

# HPLC Application

ID No.: 14450

## Extraction of Antidepressants on Strata™ X

**Column:** Luna® 5 µm C8(2) 100 Å, LC Column 150 x 4.6 mm, Ea

**Dimensions:** 150 x 4.6 mm ID

**Order No:** 00F-4249-E0

**Elution Type:** Isocratic

**Eluent A:** 20 mM KH<sub>2</sub>PO<sub>4</sub>, pH 7.0

**Eluent B:** Acetonitrile

**Eluent C:** Methanol

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B	Pct C
1	1	0	40	30	30

**Flow Rate:** 2 mL/min

**Col. Temp.:** ambient

**Detection:** UV-Vis Abs.-Fixed Wave. (UV) @ 210 nm (ambient)

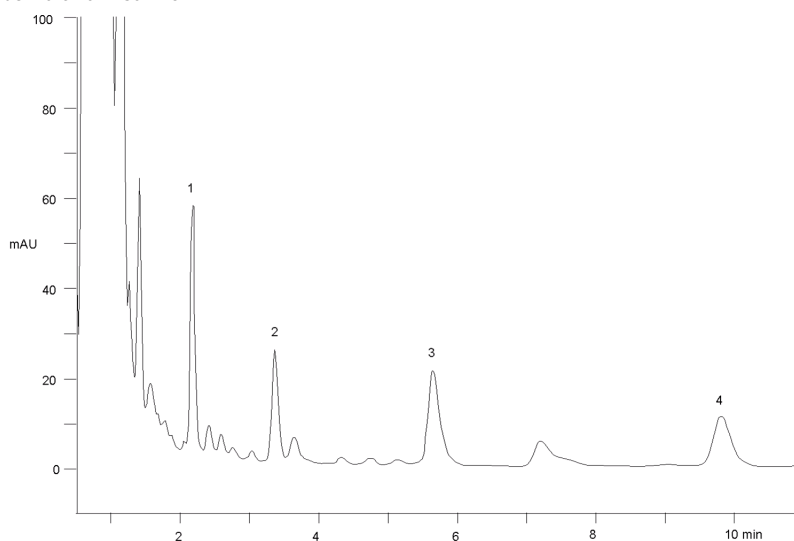
**Analyst Note:** SecurityGuard™ Guard Cartridge System extends column lifetime.

- SecurityGuard Cartridges, C8 4 x 3.0mm, 10/Pk Part No.: AJ0-4290

- Holder Part No.: KJ0-4282



Products used in this application:



### ANALYTES:

- 1 Nordoxepin (internal standard)
- 2 Nortriptyline
- 3 Doxepin
- 4 Amitriptyline



# Sample Preparation Details

for HPLC Application ID No.: 14450

## Extraction of Antidepressants on Strata™ X

### PRODUCT DESCRIPTION:

Strata™-X 33 µm Polymeric Reversed Phase, 30 mg / 1 mL, Tubes , 100/Pk

Order No.: 8B-S100-TAK

### SOLID PHASE EXTRACTION (SPE) PROCEDURE:

**Note:** The solvent volumes shown below are for a 30 mg bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

#### Condition:

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#### Load:

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Load: 1 ml of analyte and internal standard (1.0 ug/ml) spiked Porcine serum acidified with 2% conc.H3PO4 .  
(Dilute the serum spiked analyte 1: 1 with buffer or water as needed)

#### Wash:

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#### Dry:

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1min at 10" of Hg

#### Elute:

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#### Final Prep and Analysis:

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Blow down to almost dryness under slow stream of N2. Reconstitute with mobile phase to 200 µL.

Inject 50 uL on HPLC

Inject: 50 µL on HPLC UV-Vis Abs.-Fixed Wave. (UV) @ 210 nm (ambient)

#### ANALYTES:

	Spiked Conc. (ng/mL)	Log P	pKa	% Rec	%RSC (n=0)
1 Nordoxepin	1			103	
2 Nortriptyline	1			97	
3 Doxepin	1			93	
4 Amitriptyline	1			101	

**Note:** This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals.  
Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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For more information contact your Phenomenex Representative at info@phenomenex.com



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[info@phenomenex.com](mailto:info@phenomenex.com)