

Application Note

Analysis of Caffeine Metabolites Using XBridge Phenyl

Waters Corporation

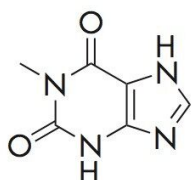


This is an Application Brief and does not contain a detailed Experimental section.

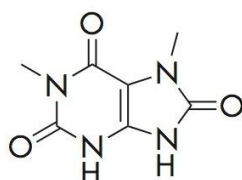
Abstract

This application brief highlights the analysis of caffeine metabolites using XBridge Phenyl columns.

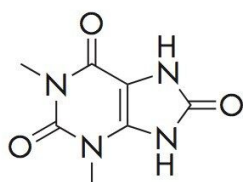
Introduction



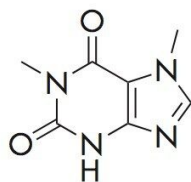
1. 1-Methylxanthine



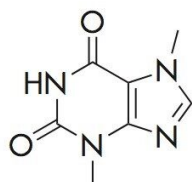
2. 1,7-Dimethyluric acid



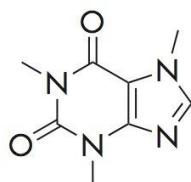
3. 1,3-Dimethyluric acid



4. 1,7-Dimethylxanthine



5. Theobromine



6. Caffeine

Experimental

Test Conditions

Columns:

XBridge Phenyl, 4.6 x 100 mm, 3.5 μ m p/n:

186003334

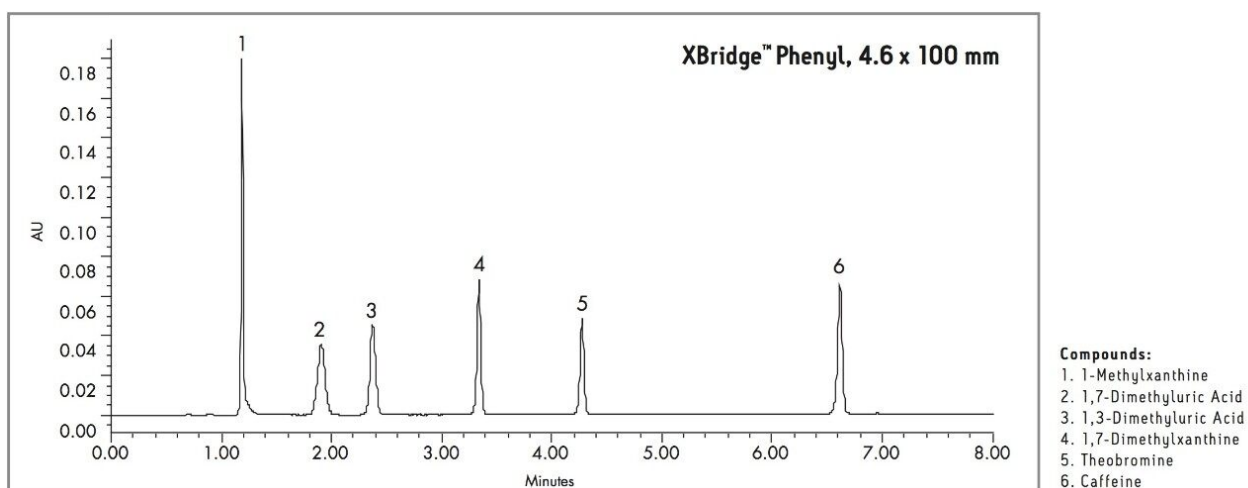
Mobile phase A:	H ₂ O
Mobile phase B:	ACN
Mobile phase C:	100 mM NH ₄ HCO ₃
Flow rate:	1.0 mL/min
Injection volume:	10 µL
Sample:	Caffeine (10 µg/mL), Theobromine (10 µg/mL), 1-Methyl xanthine (10 µg/mL), 1,3-Dimethyluric Acid (10 µg/mL), 1,7-Dimethyl xanthine (10 µg/mL), 1,7-Dimethyluric Acid (10 µg/mL) in H ₂ O/NH ₄ HCO ₃ (90/10)
Column temp.:	30 °C
Sampling temp.:	15 °C
Detection:	UV @ 280 nm
Sampling Rate:	5 points/sec
Filter Response:	0.2
Instrument:	Alliance 2695 with 2996 PDA

Gradient

Time(min)	%A	%B	%C
0	89	1	10

Time(min)	%A	%B	%C
9	66	24	10
10	89	1	10
20	89	1	10

Results and Discussion



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Alliance HPLC <<https://www.waters.com/514248>>

WA60187, June 2007

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