*Pharmaceutical Sciences*
*September 2018, 24, 235-239*
doi: 10.15171/PS.2018.34
*http://journals.tbzmed.ac.ir/PHARM*

*Research Article*

# Nanoporous Silica-Polypyrrole/SBA-15 as Fiber Coated in the Solid-Phase Microextraction for Determination of *Salvia hydrangea* DC. Essential Oil

**Roghayeh Hendi[1], Marzieh Piryaei[2*], Marzieh Babashpour Asl[1], Mir Mahdi Abolghasemi[3]**

[1]*Department of Horticulture, College of Agriculture, Maragheh Branch, Islamic Azad University, Maragheh, Iran.*
[2]*Maragheh University of Medical Sciences, Maragheh, Iran.*
[3]*Department of Chemistry, Faculty of Science, University of Maragheh, Maragheh, Iran.*

**Article Info**

***Article History:***
Received: 26 December 2017
Revised: 27 March 2018
Accepted: 2 April 2018
ePublished: 23 September 2018

***Keywords:***
-Nanoporous
-Silica-polypyrrole/sba-15
-Microextraction
-*Salvia hydrangea*

**ABSTRACT**

***Background:*** In the current study, simultaneous extraction and concentration of the analytes from the headspace on the solid phase microextraction (SPME) fiber were followed by transferring the essential oil components of *Salvia hydrangea* DC to the headspace with the help of a heating source.
***Methods:*** The essential oil components of the plant were extracted and concentrated in a single step. A one-at-the-time optimization procedure was applied to the microextraction conditions by using the nanocomposite fiber of polypyrrole/SBA-15.
***Results:*** The results of the essential oil extraction of Salvia hydrangea clearly demonstrated that polypyrrole/SBA-15 fibers were efficient with SPME technique. The acceptable SPME precision in our study was corroborated by the relative standard deviation (RSD) values of less than 15%.
***Conclusion:*** In comparison to the hydro-distillation (HD) method, the SPME technique could equally provide monitoring of almost all the constituents of the studied essential oil in an easier way and shorter time with much lower amounts of the plant sample. The results of the essential oil extraction of *Salvia hydrangea* clearly demonstrated that polypyrrole/SBA-15 fibers were efficient with SPME technique.

## Introduction

*Salvia* L., (sage), which is one of the largest Lamiaceae genera, includes 1000 species worldwide, 55 species of which occur in Iran. This genus has almost a cosmopolitan distribution and can be found all around the world, i.e., in tropical, subtropical, temperate, arctic, and subarctic zones.[1,2] The aerial parts of *Salvia* genus usually contain bioactive compounds: flavonoids (such as luteolin-7-glucoside and apigenin), phenolic acids (such as rosmarinic acid, caffeic acid, and ferulic acid), and triterpenoids (such as ursolic acid), as well as essential oils (such as monoterpenoids).[3] Some members of this genus serve as flavoring agents in perfumery and cosmetics, as well as in food industry.[4] Being endemic to Iran, *Salvia hydrangea* DC. has been widely utilized in traditional Iranian medicine as carminative, spasmolytic, and anodyne, as well as for its anti-inflammatory effect.[5,6] In recent years, studies on the chemical compounds of medicinal plants have generally focused on the phenolic compounds and volatile oils. As a recommended approach employed in Chinese pharmacopoeia, Hydro-Distillation (HD) method associated with GC/MS can provide volatile oil analyses in medicinal plants. Nonetheless, a great quantity of plant samples, much energy, and a long time is often needed to perform HD method.[7] Compared to the traditional techniques of sample preparation, (SPME) provides a simple, quick, and efficient approach to prepare green samples. This method allows sampling, extraction, and enrichment of medicinal plants in one single step.[8,9] Although varied types of commercial SPME fibers have been applied in this process,[10] they have not usually had favorable chemical and thermal stabilities in addition to having a poor reusability and high cost. To obtain the desired analysis results, a suitable fiber coating for SPME should be selected. Several nanostructured metal oxides have been recently synthesized to coat and fabricate fibers.[11,12] A number of commercially available SPME fibers, including calyx open-chain crown ether, and polymer materials coupled with new fiber coating techniques, such as solid sorbent utilization and HPLC stationary phases for the chemical bonding of silica have been introduced in the literature to extract a variety of compounds. Moreover, to prepare SPME fibers, various methods like vapor deposition, physical deposition, electrodeposition, electrophoretic deposition, sol–gel technology, and direct

***Corresponding Author:** Marzieh Piryaei, E-mail: m.piriyaei@gmail.com
©2018 The Authors. This is an open access article and applies the Creative Commons Attribution (CC BY), which permits unrestricted use, distribution and reproduction in any medium, as long as the original authors and source are cited. No permission is required from the authors or the publishers.

application of uncoated fibers have been suggested.[13] As a promising versatile material, silica fiber can provide a supporting substrate fiber for SPME though its utilization has been limited by its fragility and easy functionalization. Nevertheless, silica-polypyrrole / SBA-15 as a nanoporous type of silica has been shown to attach to copper wire after functionalizing with 3-[bis (2-hydroxyethyl)amino] propyl-triethoxysilane and subsequently provide a new SPME fiber with a significantly higher Energy Equivalent Speed (EES) for phenol removal compared to the commercial polydimethylsiloxane (PDMS) fibers.[14] Therefore, the current study aimed at investigating the extraction efficiency of coated nanoporous silica-polypyrrole/SBA-15 fibers for determining the essential oil constituents of *Salvia hydrangea* DC. in SPME as compared to the traditional HD approach.

## Materials and Methods

### *Plant materials*

The aerial parts of *Salvia hydrangea* were harvested from north-west of Iran during the flowering season in 2015. The plant materials were dried in air and stored in sealed bags in a cool place. A voucher specimen was deposited at the herbarium of the Department of Chemistry, Faculty of Science, University of Maragheh under accession code 1375SH.

### *Chemicals and reagents*

All the chemicals and reagents were purchased from Merck Company (Darmstadt, Germany). Tetraethylorthosilicate (TEOS) as a silica source and poly (ethylene glycol)-block-poly (propylene glycol)-block-poly (ethylene glycol) (EO20-PO70-EO20 or Pluronic P123) as a surfactant were prepared from Sigma-Aldrich Company (Steinheim, Germany). Double-distilled water was used throughout the experiment.

### *HD apparatus and procedure*

After grinding 100 g of the air-dried aerial parts of *S. hydrangea*, the plant sample was subjected to hydro-distillation for 2 h with the help of a Clevenger-type apparatus according to the recommended guidelines. Upon immersion of the plant into water and its heating to the boiling point, the evaporated volatile oil was collected in a condenser together with water vapor. The isolated distillate was dried over anhydrous sodium sulfate. A dry weight of 0.32% (w/w) of the yellowish oil of the plant aerial parts was yielded, which was then stored at 4˚C to be later evaluated through GC/MS analysis.

### *GC/MS analysis*

The chemical composition of the oil was analyzed by using a Hewlett-Packard Agilent 7890A series GC equipped with a split/splitless injector. An Agilent 5975C mass-selective detector system was used for the determination. The injection volume was 0.1 µL in *n*-hexane and the injector temperature was set at 260°C in the splitless mode for 2 min. The oven temperature was programmed at 50˚C, which was increased to 180˚C at a rate of 15˚C $min^{-1}$. Then, it was raised to 260˚C at a rate of 20˚C $min^{-1}$ and maintained for 5 min. Helium (99.999%) was utilized as a carrier gas at a flow rate of 1 mL $min^{-1}$ for the analysis. The MS was taken at 70 eV in the EI mode. The GC/MS interface, quadrupole, and ion source temperatures were set at 280˚C, 150˚C, and 230˚C, respectively. The components of the essential oil were identified using the Wiley 7N (Wiley, New York, NY, USA) Mass Spectral Library. The fiber was conditioned in the injection port of the GC for 1 h. A homemade SPME device was employed for holding and injecting the fabricated fiber into the GC/MS injection port.

### *Preparation of polypyrrole/SBA-15 nanocomposites*

The polypyrrole/SBA-15 nanocomposite fiber was prepared as reported earlier. Briefly, polypyrrole/SBA-15 particles were prepared by a chemical polymerization method. To prepare the polypyrrole/SBA-15 nanocomposites, calcined SBA-15 material was dried under vacuum at 250°C for 6 h to remove air and water from the channels. Then, monomer pyrrole was adsorbed into the pores of SBA-15 through vapor at room temperature for 24 h. After wards, SBA-15 containing pyrrole was immersed in 0.25 M of $FeCl_3$.6H2O aqueous solution and stirred at room temperature for 24 h. Finally, the products were washed several times with deionized water and acetone and then dried in a vacuum at 40˚C for 24 h.

## Results and Discussion

In the current study, simultaneous extraction and concentration of the analytes from the headspace on the SPME fiber were followed by transferring the essential oil components of *Salvia hydrangea* DC to the headspace with the help of a heating source. Therefore, the essential oil components of the plant were extracted and concentrated in a single step. A one-at-the-time optimization procedure was applied to the microextraction conditions by using the nanocomposite fiber of polypyrrole/SBA-15. A varied extraction temperature of 50-90˚C was considered as shown in Figure 1. Either the total or individual peak areas were found to enhance with the temperatures of up to 80˚C, which then led to a leveling-off state. The significant impact of the extraction temperature on the extraction process was evidenced due to its effect on the compound distribution between the headspace and the fiber/sample. These results made us choose the final temperature of 80˚C for this work. The profiles of the total and individual peak areas revealed the highest peak area for the target compounds to occur at 40 min. Any longer times of extraction had no impacts on the extraction efficiency. The results of the extraction time range of 15-50 min are displayed in Figure 2. Increasing the amount of a sample generally leads to elevated analyte signals; however, extraction efficiency may be influenced by its large amounts. This does not mean that better results can be achieved by larger amounts of a sample. In the current

research, a range of 1-4 g of the sample amount was utilized. The results of their effects on the total and individual peak areas of the 4 studied compounds are presented in Figure 3. As expected, the mentioned peak areas leveled off with the amounts of more than 3 g after augmenting up to this amount. It has been reported that the extraction efficiency may be significantly affected by humid samples in the SPME experiment.

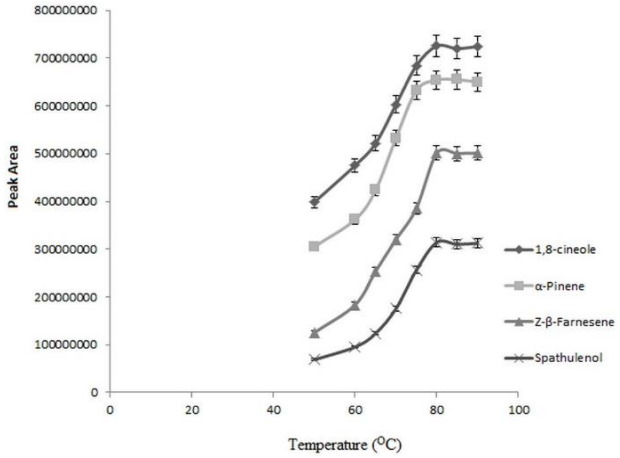


**Figure 1.** Effect of extraction temperature on the extraction efficiency.

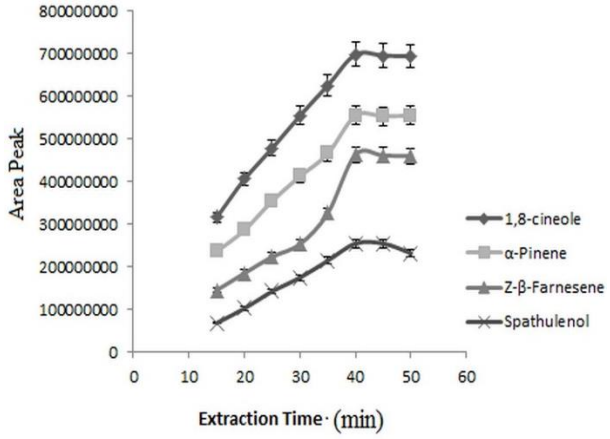


**Figure 2.** Effect of extraction time on the extraction efficiency.

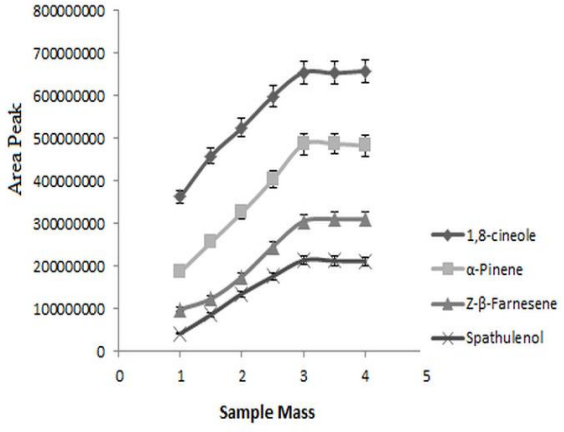


**Figure 3.** Effect of sample weight on the extraction efficiency.

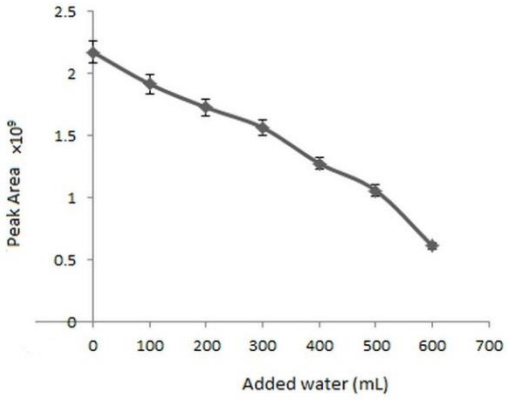


**Figure 4.** Effect of added water on the extraction efficiency.

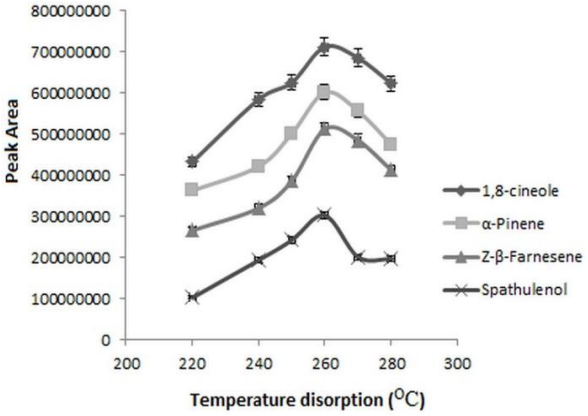


**Figure 5.** Effect of desorption temperatures on the extraction efficiency.

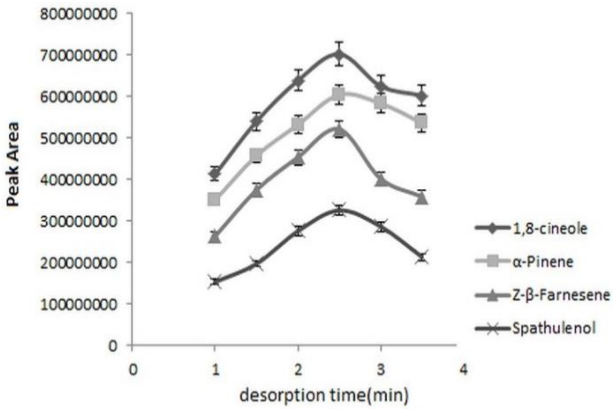


**Figure 6.** Effect of time temperatures on the extraction efficiency.

Hence, varied amounts of water were added to the samples under optimized conditions to investigate humidity effects on the 4 compounds, the results of which are exhibited in Figure 4. The previous reports could be verified by the results of this study, which were indicative of a reduction in the total and individual peak areas induced by water vapor in the headspace atmosphere. This was implicative of the fiber surface deactivation caused by water molecules blocking the active sites. The proposed fiber showed a good adsorption for the dried samples.

**Table 1.** Chemical composition of *S. hydrangea* DC. essential oil.

| No | Compounds | RI[a] | HD, Area%[b] | HS-SPME, Area%[c] | Repeatability R.S.D.%[d] |
|---|---|---|---|---|---|
| **1** | **α-Thujene** | 928 | 1.2 | 0.8 | 10.5 |
| **2** | **α-Pinene** | 937 | 7.2 | 5.5 | 6.4 |
| **3** | **Camphene** | 955 | 2.8 | 2.6 | 5.2 |
| **4** | **Sabinene** | 975 | 1.6 | 1.5 | 9.3 |
| **5** | **β-pinene** | 978 | 5.5 | 6.2 | 7.1 |
| **6** | **Myrcene** | 996 | 0.7 | 0.2 | 10.2 |
| **7** | **δ-3-Caren** | 1010 | 0.8 | 0.2 | 11.3 |
| **8** | **α-Terpinene** | 1020 | 1.1 | 0.3 | 8.5 |
| **9** | **p-cymene** | 1025 | 0.9 | 0.2 | 8.2 |
| **10** | **1,8-cineole** | 1035 | 12.7 | 10.5 | 4.3 |
| **11** | **Z- β -Ocimene** | 1038 | 5.4 | 3.7 | 7.6 |
| **12** | **E- β-Ocimene** | 1045 | 2.5 | 1.2 | 10.6 |
| **13** | **γ-Terpinene** | 1062 | 1.3 | 1.6 | 5.3 |
| **14** | **Terpinolene** | 1081 | 0.7 | 0.2 | 11.4 |
| **15** | **linalool** | 1102 | 0.6 | 0.1 | 9.5 |
| **16** | **Camphor** | 1139 | 0.3 | 0.5 | 7.4 |
| **17** | **borneol** | 1160 | 2.6 | 2.1 | 10.6 |
| **18** | **Terpinen-4-ol** | 1173 | 0.2 | 0.1 | 11.3 |
| **19** | **α-Terpineol** | 1190 | 3.4 | 3.5 | 9.4 |
| **20** | **1-dodecene** | 1192 | 0.1 | 0.2 | 6.1 |
| **21** | **Verbanol** | 1196 | 0.3 | 0 | - |
| **22** | **Gerani** | 1253 | 0.1 | 0.1 | 8.5 |
| **23** | **Bornyl acetate** | 1286 | 0.1 | 0 | - |
| **24** | **Piperonal** | 1325 | 1.4 | 0.6 | 6.8 |
| **25** | **Eugenol** | 1355 | 1.5 | 0.7 | 10.1 |
| **26** | **α-Copaene** | 1378 | 0.2 | 0 | - |
| **27** | **β-bourbonene** | 1384 | 0.4 | 0.3 | 11.2 |
| **28** | **Cubebene** | 1390 | 0.2 | 0.1 | 11.6 |
| **29** | **Z-jasmone** | 1391 | 2.7 | 2.1 | 9.4 |
| **30** | **E-caryophyllene** | 1420 | 21.3 | 19.8 | 10.5 |
| **31** | **Z-β-Farnesene** | 1442 | 4.4 | 4.3 | 9.6 |
| **32** | **α-humulene** | 1453 | 1.1 | 1.3 | 8.5 |
| **33** | **γ-muurolene** | 1475 | 0.5 | 0.2 | 9.1 |
| **34** | **Germacrene D** | 1480 | 4.2 | 3.1 | 9.6 |
| **35** | **Bicyclogermacrene** | 1493 | 1.3 | 0.6 | 10.1 |
| **36** | **β-bisabolene** | 1506 | 0.5 | 0.2 | 6.7 |
| **37** | **γ-cadinene** | 1510 | 0.6 | 0 | - |
| **38** | **δ-cadinene** | 1523 | 2.1 | 2.0 | 10.3 |
| **39** | **Spathulenol** | 1575 | 2.3 | 1.2 | 9.5 |
| **40** | **Cedrol** | 1596 | 0.6 | 1.1 | 4.8 |
| **41** | **α-cadinol** | 1653 | 0.2 | 0.3 | 7.3 |

a) Retention indices using a HP-5MS column (relative retention times normalize to closely eluting *n*-alkanes).
b) Relative area (peak area relative to total peak area) for hydrodistillation method.
c) Relative area (peak area relative to total peak area) for HS-SPME method.
d) RSD values for HS-SPME method ($n$ = 5).

Different times and temperatures were tested to determine the optimal desorption conditions based on the desorption of all the analytes in the fiber coating process with minimal carry-over impacts on the analyses. Consequently, the optimized desorption temperatures and times for polypyrrole/SBA-15 SPME fiber were selected to be 220-280°C (Figure 5) and 1-3.5 min (Figure 6), respectively. After stabilizing the chromatograms, the peak areas were reproduced for the sample at 260°C with the desorption time of 2.5 min. Our subsequent studies were based on the obtained temperature and desorption time values. The replicate analyses provided us with the peak areas to calculate the values of Relative Standard Deviation (RSD) (Table 1). The acceptable SPME precision in our study was corroborated by the RSD values of less than 15% as depicted in Table 1.

## Conclusion

In comparison to the HD method, the SPME technique could equally provide monitoring of almost all the constituents of the studied essential oil in an easier way and shorter time with much lower amounts of the plant sample. The results of the essential oil extraction of *Salvia hydrangea* clearly demonstrated that polypyrrole/SBA-15 fibers were efficient with SPME technique.

## Conflict of Interests

The authors claim that there is no conflict of interest.

## References

1. Hedge IC. Labiateae. In: Flora Iranica, Rechinger KH editors. Graz, Austria: Akademische Druckund Verlagsanstalt; 1982. p. 403- 76.
2. Walker JB, Sytsma KJ, Treutlein J, Wink M. *Salvia* (Lamiaceae) is not monophyletic: implication for the systematics, radiation, and ecological specialization of *Salvia* and Tribe Mentheae. Am J Bot. 2004;91(7):1115-25. doi:10.3732/ajb.91.7.1115

3. Lima CF, Andrade PB, Seabra RM, Fernandes-Ferreira M, Pereira-Wilson C. The drinking of a *Salvia officinalis* infusion improves liver antioxidant status in mice and rats. J Ethnopharmacol. 2005; 97(2): 383-9. doi:10.1016/j.jep.2004.11.029
4. Kotan R, Kordali S, Cakir A, Kesdek M, Kaya Y, Kilic H. Antimicrobial and insecticidal activities of essential oil isolated from Turkish *Salvia hydrangea* DC. Ex Benth. Biochem Syst Ecol. 2008;36(5-6):360-8. doi:10.1016/j.bse.2007.12.003
5. Ji XY, Tan BK, Zhu YZ. *Salvia miltiorriza* and ischemic diseases. Acta Pharmacol Sin. 2000;21(12):1089-94.
6. Lu Y, Foo LY. Polyphenolic in *Salvia*. Phytochemistry: a review. 2002;59(2):117-40. doi:10.1016/s0031-9422(01)00415-0
7. Wang L, Wang Z, Zhang H, Li X, Zhang H. Ultrasonic nebulization extraction coupled with headspace single drop microextraction and gas chromatography-mass spectrometry for analysis of the essential oil in *Cuminum cyminum* L. Anal Chim Acta. 2009;647(1):72-7. doi:10.1016/j.aca.2009.05.030
8. Arthur CL, Killam LM, Buchholz KD, Pawliszyn J, Berg JR. Automation and optimization of solid-phase microextraction. Anal Chem. 1992;64(17):1960-6. doi:10.1021/ac00041a034
9. Xia XR, Leidy RB. Preparation and characterization of porous silica-coated multifibers for solid-phase microextraction. Anal Chem. 2001;73(9):2041-7. doi:10.1021/ac001273f
10. Pawliszyn J. (Ed.), Applications of Solid-phase Microextraction, RSC, Cornwall, UK; 1999. doi.org/10.1016/S0021-9673(00)00309-5
11. Alizadeh R, Najafi NM, Kharrazi Sh. A new solid phase micro extraction for simultaneous head space extraction of ultra traces of polar and nono-polar compounds. Anal Chim Acta. 2011;689(1):117-21. doi:10.1016/j.aca.2011.01.013
12. Mehdinia A, Mousavi MF, Shamsipur M. Nano-structured lead dioxide as a novel stationary phase for solid-phase microextraction. J Chromatogr A. 2006;1134(1-2):24-31. doi:10.1016/j.chroma.2006.08.087
13. Wang JX, Jiang DQ, Gu ZY, Yan XP. Multiwalled carbon nanotubes coated fibers for solid-phase microextraction of polybrominated diphenyl ethers in water and milk samples before gas chromatography with electron-capture detection. J Chromatogr A. 2006;1137(1):8-14. doi:10.1016/j.chroma.2006.10.003
14. Abolghasemi MM, Piryaei M. Fast determination of *Prangos uloptera* essential oil by nanoporous silica-polypyrrole SPME fiber. Chemija. 2012;23(3):244-9.