Section 4. Chemistry

https://doi.org/10.29013/EJTNS-23-3-32-37

Jona Keri,
Department of Pharmacy, Faculty of Medical Sciences,
Aldent University, Tirana, Albania
Lindita Vrushi,
Esencial Herbalist and Producer,
Elbasan-Librazhd Road no, 3401 Elbasan, Albania

THE HEAVY METAL DETECTION IN THE ALBANIAN SALVIA OFFICINALIS, WITH ICP-MS AND UV-VIS SPECTROPHOTOMETER

Abstract. This work aims to study the impact of the method of extracting the essential oil of the sage plant. The identification of heavy metals both in plants and in oil, but in oil extracted with 2 methods, with the classic one using Clevenger's apparatus and with supercritical extraction using CO_2 as a solvent. The results are different.

The results clearly show how the extraction and analysis methods affect the values obtained. Lead was found to be the most abundant metal in the Salvia Officinalis oil, with a concentration of 0.028 parts per million, followed by mercury at 0.015 ppm, according to an analysis utilising the Clevenger equipment and ICP-MS.

After CO_2 extraction, the extract was diluted 1:2 with n-hexane, and ISO 11212 Spectrophotometry was used to check for the presence of heavy metals. Lead, which was detected at the highest concentration (0.22 ppm), was followed by cadmium (0.12 ppm).

All of the values obtained meet the WHO requirements for fruits and vegetables.

Keywords: extraction, heavy metals, ICP-MS, UV–VIS spectrophotometer.

1. Introduction

Salvia officinalis originates from a Latin term for culinary and therapeutic applications. There are different species called Salvia, which are mostly found in Mediterranean European countries and have long been used to cure various diseases such as digestive and blood circulation difficulties, bronchitis, cough, asthma, memory problems, angina, inflammation of the mouth and throat, depression and excessive sweating [1].

Salvia plants have long been known for their antioxidant properties as well as their potential to improve head and brain function, improve memory, sharpen the senses, and slow age-related cognitive decline. Various studies conducted by researchers for Alzheimer's disease have considered sage and Melissa officinalis as some of the plants with 'curative' properties, in improving memory functioning and cholinergic properties [2].

Figure 1. The Salvia officinalis

Although present in different amounts in all examined specimens of S. Officinalis, each of the following essential elements are present: 1,8-cineole, camphor, borneol, bornyl acetate, camphene,– and -thujone, linalool,– and -caryophyllene, -humulene,– and -pinene, pimaradiene, salvianolic acid, rosmarinic etc [3]. Some of the major compounds of sage are presented in the figure below (figure 2).

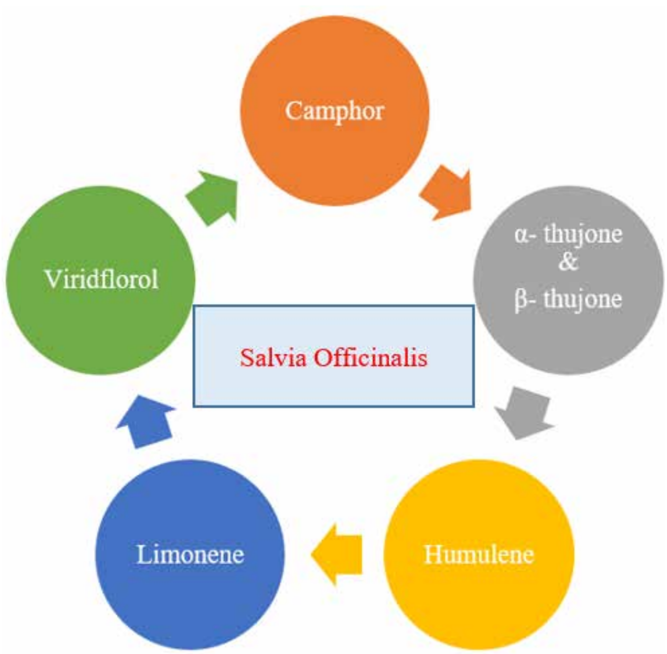

Figure 1. The Salvia Officinalis major compounds

According to research, some biological properties of Salvia essential oil depend on camphor, 1,8-cineole, -thujone, and -thujone [4]. Sage essential oil contains roughly 20% camphor, increasing the camphor level as the leaves expand [5]. According to one study, essential oil's most potent scavenging chemicals are -thujone and -thujone, bornyl acetate, camphor, menthone, and 1,8-cineol. In the same study, the essential oils of Melissa officinalis and Salvia officinalis were found to have higher antioxidant activity than other Lamiaceae species [6].

Camphor is well known for its anti-inflammatory and decongestive effects. Camphor, it could help with nose blockage and congestion, and also it may act as a cough suppressor on the nerves and lessen cough. It may be beneficial against a variety of respiratory diseases.

Alpha and beta thujone as ketones, studies have shown that in moderate concentration they may present beneficial effects [7; 8].

Salvia Officinalis essential oil contains terpene **viridiflorol**, which is utilized in perfumes and flavourings in medicines. Recent research has demonstrated that it has antibacterial properties.

Because of the sunny weather and geographical conditions in Albania, this plant can be found in a variety of locations. It has numerous applications. It is used by women because it aids in the physiological process of organization during the early stages of menopause, and it is thought to aid in the fight against bone porosity. Another reason it is used is because it aids in the prevention of osteoporosis and osteopenia.

It is most commonly used during the winter season in the form of tea, blended with the laurel plant, since it provides respiratory comfort. Sage is also a natural source of flavonoids and polyphenolic chemicals (e.g., carnosic acid, rosmarinic acid, and caffeic acid) with antioxidant, radical-scavenging, and antibacterial properties [9].

In this study, we can evaluate and compare the presence of the heavy metals in salvia officinalis in 2 methods, with the classical extraction using the Clevenger apparatus and using the SC–CO_2 extraction method. After obtaining the essential oil we were able to evaluate their presence using the ICP-MS for the classical extraction and using the UV–VIS Spectrophotometer for the examination of the essential oil obtained with the SC–CO_2.

Heavy metal poisoning can be caused by ingesting foods, medications, improperly sealed food containers, occupational exposure to heavy metals, contaminated air or water, or lead-based paint. However, if these metals accumulate to a dangerous level in the body, they could cause serious harm. The heavy metals that have been linked to human poisoning the most common are *lead, mercury, cadmium, tin, and arsenic.*

Because they are toxic, thermally and biologically inert, heavy metals typically build up in living things. Due to these metals' extreme toxicity, capacity to bioaccumulate throughout the food chain, ubiquity, and durability in the environment, numerous health problems, including organ failure, have been linked to their consumption of food and water [10].

2. Materials and methods

The following materials were required for the completion of this work: an SC–CO_2, hexane, sage, hydrochloric acid, nitric acid, and nitric acid Clevenger's apparatus.

This method was used in this study to identify metals in plants and essential oils.

2.1 The hydro distillation distillation

This method is used to extract essential oils.– Steam or boiling water is employed in hydro distillation to remove the essential oil from the aromatic plant. Hot water or steam is used to extract the essential oils from the oil glands, which are found in the plant tissue. The separated water and oil (vapour combination) are converted into liquid in the condenser and then transported to the separator to separate the essential oil from the water.

The multi-element stock standard solution (10 LG mL-1) was used to construct the elemental calibration standard. H_2O_2 (30%) and HNO3 (Suprapure grade, 65%) were purchased from Merck [11].

The process of cleaning the dry leaves of contaminants and other solid substances removed them from their natural packaging. The samples were comminuted prior to hydrodistillation extraction, and then the hydrodistillation procedure started. A distillation flask heater and a Clevenger-style apparatus were used to boil the sample, which contained 50 grammes of tea, for three hours at boiling point. To prevent any polyphenols or other vital oil components from coming into contact with the water during the first 30 minutes after the oil began to gather in the Clevenger column, around 1 mL of hexane was added to the condenser.

2.2 Supercritical CO_2 extraction

Carbon dioxide is turned into a liquid at high pressure and passed over the natural substance using a process called supercritical CO_2 extraction to remove all the smell components without harming the material. The CO_2 reaches the supercritical state at 31.1^0 C and 72.87723 atm of pressure. SC–CO_2, just like with natural products. More effectively extracts the scents than steam distillation, but maybe more significantly, when the environment is restored, the CO_2 turns into a gas and disappears, leaving no trace behind. The smell is pure and undisturbed. No taste or odour of a solvent is present.

Before entering the separators, the supercritical CO_2 travels through a series of pressure-regulating valves to separate the oils from the plant (Salvia officinalis). To separate the various extract components, the separator's two pressures are independently regulated. After the oil is removed and the CO_2 that is emitted as gas is separated, the CO_2 is recycled by condensing and storing it as a liquid in the tank. The oil is then put into containers for collection. This CO_2 is then used throughout the batch to repeat the complete cycle.

3. Results and discussions

By evaluating specificity, linearity, the limit of detection and quantification, precision, and accuracy, the devised assay analytical method was put through validation.

Using the digested and extracted oil samples, ICP-MS was used to determine the presence of heavy metals in the herbal tea sample (Salvia officinalis L.), which was obtained from an herbalist in North Albania [11].

Table 1.– Detection of heavy metals in herbal leaves of *Salvia officinalis L.*

Heavy Metals	Results	UdM	Method
Arsenic	0.055	mg/kg	Meth. (151)
Cadmium	0.005		
Mercury	0.023		
Lead	0.242		

The first analysis performed was to detect the presence of heavy metals in herbal leaves, From the results obtained it can be seen clearly that the leaves contain lead in higher concentrations, at 0.242 ppm, followed by arsenic at 0.055 ppm.

3.1 Presence of the heavy metals in the essential oil:

3.1.1 Essential oil of the salvia officinalis with classical extraction

The percentage of oil after extraction was 3.7%.

Table 2.– Detection of heavy metals in the extracted oil of *Salvia officinalis L.*

Heavy Metals	Results	UdM	Methods
Arsenic	<0.001	mg/kg	Met. (151)
Cadmium	0.002		
Mercury	0.015		
Lead	0.028		

The most abundant metal present in the extracted oil of *Salvia Officinalis* is Lead at 0.028 ppm, followed by mercury at 0.015 ppm.

3.2 Essential oil of the salvia officinalis with classical extraction, CO_2 extraction

Table 3.– Detection of heavy metals in the extracted oil of *Salvia officinalis L,* using the CO_2 extraction

Heavy Metals	Results	Specifications	Methods
1	*2*	*3*	*4*
Arsenic	0.03 ppm	< 1.50	ISO 11212 SPECROFOTOMETRY

1	2	3	*4*
Cadmium	0.12 ppm	< 1.00	
Mercury	< 0.06 ppm	< 0.10	
Lead	0.22 ppm	< 5.00	

After the CO_2 extraction, the extract was diluted with n-hexane at a ratio of 1:2, and the analysis with the ISO 11212 Spectrophotometry have shown the presence of the heavy metals, where lead is the most abundant metal present at 0.22ppm, followed by cadmium at 0.12 ppm.

4. Conclusions

The first investigation was to examine for the presence of heavy metals in herbal plants, and the results show that lead is present in the leaves in higher amounts than arsenic, at 0.242 ppm, obviously.

The examination of the Salvia Officinalis oil using the Clevenger apparatus and ICP-MS revealed that lead was the most prevalent metal present at 0.028 ppm, followed by mercury at 0.015 parts per million.

Following CO_2 extraction, the extract was diluted 1:2 with n-hexane, and the presence of heavy metals was detected using ISO 11212 Spectrophotometry. Lead was the most abundant metal, present at 0.22 ppm, followed by cadmium at 0.12 ppm.

The concentrations of lead decreased from the plant to the essential oil obtained from the hydro distillation and the values were detected with ICP-MS, from 0.242 ppm to 0.028 ppm, which reflects the properties of the microwave and then the ICP-MS properties and advantages for the detection.

But apart from these values, the presence of lead at 0.22ppm is present even in the essential oil obtained from the SC CO_2 extraction. Furthermore, it is significant to highlight that compared to other methods, CO_2 extracts are more similar in composition to the oil found naturally in botanical plants. Best as a flavoring and fantastic fragrance ingredient: better solubility, more top notes, more back notes, and no solvent residue or off notes.

All the values obtained conform to the WHO standards for vegetables and fruits.

References:

1. Salvia (Sage): A Review of its Potential Cognitive-Enhancing and Protective Effects Adrian L. Lopresti, Drugs R D. Mar; – 17(1). 2017.– P. 53–64. Doi: 10.1007/s40268–016–0157–5
2. Pickering A. T., Wang W. W., Houghton P. J., Perry N. S. 'Medicinal plants and Alzheimer's disease: from ethnobotany to phytotherapy' Affiliations expand, PMID: 10411211. DOI: 10.1211/0022357991772808
3. Mohsen Hamidpour, Rafie Hamidpour, Soheila Hamidpour, and Mina Shahlari, J Tradit. Complement Med. 'Chemistry, Pharmacology, and Medicinal Property of Sage (Salvia) to Prevent and Cure Illnesses such as Obesity, Diabetes, Depression, Dementia, Lupus, Autism, Heart Disease, and Cancer', Apr-Jun; – 4(2). 2014.– P. 82–88. Doi: 10.4103/2225–4110.130373
4. Radulescu V., Chiliment S., Oprea E. Capillary gas chromatography-mass spectrometry of volatile and semi-volatile compounds of Salvia officinalis. J Chromatogr.– 1027. 2004.– P. 121–6. [PubMed] [Google Scholar].

5. Avato P., Fortunato I., Ruta C., D'., Elia R. Glandular hairs and essential oils in micro propagated plants of Salvia officinalis L. Plant Sci.– 169. 2005.– P. 29–36. [Google Scholar].
6. Hussain A., Anwar F., Iqbal T., Bhatti I. Antioxidant attributes of four Lamiaceae essential oils. Pak J Bot.– 43. 2011.– P. 1315–21. [Google Scholar].
7. Scott R. P.W. 'Essential Oils', in Encyclopedia of Analytical Science (Second Edition), 2005.
8. Karin M. Höld,* Nilantha S. Sirisoma,* Tomoko Ikeda,† Toshio Narahashi,† and John E. Casida. 'α-Thujone (the active component of absinthe): γ-Aminobutyric acid type A receptor modulation and metabolic detoxification'.
9. Baranauskiene R., Dambrauskiene E., Venskutonis P. Influence of harvesting time on the yield and chemical composition of sage (Salvia officinalis L.) Foodbalt. 2011.– P. 105–9. [Google Scholar].
10. The use of FNMs-based electrochemical sensors in the food and beverage industry. Masoud Reza Shishehbore, Mohadeseh Safaei, in Functionalized Nanomaterial-Based Electrochemical Sensors, 2022.
11. Keri Jona, Memushaj Lorena, Xhangoli Ina, Hasa Evelina. 'Detection of heavy metals in the sideritis raeseri (çaj mali) provided at albanian pharmacies'. New trends and strategies in the chemistry of advanced materials with relevance in biological systems, technique and environmental protection,– 14th edition, October 20–21, 2022. Timisoara, – Romania,– 46 p.