triacylglycerols

n-alkanes

Minor
components
(MCs)



Classes of Analytes

Acids
Alcohol
Aldehydes
Ethers
Monoglycerols
Tocopherol
Steroids



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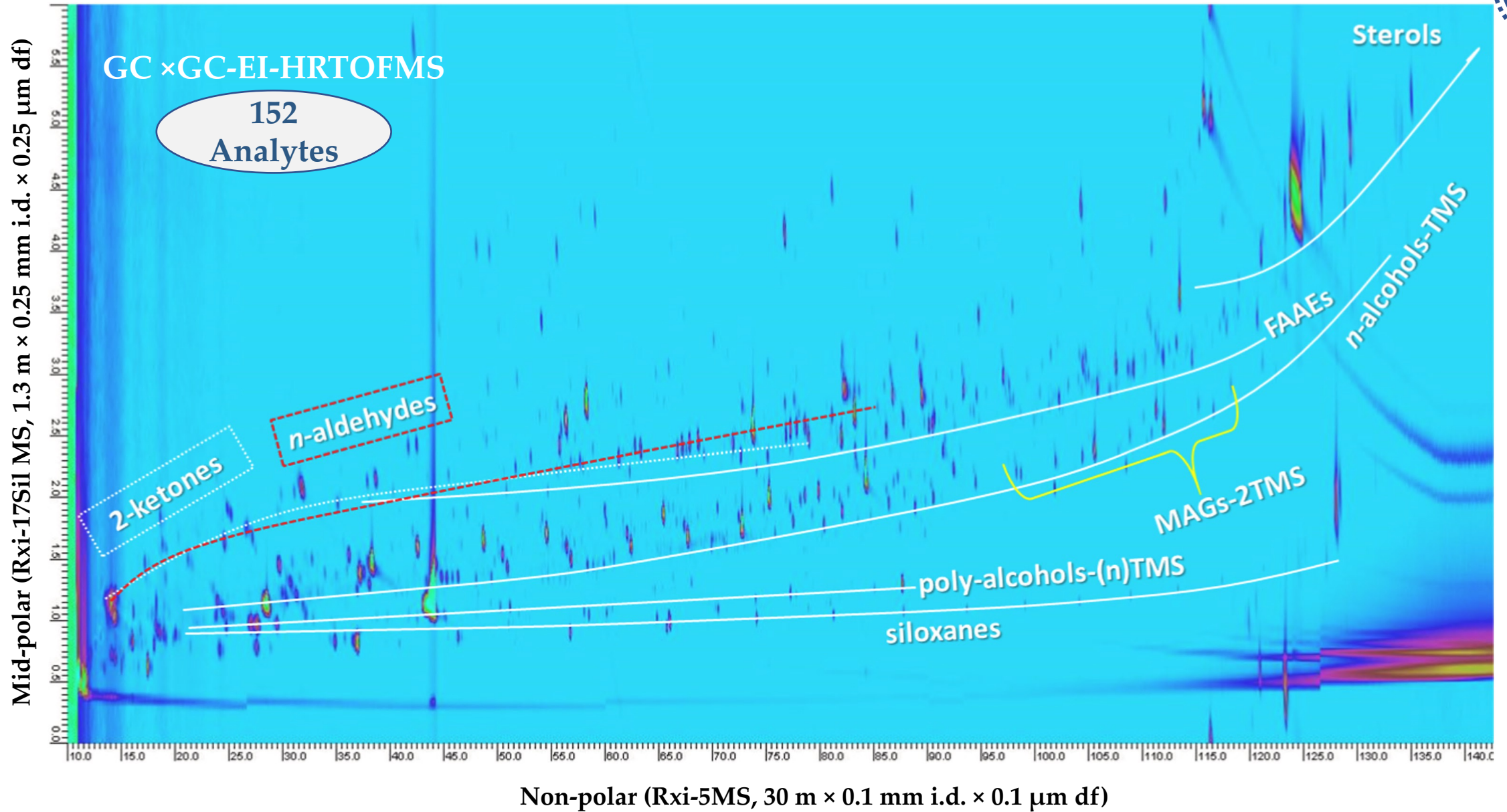
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^e International Joint Laboratory - iC2MC: Complex Matrices Molecular Characterization, TRIG, Harfleur, France

^f AgrobiChem Department, Laboratory of Analytical Chemistry, University of Liège, Gembloux Agro-Bio Tech, Passage des Deportes 2, 5030 Gembloux, Belgium

GC×GC fingerprinting of minor components of pig fat for Bio-Diesel



Criteria for identification of analytes

EI Mass spectra database match
70ev ($\geq 800/1000$)



Accurate mass (± 3 ppm)
HR-TOFMS

Linear Retention Indices
(1t_R)

Structured chromatographic
separation
($^1t_R, ^2t_R$)

Criteria for identification of analytes

152 Analytes identified

EI Mass spectra
database match
70ev ($\geq 800/1000$)

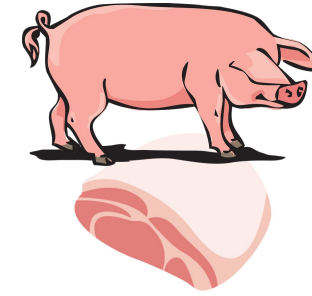
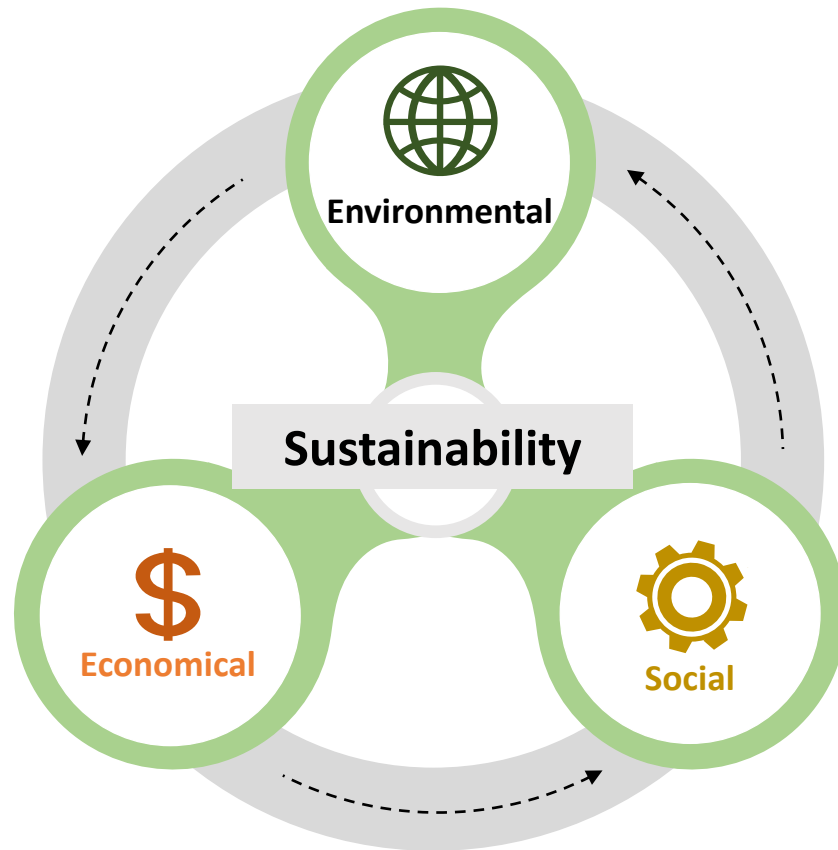
Accurate mass (± 3 ppm)
HR-TOFMS



Linear Retention
Indices (1t_R)

Structured
chromatographic
separation
($^1t_R, ^2t_R$)

Acids	22
Alcohol (mono)	18
Alcohol (poly)	14
Aldehyde	23
Esters	28
Ether	1
Hydrocarbon	4
Ketones	10
Monacylglycerol	9
Nitrogen containing analytes	2
Tocopherol	2
Sterols	19



Animal Wastage



Plastic Wastage

370 million tons of plastic were produced in the world in 2020



Characterization of mixed plastic pyrolysis oil



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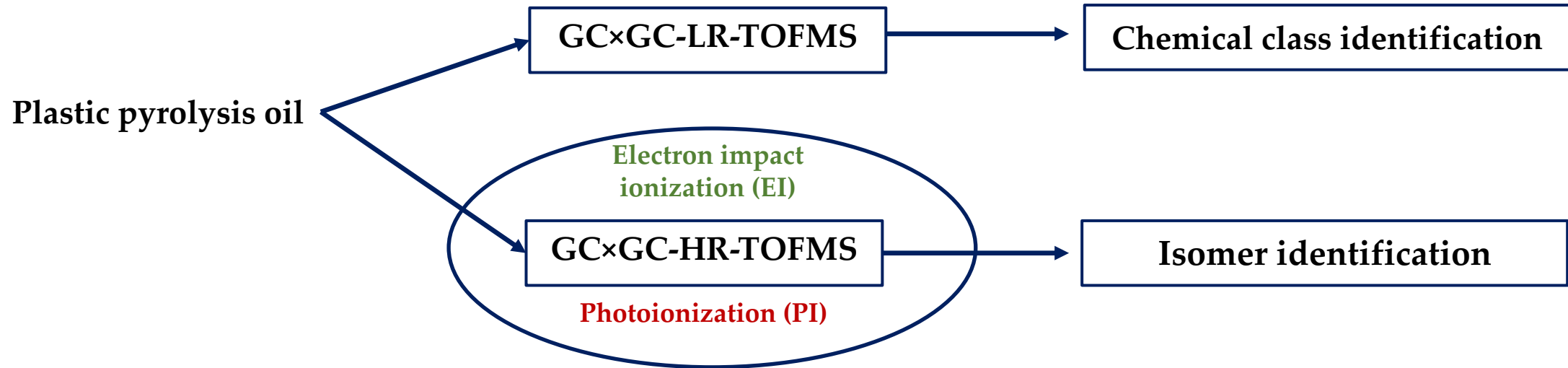
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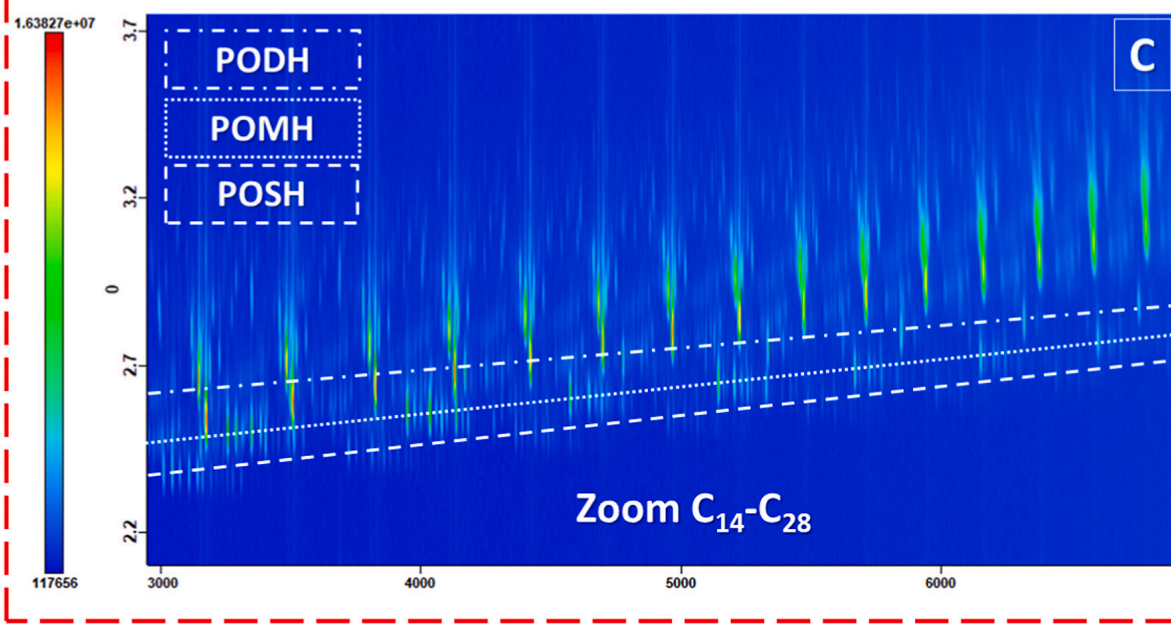
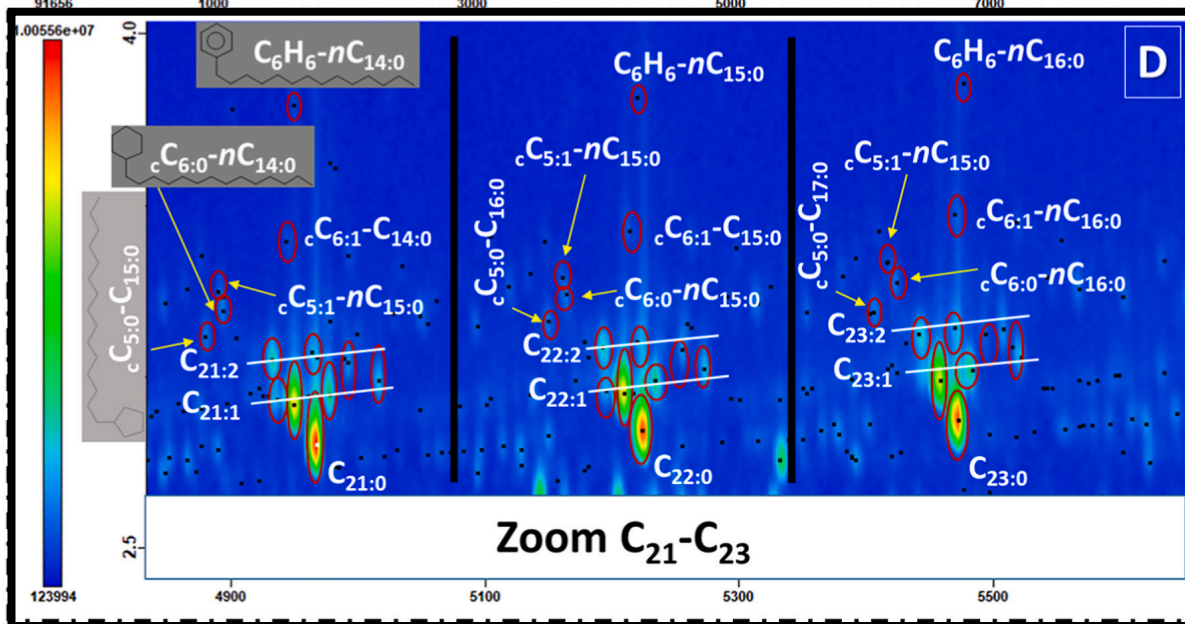
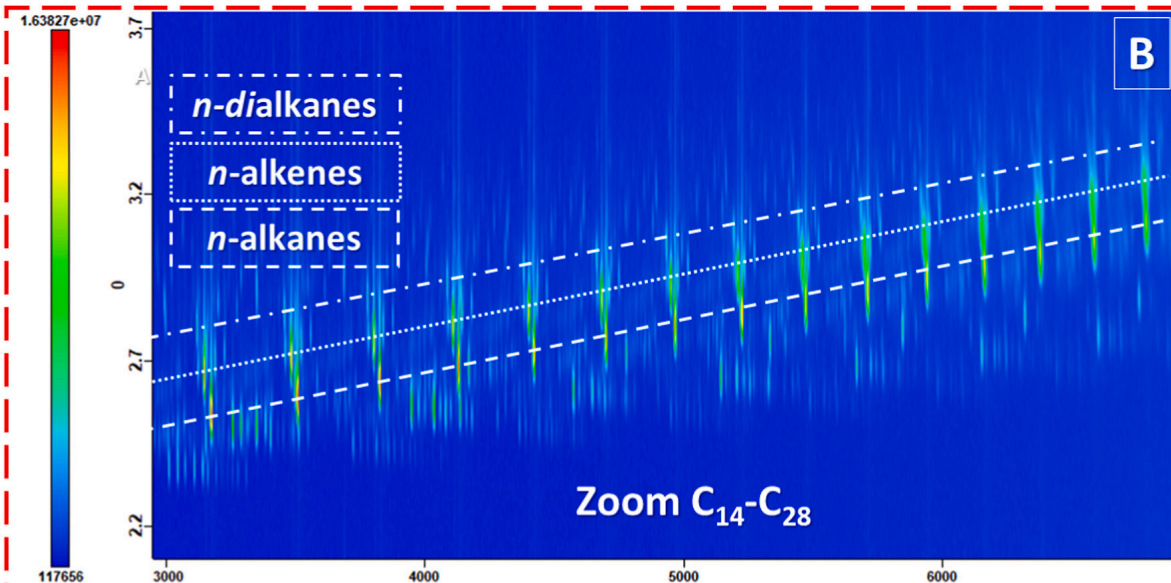
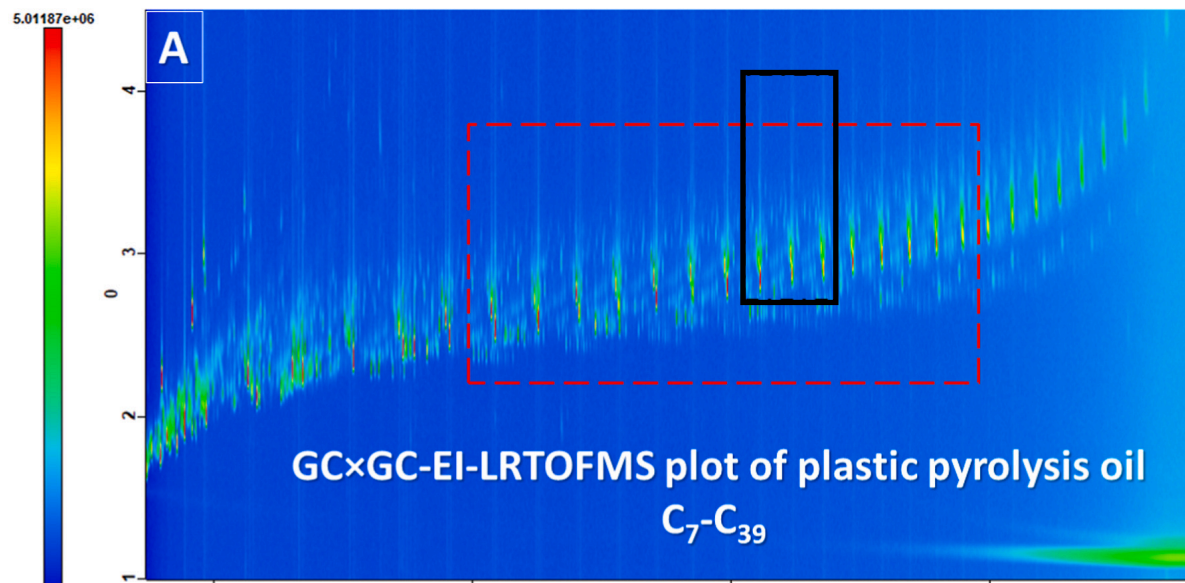
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Analysis of mixed plastic pyrolysis oil by comprehensive two-dimensional gas chromatography coupled with low- and high-resolution time-of-flight mass spectrometry with the support of soft ionization



- HRMS (EI,PI) helps predicting exact molecular mass, enables prediction of elemental composition
- Helps to identify compounds not yet included in MS libraries, accelerating understanding of plastic pyrolysis oil and mechanisms



Take home message



**Looking for renewable energy sources while solving the existing world problems
Win! Win!**

Diverse mass composition
of raw materials requires
advanced characterization



Take home message



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Diverse mass composition
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Optimization of sample preparation is a key
to focalize hidden information in a sample

Take home message



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Win! Win!**

Diverse mass composition
of raw materials requires
advanced characterization



Separation power of GC×GC
is essential for analysis of
complex sample mixture

Optimization of sample preparation is a key
to focalize hidden information in a sample



Thank you for your attention

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ikinjalbhatt@gmail.com

