

Automated Extraction of Tricyclic Antidepressants by Perkin Elmer Multiprobe II

Column: Kinetex® 2.6 µm C18 100 Å, LC Column 50 x 2.1 mm, Ea**Dimensions:** 50 x 2.1 mm ID**Order No:** 00B-4462-AN**Elution Type:** Gradient**Eluent A:** 0.1% formic acid/water**Eluent B:** 0.1% Formic acid/methanol

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	60	40
	2	3.5	20	80
	3	4	20	80
	4	4.01	60	40
	5	5	60	40

Flow Rate: 0.4 mL/min**Col. Temp.:** ambient**Detection:** Mass Spectrometer (MS) @ amu (ambient)**Detector Info:** SCIEX<

Compound	Q1	Q3	Dwell time	DP	EP	CE	CXP
Protrip	264.2	191.1	35	60	10	35	10
Notrip	264.3	191.2	35	60	10	35	10
DM-Dox	266.2	107.1	25	60	10	35	10
Desip	267.2	208.2	35	60	10	35	10
Protrip-D3	267.2	191.1	35	60	10	35	10
Notrip-D3	267.3	191.2	35	60	10	35	10
Desip-D3	270.2	208.2	35	60	10	35	10
Amitrip	278.2	191.2	35	60	10	35	10
Dox	280.3	107.1	25	60	10	35	10
Imip	281.3	208.2	35	60	10	35	10
Imip-D3	284.2	208.1	35	60	10	35	10
DM-Clomip	301.2	242.2	35	60	10	35	10
Clomip	315.2	242.2	35	60	10	35	10
Clomip-D3	318.2	242.2	35	60	10	35	10

Ionization Parameters

ESI in positive polarity

Gas 1	50
Gas 2	50
CAD	8
Cur	20
IS	5500
Temp	600

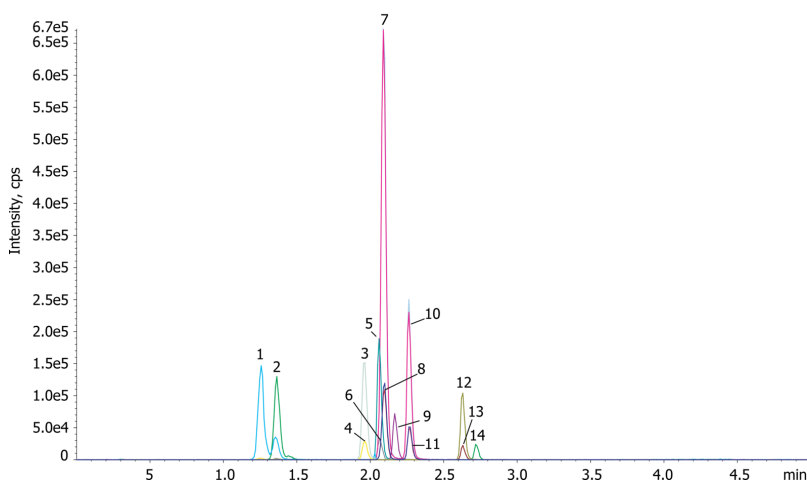


Products used in this application:



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20937



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ANALYTES:

- 1** Doxepin
- 2** DM-Doxepin
- 3** Imipramine-d3
- 4** Imipramine
- 5** Desipramine-d3
- 6** Desipramine
- 7** Nortriptyline-d3
- 8** Nortriptyline
- 9** Amitriptyline
- 10** Protriptyline-d3
- 11** Protriptyline
- 12** Clomipramine-d3
- 13** Clomipramine
- 14** DM-Clomipramine



Sample Preparation Details

for HPLC Application ID No.: 20937

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PRODUCT DESCRIPTION:

StrataTM-X-Drug B 33 µm Polymeric Strong Cation, 30 mg / well, 96-Well Plates , 2/Pk

Order No.: 8E-S128-TGB

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:

Load:

500 µL Plasma (Human) diluted with 1 mL 100mM sodium acetate buffer (pH5.0) spiked with TCAs (25ng/mL)

Wash:

Dry:

8-10 mins at 10" of Hg under N₂ at 45 degC

Elute:

Final Prep and Analysis:

To do % absolute recovery, the IS was spiked in the initial mobile phase in the reconstitution step

Inject: 10 µL on HPLC Mass Spectrometer (MS) @ amu (ambient)

ANALYTES:	Spiked Conc. (ng/mL)	Log P	pKa	% Rec	%RSC (n=0)
1 Doxepin	25			93	
2 DM-Doxepin	25			96	
3 Imipramine-d3	200				
4 Imipramine	25			90	
5 Desipramine-d3	200				
6 Desipramine	25			85	
7 Nortriptyline-d3	200				
8 Nortriptyline	25			75	
9 Amitriptyline	25			82	
10 Protriptyline-d3	200				
11 Protriptyline	25			94	
12 Clomipramine-d3	200				
13 Clomipramine	25			92	
14 DM-Clomipramine	20			89	

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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