

Nota de aplicación

## Analysis of Herbal Medicine on an XBridge HPLC Amide Column

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Waters Corporation



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

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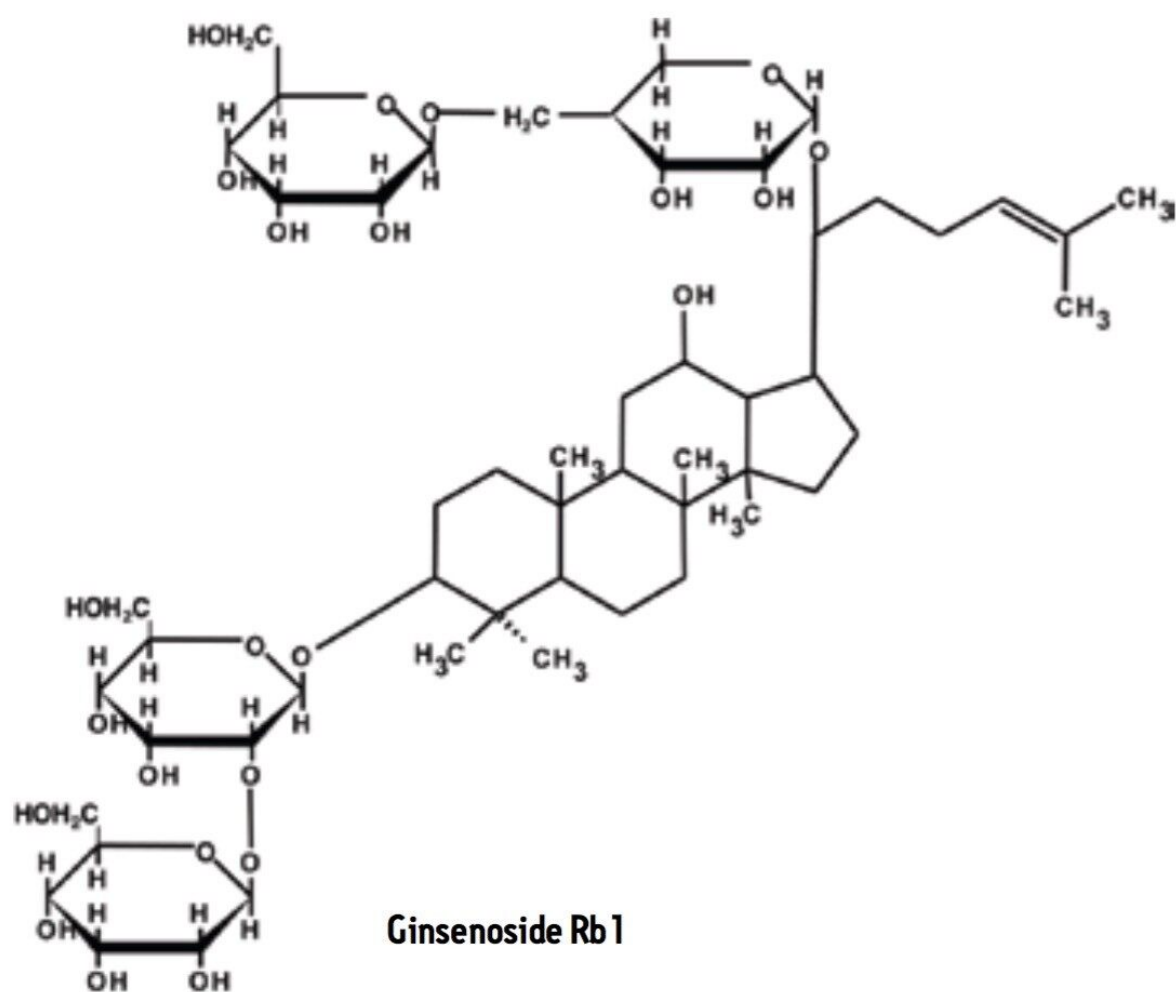
### Abstract

This application brief demonstrates analysis of herbal medicine on an XBridge HPLC amide column.

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## Introduction

### Compound



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## Experimental

## HPLC Conditions

|                       |   |
|-----------------------|---|
| Column:               | XBridge Amide, 3.5 $\mu$ m, 4.6 x<br>250 mm |
| Part Number:          | 186004870                                   |
| Mobile Phase:         | 80:20 MeCN:H <sub>2</sub> O                 |
| Isocratic Flow Rate:  | 0.8 mL/min                                  |
| Column Temp.:         | 60 °C                                       |
| Sample Temp.:         | 10 °C                                       |
| Injection Vol.:       | 20 $\mu$ L                                  |
| Needle Wash:          | 95:5 MeCN:H <sub>2</sub> O                  |
| Seal Wash:            | 10:90 MeOH:H <sub>2</sub> O                 |
| UV:                   | 203 nm                                      |
| Sampling Rate:        | 20 Hz                                       |
| Filter Time Constant: | 0.1 sec                                     |
| Total Run Time:       | 18 min                                      |
| Instrument:           | Alliance 2695 and 2998 PDA                  |

## Pretreatment

1. Weigh 2 g of herbal medicine powder into a centrifuge tube.
2. Add 30 mL of 60% MeOH/40% H<sub>2</sub>O.

3. Shake for 15 min.
4. Centrifuge at 4,000 rpm for 10 min.
5. Obtain the supernatant.
6. Repeat steps 2-5 with the residue using 15 mL of 60% MeOH/40% H<sub>2</sub>O.
7. Combine the supernatant, and make exactly 50 mL by adding 60% MeOH/40% H<sub>2</sub>O.
8. Take 10 mL of this solution and add 3 mL of NaOH test solution (1 mol/L).
9. Let stand for 30 min.
10. Add 3 mL of HCl test solution (1 mol/L).
11. Add 60% MeOH/40% H<sub>2</sub>O to make exactly 20 mL.

## Solid-Phase Extraction

SPE Device: Sep-Pak Plus C<sub>18</sub> cartridge  
360 mg (55-105 µm)

Part Number: WAT020515

1. Condition with 2 mL MeOH.
2. Equilibrate with 2 mL of 30% MeOH/70% H<sub>2</sub>O just before loading.
3. Load 5 mL of the solution from step 11 in the pretreatment stage.
4. Wash with 2 mL of 30% MeOH/70% H<sub>2</sub>O.
5. Wash with 1 mL of Na<sub>2</sub>CO<sub>3</sub> test solution (1 mol/L).
6. Wash with 10 mL of 30% MeOH/70% H<sub>2</sub>O.
7. Elute with 5 mL MeOH (this is the injection solution).

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## Featured Products

Alliance HPLC System <<https://www.waters.com/534293>>

2998 Photodiode Array (PDA) Detector <<https://www.waters.com/1001362>>

WA64084, February 2010

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