# Considerations for Residual Solvent Analysis – USP Method 467

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# The Role of Solvents?

- They may be critical to the synthetic process:
  - Enhance yields
  - Improve crystallization
  - Increase solubility
- The list of regulated solvents will most likely grow
  - Improved toxicological testing
  - New, unknown toxic affects

# <467> Organic Volatile Impurities

- Solvents classified by risk assessment
  - Class 1: Solvents to be avoided
  - Class 2: Solvents to be limited
  - Class 3: Solvents with low toxic potential
- Drug formulations containing these solvents must be tested
- Only the solvents used or produced in the manufacturing and/or purification process must be evaluated

# <467> Methodology Overview

- Drug product is dissolved in solution
  - Water Soluble Articles: Water
  - Water Insoluble Articles: DMF, DMI
- Headspace injection
- GC analysis with FID detection
  - Procedure A: G43 (ZB-624) Phase
  - Procedure B: G16 (ZB-WAX*plus*) Phase

# GC Conditions

**Column:** Zebron ZB-624

**Dimensions:** 30 meters x 0.32 mm x 1.80 µm

**Order No:** 7HM-G005-31

**Injection:** Split 5:1 1 mL @ 140 °C

**Oven Profile:** 40°C for 20min to 240°C at 10°C/min for 20min

**Carrier Gas:** Constant Flow Helium, 35 cm/sec

**Detection:** Flame Ionization @ 250 °C

# Class 1 Solvents

- Testing required
- Solvent levels must meet *Concentration Limit*

| Solvent | Concentration Limit (ppm) | Concern |
|---|---|---|
| Benzene | 2 | Carcinogen |
| Carbon tetrachloride | 4 | Toxic and Environmental Hazard |
| 1,2-Dichloroethane | 5 | Toxic |
| 1,1-Dichloroethene | 8 | Toxic |
| 1,1,1-Trichloroethane | 1500 | Environmental Hazard |

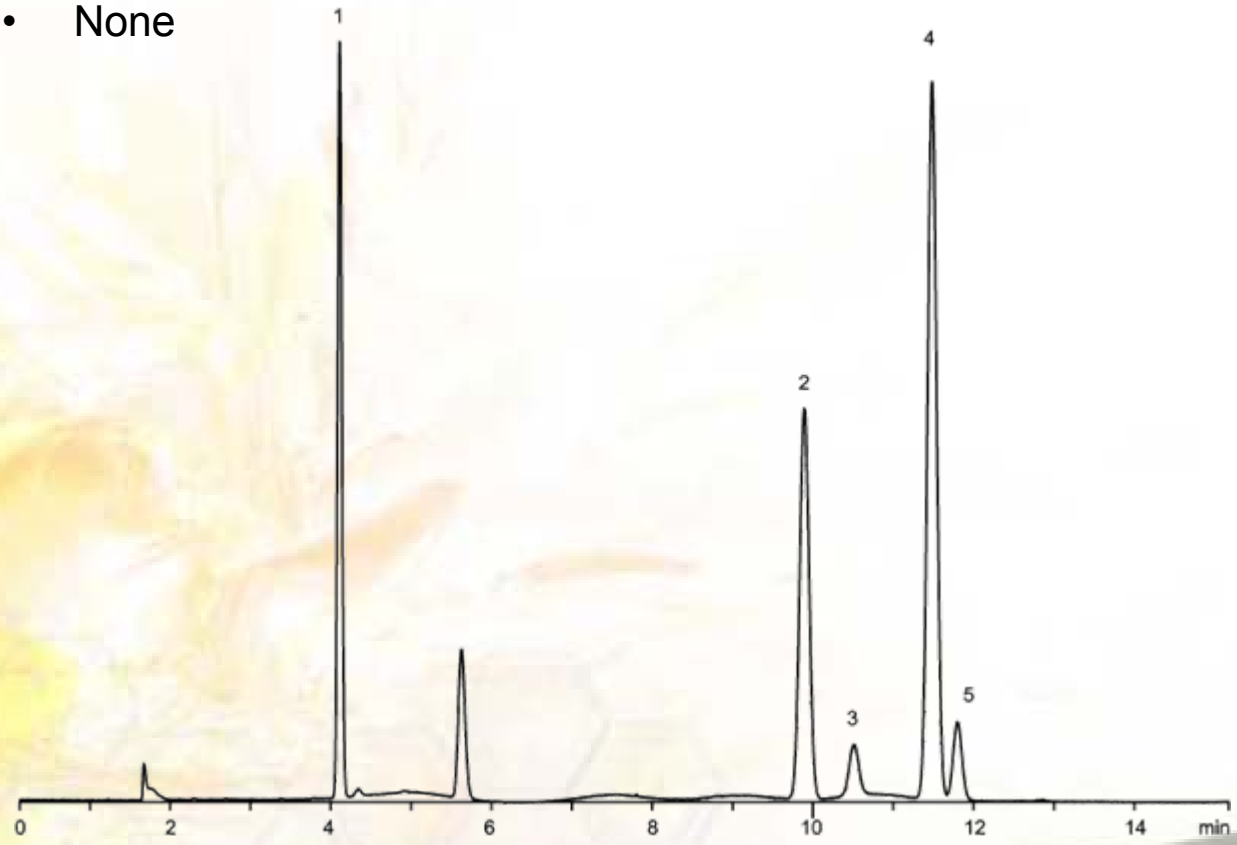
# Class 1 Solvents As Impurities

- Class 1 solvents are highly regulated
- *Concentration Limit* requires very low level
- These levels may be present in Class 2 or 3 solvents as impurities
- Example: Toluene possible impurity – Benzene
- Cross contamination possible

# Class 1:
# ZB-624 30m x 0.32mm x 1.80µm

**Co-elution's:**

- None



# Class 2 Solvents

- Solvents are assigned a permitted daily exposure (PDE) limit
- Each solvent is assigned a *Concentration Limit* allowable in any component of the drug product

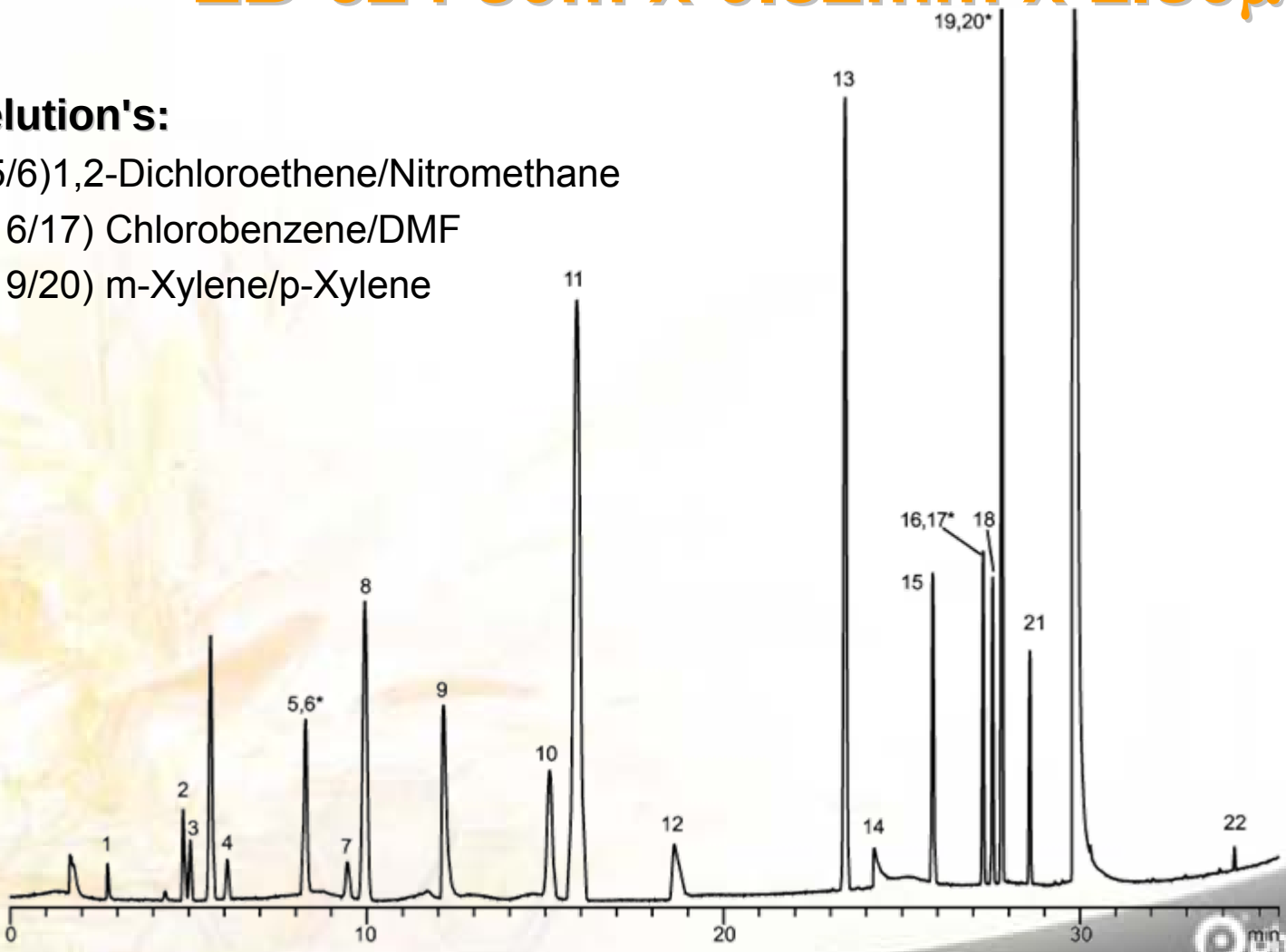
**Example 1:**

| | PDE (mg/day) | Concentration Limit (ppm) |
|---|---|---|
| Acetonitrile (ACN) | 4.1 | 410 |

# Class 2:
# ZB-624 30m x 0.32mm x 1.80µm

**Co-elution's:**

- (5/6)1,2-Dichloroethene/Nitromethane
- (16/17) Chlorobenzene/DMF
- (19/20) m-Xylene/p-Xylene



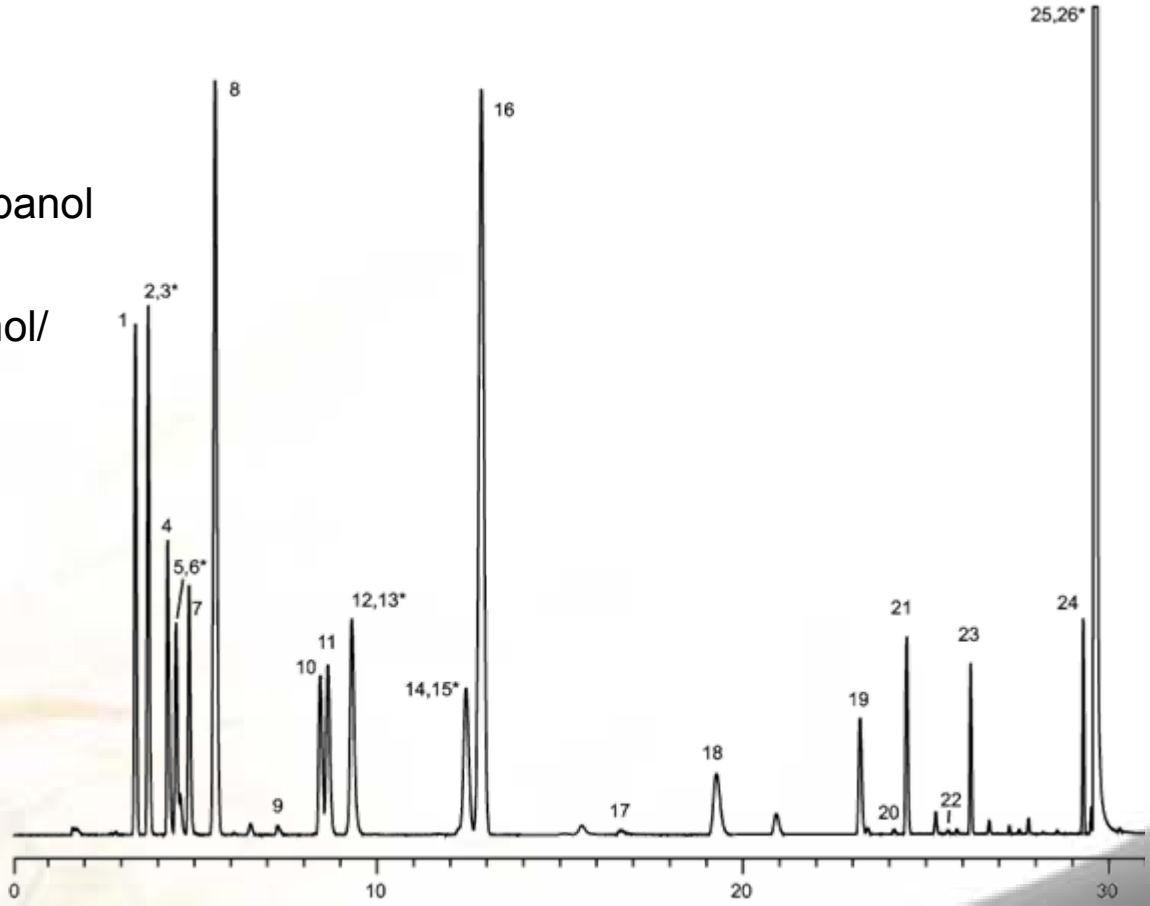
# Class 3 Solvents

- If only Class 3 solvents are present…
- Solvent level is determined by <731> *Loss on Drying*
- The **PDE** limit, unless otherwise specified, is 50mg/day or 5,00ppm
- If solvent level is above the **PDE** limit, it must be identified and quantified

# Class 3:
# ZB-624 30m x 0.32mm x 1.80µm

**Co-elution's:**

- (2/3) Ethanol/Ethyl Ether
- (5/6) Ethyl Formate/2-Propanol
- (12/13) THF/2-Butanol
- (14/15) 2-Methyl-1-propanol/ Isopropyl Acetate
- (25/26) Anisole/DMSO



# Limitations of the Method <467>

- Long analysis time >30min
- Poor resolution of some compounds
- Long wait equilibration time for headspace samples
- Poor detection of some compounds
- No definitive identification of contaminates
  - FID does not give information about the peak

# Single Solvent Method

Fast, Accurate, Sensitive, Definitive

## Starting Point:

1. Injection Techniques
2. Column Phase
3. Analysis Time
4. Detector
5. Example Method

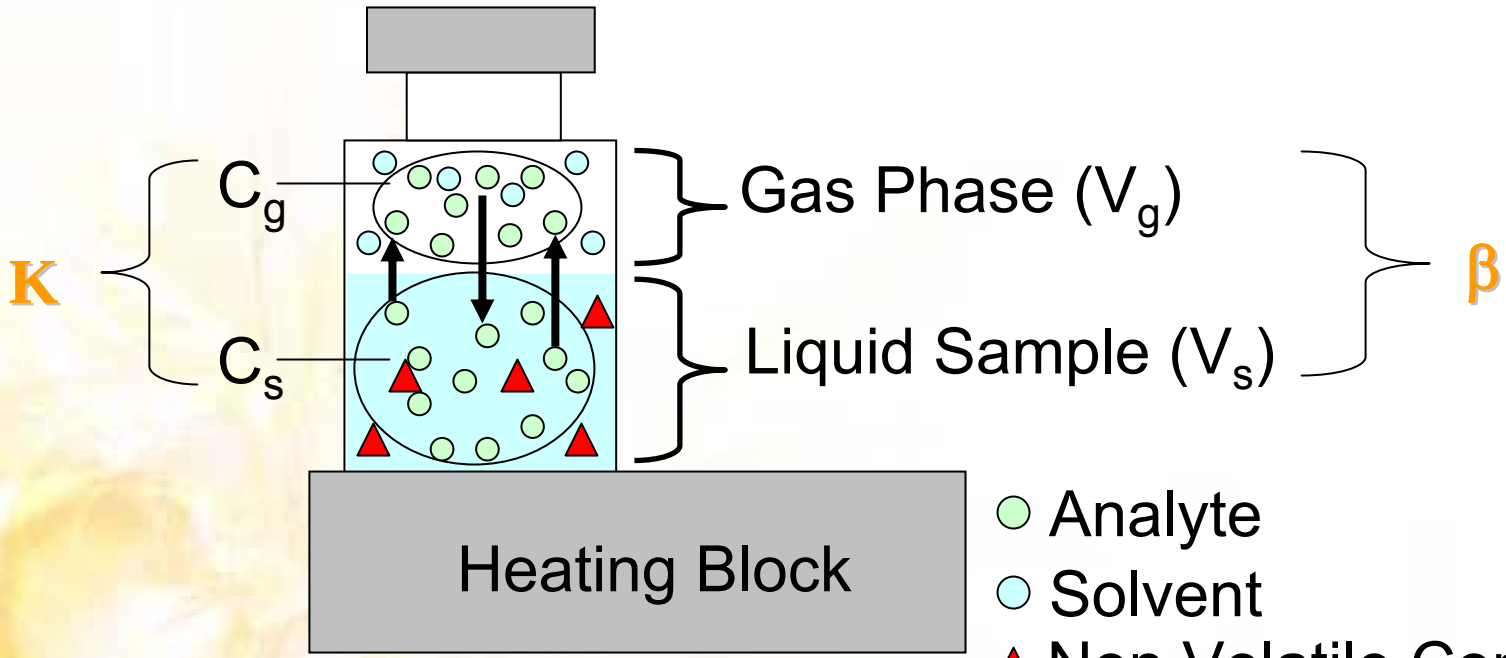
# 1. Injection Techniques

1. Headspace
2. Split

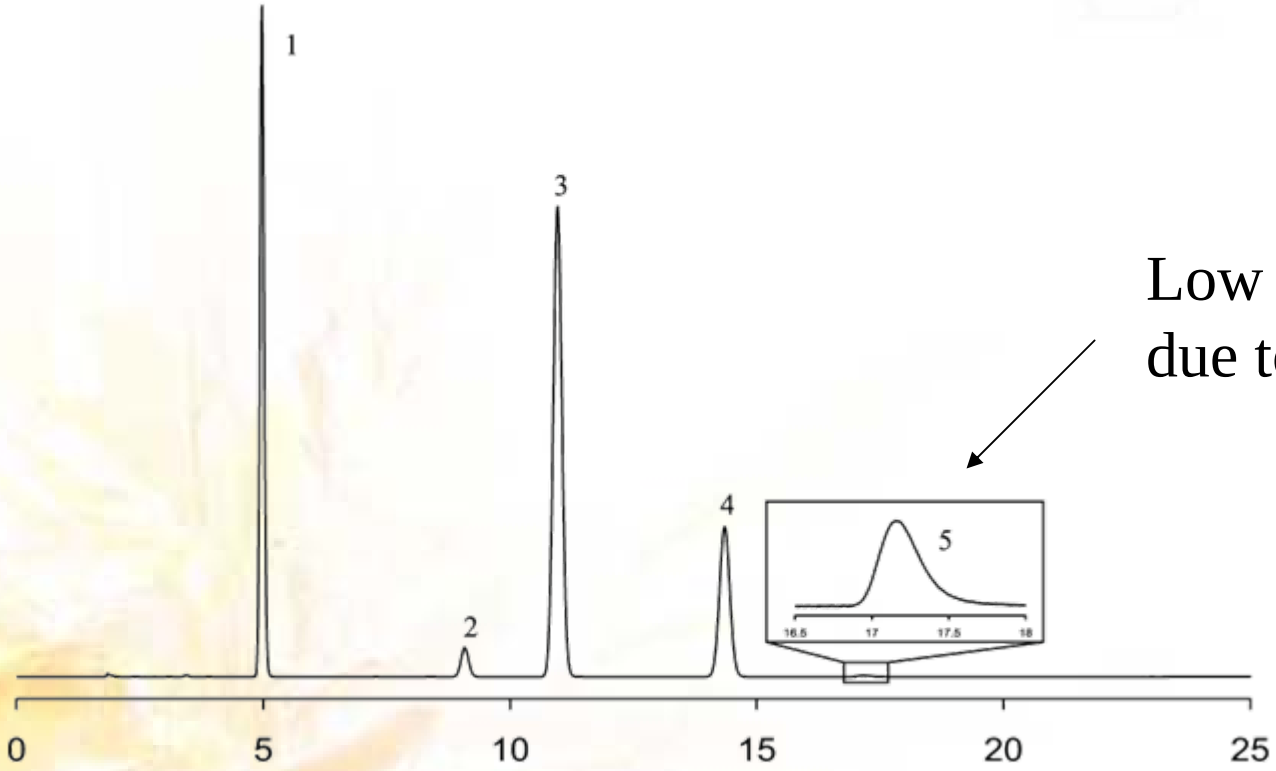
# USP Method 467

- Method specifies headspace
- Reduces matrix interference
  - Most drug formulation products aren't volatile
- Negative: some compounds not detected
  - Class 2: formamide, 2-ethoxyethanol, 2-methoxyethanol, ethylene glycol, N-methylpyrrolidone, and sulfolane

# Headspace Basics



# The Effect of K on Sensitivity



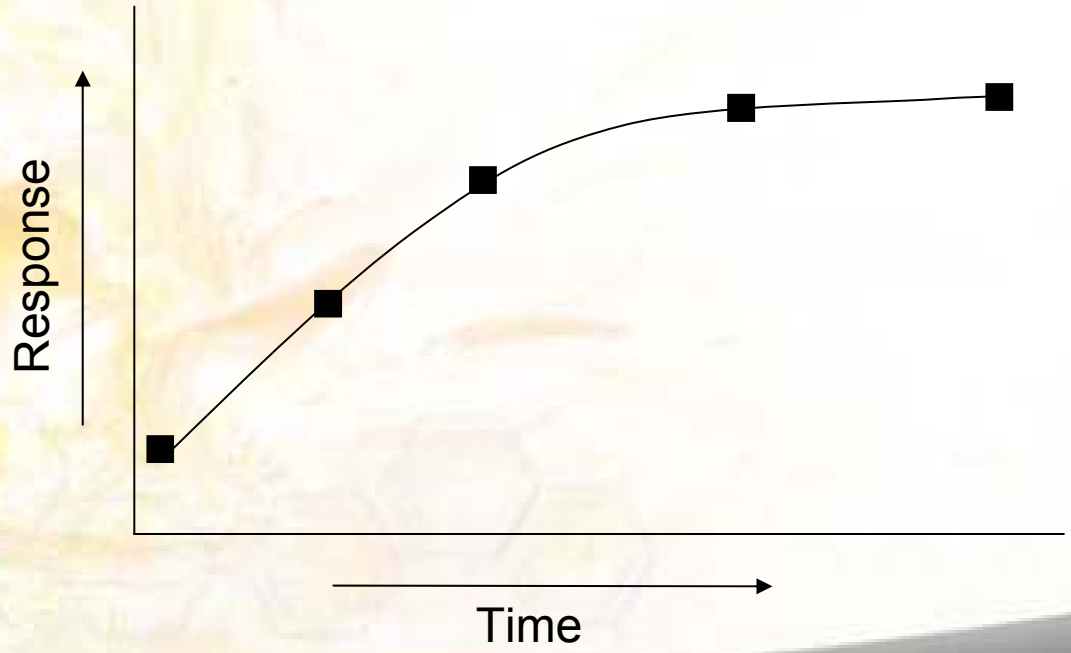

Low response due to high K

| Peak | Solvent | Conc. | K Value* |
|---|---|---|---|
| 1 | Methylene Chloride | 10mg/mL | 5.65 |
| 3 | Benzene | 1mg/mL | 2.9 |
| 5 | 1,4-dioxane | 2mg/mL | 1618 |

**Compounds dissolved in water at 40°C*

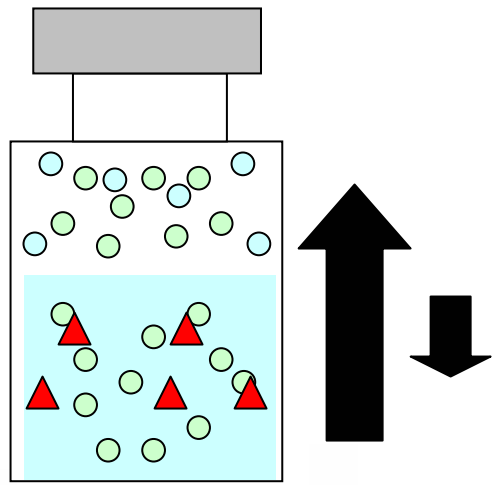
# Equilibration Time

- Equilibration time is unique for sample
- Sample matrix will effect equilibration time
- Ideal time must be determined experimentally



# Optimizing K

- Addition of matrix modifiers:
  - Ammonium Chloride
  - Ammonium Sulfate
  - Sodium Chloride
  - Sodium Citrate
  - Sodium Sulfate
  - Potassium Carbonate
- Change the dilution solvent
- Increase vial temperature



# Headspace Systems

## Gas-Tight Syringe Injection

- Manual
- Autosampler

## Sample Loop Systems

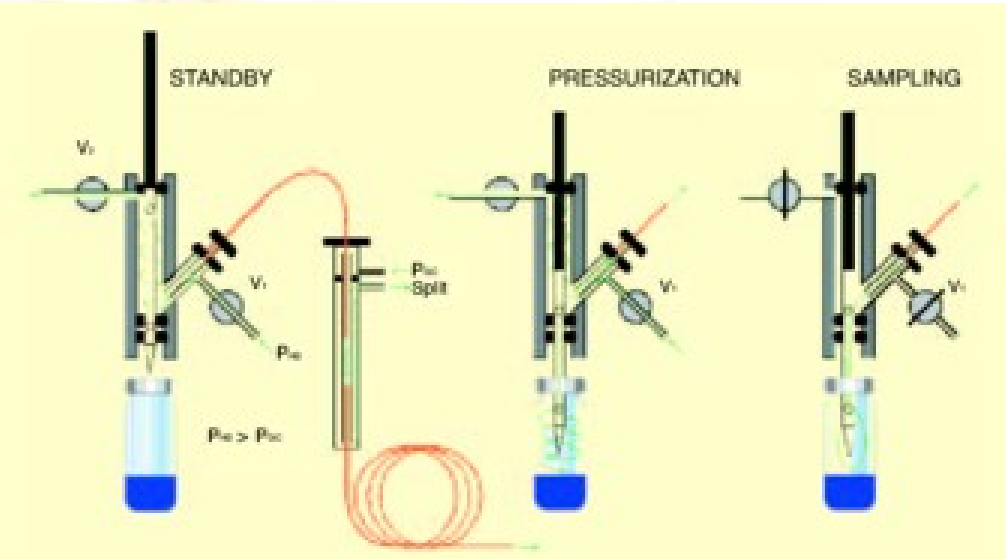
- Pressure-Balanced
- Pressurized Loop

# Syringe Injection Systems

- Available from many different manufacturers
- Sample vials are heated from 35-200°C
- Syringe is heated from 35-150°C (limited by syringe)
- Syringe can be flushed with Nitrogen between runs to reduce carryover

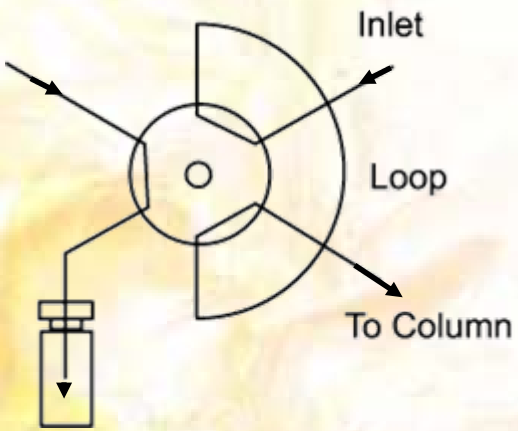
# Balanced Pressure Loop

- Headspace sample introduced without a syringe
  - Reduces fractionation caused by pressure changes
  - Closed system prevents any sample loss
- No valve system

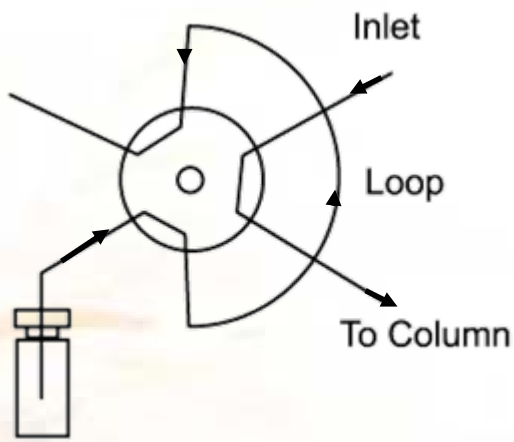


# Pressurized Loop System

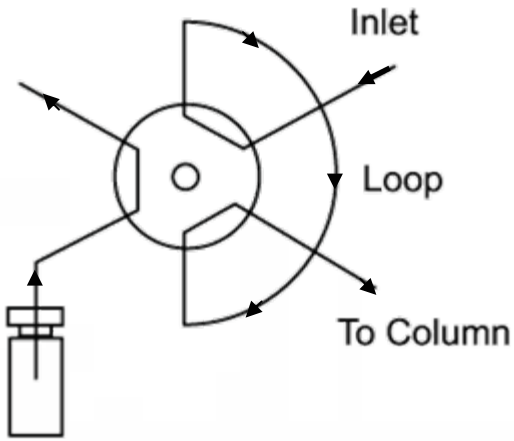
- Sample loop filled with defined amount
- Important to fill sample loop (no partial injections)
- Sample loop can be heated to high temperatures




1. Vial Pressurization



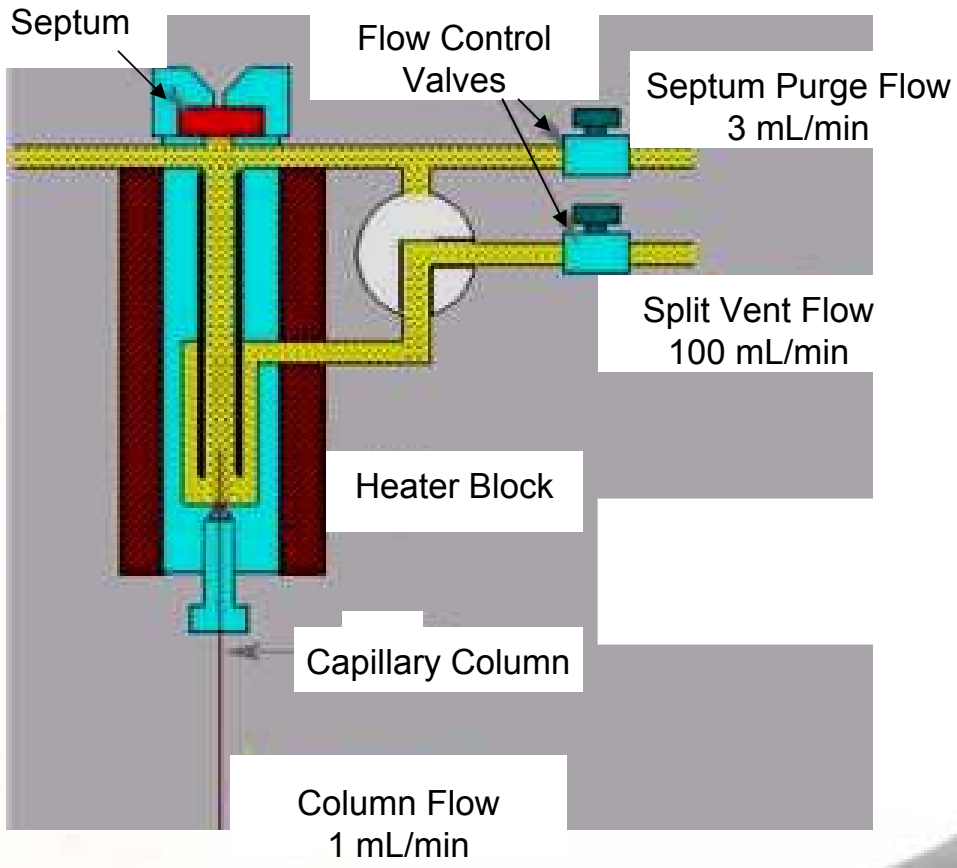

2. Loop Fill




3. Injection

# Split Injection

- Most commonly used
- 1-5 µL injection
- Sample vaporized
- Small fraction enters the column
- Most of sample vapor is vented by the inlet purge stream



# Advantage of Split

- All compounds detected:
  - Class 2: formamide, 2-ethoxyethanol, 2-methoxyethanol, ethylene glycol, N-methylpyrrolidone, and sulfolane
- Simple to operate
- Reduces the amount of contamination on the column

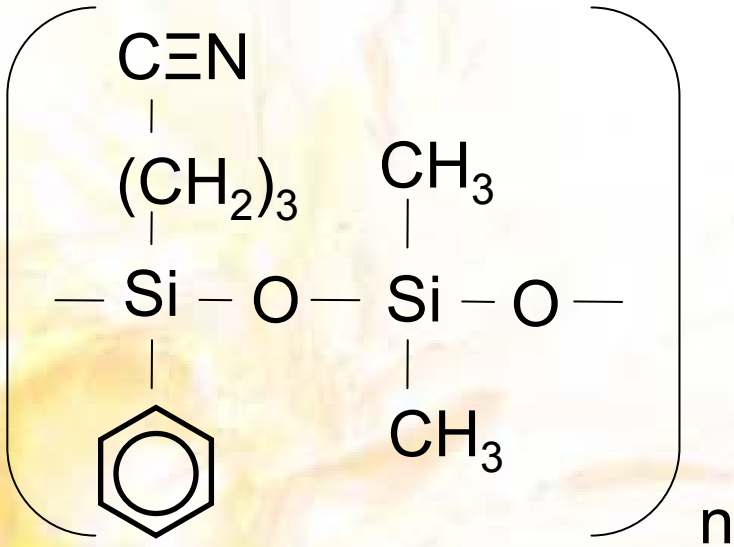
# 2. Column Phases

What is the ideal phase?

- Depends on the goal of the separation…what are the target analytes

# G43 Phase

## Phase Structure




6%-Cyanopropyl-Phenyl - 94% Dimethylpolysiloxane

## Column:

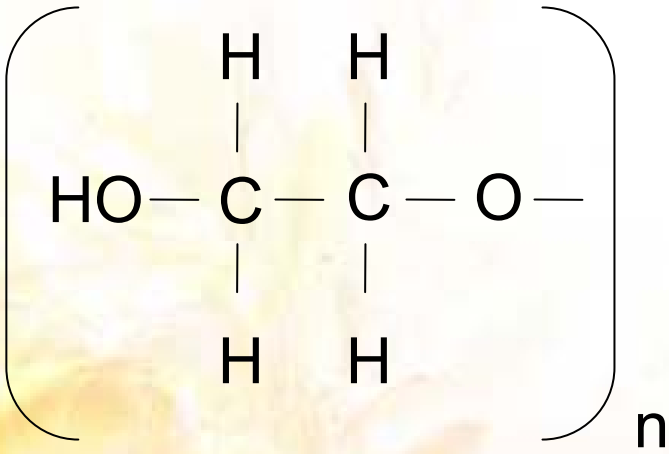
ZB-624

## Retention Mechanism:

- London Dispersion
- Permanent Dipole
- Induced Dipole
- Pi-Pi

# G16 Phase

## Phase Structure




Polyethylene Glycol (PEG)

## Columns:

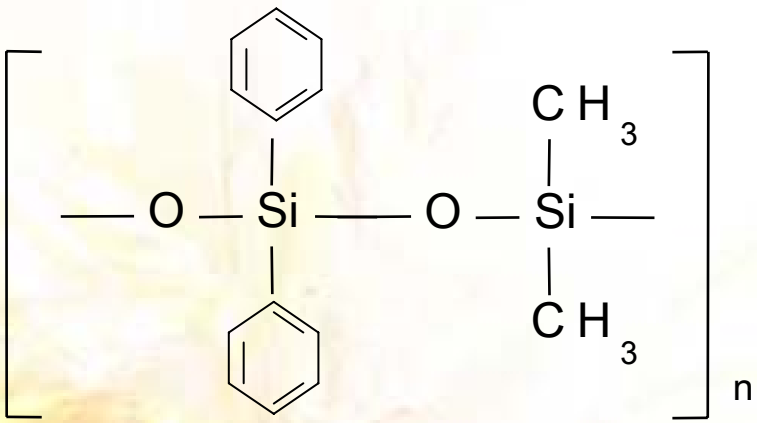
- ZB-WAX
- ZB-WAX*plus*

## Retention Mechanism:

- London Dispersion
- H-Bonding
- Permanent Dipole
- Induced Dipole

# Phenylmethyl Polysiloxane

## Phase Structure




Phenyl Dimethylsiloxane

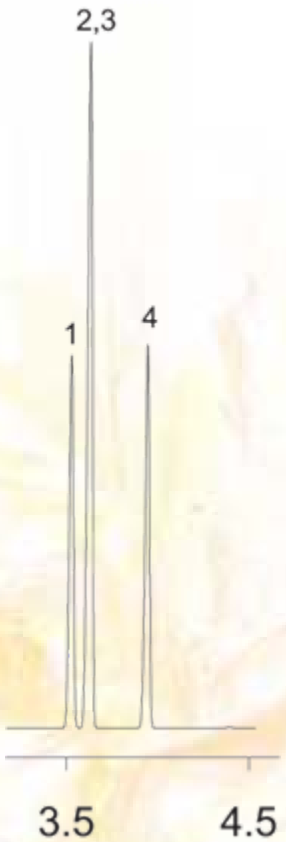
## Columns:

- ZB-5
- ZB-35
- ZB-50

## Retention Mechanism:

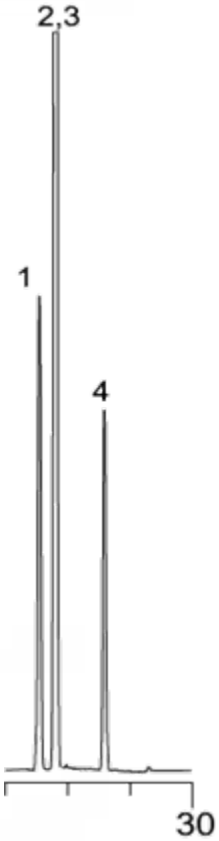
- London Dispersion
- Induced Dipole
- Pi-Pi

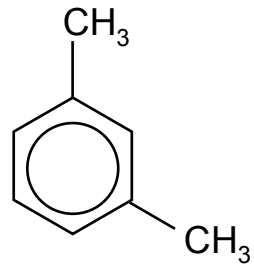
# Selectivity – Xylene Isomers




ZB-5 30m x 0.32mm x 0.25µm

1. Ethylbenzene
2. m-Xylene
3. p-Xylene
4. o-Xylene

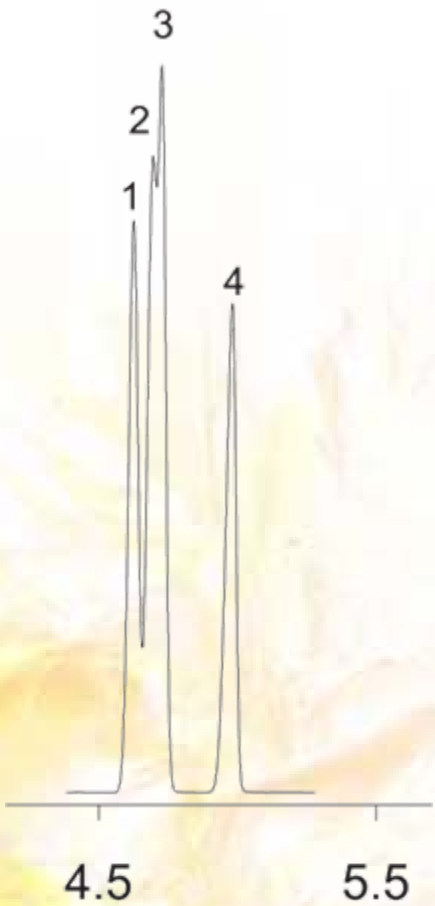



m-Xylene

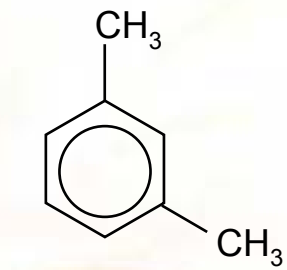
ZB-624 30m x 0.32mm x 1.80µm

# Xylene Isomers Cont.

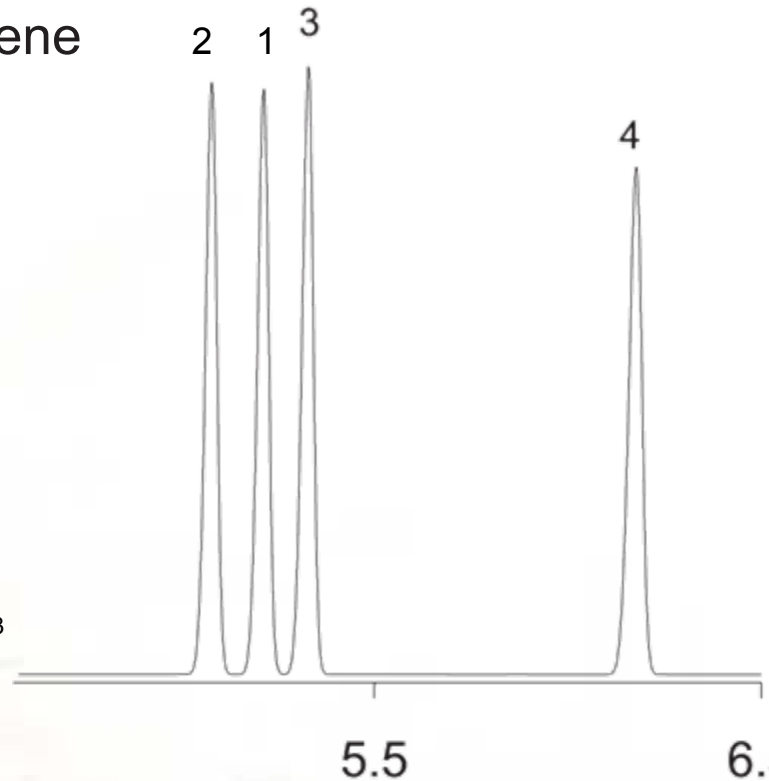



ZB-50 30m x 0.32mm x 0.25µm

1. Ethylbenzene
2. m-Xylene
3. p-Xylene
4. o-Xylene




m-Xylene




ZB-WAX 30m x 0.32mm x 0.25µm

# 3. Analysis Time

As fast as possible with the best accuracy & precision!

# 4. Detector

The USP Method specifies FID

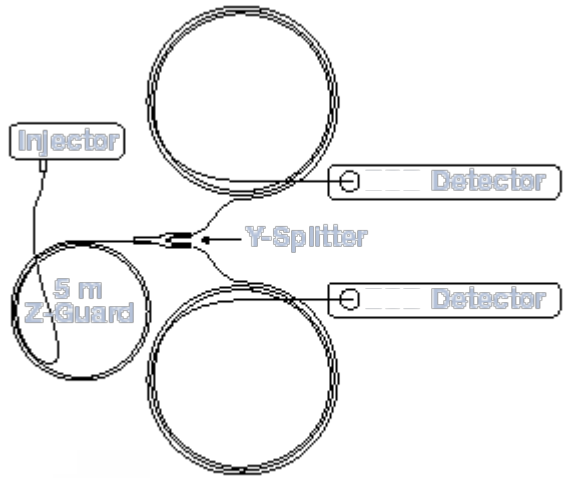
- Benefits:
  - Responds to a wide range of compounds
  - Large dynamic range
  - Inexpensive
  - Stable and easy to use
- Negatives:
  - No information about the analyte
  - Poor response for highly chlorinated compounds

# Mass Spectrometer

- Benefits
  - Spectral confirmation
  - Highly sensitive
  - The “Gold Standard” for many other industries (toxicology, environmental, etc.)
- Negatives
  - Expensive
  - Requires specialized training

# Alternatives

- Dual column GC-FID analysis
- Similar to EPA methodologies
- Two columns of dissimilar polarity used in parallel
- Confirmation is made by having a peak at the specified retention time on each phase

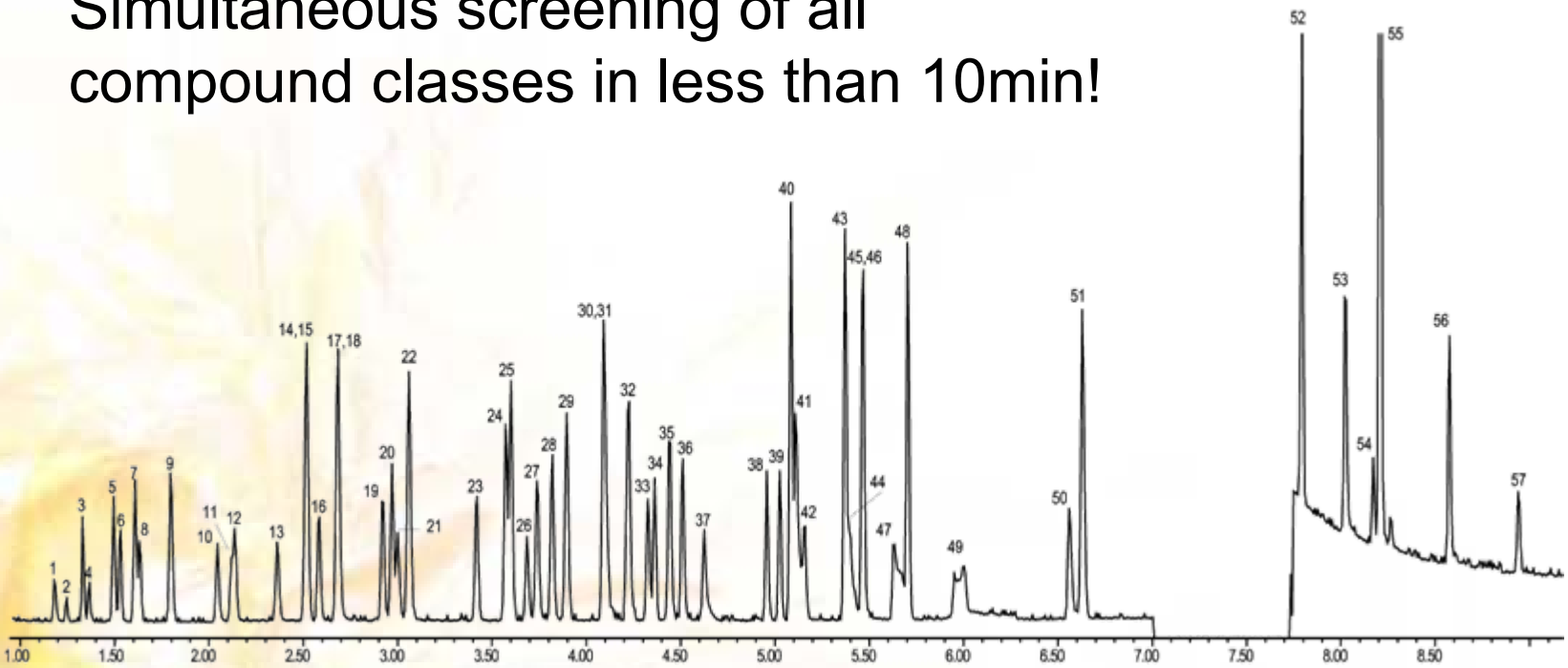


# 5. Example Method

| | |
|---|---|
| **Column:** | Zebron ZB-WAX*plus* |
| **Dimensions:** | 30 meters x 0.25 mm x 0.25 µm |
| **Order No:** | 7HG-G013-11 |
| **Injection:** | Split 50:1 0.2 µL @ 220 °C |
| **Carrier Gas:** | Constant Flow Helium, 1.2 mL/min |
| **Oven Profile:** | 30°C for 1min to 70°C at 14°C/min to 220°C at 25°C/min for 3 min |
| **Detection:** | MSD 18-350 amu @ 250 °C |

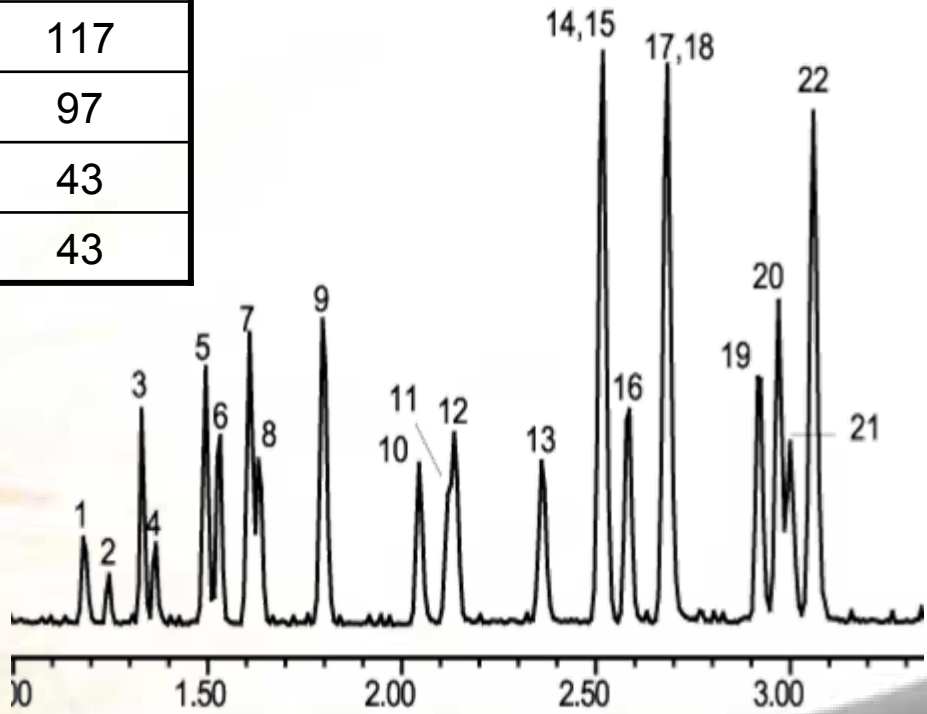
# Class 1, 2, and 3 Solvents

Simultaneous screening of all compound classes in less than 10min!



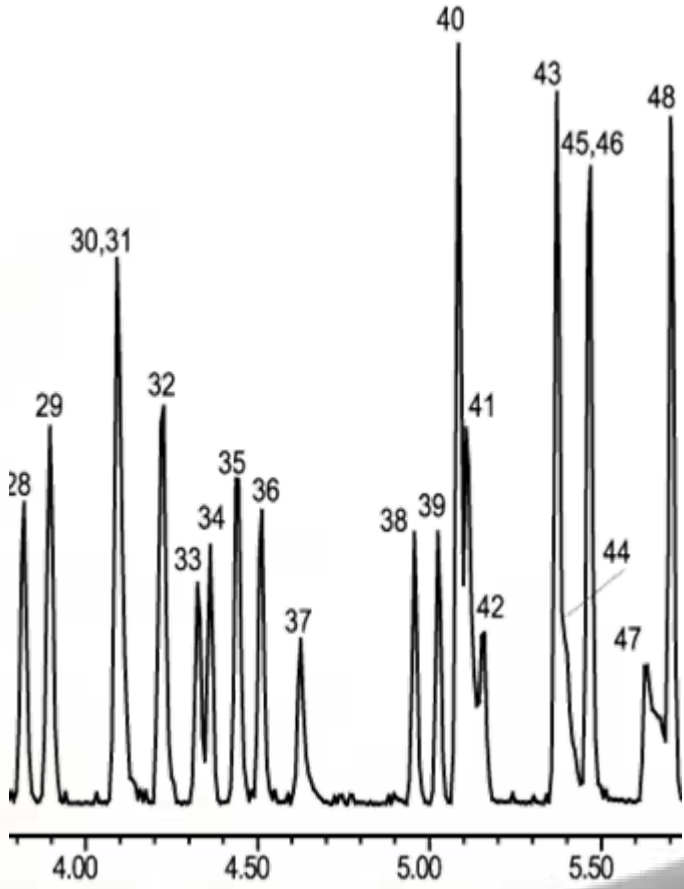
# A Closer Look

| Peak | Compound | Mass Ion |
|---|---|---|
| 11 | Ethyl Formate | 31 |
| 12 | Methyl Acetate | 43 |
| 14 | Carbon Tetrachloride | 117 |
| 15 | 1,1,1-Trichloroethane | 97 |
| 17 | Isopropyl Acetate | 43 |
| 18 | MEK | 43 |



# A Closer Look Cont.

| Peak | Compound | Mass Ion |
|---|---|---|
| 30 | Toluene | 91 |
| 31 | N-Propanol | 31 |
| 40 | m-Xylene | 91/106 |
| 41 | Butanol | 56 |
| 42 | Nitromethane | 30 |
| 43 | Cumene | 105 |
| 44 | 2-Methoxyethanol | 45 |
| 45 | o-Xylene | 91/106 |
| 46 | Pyridine | 79 |



# Summary - USP Method 467

- Current <467> Method has some limitation
  - Long analysis time
  - Co-elution
  - Some compounds are not detected via Headspace
  - No spectral confirmation of target analytes
- By changing various parameters, method performance can be improved
- The final method needs to simple & easy to use

# Thank You!

Questions?