

**Example 1** – Analysis of Chlorophenoxy Acid Herbicides in Water (from Thermo SPE library)

1. Why adjust sample pH to 1.0?
2. Why condition with hexane/acetone?
3. Why condition with acidified methanol?
4. Why wash with pH 1 water?
5. Why dry the material at this point?
6. Why elute with hexane/acetone mixture? Why not elute with just hexane?
7. What is the purpose of the 'keeper solvent'?

**Example 2** – Analysis of Amphetamines in Urine by GC/MS (from Thermo SPE library)

1. Why adjust the sample pH to 6.0?
2. Why condition first with MeOH?
3. Why condition with water prior to the phosphate buffer?
4. Why condition with phosphate buffer at pH 6.0?
5. Why is it important to prevent drying of the material at this point?
6. Why wash with 100 mM acetic acid (what is the pH)?
7. What will elute during the MeOH washing step?
8. Why dry the material at this point?
9. What is the role of the ammonia in the elution step?
10. What is the purpose of the fluoroacylation step?
11. What is the purpose of drying the extract at this point?

# Amphetamines in Urine, Oxidation with Periodate for GC or GC/MS Confirmations

Using 200mg 10mL HyperSep Verify-CX Extraction Column (Part Number: 60108-742)

## 1 Prepare Sample

To 2mL of urine add internal standard(s)\*, 1mL of 100mM phosphate buffer (pH 6.0) and 1mL of 0.35 M sodium periodate

Mix/vortex

Incubate at room temperature for 20 minutes

Sample pH should be 6.0±0.5

Adjust pH accordingly with 100mM monobasic or dibasic sodium phosphate

## 2 Condition Verify-CX Extraction Column

3mL of CH<sub>3</sub>OH then aspirate

3mL of DI H<sub>2</sub>O then aspirate

1mL of 100mM phosphate buffer (pH 6.0) then aspirate

**NOTE:** Aspirate at <3" Hg to prevent sorbent drying

## 4 Apply Sample

Load sample at 1 to 2mL/minute

## 6 Wash Column

3mL of DI H<sub>2</sub>O

1mL of 100mM acetic acid

3mL of CH<sub>3</sub>OH

Dry column (5 minutes at >10" Hg)

## 8 Elute Amphetamines

3mL of CH<sub>2</sub>Cl<sub>2</sub>/IPA/NH<sub>4</sub>OH (78:20:2)

Collect eluate at 1 to 2mL/min

**NOTE:** Prepare elution solvent fresh daily; add IPA/NH<sub>4</sub>OH mix, then add CH<sub>2</sub>Cl<sub>2</sub> (pH 11-12)

## 10 Concentrate Eluate

Add 30µL silylation grade DMF\*\*\* to eluate

Evaporate to 30µL at <40°C

## 11 Fluoroacylate with PFPA (PFAA)

Add 50µL PFPA (PFAA)\*\*\*\*

Overlay with N<sub>2</sub> and cap

Improve derivatization by addition of 50µL PFPOH

React for 20 minutes at 70°C

Evaporate to dryness at <40°C

## Quantitate

Inject 1 to 2µL onto gas chromatograph

For mass spectrometry monitor the following ions:

Analyte (TMS)	Primary Ion**	Secondary	Tertiary
D <sub>5</sub> -amphetamine*	194	92	123
Amphetamine	190	91	118
D <sub>5</sub> -methamphetamine*	208	92	163
Methamphetamine	204	91	160

\* Suggested internal standards

\*\* Quantitation ion

\*\*\* Part number TS-20672 (50mL vial)

\*\*\*\* Part number TS-65193 (10 x 1mL ampules)

## Recommended GC Columns

## Part Number

TRACE TR-DoA5 Column, 15m x 0.25mm x 0.25µm	26AF130P
TRACE TR-DoA35 Column, 15m x 0.20mm x 0.33µm	26AC497P

## Chlorophenoxy Acid Herbicides in Water

Using 1g 6mL HyperSep C18 Extraction Column (Part Number: 60108-301)

①

### Sample Preparation

Adjust pH of 1L of water sample to pH 1.0 with hydrochloric acid

②

### Condition C18 Extraction Column

10mL of hexane/acetone (50:50)  
10mL of acidified methanol (5% HCl in methanol)  
10mL of DI H<sub>2</sub>O

③

### Apply Sample

Load 1 liter of sample at a rate of 8 to 10mL/minute

### Wash Column

10mL of DI H<sub>2</sub>O adjusted to pH 1.0 with HCl

④

### Dry Column

Use maximum vacuum pressure for 15 to 30 minutes

⑤

### Elute Chlorophenoxy Acid Herbicides

10mL of hexane/acetone (50:50)

⑥

### Concentrate/Evaporate

Add 500µL of a keeper solvent (methanol, DMF, other)  
Evaporate to 500µL under a nitrogen stream at room temperature

⑦

### Injection/Analysis

Reconstitute with 100µL of TCTEF and inject at 1 to 2µL onto GC column

### Chlorophenoxy Acid Herbicides Extracted

2,4-D acid  
2,4,5-trichloro phenoxy propionic acid (Silvex)  
Dicamba  
Dinitro-sec-butyl phenol

## Chlorophenol in Water

Using 500mg 6mL HyperSep Retain PEP Extraction Column (Part Number: 60107-206)

### Sample Preparation

Collect 500mL of H<sub>2</sub>O  
Adjust pH to 2 with H<sub>2</sub>SO<sub>4</sub>

### Condition HyperSep Retain PEP Extraction Column

1 x 5mL CH<sub>3</sub>OH  
1 x 5mL H<sub>2</sub>O

**NOTE:** Do not allow the cartridge to dry out

### Apply Sample

Load 1 x 500mL H<sub>2</sub>O  
Elute at 5mL/minute

### Wash Column

1 x 5mL H<sub>2</sub>O  
1 x 1mL CH<sub>3</sub>OH

### Elute

1 x 3mL THF  
Concentrate sample to 3mL under N<sub>2</sub>

### Analysis

Inject onto HPLC

### Recommended HPLC Column

Hypersil GOLD PFP 3µm, 150 x 4.6mm

### Part Number

25403-154630

## 2,4-Dichlorophenol in Water

Using 500mg 6mL HyperSep Retain PEP Extraction Column (Part Number: 60107-206)

### Sample Preparation

Adjust pH to 2 with H<sub>2</sub>SO<sub>4</sub>

### Condition HyperSep Retain PEP Extraction Column

1 x 5mL CH<sub>3</sub>OH  
1 x 5mL H<sub>2</sub>O

**NOTE:** Do not allow the cartridge to dry out

### Apply Sample

Load 1 x 500mL H<sub>2</sub>O  
Elute at 5mL/minute

### Wash Column

1 x 5mL H<sub>2</sub>O  
1 x 1mL CH<sub>3</sub>OH  
Dry column (20 minutes under N<sub>2</sub>)

### Elute

1 x 3mL THF  
Concentrate sample to 3mL under N<sub>2</sub>

### Analysis

Inject onto HPLC

### Recommended HPLC Column

Hypersil GOLD PFP 3µm, 150 x 4.6mm

### Part Number

25403-154630