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Application Note

Tricyclic Antidepressants in Plasma

Waters Corporation



This is an Application Brief and does not contain a detailed Experimental section.

Abstract

This application brief demonstrates analysis of tricyclic antidepressants in plasma.

Introduction

The compounds used in this study are -

Spiked at 500 ng/mL versus Blank:

- 1. Nordoxepin (I.S.)
- 2. Nortriptyline
- 3. Doxepin
- 4. Imipramine
- 5. Amitriptyline
- 6. Trimipramine

1. Nordoxepin

2. Nortriptyline

3. Doxepin

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Experimental

HPLC Method

Column:	Symmetry Shield RP ₈ , 4.6 x 75 mm, 3.5 μ m	
Part numbers:	WAT094263	
Mobile phase:	50 mM phosphate, pH 7/methanol 26:74	
Flow rate:	1.4 mL/min	
Injection volume:	1 mL spiked plasma plus 20 μL concentrated phosphoric acid, 100 μL injection	
Temperature:	29 °C	
Detection:	UV @ 254 nm	

Oasis® HLB Extraction Method

Oasis® HLB Extraction Plate, 30 mg/96-well Part Number WAT058951

Condition

1 mL methanol/1 mL water

Load

1 mL spiked plasma plus 20 µL concentrated phosphoric acid

Wash

A. 1 mL 2% ammonium hydroxide in 5% methanol

B. 1 mL 2% ammonium hydroxidein 65% methanol

C. 1 mL 2% acetic acid in 5% methanol

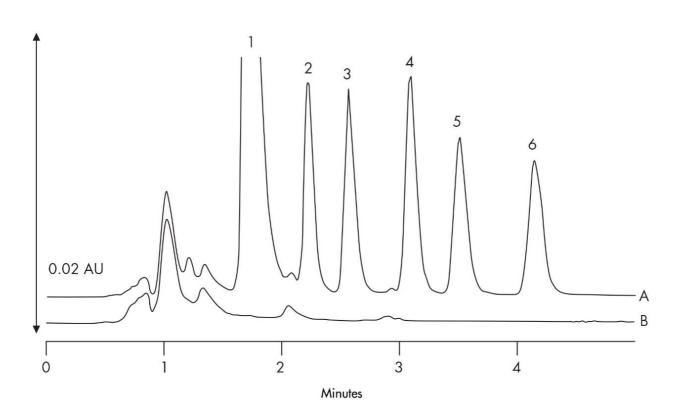
Elute

600 µL 65% methanol

Add internal standard (I.S.)
60 µL 36 µg/mL nordoxepin in
10% ammonium hydroxide

	500 ng/mL		100 ng/mL	
Compound	Recovery	RSD (n=96)	Recovery	%RSD (n=95)
Nortriptyline	92.3%	1.4%	90.8%	5.7%
Doxepin	90.6%	1.4%	90.4%	4.7%
Imipramine	92.2%	1.7%	86.4%	5.3%
Amitriptyline	90.2%	1.6%	85.3%	5.8%
Trimipramine	90.3%	1.9%	89.8%	6.1%

Chromatogram of Plasma Extracts: A) Blank B) Spiked Sample



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