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Nota de aplicación

Chlordiazepoxide and Metabolites in Serum

Waters Corporation



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

Abstract

This application brief demonstrates analysis of chlordiazepoxide and metabolites in serum.

Introduction

The compounds used in this study are -

- 1. Norchlordiazepoxide
- 2. Oxazepam
- 3. Nordazepam
- 4. Chlordiazepoxide
- 5. Demoxepam (I.S.)

Experimental

HPLC Method

Column:

Mobile phase:

Guard column:	Sentry Guard Column, 3.9 x 20 mm, 5 µm
Part number:	Column - WAT046980, Guard - WAT054225

Symmetry C_{18} , 3.9 x 150 mm, 5 μm

20 mM potassium phosphate, pH 7.0/acetonitrile/methanol 56:21:23

Flow rate: 1.0 mL/min

Detection:

UV @ 240 nm

Oasis® HLB Extraction Method

Oasis® HLB 1 cc/30 mg Extraction Cartridge Part Number WAT094225

Condition:

1 mL methanol/1 mL water

Load:

1 mL spiked porcine serum with 1.5 µg/mL demoxepam (I.S.)

Wash:

1 mL 5% methanol in water

Elute:

1 mL methanol

Evaporate and Reconstitute:

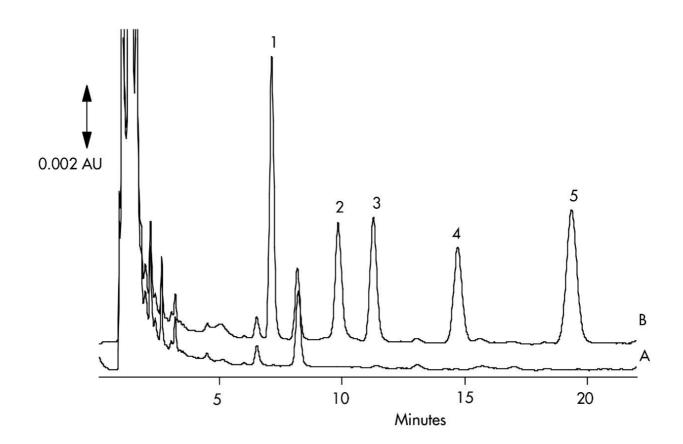
40 °C under nitrogen stream 200 µL of 20 mM potassium phosphate pH 7/methanol 80:20 (v/v)

Results and Discussion

Compound	Concentration µg/mL	% Recovery	%RSD (n-6)
1. Norchlordiazepoxide	0.200	97.8%	4.3%
	0.040	96.5%	5.5%
2. Oxazepam	0.200	104.0%	2.6%
	0.040	90.0%	2.5%
3. Nordazepam	0.200	101.0%	3.0%
	0.040	98.9%	1.2%
4. Chlordiazepoxide	0.200	90.0%	2.4%
	0.040	100.0%	4.1%

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



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