Fast Analysis of Eleutherosides in Siberian Ginseng Using the Agilent 1290 Infinity LC and Eclipse 1.8 µm Column

Application Note

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Abstract

Analysis of eleutherosides in *Eleutherococcus senticosus* (often called Siberian Ginseng) extract is typically performed using parameters outlined by the United States Pharmacopeia [1]. A method has been developed using an Agilent 1290 Infinity LC with an Agilent ZORBAX EclipsePlus 3 × 100 mm, 1.8 µm UHPLC column. This new method reduces solvent consumption and improves peak resolution.

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Introduction

Siberian ginseng (*Eleutherococcus senticosus*) has been used for centuries to prevent colds and flu, and to increase energy, longevity, and vitality. The Siberian ginseng is originally native to the far East countries, and its roots are harvested for medicinal purposes. The proposed active ingredients in Siberian ginseng, eleutherosides, are believed to stimulate the immune system [2]. Other ginseng types, particularly panax ginseng, are normally characterized for ginsenosides, making the Siberian variant unique with respect to the active components.

Experimental

Eleuthero extract was purchased from Draco, 539 Parrott St. San Jose, CA 95112

Sample Preparation

A 500 mg amount of extracted sample was dissolved in 100 mL of 50/50 methanol/water by sonication for 30 minutes.

Results and Discussion

Figure 1 shows the chromatogram produced when analyzing for Siberian ginseng using an Agilent ZORBAX Eclipse Plus column and the instrument conditions listed in Table 1. Figure 2 shows the chromatogram produced by Siberian ginseng analysis using the USP method, listed in Table 2, with an Agilent C18 column. Both methods used the same mobile phase, flow rate, and temperature. In addition, both chromatograms show good separation and peak shape. However, comparison of the results illustrates that the Agilent method results in an overall cycle time savings of 25 minutes, as well as a reduction of solvent consumption.

Peak confirmation is accomplished using certified reference standards with the same separation conditions used for sample extracts. This separation, shown in Figure 3, gives excellent retention time matching when compared to the extracts in Figure 1.

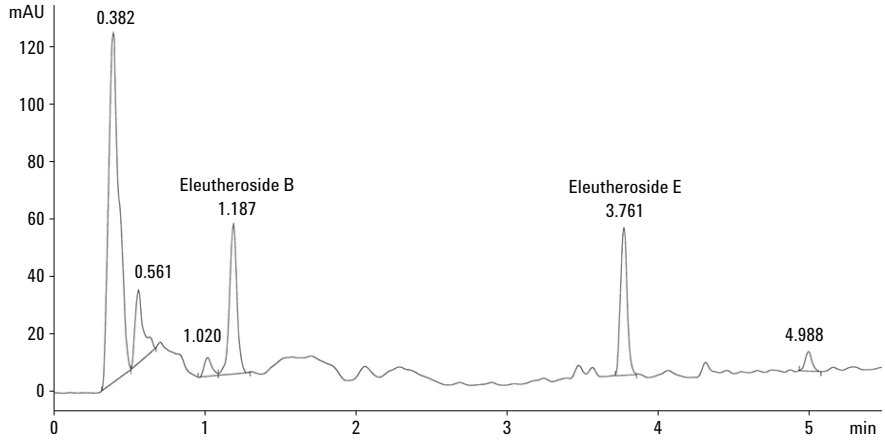


Figure 1. Eleutherosides in Siberian ginseng using an Agilent 1290 Infinity LC with an Agilent ZORBAX Eclipse Plus, 3 × 100 mm, 1.8 µm column.

Table 1. Instrument Parameters for Siberian Ginseng Detection using an Agilent 1290 Infinity LC/MS

Temperature	25 °C
Flow rate	1.0 mL/minute
Detection	UV, 220 nm
Mobile Phase A	5% acetonitrile/95% water
Mobile Phase B	60% acetonitrile/40% water

Gradient

Time (min)	% A	%B
0.00	85	15
2.00	85	15
5.14	60	40
5.34	5	95
7.71	5	95
7.72	85	15
9.44	85	15

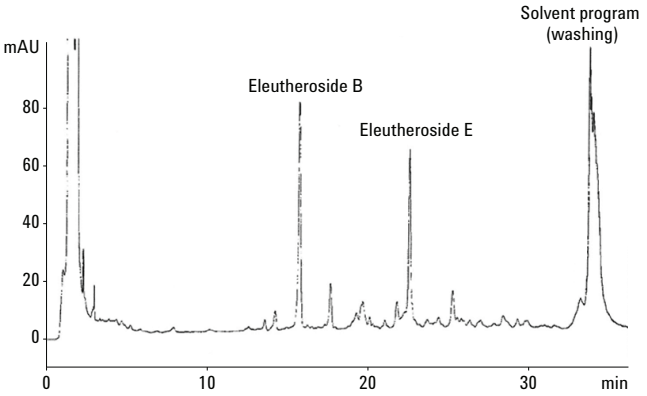


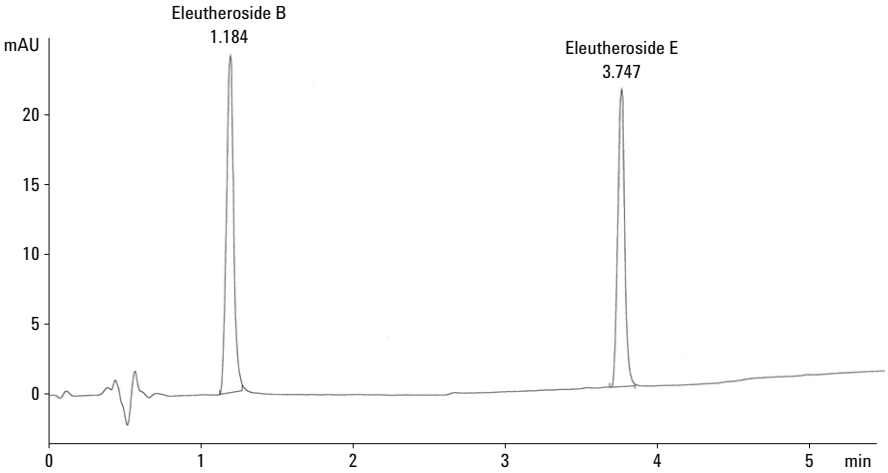
Figure 2. Eleutherosides in powdered eleuthero extract; USP method using an Agilent C-18, 250 × 4.6 mm, 5.0 µm column [2].

Table 2. Instrument Conditions for Siberian Ginseng Detection using the USP Method

Temperature	25 °C
Flow rate	1.0 mL/minute
Detection	UV, 220 nm
Mobile Phase A	5% acetonitrile/95% water
Mobile Phase B	60% acetonitrile/40% water

Gradient

Time (min)	% A	%B
0	97	3
5	97	3
30	60	40
31	5	95
45	5	95
45.1	97	3
60	97	3



Eleutheroside B	Chromadex #ASB-0005060 – Lot # 00005060-004
Calibration amount	0.004786 mg/mL
%RSD of 5 injections	0.2467
Eleutheroside E	Sigma-Aldrich #08198 – Lot # BCBH6095V
Calibration amount	0.012724 mg/mL
%RSD of 5 injections	0.1428

Figure 3. Eleutherosides reference standards.

Conclusions

The Agilent 1290 Infinity LC/MS with an Agilent ZORBAX Eclipse Plus sub-2 µm column provides excellent results when analyzing complex botanical extracts such as Siberian ginseng. This method provides a reduction in cycle time, solvent use, and labor costs without loss of resolution.

References

1. Powdered Eleuthero Extract Monograph, United States Pharmacopeia, USP35, NF30 p.1287, ISBN reference 978-1-936424-00-9.
2. Siberian Ginseng, University of Maryland Medical Center, 2011. http://umm.edu/health/medical/altmed/herb/siberian-ginseng

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