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Nota de aplicación

Caffeine and Metabolites in Serum

Waters Corporation



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

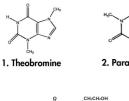
Abstract

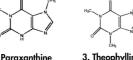
This application brief demonstrates analysis of caffeine and metabolites in serum.

Introduction

The compounds used in this study are -

- 1. Theobromine
- 2. Paraxanthine
- 3. Theophylline
- 4. β-hydroxyethyltheophylline (I.S.)
- 5. Caffeine







H₂C CH₃

4. B-hydroxyethyltheophylline (I.S.)

5. Caffeine

Experimental

HPLC Method

Column: Symmetry Shield RP8, 3.0 x 150 mm, 5 μ m

Guard column: Sentry Guard Column, 3.9 x 20 mm, 5 µm

Part number: Column - WAT094243, Guard - WAT200675

Mobile phase: 20 mM ammonium acetate, pH 5/acetonitrile,

95:5

Flow rate: 0.6 mL/min

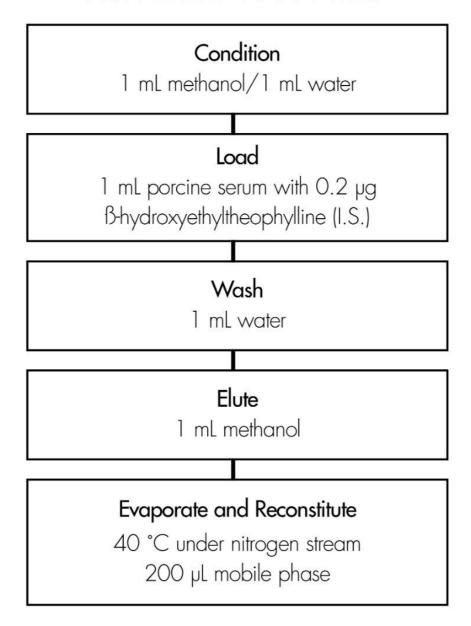
Injection volume: 20 μ L of reconstituted porcine serum extract

Temperature: 25 °C

Detection: UV @ 273 nm

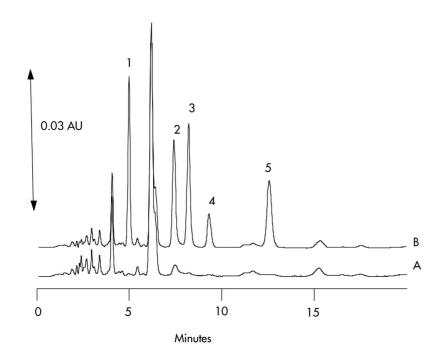
Oasis® HLB Extraction Method

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



Compound	Concentration µg/mL	% Recovery	%RSD (n-6)
Theobromine	0.100	103.0%	3.4%
	0.500	104.0%	2.4%
Paraxanthine	0.100	101.0%	6.2%
	0.500	98.9%	2.3%
Theophylline	0.100	100.0%	5.2%
	0.500	101.0%	2.1%
Caffeine	0.100	92.6%	4.4%
	0.500	97.5%	3.2%

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



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