Rapid Simultaneous Analysis of Valerenic Acids Using the Agilent 1290 Infinity LC System and Sub-2 Micron Columns

Application Note

Author
Steven Brauer, QC Chemist
Schwabe North America

Introduction
Advancements in LC instrumentation and column design have drastically increased the potential efficiency of many LC analyses. The instrumental quantification of valerenic acids can be completed approximately 4x faster than the current USP instrument method using a sub-2 µm (STM) column, the Agilent 1290 Infinity LC System, and an alternative instrument method developed at Schwabe North America. It also reduces solvent consumption significantly.
**Valerenic acids**

Valerenic acid and the analogs hydroxyvalerenic and acetoxyvalerenic acid are the active components of the herb *Valeriana officinalis*. It has been used to treat insomnia and other sleep disorders. Though not fully understood, there are some basic theories about the physiological pathways in which the herb works. Powdered valerian root extracts typically have a distinct unpleasant odor not characteristic of the valerenic acids themselves.

**Extraction procedure**

USP Method: Sonicated in 25/75 aqueous 0.1 % o-phosphoric acid/methanol.

---

**Results and Discussion**

The new method and instrumentation can separate valerenic acid and its analogs for quantification in 10 minutes (Figure 1). This is faster than the USP analysis, which takes 37 minutes (Figure 2). The USP analysis uses a 4.6 × 250 mm, 5 µm C-18 column, while the new Schwabe North America method uses an Agilent ZORBAX Eclipse C-18, 2.1 × 50 mm, 1.8 µm column.

---

![Diagram of valerenic acids]
Conclusion
In analyzing complex herbal blends, the Agilent 1290 Infinity LC System coupled with STM columns can speed up analysis time, thus saving cost in labor and solvent use. This is done without any loss in resolution.

Figure 2. Valerenic acids in a multicomponent matrix using the USP method with an Agilent ZORBAX Eclipse C-18, 4.6 × 250 mm, 5.0 µm column.

Instrument parameters
Temperature 40 °C
Injection amount 15.0 µL
Detection UV (DAD), 225 nm
Flow rate 1.0 mL/minute
Mobile phases A) Water with 0.3 % o-phosphoric acid
B) Methanol with 0.3 % o-phosphoric acid
Gradient

<table>
<thead>
<tr>
<th>Time</th>
<th>% A</th>
<th>% B</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>15.0</td>
<td>5</td>
<td>95</td>
</tr>
<tr>
<td>25.0</td>
<td>5</td>
<td>95</td>
</tr>
<tr>
<td>30.0</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>37.0</td>
<td>40</td>
<td>60</td>
</tr>
</tbody>
</table>

Figure 3. Reference standard, valerenic acid: Indofine (023801S) – Lot# 12121035, concentration: 20.7 µg/mL, % RSD (five injections): 0.048 %.
Reference


For More Information

These data represent typical results. For more information on our products and services, visit our Web site at www.agilent.com/chem.