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Research Article

Determination of Lead in Eleuthero Root Extract by Atomic Absorption Spectrophotometry (AAS)

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Abstract

Eleuthero root extract (also known as Siberian ginseng) is an herbal supplement derived from the root of *Eleutherococcus senticosus*, a shrub native to north eastern China, Japan, Russia, particularly Siberia. Eleuthero has been used for over 2,000 years in traditional medicine, particularly in Chinese and Russian healing systems. American and Asian ginseng vary in their concentration of active compounds and effects on the body. Ginseng is an herb rich in antioxidants. It may offer benefits for brain health, immune function, blood sugar management, and more. However, more research is necessary. In this present study, explores the application of various analytical techniques for the quantification of lead (Pb) in eleuthero root extract samples. We measured the concentration of eleuthero root extract by using AAS, atomic absorption spectrophotometry. A pretreatment method of the samples, digested the analyte with a mixed acid solution. The AAS method was successfully used for the determination of lead in eleuthero root extract sample. Spectrophotometric determination of lead in eleuthero root extract sample was found to be adequately sensitive in terms of linearity, repeatability, and accuracy. The correlation coefficient (R^2) was found to be 0.99866, average percent recovery was 97.1%, spiked sample, and 98.6% for the duplicate; and average sample result have been found 1.27 mg/Kg. The results were within the specification of not more than 10 mg/Kg. maximum.

Keywords

Eleuthero Root Extract, Siberian Ginseng, Lead Determination, Atomic Absorption, Spectrophotometry (AAS)

1. Introduction

Paradise Herbs Eleuthero (*Eleutherococcus senticosus)* is a species of small, a genus of 38 species of thorny shrub in the family Araliaceae native Northern Asia [1, 2]. It may be colloquially called devil's bush [3], Siberian ginseng, eleuthero, wild pepper, or kanjang [4]. *E. senticosus* has a history of use in Traditional Chinese medicine [1]. Root extracts of E. senticosus are sold as a dietary supplement or cosmetic, usually under the name *Siberian ginseng* [2].

Roots of E. senticosus are cylindrical, up to 0.5 centimeters (0.20 in) in diameter, straight, or branched, dark brown, and have a smooth surface with bark fixed closely to the xylem [2]. The derived extract from the roots has been characterized for its major constituents, including lignans, sesamin (eleutheroside B4), syringaresinol, phenylpropanoids, cou-

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marins, beta sitosterol and daucosterol [2].

Eleutherococcus senticosus is used as a medicinal plant in case of asthenia with stress and fatigue reduction, immune sport, and weakness. See structural formula of eleuthero, Figure 1. There is some evidence that eleuthero can support immune function, blood sugar regulation, hormonal balance, arterial hypertension and potentially helping to prevent colds and infection. Siberian ginseng, or eleuthero, adoptogen, performance enhancer, and immunostimulant [5]. Active components include eleutherosides and polysaccharides [6].

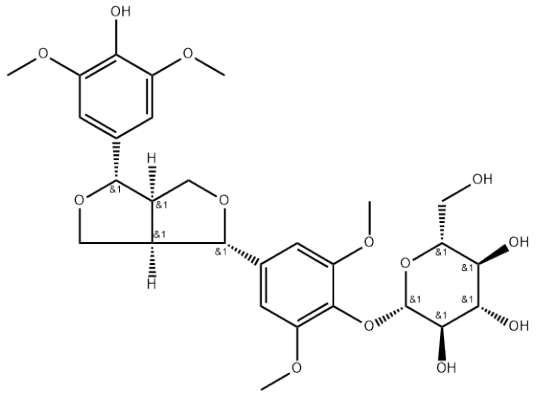

Figure 1. *Structural formula Siberian Ginseng (Eleuthero Root) Extract BP EP USP CAS 7374-79-0.*

Eleuthero might cause a pounding heart, irregular heartbeat, or high blood pressure in people who have heart disorder. It is sensitive to conditions such as breast cancer, uterine cancer, ovarian cancer, and endometriosis. Eleuthero might act like estrogen. The contraindication "arterial hypertension" is therefore highly restrictive for the clinical use of eleuthero. Fatigue, poor stamina, and stress intolerance. Used for adrenal support, nervous and mood disorders such as depression, mental fatigue, and poor concentration and is a traditional remedy to improve immune function in both chronic and acute infections.

2. Materials and Method

2.1. Equipment /Apparatus

A Perkin Elmer (Germany) 5100 PC Atomic Spectrophotometer with lead (Pb) lamp and air-acetylene flame. The effects of ionization may be substantially overcome by the addition of excess (1000-2000 µg/mL) of another alkali to standards and samples. The model 5100 combines the spectrometer of the model 5000 with at that new possibilities by using modern computers [7, 8].

Analytical balance XPR106DUHQ for automatic weighing. Maximum Capacity 120 g/41 readability 0.005 mg, 0.002 m, minimum weight (USP, 0.1%, typical) 4 mg [9]. Magnetic stirrer, thermal insulated. Fluoropolymer coated magnetic bar. Timer. Pipettes: 5, 10, 25, 50, and 100-mL pipettes. Volumetric Glassware (pipettes, flasks, and burettes) 50 mL, 100 mL, 500 mL volumetric flask *Glassware:* All glassware is washed in the following sequence: Alconox detergent solution, tap water; 1:1 nitric acid, 1:1 hydrochloric acid, and final rinse with deionized water both before and after use.

2.2. Reagents

All chemicals were used of analytical grade, doubly distilled deionized water (18M Ω cm-1) from a Milli-Q (MQ) was used in the preparations of solutions and used throughout. Good water, good results. Laboratory water purification systems play a crucial role in everyday work [10].

Lead (Pb) Stock Standard Solution, Fisher Scientific, lead Reference Solution (1000 ± 1% Certified), Fisher Chemical. CAS# 10099-74-8 Lead (II) nitrate $Pb(NO_3)_2$.

Lead Standard Solutions (various concentrations of lead, typically in the range of 0.1-10 µg/mL) Eleuthero Root Extract (sample). Commercial sample of root eleuthero senticosus its form of Cut root, was purchased by online Nitric Acid (HNO_3) (for digestion) were general purpose reagent. J. T. Baker ACS reagent, Lot# EO2056, concentration: 69-70%.

Sulfuric acid, H_2SO_4, Fischer Chemicals, Analytical-Trace metal grade, lot# 3112052 Hydrochloric Acid (HCl) (optional, for sample matrix adjustments) Pharmco-Aaper, ACS reagent grade, Lot # PB006406HAG

3. Experimental

3.1. Contamination Control

All reagent were of a high purity grade. Eppendorf pipettes with plastic tips were used for all pipetting. All sample handling operations were conduct under a laminar flow hood to reduce atmospheric contamination.

3.2. Operating Parameters

Flame AAS: Flame atomic absorption spectrometry is a globally recognized analytical technique used for analyzing metals in many industries. In FAAS, the sample solution is aspirates into a flame, where the lead atoms are excited and absorb light at a specific wavelength, 283.2 nm. FAAS is relatively simple and cost-effective.

Atomic absorption spectrophotometer provided with background corrector and having following parameters [7].

Instrument Setup:

Atomic Absorption Spectrophotometer: Set up the AAS with a lead hollow cathode lamp.

Wavelength: Set the wavelength to 283.3 nm, the characteristic absorption line for lead.

AAS Calibration: Run the calibration standards on the AAS to create a calibration curve. Plot the absorbance vs. the

concentration for the standards.

Aspirator Adjustment: Perkin Elmer instructs to optimize nebulizer using lead solution & lamp. That may or may not be crucial. It's most convenient to optimize it for elements using air-acetylene burner heads so carbon build up does not become an issue. Optimize the lamp position, burner position and nebulizer instrument parameters. Aspirate the highest standard. Unlock the locking nut on the aspirator. Turn the aspirator counter-clockwise until bubbles exit from the tube into standard solution. Turn aspirator clockwise and observe signal on continuous graphics. Signal will go through a maximum. Turn the aspirator to the place of highest signal. This is the optimized position. Lock in place lock ring [8].

Table 1. *Perkin-Elmer Instrument Setting.*

Instrument	**Perkin Elmer AAS**
Wavelength	283.3 nm
Relative Sensitivity	0.5 µg/mL Pb for 1% absorption.
Split Setting	4(0.7 nm)
Light Source	Electrodeless Discharge Lamp or Hallow Cathode Lamp
Lamps	Pb element hallow cathode lamp
Burning Head	Air-acetylene flame
Flame Type	Air-Acetylene flame oxidizing (lean, blue)

3.3. Sample Preparation

Drying (if using whole root): If you have dried Eleuthero root, grind it into a fine powder using a mortar and pestle or a grinder.

Extraction: Extract the active compounds from the powdered root using a suitable solvent (e.g., ethanol, water, or a mixture). This step may vary depending on the type of analysis you need to conduct. In most cases, a solvent extraction process is followed by concentration via evaporation.

3.4. Digestion of the Sample

1) Accurately weigh about 0.5–1.0 g (1.0025 g taken) of the Eleuthero root extract (or the dried root powder) and place it into a clean digestion vessel.
2) Add 5 mL of concentrated nitric acid (HNO_3) to the sample, and heat the mixture on a hotplate until the sample is fully dissolved.
3) After digestion, dilute the resulting solution with deionized water to a known volume (e.g., 50 or 100 mL). This is your sample solution.

3.5. Preparation of Calibration Standards

The aqueous calibration standard were prepared in pre-cleaned 100-mL volumetric flask using glass pipettes and fixed-volume Effendorf pipette for volume transfers.

1) Prepare a series of lead standards by diluting a lead stock solution with deionized water to create a set of standards of known concentration (e.g., 0.0, 0.5, 1.0, 5.0, 10.0 µg/mL).
2) The lead concentration of the standards should encompass the expected concentration of lead in your sample extract

3.6. Analysis of Sample

1) Aspirate the prepared sample solution into the flame and record the absorbance of the sample at 283.3 nm.
2) Using the calibration curve, determine the lead concentration in the sample based on its absorbance.

Table 2. *Lead Calibration Data-Absorbance of Pb standards.*

Lead (Pb) in Eleuthero Root Extract			
Calibration Data:			
[Pb] mg/L	AA-BG	R^2	0.99866
0.00	-0.0008	m	0.00756
0.50	0.0036	b	0.00015
1.00	0.0081		
5.00	0.0398		
10.00	0.0748		

Bracketing Standards: Linear Calibration Algorithms

The bracketing standards calibration optional is applicable over a restricted concentration range at higher concentration levels. The calibration is performed as usual with a blank and at least two calibration standards that bracket the concentration range of interest. The equation used for this option is: The coefficients since it is normally not close to the limited concentration range of interest [8].

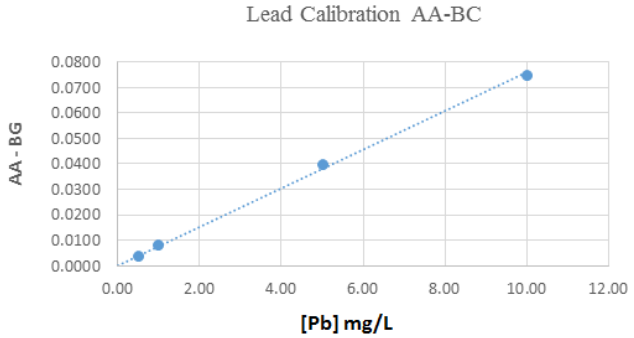

Figure 2. *Linear Calibration- Calculated in Excel.*

Table 3. *Results for lead in eleuthero root- Calculated in Excel.*

Sample mg/L	DF	Absorbance	Blank-corr. Abs	[Pb] mg/L	W (g)	V(mL)	mg/Kg
Water		0.0002					
0	1	-0.0001	-0.0008	-0.12			
Water		0.0011					
0.5 mg/L	1	0.0043	0.0122	1.593			
Water		0.0004					
1 mg/L	1	0.0085	0.0081	1.044			
Water		0.0006					
5 mg/L	1	0.0125	0.0919	4.98			
water		0.0006					
Sample	1	0.0017	0.0011	0.13	1.0025	10	1.27
Water		0.0007					
Sample dup-1	1	0.0017	0.0011	0.12	1.0025	10	1.26
Water		0.0006					
10 mg/L Check Std.	1	0.0725	0.0748	9.88			
Water		0.0008					
Sample Spiked	1	0.0014	0.0014	0.61	1.0025	10	6.12
Water		0.0006					
Sample Spiked duplicate	1	0.0021	0.0014	0.61	1.0025	10	6.12
Water		0.0008					

3.7. Calculation

1) The concentration of lead in the sample extract (in µg/mL) can be calculated using the calibration curve equation (usually in the form of y = mx + b, where y is absorbance, x is the concentration, m is the slope, and b is the intercept).
2) If the sample was diluted during preparation, account for the dilution factor in your final result.

Lead Concentration (µg/mL) = (Sample Absorbance - Intercept) / Slope × Dilution Factor

3) Convert the concentration to the desired units (e.g., µg/g) by considering the weight of the sample and the final volume after dilution.

4. Result and Discussion

Absorption spectra: The absorption spectra of the clear eleuthero solution were recorded using an atomic absorption spectrophotometer. The reagent blank exhibited negligible absorbance, despite having a wavelength in the same region. In all instances, measurements were made at 283.3 nm against a reagent blank [Table 3].

4.1. Quality Assurance / Quality Control

Before spike: sample results: 1.27 mg/Kg, duplicate sample result: 1.26 mg/Kg or ppm.

Average: 1.265 mg/Kg [Table 2].

$$\%RPD = \frac{sample\ result - dup.sample\ result}{average} \text{ x } 100\%$$

$$\%RPD = 0.80\%.$$

Actual known concentration: 5 ppm

$$\%\ \text{Spike Recovery} = \frac{\text{spiked sample} - \text{sample result}}{\text{Actual known nconcentration}} \text{x } 100\%$$

Table 4. Calculation for MS/MSD.

Matrix Spiked Recovery	Matrix Spiked dup. Recovery	Average Spiked Recovery
$\%R_{MS}$=97.0%	%RMSD= 97.2	R%= 97.1%

4.2. Quality Control

1) Run a blank sample (containing no lead) to check for instrument baseline drift.
2) Ensure that the calibration curve is linear within the concentration range used.
3) Run a spike recovery test by adding a known quantity of lead standard to the sample and analyzing to ensure accuracy.
4) Perform replicate measurements to ensure precision.

5. Conclusion

This method provides a reliable and sensitive technique to determine trace amounts of lead in Eleuthero root extracts. By using AAS, the lead concentration can be quantified precisely, ensuring the safety and quality of the herbal extract.

In the present work, a simple, sensitive, selective and inexpensive miscellar method was developed for the determination of lead in eleuthero root extract sample.

Spectrophotometric determination of lead (Pb) in eleuthero root extract sample was found to be adequately sensitive in terms of linearity, repeatability, and accuracy. The correlation coefficient (R2) was found to be 0.99866 [Table 1], average percent recovery was 97.1%, spiked sample, and 98.6% for the duplicate [Table 2]. The results were within the specification of not more than (NMT) 10.0 mg/Kg or ppm maximum, with the average concentration of lead in Eleuthero Root Extract found to be 1.27 mg/Kg. This method was successfully applied to the monitoring of trace amounts of lead in eleuthero root extract sample.

Abbreviations

AAS	Atomic Absorption Spectrometer
FAAS	Flame Atomic Absorption Spectrometer
USP	The United States Pharmacopeia
BP	British Pharmacopeia
EP	European Pharmacopeia

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Author Contributions

Yusuf Yildiz: Formal Analysis, Funding acquisition, Project administration, Writing – original draft

Recep Karadag: Formal Analysis, Project administration, Supervision

Zehra Nilsu Ergin: Formal Analysis, Project administration, Supervision

Conflicts of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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