

Nota applicativa

## Gradient Separation of Morphine and Metabolites on Atlantis HILIC Silica

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Waters Corporation

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

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### Abstract

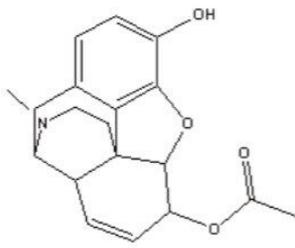
This application brief demonstrates the gradient separation of morphine and its metabolites on Atlantis HILIC Silica column.

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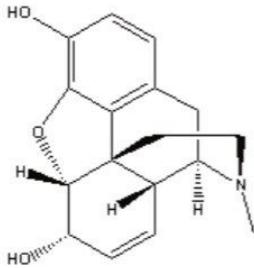
### Introduction

The compounds used in this study are-

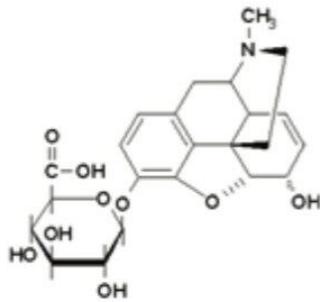
1. 6-Acetylmorphine
2. Morphine
3. Morphine-3 $\beta$ -D-glucuronide



6-Acetylmorphine



Morphine



Morphine-3β-D-glucuronide

Figure 1. Structures of the compounds used in this study.

## Experimental

### Test Conditions

Column:	Atlantis HILIC Silica, 2.1 x 50 mm, 3 μm
Part Number:	186002011
Mobile Phase A:	10 mM NH <sub>4</sub> COOH in H <sub>2</sub> O, 0.125% HCOOH in 50:50 ACN:H <sub>2</sub> O

Mobile Phase B: 10 mM NH<sub>4</sub>COOH in H<sub>2</sub>O, 0.125% HCOOH in 90:10 ACN:H<sub>2</sub>O

Flow Rate: 0.6 mL/min

Injection Volume: 5 µL

Sample Concentration: 25 µg/mL each

Sample Diluent: 75:25 ACN:MeOH with 0.2% HCOOH

Column Temperature: 30 °C

Detection: UV @ 280 nm

Sampling Rate: 20 points/sec

Time Constant: 0.1

Instrument: Waters ACQUITY UPLC with ACQUITY PDA

### Gradient:

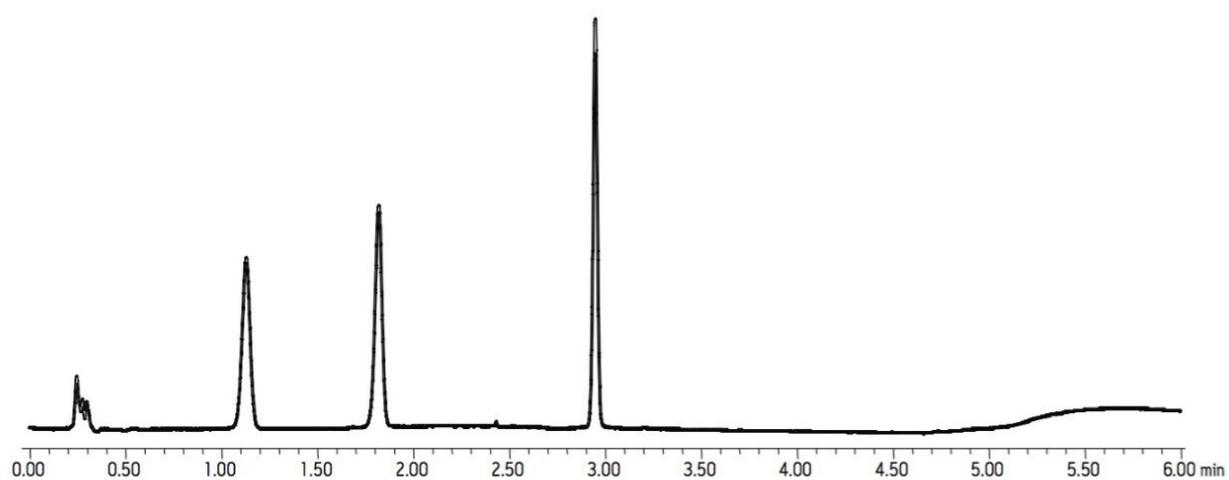
Time(min)	Profile
	%A
0.00	0.1
1.05	0.1
4.35	99.9
4.50	0.1

Time(min)          Profile

6.00                0.1

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## Results and Discussion



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[ACQUITY UPLC PDA Detector <https://www.waters.com/514225>](https://www.waters.com/514225)

WA64070, August 2009

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