Characterization of Extractables and Leachables Associated with Pharmaceutically Relevant Materials: Case Studies Outlining Analytical Approaches, Challenges and Examples

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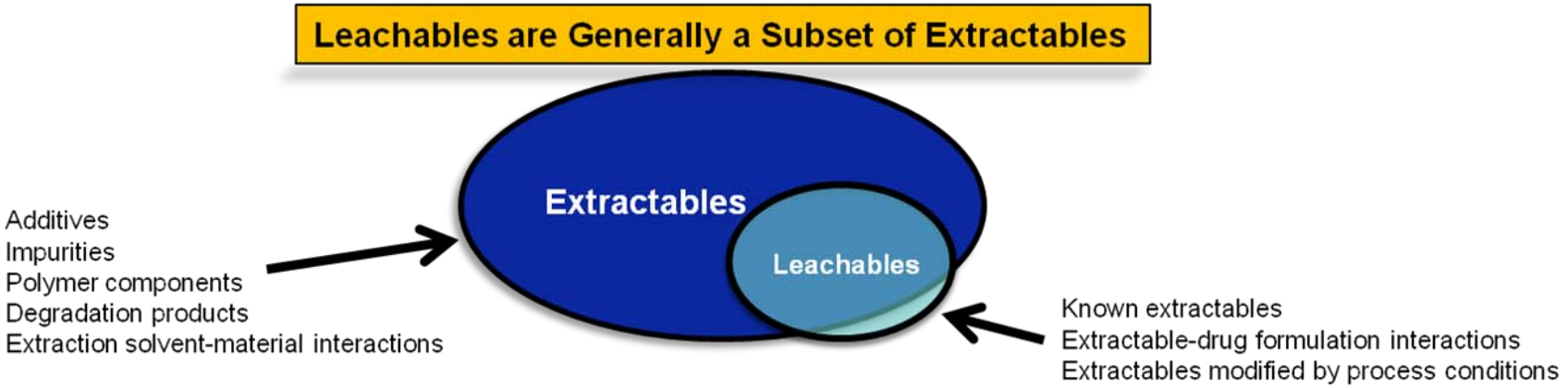
Lilly

Answers That Matter.

Some Key Definitions

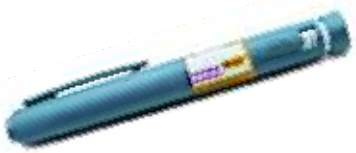
- **Extractables**
  - Chemical compounds that are extracted from any product contact material when exposed to an appropriate solvent under exaggerated conditions of time and temperature
- **Leachables**
  - Chemical compounds that migrate into a drug formulation from any product contact material as a result of direct contact under typical process or accelerated storage conditions
  - Likely to be found in the **finished drug product**
  - Arise from interaction of material or system **during intended use**

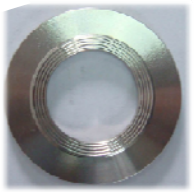


Why…

E&L evaluation is a Regulatory expectation for materials contacting or with potential to contact API or drug product

- container/closure systems
- device components
- mfg. process materials
- ICH Q3, Q6(A,B), Q7A, Q8, 21 CFR 211, EMEA/205/04





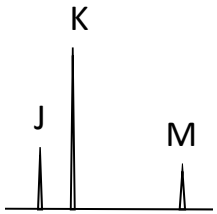
Polymer Supply Chain for Pharmaceutically Relevant Materials

Converters

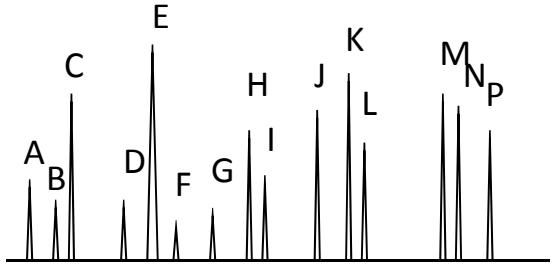
Natural Raw Material	Monomer	Polymer	Masterbatcher	End products & Suppliers
N-4	N-3	N-2	N-1	N
	• Bulk process chemicals • Storage stabilizers	• Initiators/Catalysts • Storage stabilizers • Antioxidants • Processing aids	• Antioxidants • Processing aids • Light stabilizers • Antistatic agents	• Colorants • Processing aids • Lubricants

Components for
- Packaging
- Devices
- Container/closure
- Processing

QBD and Extractable/Leachable Design Space



Control Space refers to the normal variation range of the product within conditions of intended use



Control Space
Leachables

Extractables

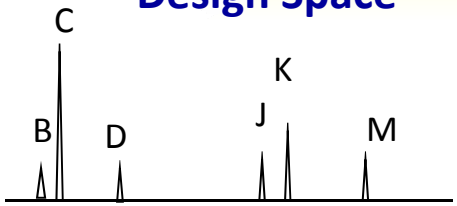
Knowledge Space

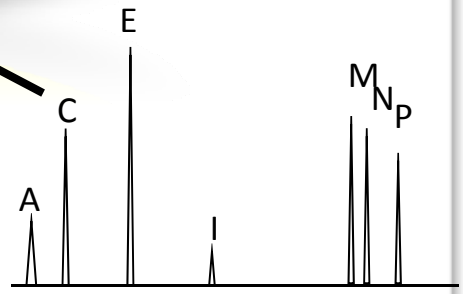
Drug Product A Leachables

Drug Product B Leachables

Product A Design Space

Product B Design Space

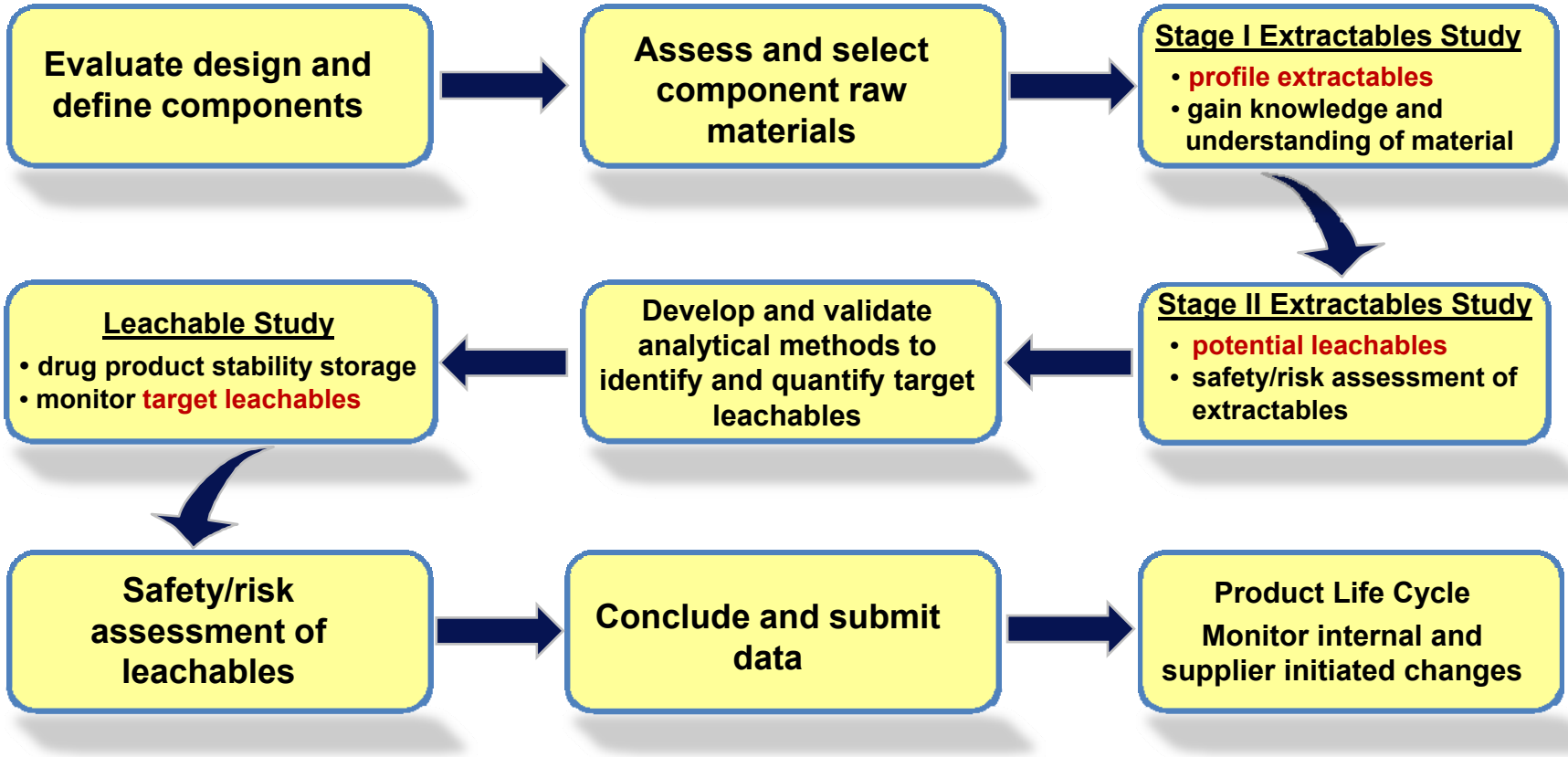




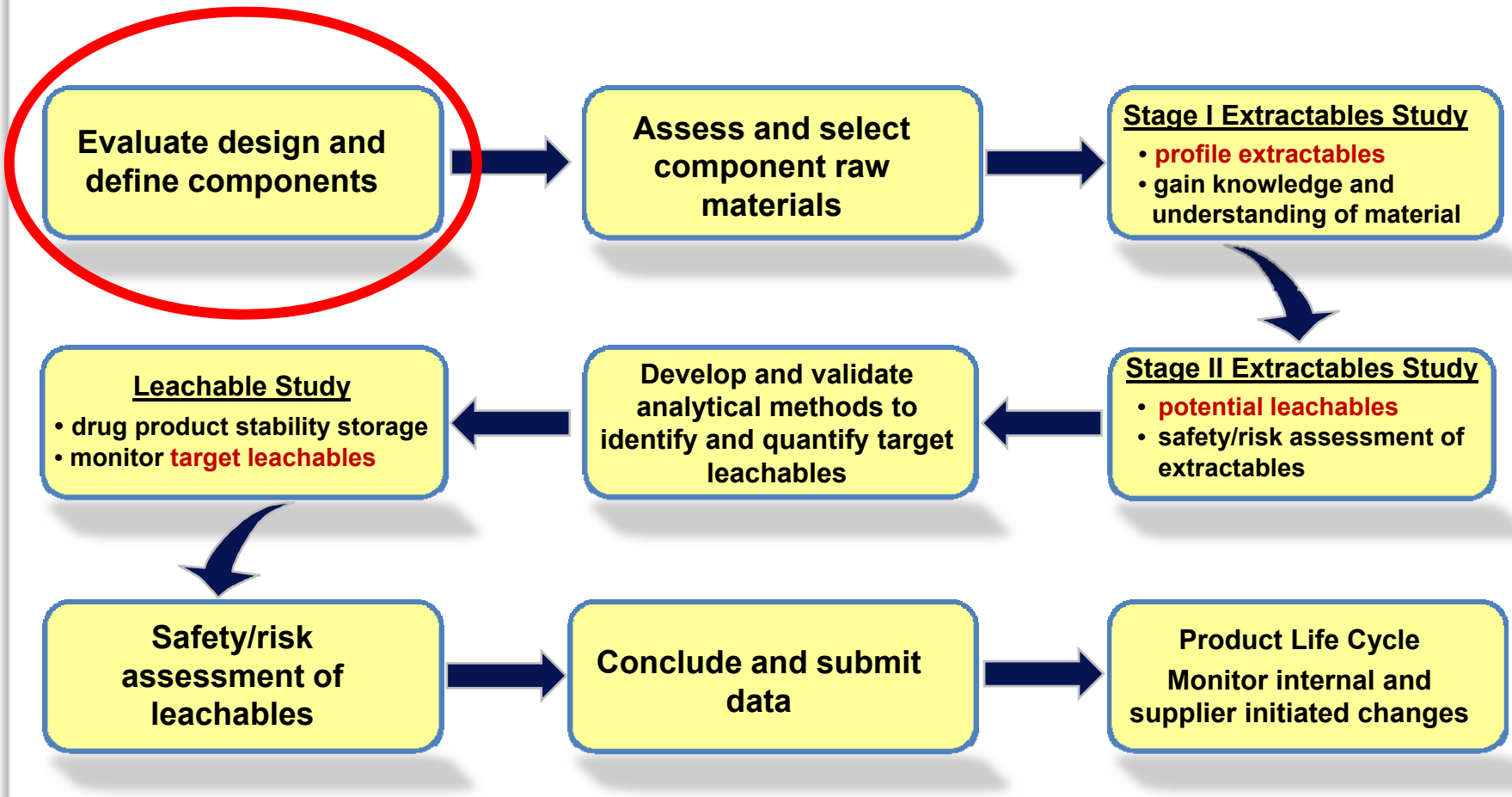
Each unique formulation will have a unique leachables design space

Figure courtesy of ELSIE

Extractable/Leachable Assessment Example



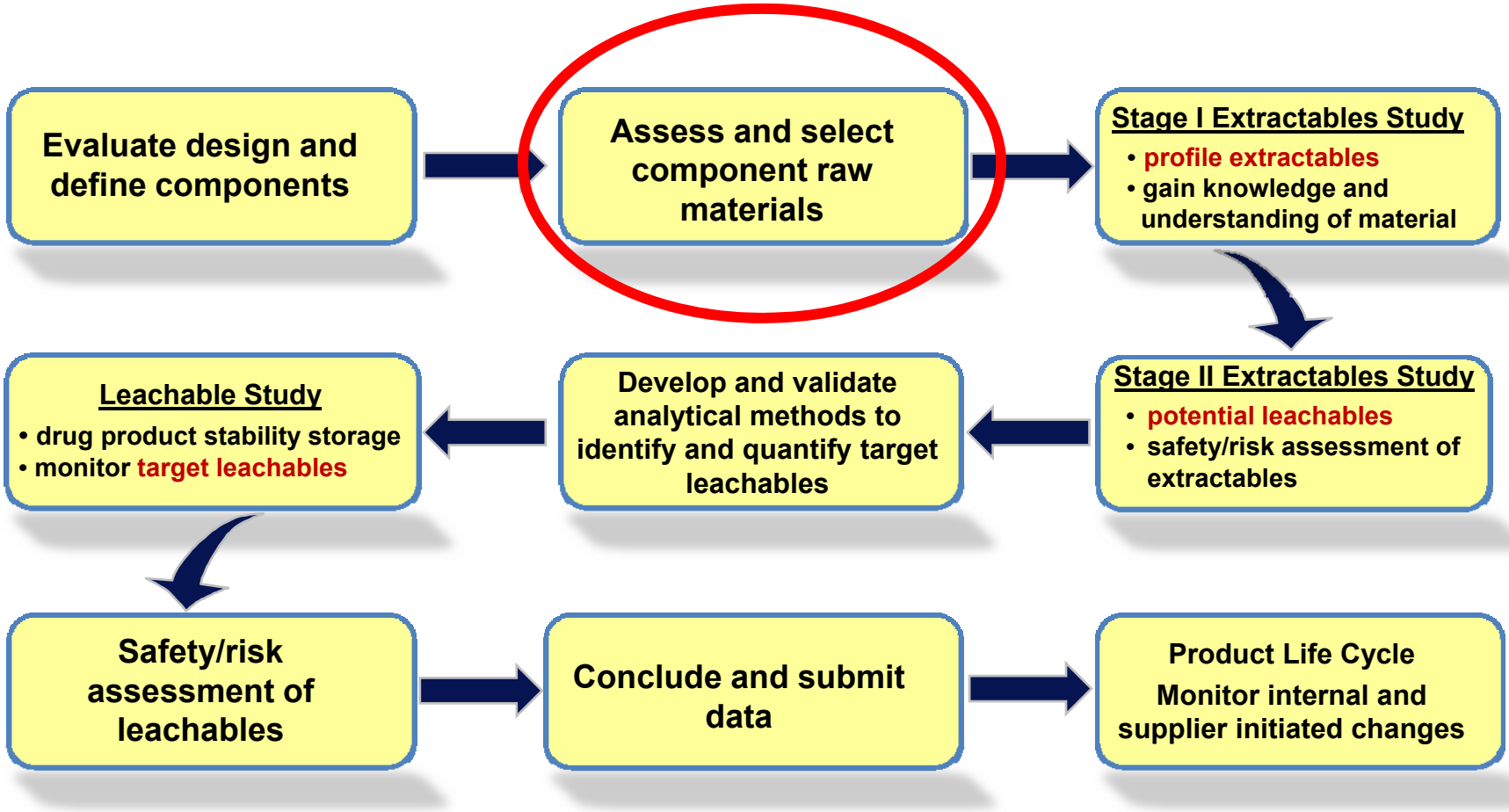
Extractable/Leachable Assessment Example



Evaluate Design and Define Components

- **The container/closure or device components are <u>part of the drug product</u>**
- **What are some desired performance characteristics of a C/C or device?**
  - **Accurate, reproducible and reliable delivery of drug product**
  - **Robust physical and mechanical operation and construction**
  - **Protection of drug product across range of intended use conditions**
- **Manufacturability—complexity, reproducibility and reliability**
- **Patient-friendly characteristics**
  - **Convenience and ease of use**
  - **Appearance and dimensions**
  - **Ruggedness across diverse patient population**

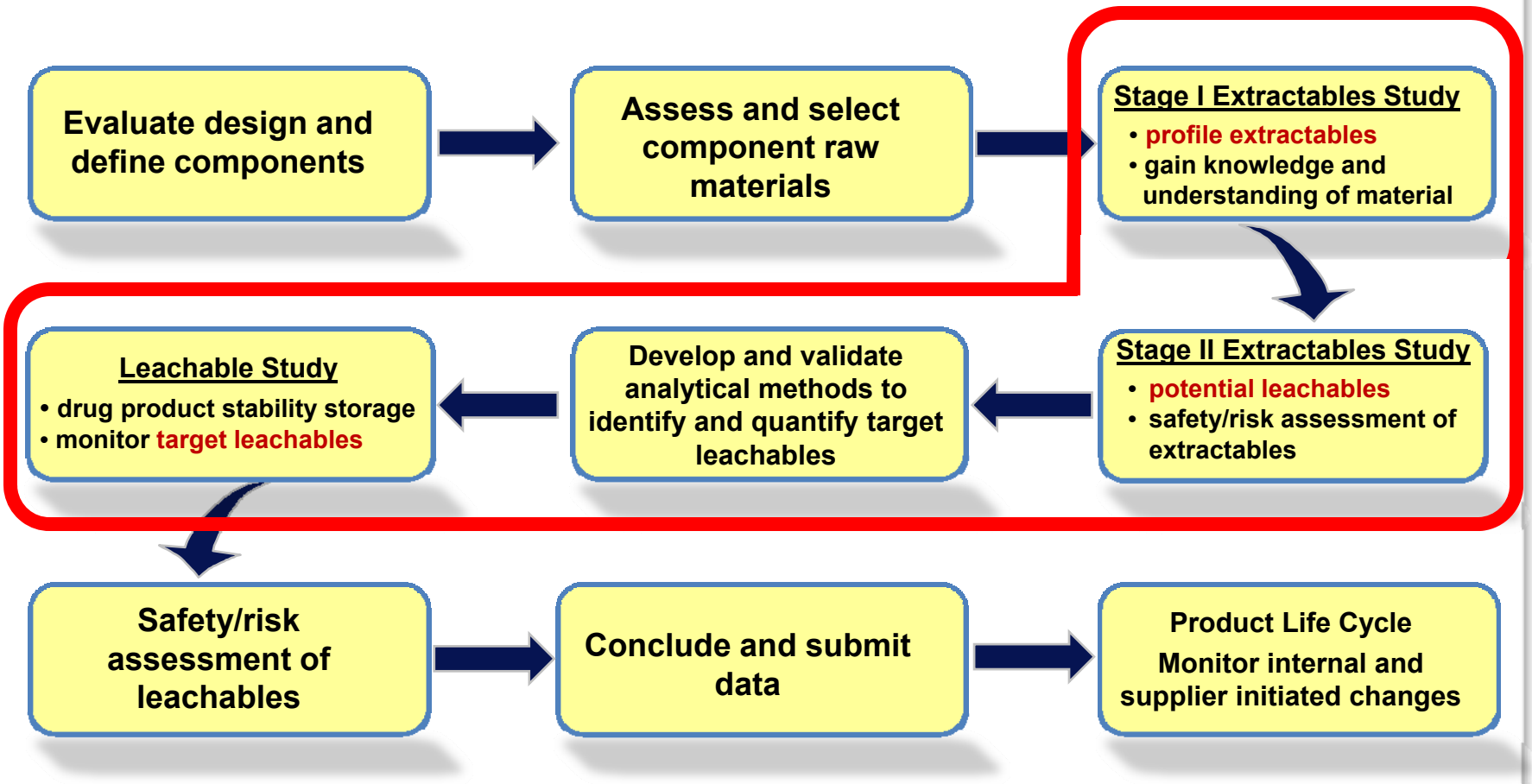
Extractable/Leachable Assessment Approach Example



Assess and Select Component Raw Materials

- Obtain as much information as possible from component vendor about raw materials and production of components
- Complete list of chemicals and additives, under CDA if necessary
- As much history from “upstream” processors as available
- List of extractables if testing completed by vendor, including conditions under which extractables were determined
  - Extraction solvents, methods and conditions
  - Analytical conditions
- In-use history of component lot in industry, if applicable

Extractable/Leachable Assessment Approach Example



Extractable/Leachable Assessment Approach Example

Study	Conditions	Purpose	Analytical Techniques
Stage I Extractables	• Aggressive • Water, IPA, Hexane • Reflux	• Establish extractables profile • Gain understanding of material	HS GC-MS GC-MS LC-MS, w/PDA ICP-MS IC
Stage II Extractables	• Milder conditions than reflux • Water, IPA, buffers, "model solvents" • May include placebo	• Identify extractables that may not be stable to reflux • Simulate worst case drug product—packaging contact • Impact of excipients	Same as above
Leachables	• Drug product on stability storage • Conditions of intended use	• Determine real-time leachables • Monitor leachables over shelf life of product	Determined with tox assessment of extractables

Extractable Compound Evaluation

- One of the most important and analytically challenging steps is the initial establishment of a comprehensive extractable compound profile

  - Unknown compound structural characterization is not trivial

- Establishing an extractable compound profile is important to enabling understanding of a material

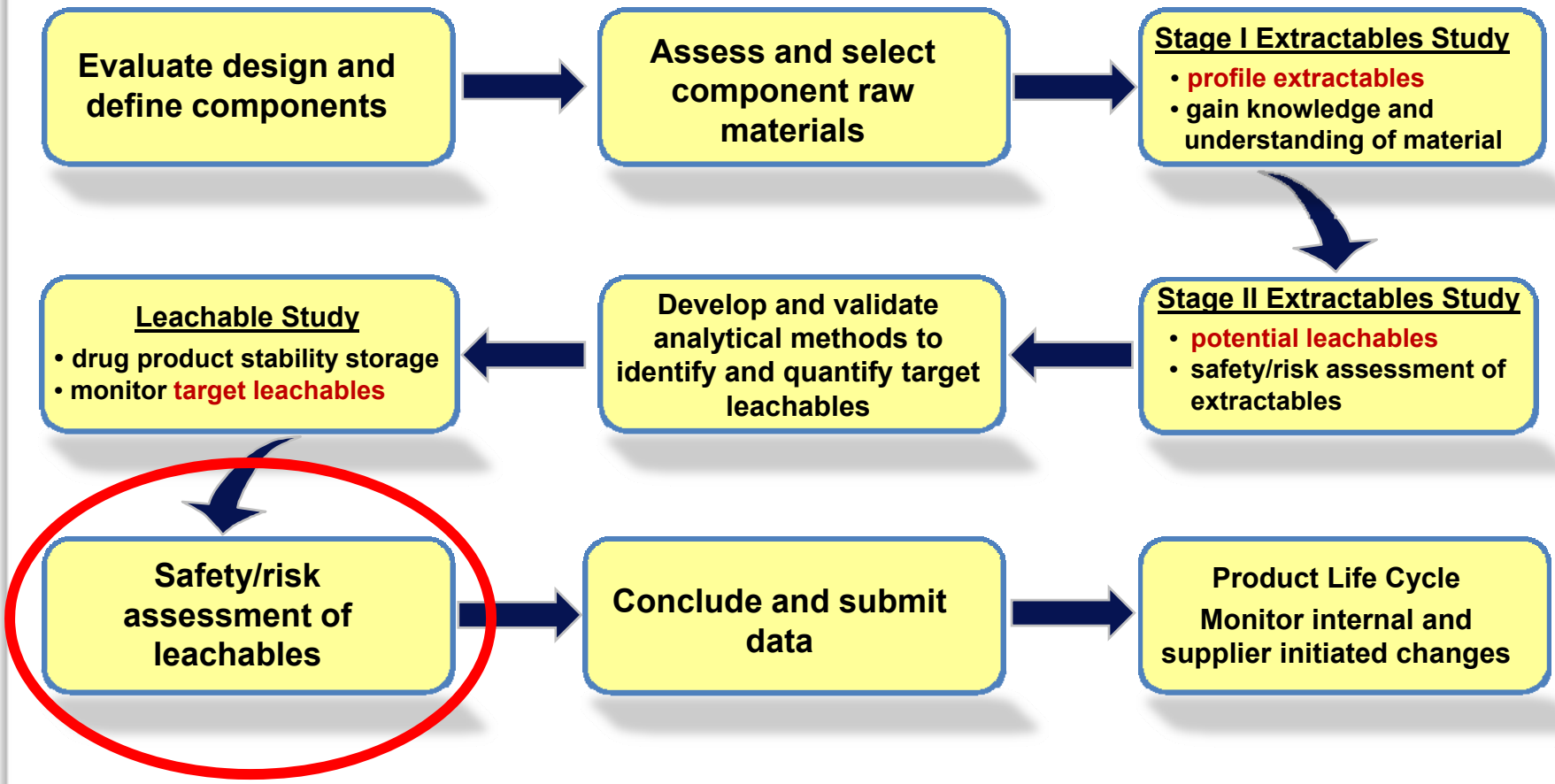
- Often involves characterization of complex mixtures of diverse organic molecules in a variety of matrices

- Multiple analytical techniques are required

Safety/Risk Assessment of Extractables

- Toxicologist screens characterized extractables for potential problematic compounds or alerting structures
- Qualitative SAR for compounds for which sufficient toxicity data is not available
- Provides indication of potential problematic compounds early enough to permit a decision involving changes to the design or materials for a component or container/closure system
- Early enough to work with supplier to mitigate changes

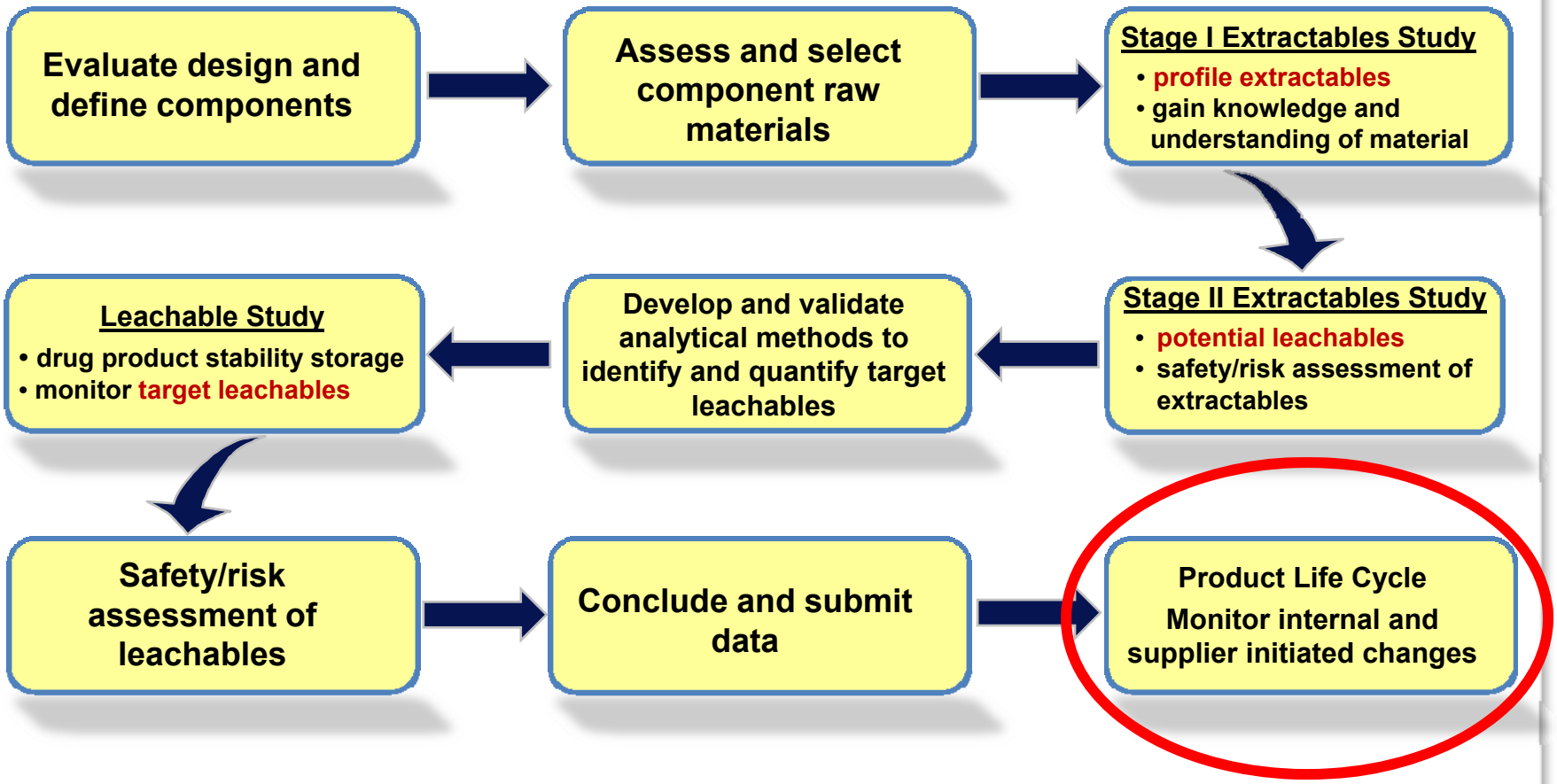
Extractable/Leachable Assessment Approach Example



Safety/Risk Assessment of Leachables

- Toxicologist evaluates qualitative and quantitative real-time storage leachables data
- Ensure that confirmed leachables are consistent with observed extractables
- Confirmed leachables are correlated with available toxicity data
- If toxicity data is not available, SAR correlation or *in-vivo*/*in-vitro* studies may be necessary
- Additional considerations may include:
  - Drug product dose
  - Administration route
  - Dose frequency
  - Treatment duration
  - Patient population

Extractable/Leachable Assessment Approach Example



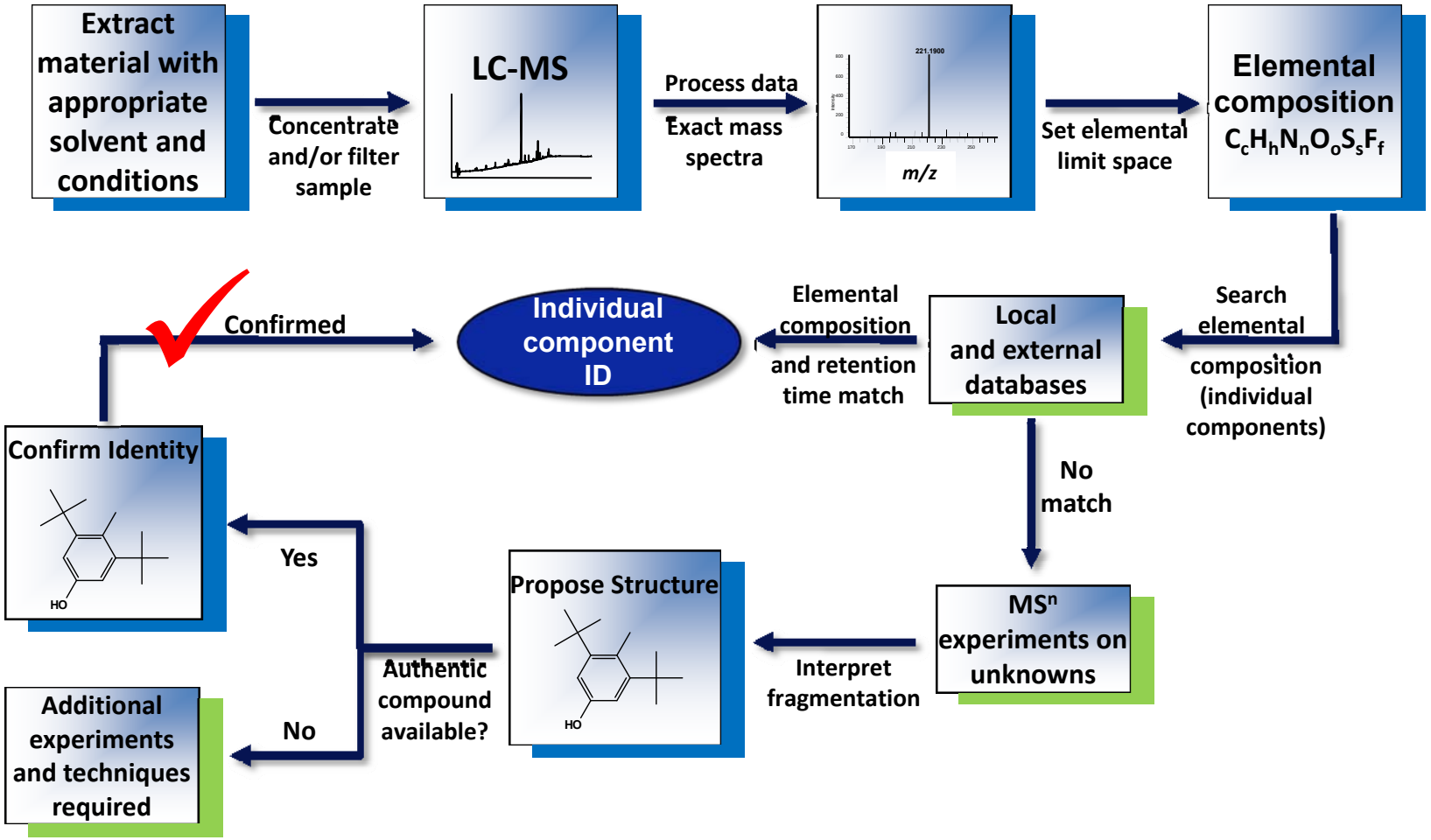
Product Life Cycle Management

- Strategy for gaining knowledge of, and addressing change should be established
- Supplier/vendor should alert customers of any changes in composition, production or construction
- Supplier/vendor should be aware of any changes initiated by “upstream” processors
- Routine extractables testing to monitor variation across lots

Extractable Compound Evaluation

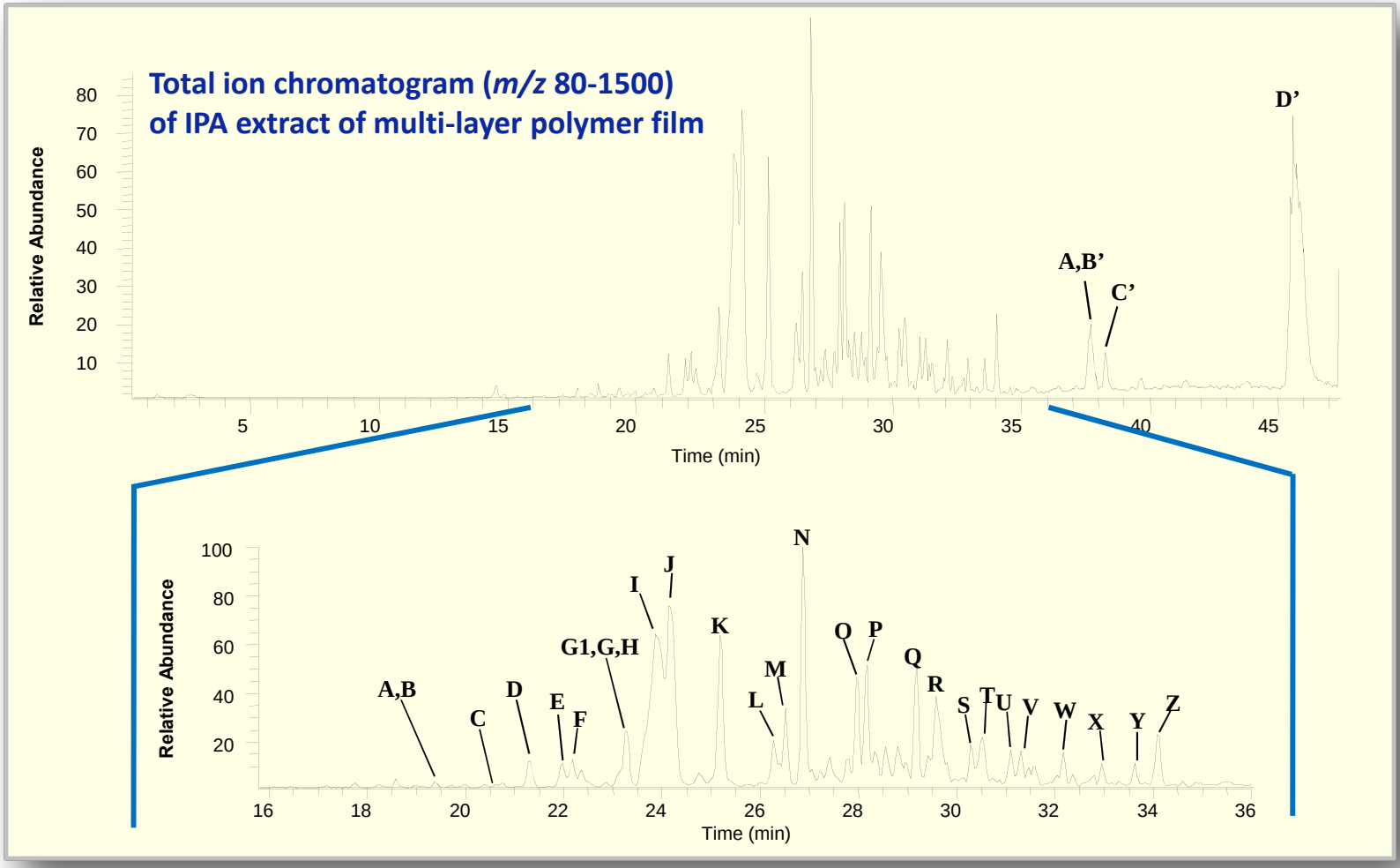
The structural characterization of diverse, unknown organic compounds, particularly if present in complex mixtures and multiple matrices, represents the greatest challenge in establishing an initial extractable compound profile for a given material. This often requires extensive effort, expertise and sophisticated capabilities.

Workflow for Identifying Unknowns with LC-MS



How Complex is "Complex?"

Chromatographic complexity

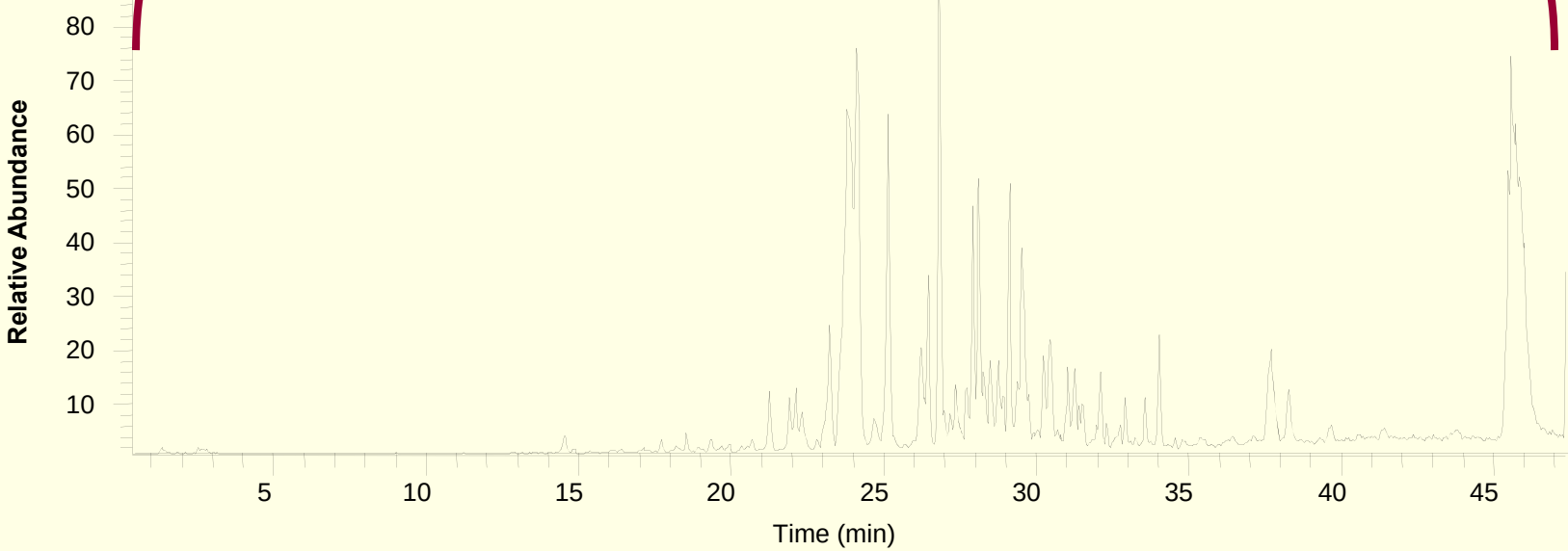


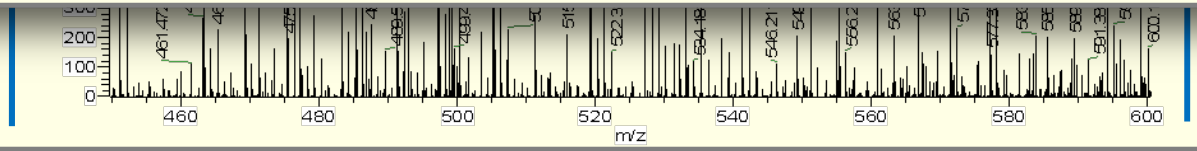
How Complex is “Complex?”

Mass spectral complexity

Composite mass spectrum of all components

Co-add mass spectra of all components in TIC





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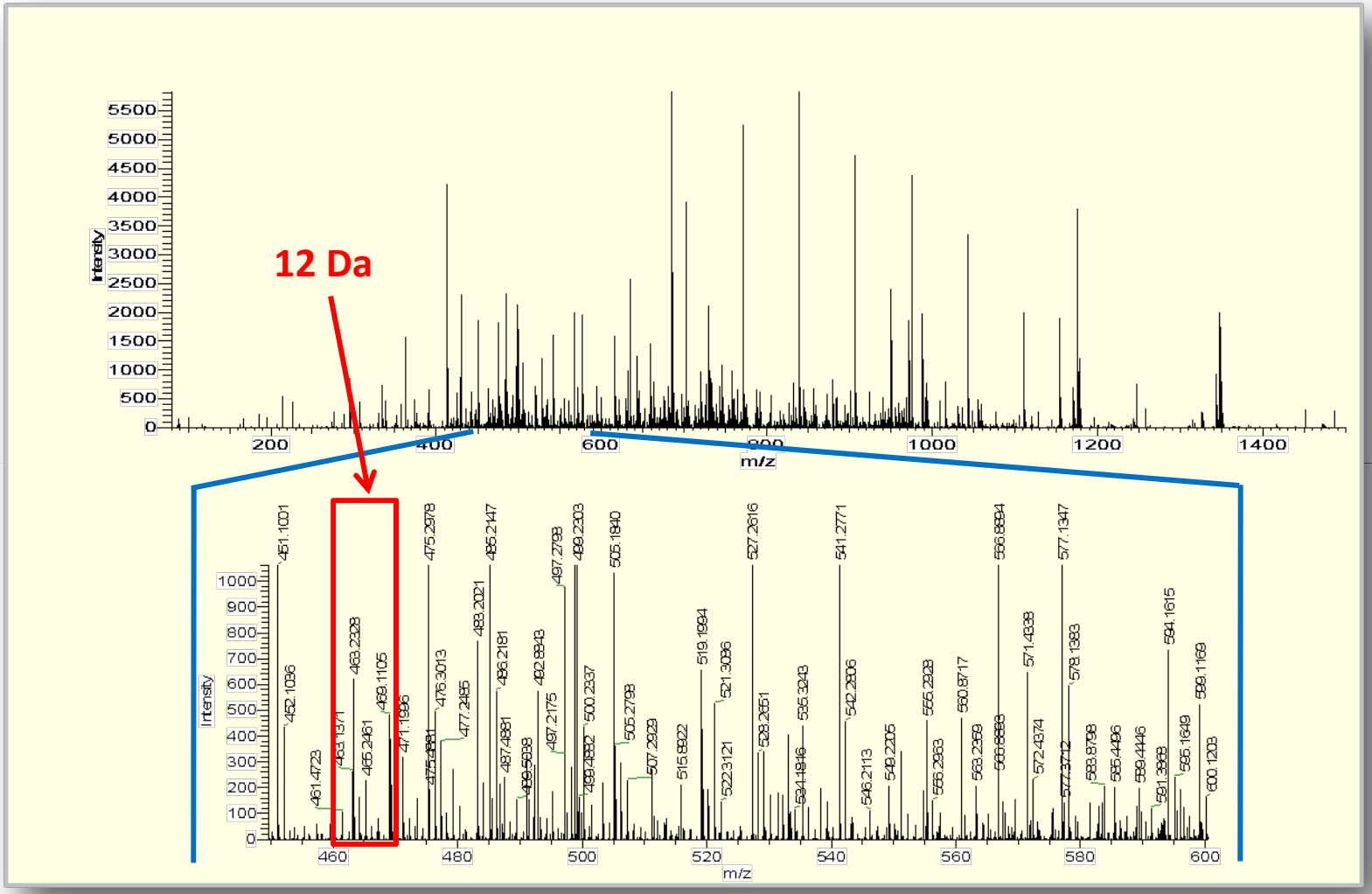
Mass spectral complexity



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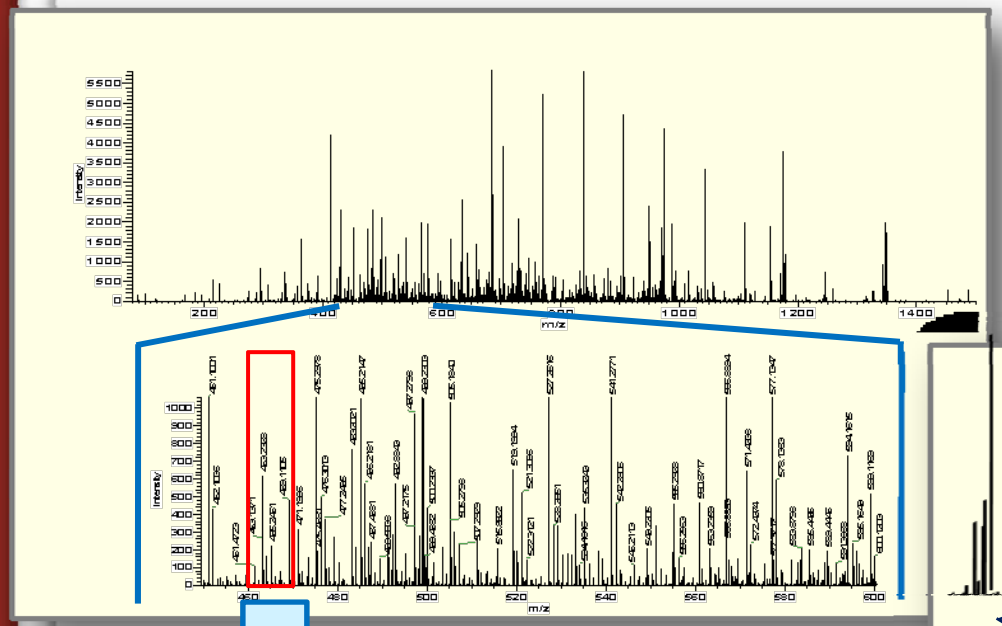
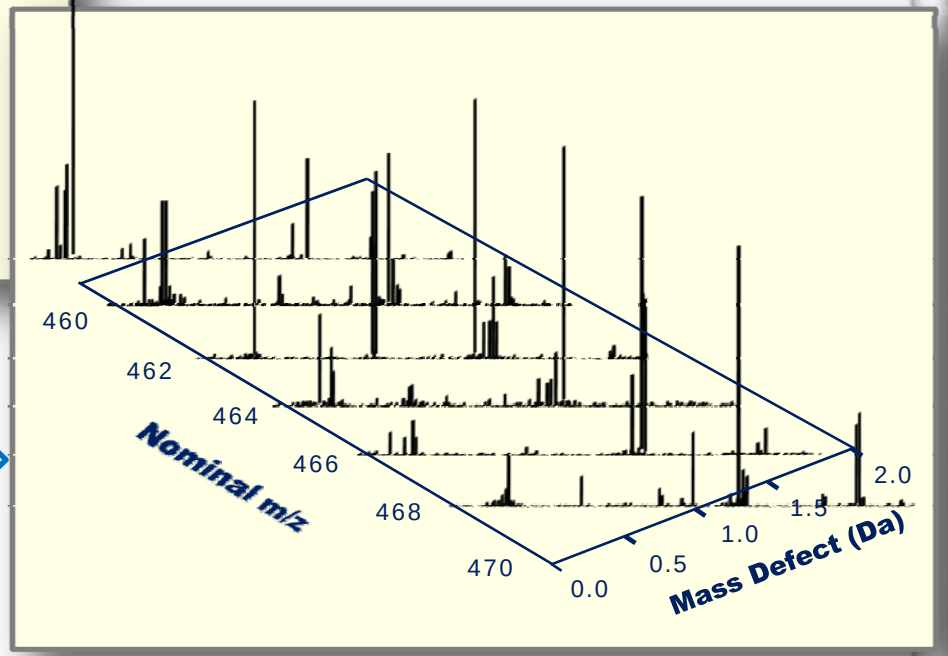
Mass spectral complexity



10-20 unique compounds per 2 Da segment

“Compressed” spectrum



FT-ICR MS

Ion accumulation, fragmentation and isolation

Linear Ion Trap MS
- MS, MS/MS and MS^n Analysis

Linear Ion Trap Data

FTICR MS
- Accurate Mass **(< 1 ppm)**
- High Resolution **(100-500K)**

FTMS Data

7 T Superconducting Magnet

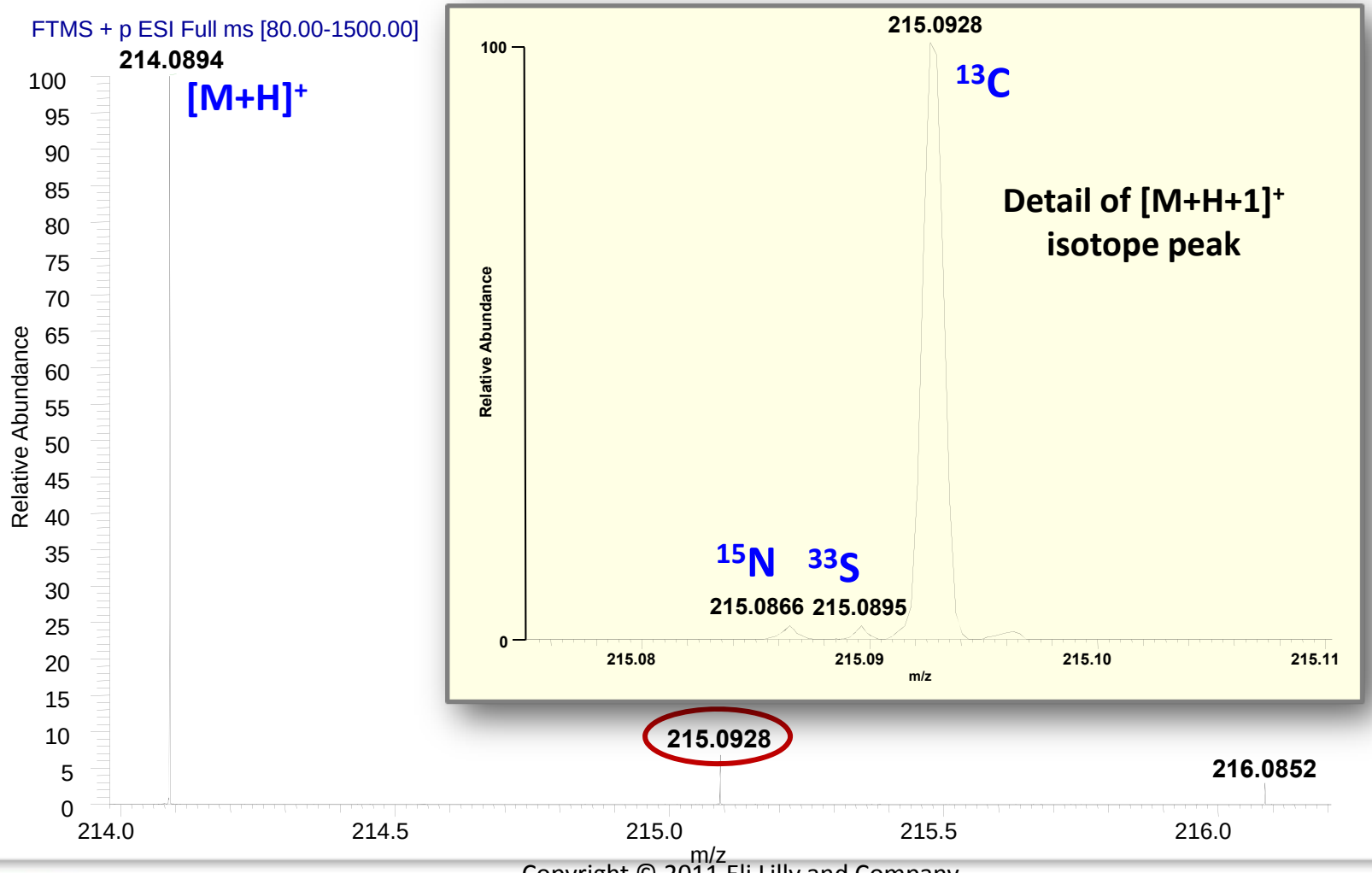
60 m^3/hr 15 L/sec 300L/sec 400L/sec 210L/sec 210L/sec

Triple Ported Turbo Pump

Image Courtesy of Thermo Scientific

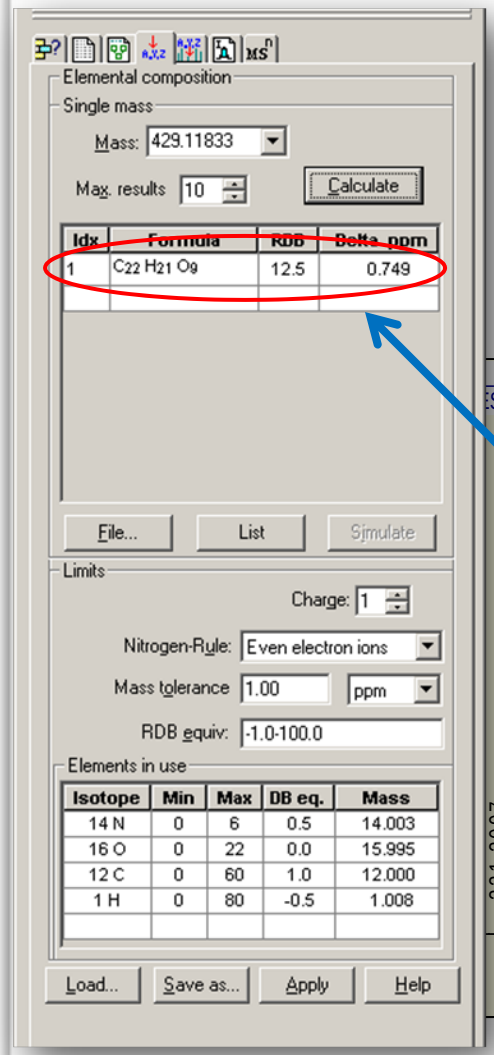
Why High Resolution MS?

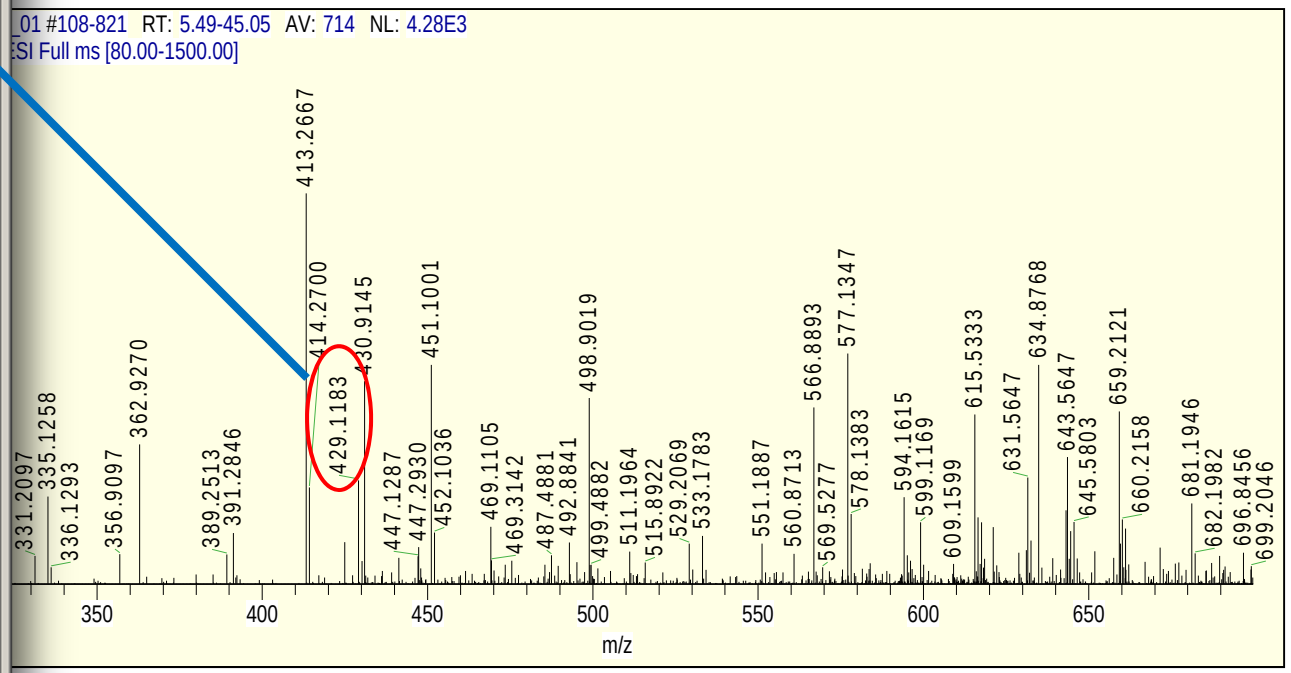
ESI FT-ICR MS mass spectrum of n-butyl benzenesulfonamide



To Extol the Virtues of Accurate Mass Measurement…

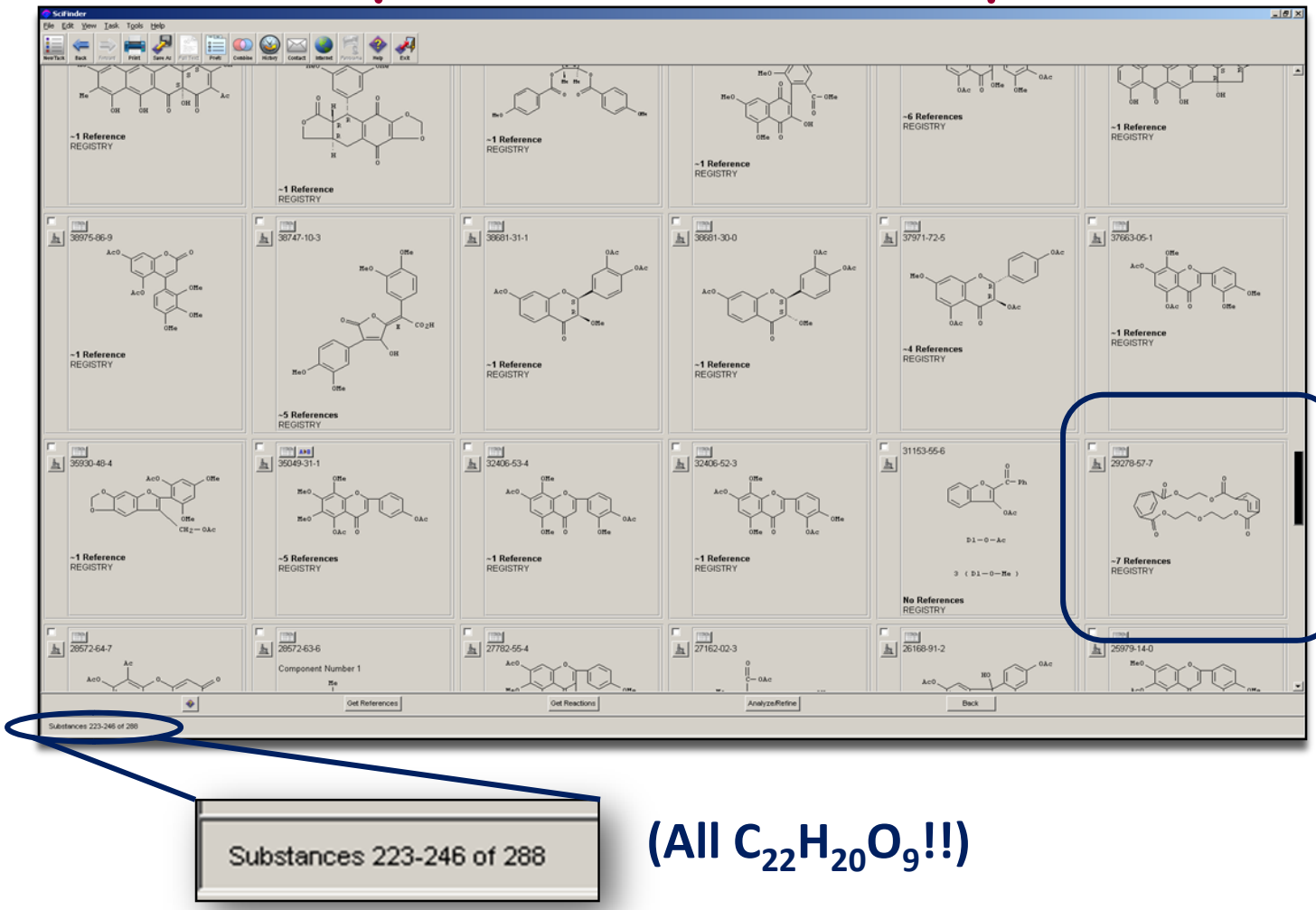
< 1 ppm mass accuracy narrows possible choices for elemental composition…





Correlating Structure with Formula

…but many structures are still possible…



Correct structure

(All $C_{22}H_{20}O_9$!!)

Five Case Studies will be examined:

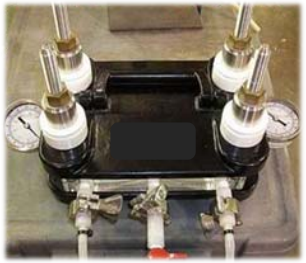
1. Identification of a packaging-related impurity detected in API material

2. Characterization of unknown extractables from PETG bottles with HDPE caps

3. Characterization of unknown extractables from an elastomer material under evaluation for device components

4. Qualitative comparison of 60 mL polypropylene syringes from two sources

5. Characterization of unknown impurities in bioprocessing equipment high volume filters

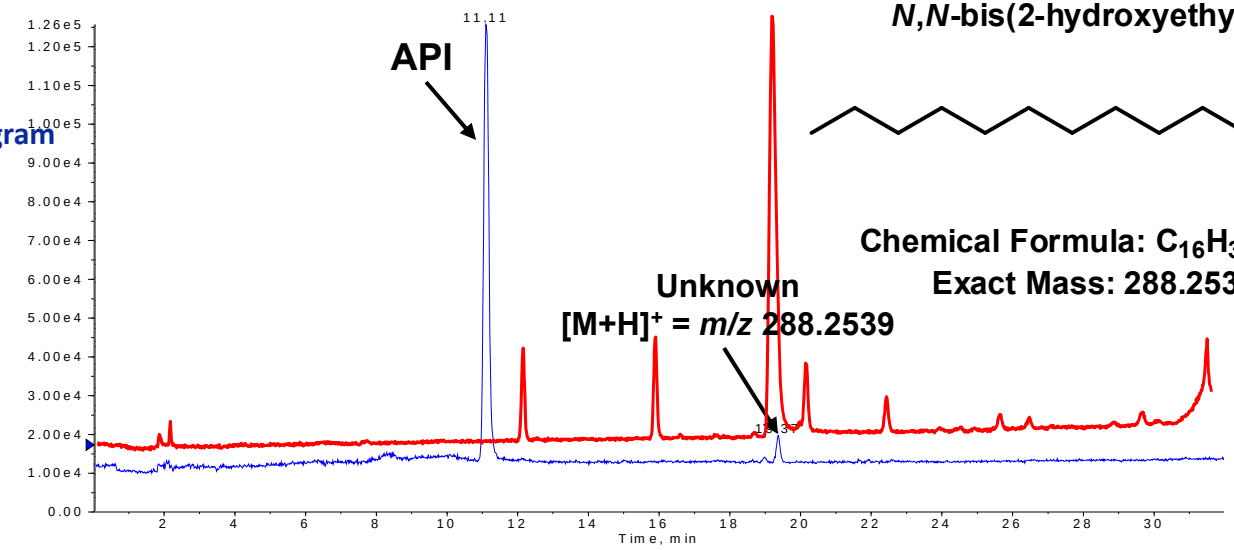


Case Study #1

- API powder on accelerated stability (40 °C, 75% RH) exhibited a previously unidentified impurity peak at 0.3% when assayed by HPLC/UV with related substances method
- Accurate mass data indicated a proposed formula of $C_{16}H_{33}NO_3$ and MS/MS data were indicative of an long-chain hydroxylated amide
- Materials composition list for the packaging material was obtained from the vendor, revealed the presence of a commercial dodecanamide anti-static agent incorporated into the film at 0.5%
- A sample of the anti-static agent was acquired and confirmed identity

Case Study #1

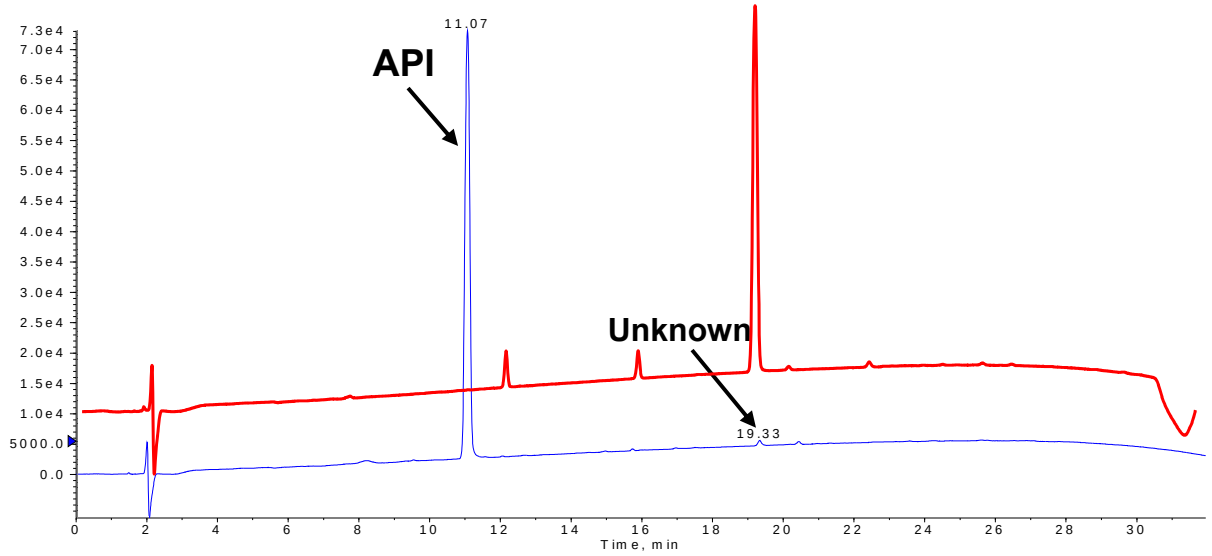
Total ion chromatogram
(*m/z* 80-1500)



N,*N*-bis(2-hydroxyethyl)dodecanamide

Chemical Formula: $C_{16}H_{34}NO_3^+$
Exact Mass: 288.2533

PDA chromatogram
(200-400 nm)



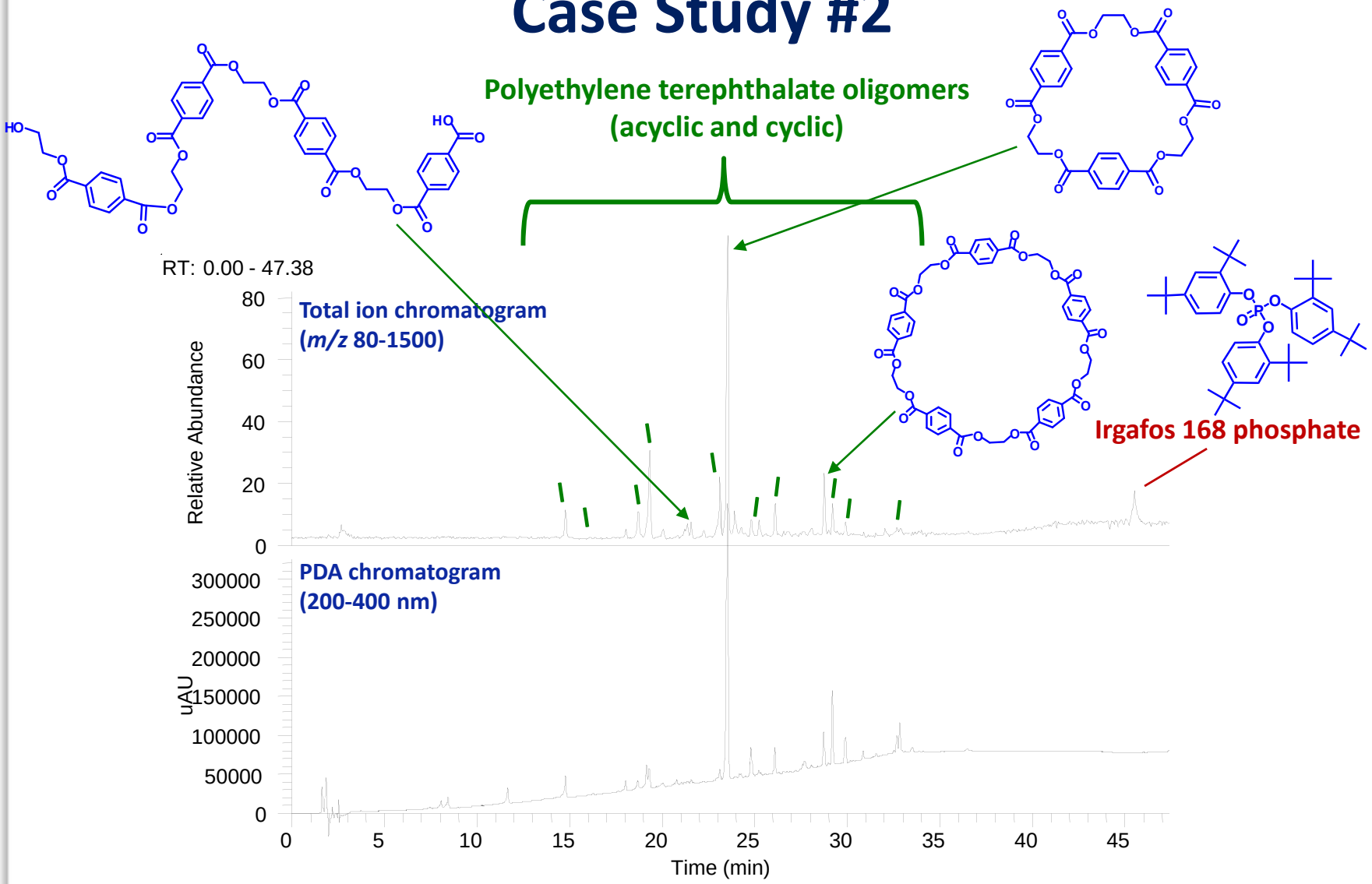
Case Study #1

- **Anti-static compound N,N-bis(2-hydroxyethyl)dodecanamide apparently migrating from packaging film into the API at the contact interface**
- **An alternate packaging film was available without incorporated anti-static compounds**
- **Utilization of the alternate packaging material resulted in no additional impurity peaks**

Case Study #2

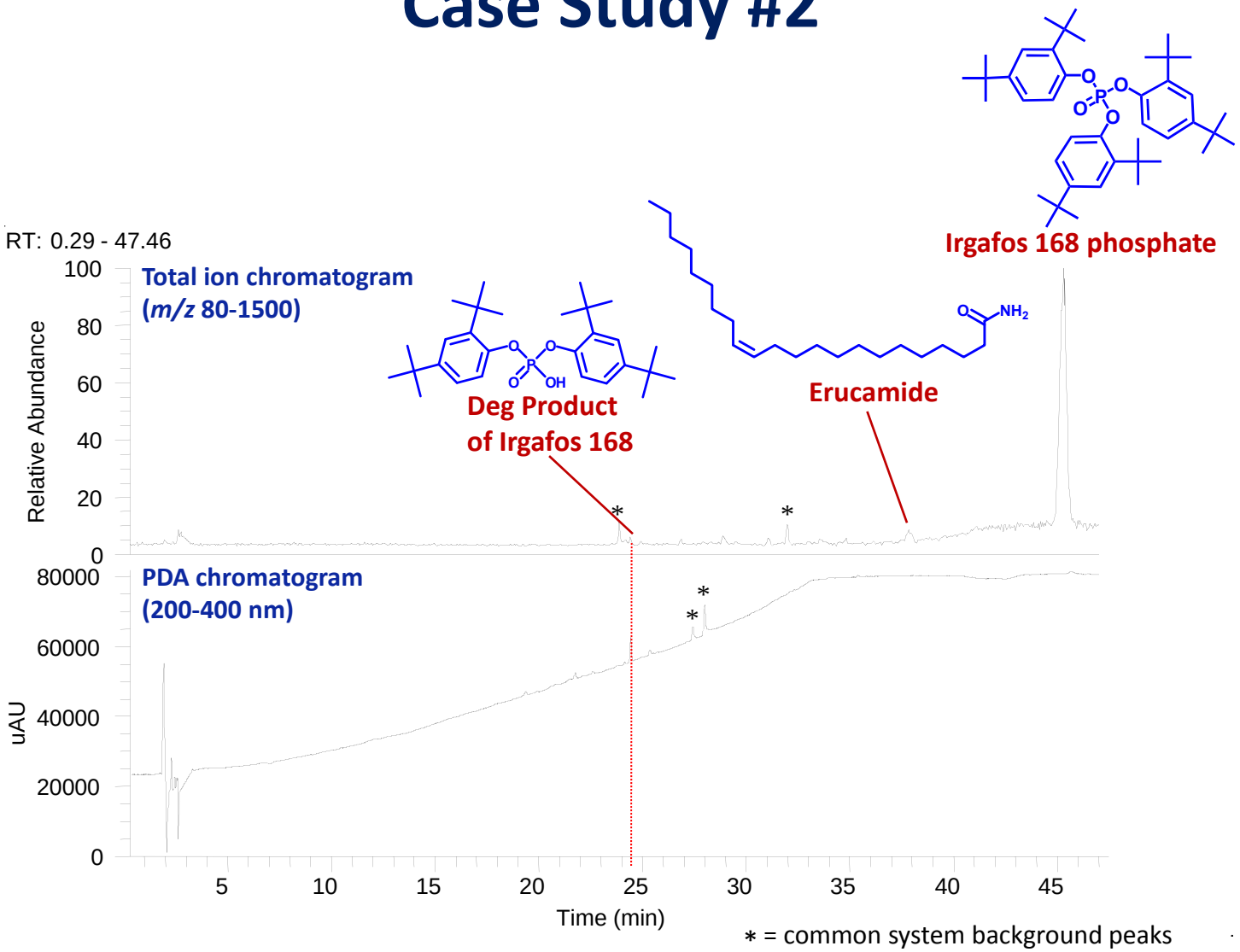
- Polyethylene terephthalate glycol (PETG) bottles and corresponding HDPE caps (no liner) were exposed to IPA at 50 °C for 14 days
- GC-MS and HPLC/PDA analysis indicated a number of unknown compounds in the IPA extracts
- IPA solution was submitted to the Characterization MS lab for evaluation of unknown compounds by Orbitrap LC-MS
- PETG bottles: accurate mass data confirmed the presence of a commercial antioxidant. The majority of compounds observed represent cyclic and acyclic PETG oligomers.
- HDPE caps: accurate mass data confirmed the presence of a commercial antioxidant and a related degradation product, as well as erucamide (a slip agent)

Case Study #2



PETG bottles, IPA extract

Case Study #2



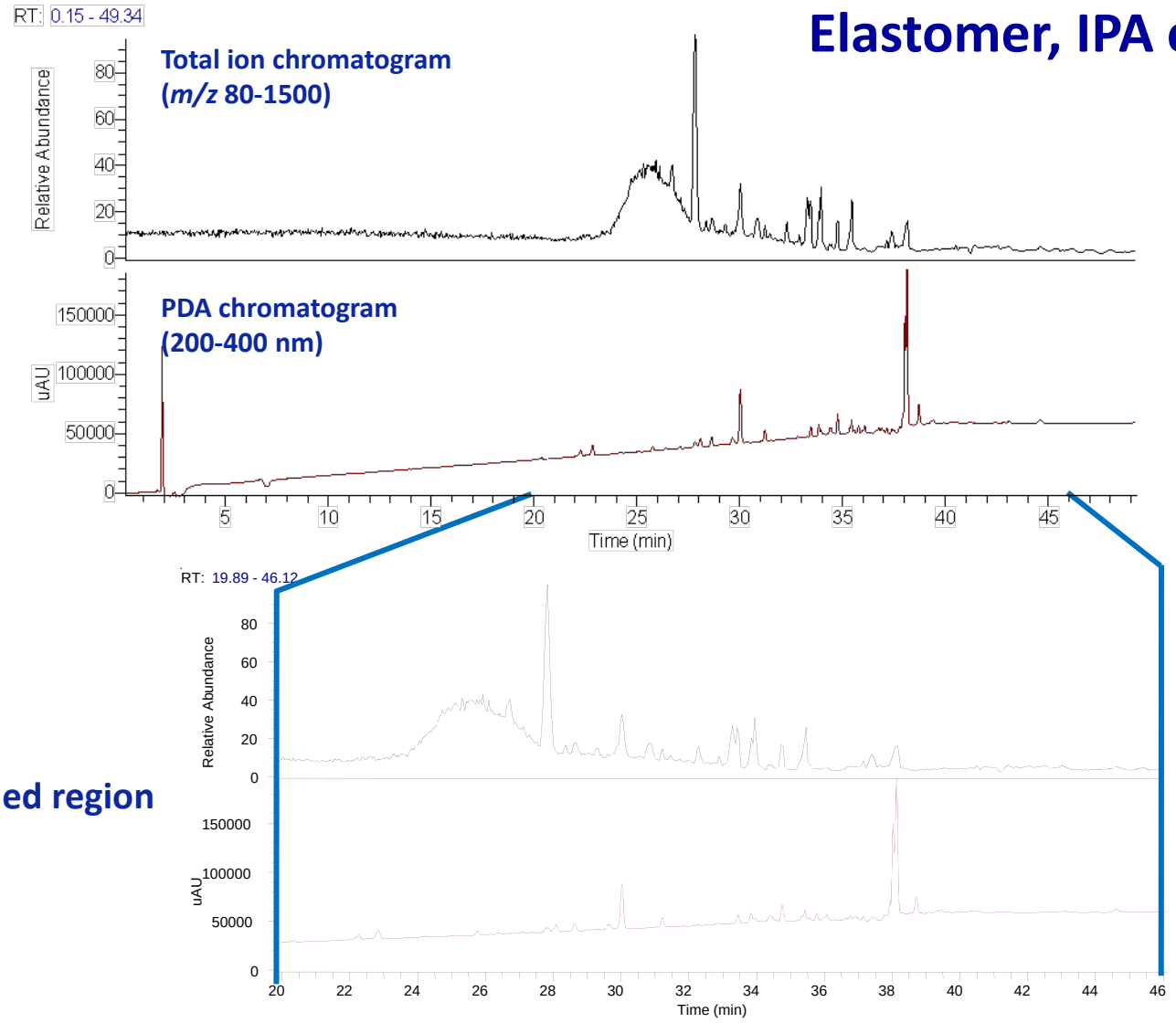
HDPE caps, IPA extract

Case Study #3

- Elastomer material under evaluation for fabrication of drug delivery device components
- Coupons of elastomer were refluxed for 60 min in IPA and H_2O
- The IPA and H_2O extracts were evaluated using FT-ICR LC-MS
- Compounds characterized in the IPA extracts included several octylsulfinylmethylphenol and other antioxidans and stabilizers, aminocrotonic acid esters, and a series of alkyl-substituted PEG homologs
- The H_2O extracts were did not reveal the presence of any detectable extractable compounds, and were consistent with blank H_2O injections

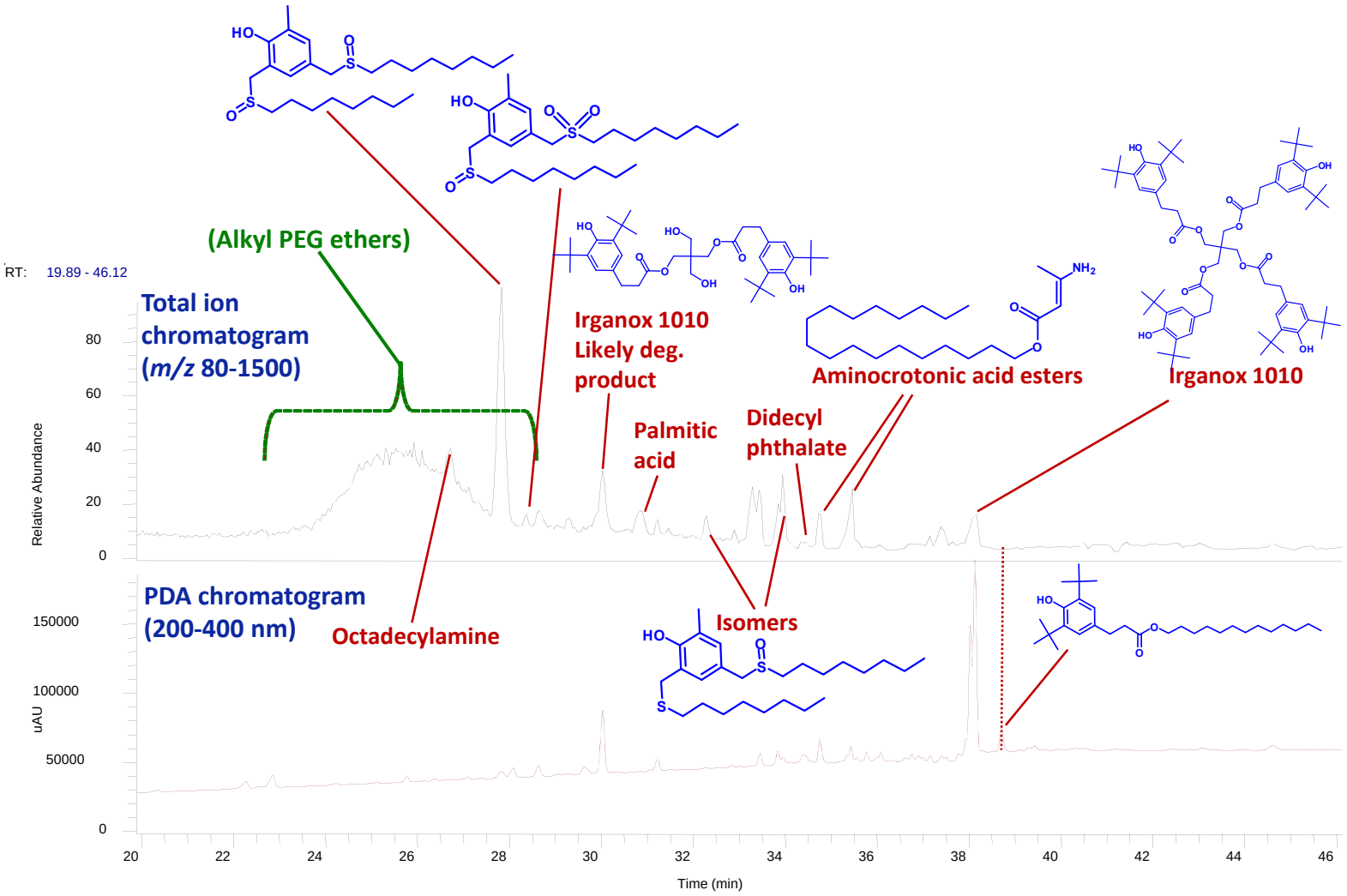
Case Study #3

Elastomer, IPA extract



Expanded region

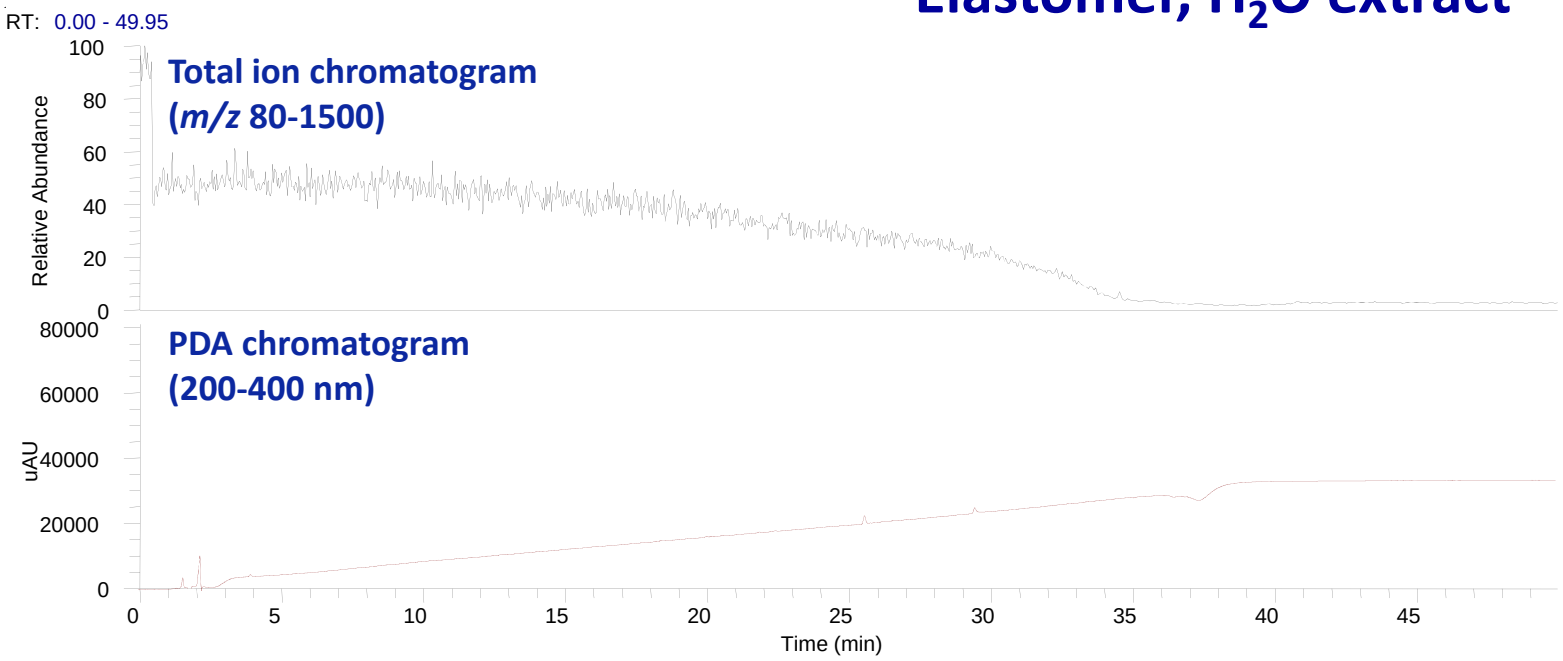
Case Study #3



Elastomer, IPA extract

Case Study #3

Elastomer, H_2O extract

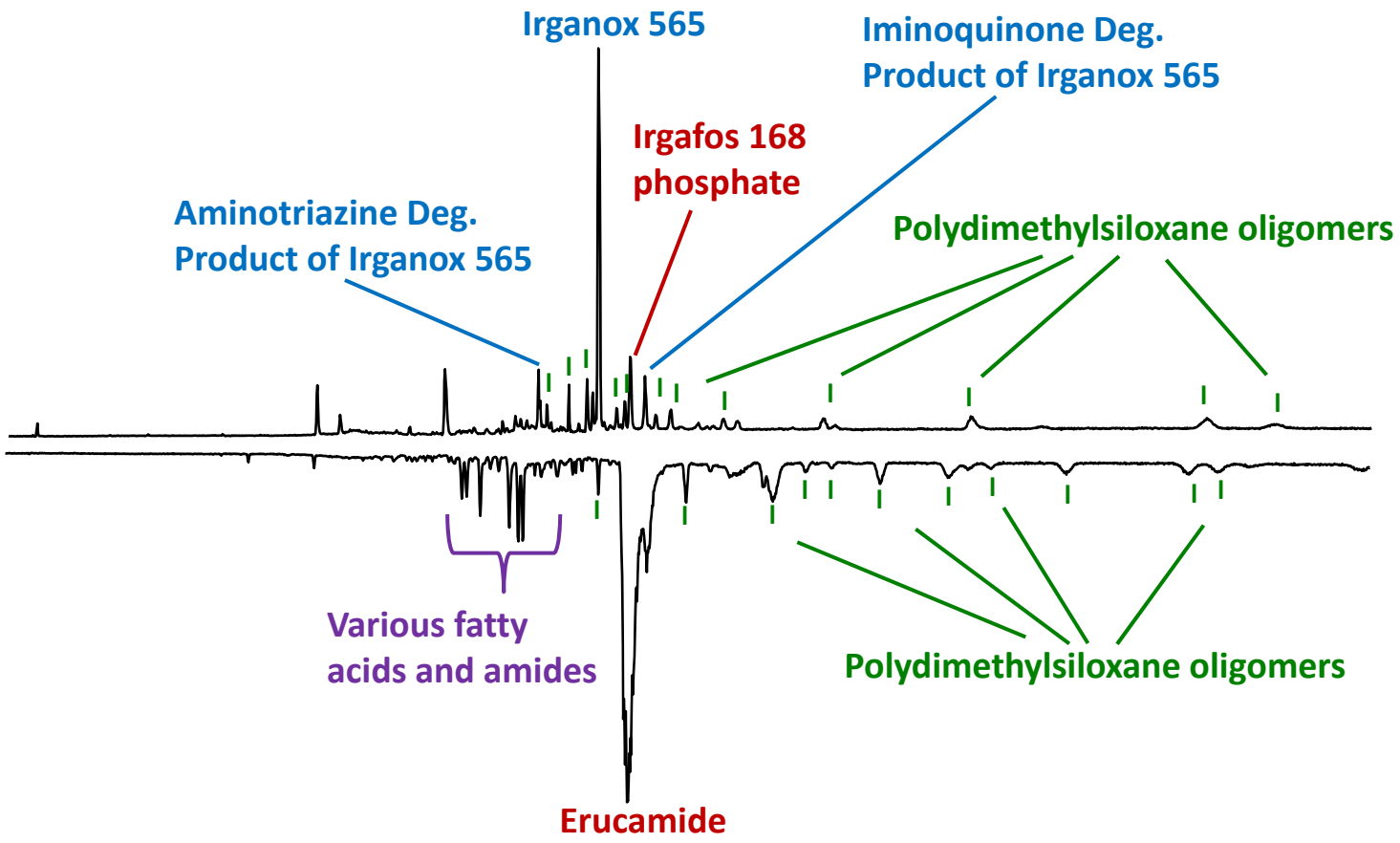


Case Study #4

- Qualitative evaluation of 60 mL polypropylene syringes from two sources for any apparent/overt differences in plunger lubricant and barrels
- Syringes were extracted with CH_2Cl_2, extracts evaluated using FT-ICR LC-MS
- Both syringes revealed presence of PDMS oligomers indicative of silicone oil
- One syringe indicated presence of large amount of erucamide and various fatty acids and amides
- The other syringe revealed presence of two commercial antioxidant additives, as well as related degradation products
- Information permitted a quick, high-level decision regarding syringe choice before commencing a formal extractables evaluation

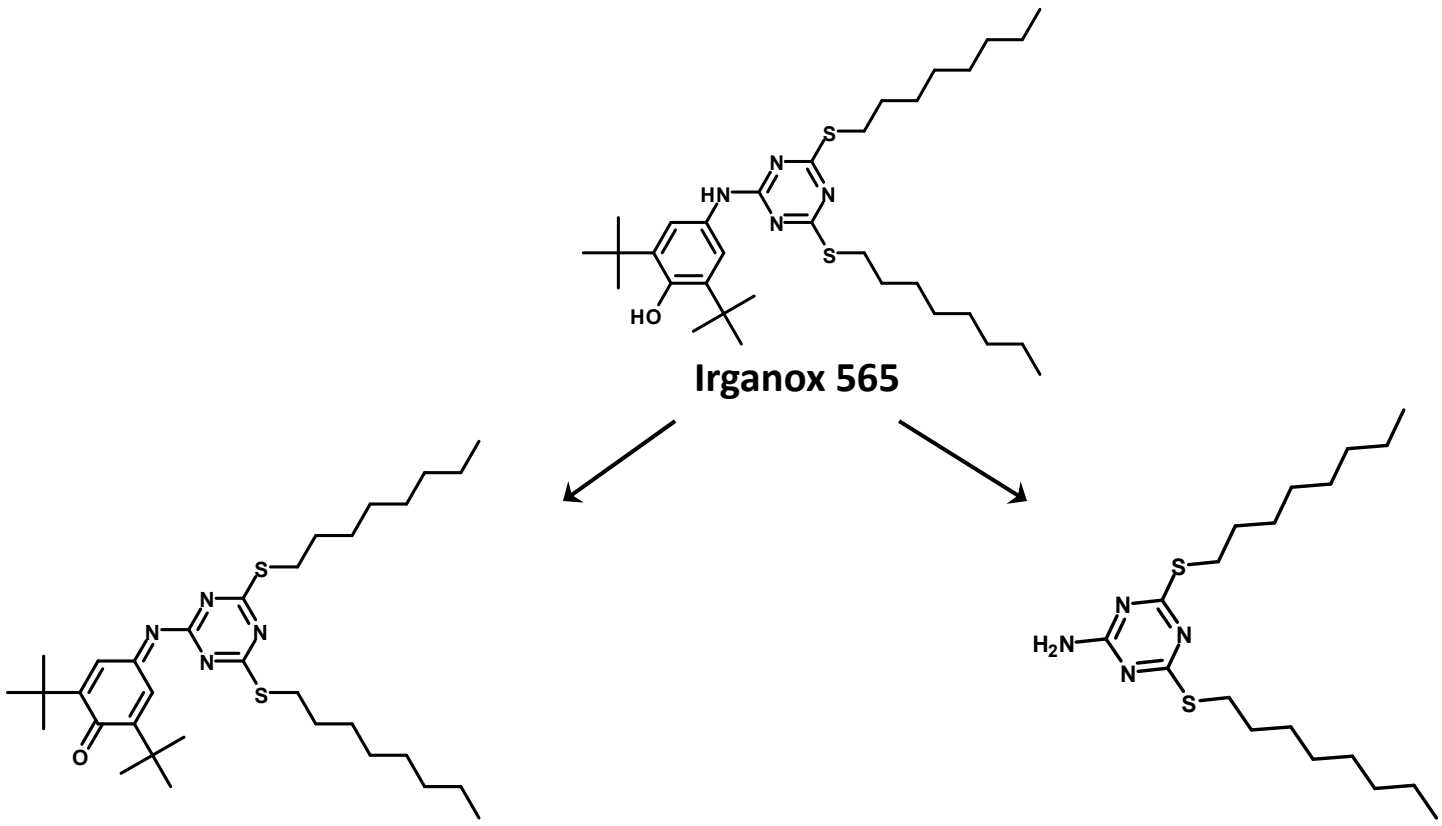
Case Study #4

FT-ICR LC-MS total ion chromatograms (m/z 80-1500) of combined CH_2Cl_2 extracts of polypropylene syringes from two different sources



Case Study #4

Irganox 565 and observed degradation products



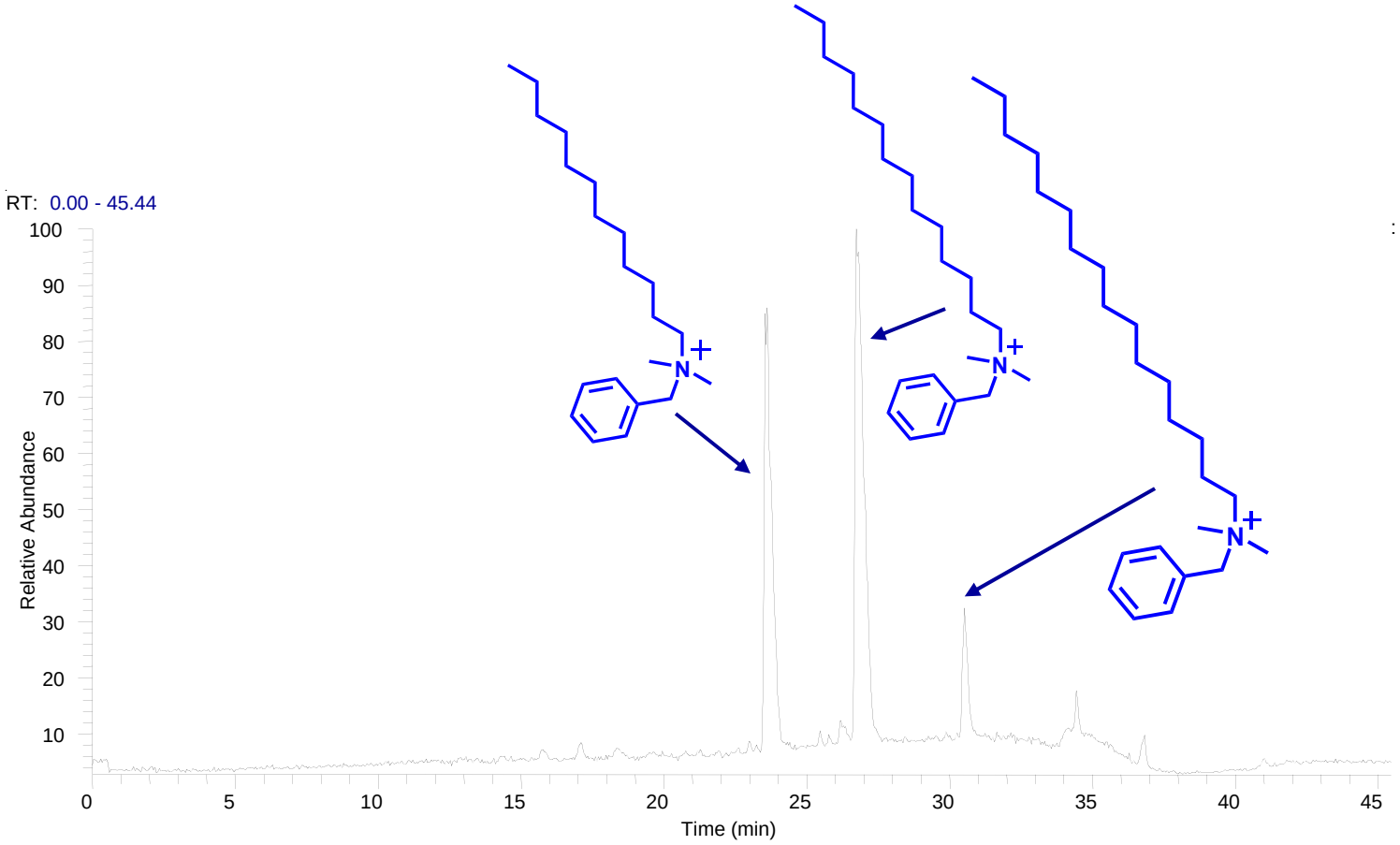
Allen, David W.; Leathard, David A.; Smith, Christine, Chemistry & Industry (1989), 2, 38-9

Case Study #5 (Residual Impurities)

- Inspection of a high flow filtration unit for bioproduct processing revealed the presence of an unidentified precipitate
- Accurate mass data (FT-ICR LC-MS) and MS/MS fragmentation for the precipitate were consistent with a homologous series of benzalkonium surfactant compounds, common in many commercial disinfectant solutions
- Materials composition list and MSDS for all components in the system were requested from vendors. A series of high-flow cartridge filters were found to be shipped in a solution containing approx. 0.1% benzalkonium chloride (C12-C16 chain length)
- Prescribed pre-wash procedure from vendor not effective
- Altering pre-wash procedure to ensure complete removal of the benzalkonium compounds resolved the issue

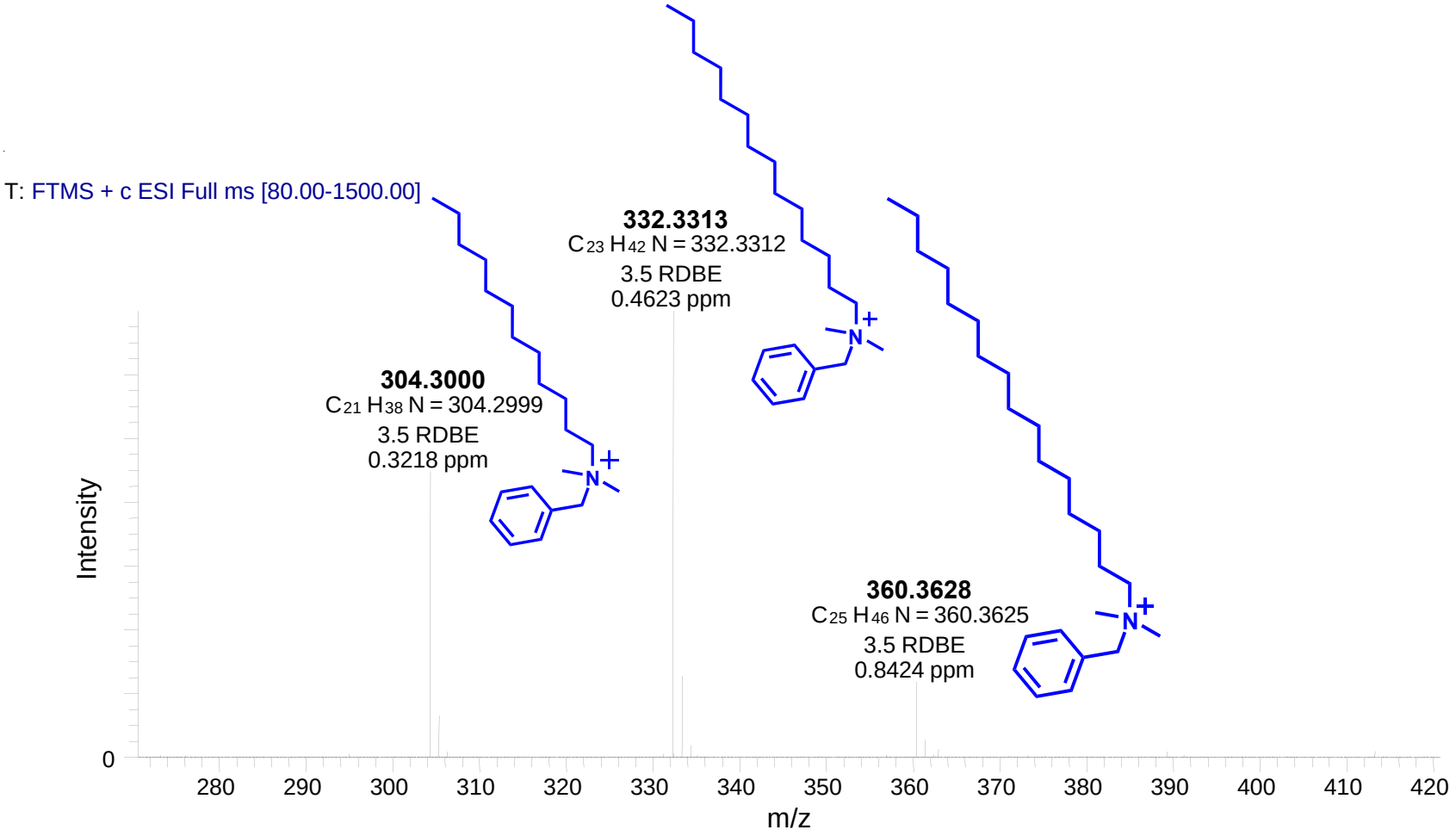
Case Study #5

FT-ICR LC-MS total ion chromatogram (m/z 80-1500) of unknown precipitate in ACN



Case Study #5

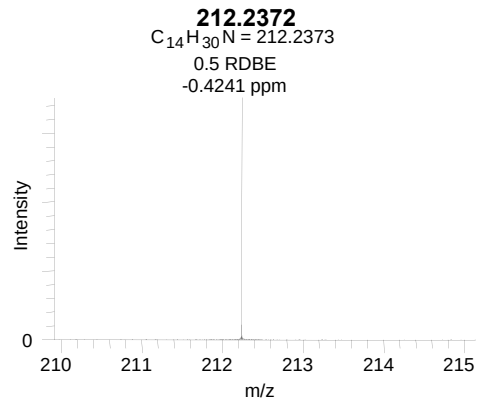
ESI FT-ICR full scan mass spectrum of unknown precipitate in ACN

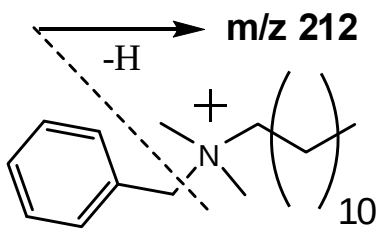


Case Study #5

ESI FT-ICR MS^2 mass spectra of unknown precipitate in ACN

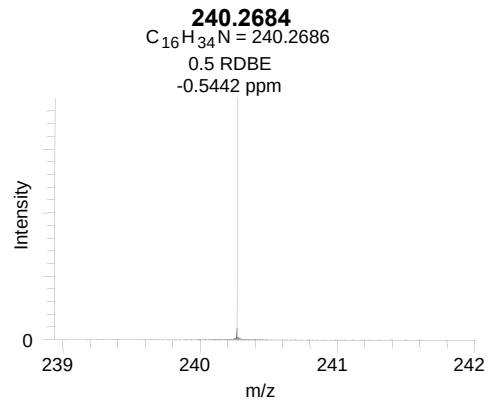
FTMS + c ESI d Full ms2 304.30@cid35.00 [70.00-315.00]

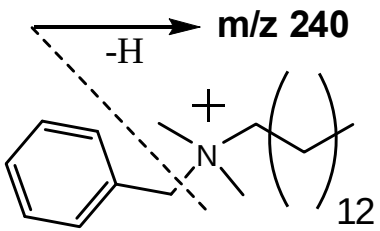




m/z 304.3

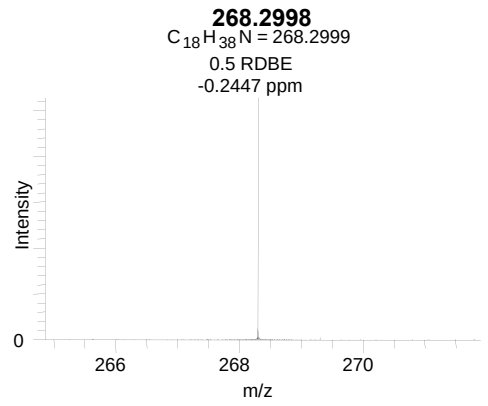
FTMS + c ESI d Full ms2 332.30@cid35.00 [80.00-345.00]

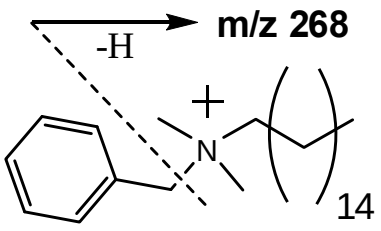




m/z 332.3

FTMS + c ESI d Full ms2 360.30@cid35.00 [85.00-375.00]





m/z 360.3

Conclusions

- ✓ A key step in an extractables/leachables assessment is the establishment of a comprehensive extractable compound profile for materials and components
- ✓ Characterization of unknown extractable compounds, particularly if present in complex mixtures of organic molecules, is not a trivial exercise and often requires sophisticated capabilities, deep expertise, and knowledge of material and component composition and history
- ✓ Extractable and leachable assessment of given materials represents a dynamic, rather than static target since development and use of new polymers, elastomers, processing agents and additives, as well as upstream changes, affect lot-to-lot material composition and properties
- ✓ Establishment of comprehensive, accessible exact mass spectral libraries will aid greatly with screening polymer and elastomer extracts for common additives and extractables (creation of a comprehensive library via a Lilly-Thermo Scientific collaboration is currently in progress using Q-Exactive technology)

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Thank you!

Back-up slides

Definitions - Thresholds

- **Safety Concern Threshold (SCT): Total Daily Intake (TDI) threshold below which a leachable present negligible safety concerns from carcinogenic and noncarcinogenic toxic effects**
- **Analytical Evaluation Threshold (AET): Threshold at or above which extractables or leachables need to be identified, quantitated, and reported for a toxicological assessment**
- **Qualification Threshold (QT): Threshold below which a given non-carcinogenic leachable would present negligible safety concerns, unless the leachable presents structure-activity relationship (SAR) concerns**

$$\text{AET } \mu g/ml = \frac{\text{SCT } \mu g/day}{\text{Dose ml/day}}$$