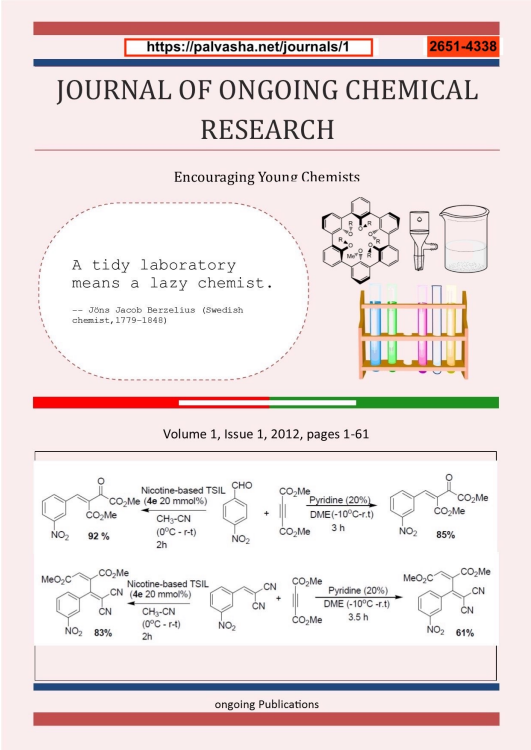
COMPONENTS AND ANTIMICROBIAL ACTIVITY OF ESSENTIAL OIL OF AERIAL PARTS OF SAGE (*SALVIA OFFICINALIS* L.)



JOURNAL OF ONGOING CHEMICAL RESEARCH

2018
Volume: 3
Issue: 2
Pages: 63-66
Document ID: 2018JOCR26

doi:10.5281/zenodo.5517838

J Ong Chem Res (2018), Volume: 3, Issue: 2, pages: 63-66, Document ID: 2018JOCR26
Research Article

Components And Antimicrobial Activity Of Essential Oil Of Aerial Parts Of Sage (*Salvia Officinalis* L.)

Elif Ayşe Erdoğan Eliuz*, Yusuf Sıcak,
For affiliations and correspondence, see the last page.

Abstract

In the present study, the volatile compounds of aerial parts of *Salvia officinalis* L. (sage) were extracted and analysed by gas chromatography-mass spectrometry (GC-MS) using the Nist and Willey libraries. It was determined that the main components of sage were camphor (26.05%), *α*-thujone (17.46%), 1.8-cineole (12.11%), viridiflorol (6.95%) and *β*-thujone (3.57%). Then, antimicrobial activity of essential oil of sage against *Escherichia coli* (ATCC 25293), *Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 25925), *Pseudomonas aureginosa*, *Candida albicans* and *Candida parapsilosis* were examined by using spectrophotometric broth microdilution method. Accordingly, the highest MIC value of *S. officinalis* essential oil was against *S. aureus* (11.7 µg/mL), while the lowest activity was *P. aureginosa* (109.9 µg/mL). Furthermore, MIC values for the other microorganisms were determined as 11.8 µg/mL for *B. subtilis*, 59.0 µg/mL for *E. coli*, 25.3 µg/mL for *C. albicans* and 15.4 µg/mL for *C. parapsilosis.* As a result, essential oil of aerial parts of *S. officinalis* were noted to be high antimicrobial efficiency under concentration of 110 µg/mL.

Keywords: natural products, Salvia officinalis L., Chemical composition, Antimicrobial activity, Aerial part,

INTRODUCTION

Salvia officinalis L. (sage) is a plant that has been used for medical purposes since ancient times. It is one of the most widely known species of the Labiateae family. *Salvia* are grayish-green leaves with single or perennial grasses or small bushes, stems are steep and quadrangular plants [Davis et al, 1965]. It is widely used in the food, pharmaceutical and cosmetic industry due to its pleasant scent, taste, aroma and therapeutic properties of the plant. The extracts of the plant contain important compounds for the treatment of various diseases such as asthma, bronchitis, cirrhosis, coronary heart disease and Alzheimer's. The essential oils in its composition are proven to be a strong antiseptic and used in pharmaceutical formations [Ceylan, 1976; Baricevic et al, 2001; SARICI et al, 2004;Aleksovski et al, 2007].

Dozens of studies on the antimicrobial properties of *S. officinalis* parts were recorded. However, to the best of our knowledge, there are few studies comparing the studies related to antimicrobial activities of the aerial parts of the sage. Therefore, in the present study, it is aimed to noted antimicrobial activity of essential oil from aerial parts of sage and compare with the similar studies.

RESULTS AND DISCUSSION

We extracted volatile oils of *S. officinalis* by water vapor distillation and analyzed them in order to identify the differences in oil composition by comparing the relative retention times and mass spectra from GC-MS data library. The results of the chemical composition of the essential oil were presented in Table 1.

Table 1: Essential oil composition of S. officinalis

RT (min)	Component	Quantity (%)	RT (min)	Component	Quantity (%)
8.120	Salvene	0.45	19.044	**β-Thujone**	3.57
8.843	Tricyclene	0.76	19.549	**Camphor**	26.05
9.025	α-Thujene	0.52	20.367	**Borneol**	2.16
9.547	**α-Pinene**	1.23	21.506	α-Terpineol	1.08
10.056	**Camphene**	1.44	22.725	Myrtenol	0.66
10.489	Sabinene	0.97	24.641	Carvone	0.94
11.212	β-Pinene	1.54	25.328	Sabinene hydrate acetate	0.48
11.773	Myrecene	1.90	26.700	**Bornyl acetate**	1.18
12.761	α-Terpinene	1.06	28.611	Thymol	0.67
13.036	*p*-Cymene	0.75	30.268	Carvacrol	0.94
13.770	**Limonene**	1.24	31.065	β-Bourbonene	0.82
13.953	**1,8-Cineole**	12.11	32.144	**α-Humulene**	2.09
15.269	(*Z*)- β-Ocimene	0.36	33.641	*allo*-Aromadendrene	0.63
16.364	(*E*)- β-Ocimene	0.81	35.357	γ-Gurjunene	0.90
17.184	γ-Terpinene	0.69	36.066	δ-Cadinene	0.65
17.846	Terpinolene	0.42	37.487	σ-Candinene	1.04
18.079	**α-Thujone**	17.46	38.025	Caryophyllene oxide	0.55
18.346	**Linalool**	1.51	40.367	**Viridiflorol**	6.95

RT: Retention Time

In present study, camphor (26.05%), α-thujone (17.46%), 1.8 cineole (12.11%), viridiflorol (6.95%) and β-thujone (3.57%) were the major component in the essential oil of *S. officinalis* aerial parts, followed by borneol (2.16%), *α*-humulene (2.09%), myrecene (1.90%), β-pinene (1.23), camphene (1.44%), limonene (1.24%), α-pinene (1.23%), bornyl acetate (1.18%), *α*-Terpineol (1.08%), α-terpinene (1.06%), *σ*-Candinene (1.04%) and less amounts than 1.0% with carvone, myrtenol, tricyclene.

Many *Salvia* species have commercial important and in traditional medicine and aromatherapy due to many active substances [Walch et al, 2011]. Mostly, the chemical nature of *Salvia* species have similar. However, numerous factors such as ecological,

morphological and genetic diversity have affected the composition essential oil of the plant [Li et al, 2015; Figueiredo et al, 2008]. In our study, essential oil of aerial parts of sage mainly consisted of camphor, α-thujone, 1.8 cineole, viridiflorol and β-thujone. However, the main component of essential oils of aerial parts of some *Salvia* genus were caryophyllene oxide and beta-caryophyllene for *S. hydrangea* and *S. ballotiflora* [Sonboli et al, 2006;CardenasOrtega et al, 2015]; cadinol and germacrene D and aromadendrene oxide for aerial parts of *S. miltiorrhiza;* terpineol, limonene, and eudesmol for *S. przewalskii* and thujene for *S.officinalis* [Li et al, 2015].

The antimicrobial activity of essential oil of aerial parts of *S. officinalis* were researched on several pathogens, namely *E. coli*, *B. subtilis, S. aureus, P. aureginosa, C. albicans* and *C. parapsilosis* using spectrophotometric microdilution method. Ampiciline and fluconazole antibiotics were used as positive control standards for bacteria and fungal strains, respectively. They inhibited microorganisms at all concentration more than 0.48 µg/mL. Antimicrobial activity of essential oil aerial parts of sage were showed in Figure 1. The MICs of sage results were 11.7 µg/ml for *S. aureus,* 11.8 µg/mL for *B. subtilis*, 15.4 µg/mL for *C. parapsilosis,* 25.3 µg/mL for *C. albicans,* 59.0 µg/mL for *E. coli*, 109.9 µg/mL for *P. aureginosa*.

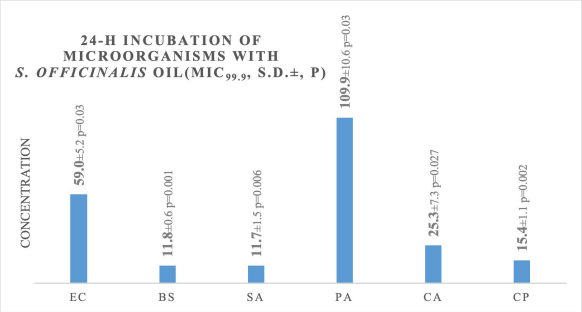


Figure 1: Statistical analysis of MICs value of *S. officinalis*against *E. coli*, *B. subtilis,S. aureus*(ATCC 25925), *P. aureginosa,C. albicans*and *C.parapsilosis*for 24 hours. The average MIC (µg/mL)values were expressed with the standard deviation (SD±) and significance level (p).

The MICs indicate that the volatile oil of sage aerial parts was rather efficient on all pathogens studied. This result demonstrated that the antimicrobial action of *S. officinalis* can be attributed to the presence of various concentrations of thujone, camphor, 1,8-cineole, and other minor components [Sur et al, 1991; Tepe et al, 2004]. It was proved that *Salvia* species have strong antimicrobial effective even in very low concentrations [Topu et al, 2007; Martins et al, 2015], as our study.

The volatile constituents and antimicrobial activities of *S. officinalis* aerial parts were revealed by some previous studies. For instance, Delamare et al., (2007) were noted that the essential oil of sage which its components were detected as α-thujone, 1,8-cineole, camphor, borneol and β-pinene, exhibited remarkable bacteriostatic and bactericidal effects on *Bacillus cereus*, *B. megatherium*, *B. subtilis*, *Aeromonas hydrophila*, *A. sobria* and *Klebsiella oxytoca* at a range of concentrations of 0.1 and 10.0 mg/ml [Delamare et al, 2007]. In another study, the essential oil of aerial parts *S. officinalis* which its major constituents were found to be 1,8-cineole, β-thujone, α-thujone, borneol, had strong antimicrobial activity for *C. albicans* (ATCC10239), *P. aeruginosa* (ATCC9027), *S. aureus* (ATCC25923) and *E. coli* (ATCC25922) by disc diffusion method. In the same study, *P. aeruginosa* (ATCC27853) were found to be resistant to oil [Hayouni et al, 2008]. Interestingly, in our study, the oil of sage were less active against to *P. aeruginosa* than other microorganisms. Pinto et al. (2007), were studied *S. officinalis* aerial parts collected from Portugal central part and identified that their major essential oil components were to be *cis*-thujone and camphor. In addition, they were reported that this oil have a broad antifungal spectrum, with high activity on *Candida* species such as *C. albicans*, *C. glabrata*, *C. krusei* [Pinto et al, 2007].

EXPERIMENTAL

Chemicals and spectral measurements

All chemicals and solvents obtained from E. Merck (Darmstadt, Germany), FlukaChemie (FlukaChemie GmbH, Sternheim, Germany, Sigma Chemical Co. (Sigma-Aldrich GmbH, Sternheim, Germany).

Plant material

S. officinalis were collected from Köyceğiz region of Muğla, Turkey, during June-July 2017, identified at the Herbarium of Biology, Faculty of Science, Muğla Sıtkı Koçman University, Turkey. The plant sample was confirmed by comparing it with the specimen voucher located at the stated herbarium.

Preparation of the extraction of essential oil

Approximately 200 g of *S. officinalis* samples were used for the essential oil extraction process. Extraction was performed by vapor distillation for 2 hours. The mixture added to water. After liqiud-liqiud extraction, the aqua in organic phase was dried over anhydrous Na_2SO_4. Organic phase was then concentrated under vacuum. Obtained essential oil was kept in desiccator. It was protected from sunlight until analysis.

Antimicrobial Activity

The antimicrobial activity of essential oil of *S. officinalis* were researched on several pathogens, namely *Escherichia coli* (ATCC 25293), *Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 25925), *Pseudomonas aureginosa, Candida albicans* and *Candida parapsilosis* using spectrophotometric microdilution technique. The inoculums of microorganisms were prepared in Triptic Soy Broth (4 mL) for bacteria, Sabouraud Dextrose Broth (4 mL) for yeasts and incubated at 37°C overnight. After 24 hours, the culture suspensions were adjusted to 0.5

McFarland Standard Turbidity (~ 10^4 for bacteria, ~ 10^3 for yeasts) and stored at +4°C until experiment [McFarland, 1987].

The 50 µL (41,5 mg/ml) of sage oil were dissolved in dimethyl sulfoxide (10% DMSO). Two-fold serial dilutions of 50 µL oil solution was made on 96-well microtiter plates which 50 µL of Mueller Hinton Broth (MHB) medium were added before. Last two columns were used as negative and positive controls (ampiciline for bacteria and fluconazole for yeast). Then, 10 µL culture of microorganisms was inoculated on all wells except medium control wells. The plates were incubated at 37°C for 24 hours, the growth (turbidity) was measured at 600 nm and 415 nm for bacteria and yeasts, respectively. For MIC analysis, the optical density was read both before (T0) and after 24 hours-incubation (T24). The OD (Optical density) for each replicate at T0 was subtracted from the OD for each replicate at T24. For each microorganism were calculated using the following formula:

The Percent growth (Cell viability) = (ODtest /OD control)x100.

Percent Inhibition = 1-(OD test well/OD of corresponding control well)x100.

MIC (the lowest concentration of test material which results in 99.9% inhibition of growth) were calculated using the R2 formula on percent inhibition of microbial growth [Patton et al, 2006].

Statistical analysis

Statistical analyses and significance were measured by Student's *t*-test (for paired samples) were performed for between MICs using SPSS 25. The experiment was repeated at least 3 times. Differences were considered significant at $p \leq 0.05$.

Acknowledgements

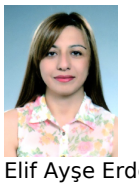
Present study was supported by Research Foundation of Muğla Sıtkı Koçman University (Project No: 17/216).

Bibliography

Aleksovski et al, 2007 Aleksovski, SA; Sovova, H (2007). *The Journal of supercritical fluids*, **40**, 239-245
Baricevic et al, 2001 Baricevic, D; Sosa, Silvio; Della Loggia, Roberto; Tubaro, Aurelia; Simonovska, B; Krasna, A; Zupancic, A (2001). *Journal of ethnopharmacology*, **75**, 125-132
CardenasOrtega et al, 2015 Cardenas-Ortega, Norma Cecilia; Gonz'alez-Ch'avez, Marco Mart'in; Figueroa-Brito, Rodolfo; Flores-Mac'ias, Antonio; Romo-Asunci'on, Diana; Mart'inez-Gonz'alez, Diana Elizabeth; P'erez-Moreno, V'ictor; Ramos-L'opez, Miguel Angel (2015). *Molecules*, **20**, 8048-8059
Ceylan, 1976 Ceylan, A (1976). *Ziraat Fakultesi dergisi. Seri: A*, **13**, 283-288
Davis et al, 1965 Davis, Peter Hadland; others (1965). *Flora of Turkey.*, **1**, 1
Delamare et al, 2007 Delamare, Ana Paula Longaray; Moschen-Pistorello, Ivete T; Artico, Liane; Atti-Serafini, Luciana; Echeverrigaray, Sergio (2007). *Food chemistry*, **100**, 603-608
Figueiredo et al, 2008 Figueiredo, A Cristina; Barroso, Jos'e G; Pedro, Luis G; Scheffer, Johannes JC (2008). *Flavour and Fragrance journal*, **23**, 213-226
Hayouni et al, 2008 Hayouni, El Akrem; Chraief, Imed; Abedrabba, Manaf; Bouix, Marielle; Leveau, Jean-Yves; Mohammed, Hammami; Hamdi, Moktar (2008). *International Journal of Food Microbiology*, **125**, 242-251
Li et al, 2015 Li, Bo; Zhang, Chenlu; Peng, Liang; Liang, Zongsuo; Yan, Xijun; Zhu, Yonghong; Liu, Yan (2015). *Industrial Crops and Products*, **69**, 329-334
Martins et al, 2015 Martins, Natalia; Barros, Lillian; Santos-Buelga, Celestino; Henriques, Mariana; Silva, S'onia; Ferreira, Isabel CFR (2015). *Food chemistry*, **170**, 378-385
McFarland, 1987 McFarland, J (1987). *J Am Med Assoc*, **49**, 1176-1178
Patton et al, 2006 Patton, Thomas; Barrett, John; Brennan, James; Moran, Noel (2006). *Journal of Microbiological Methods*, **64**, 84-95
Pinto et al, 2007 Pinto, Eug'enia; Salgueiro, L'igia Ribeiro; Cavaleiro, Carlos; Palmeira, Ana; Gonccalves, Maria Jos'e (2007). *Industrial Crops and Products*, **26**, 135-141
SARICI et al, 2004 SARICI, Serdar "Umit; KUL, Mustafa; CANDEM.IR, G"oknur; AYDIN, Halil .Ibrahim; ALPAY, Faruk; G"OKcCAY, Erdal (2004). *Gülhane Tıp Dergi*, **46**, 161-162
Sonboli et al, 2006 Sonboli, Ali; Babakhani, Babak; Mehrabian, Ahmad Reza (2006). *Zeitschrift f"ur Naturforschung C*, **61**, 160-164
Sur et al, 1991 Sur, SV; Tuljupa, FM; Sur, LI (1991). *Journal of Chromatography A*, **542**, 451-458
Tepe et al, 2004 Tepe, Bektas; Donmez, Erol; Unlu, Mehmet; Candan, Ferda; Daferera, Dimitra; Vardar-Unlu, G"ulhan; Polissiou, Moschos; Sokmen, Atalay (2004). *Food chemistry*, **84**, 519-525
Topu et al, 2007 Topçu, Gülaçti; Gören, Ahmet C (2007). *Records of Natural Products*, **1**, 1
Walch et al, 2011 Walch, Stephan G; Kuballa, Thomas; St"uhlinger, Wolf; Lachenmeier, Dirk W (2011). *Chemistry Central Journal*, **5**, 44

J Ong Chem Res (2018), Volume: 3, Issue: 2, pages: 63-66, Document ID: 2018JOCR26
Research Article

Corresponding: Elif Ayşe Erdoğan Eliuz
Email: eliferdogan81@mersin.edu.tr
Phone: +90 364 361 6772



Elif Ayşe Erdoğan Eliuz: Department of Food Technology, Technical Sciences Vocational School, Mersin University, Mersin-33343, MERSİN Turkey

Yusuf Sıcak: Department of Aromatic and Medicinal Plants, Köyceyiz Vocational School, Muğla Sıtkı Koçman University, Muğla-48000, MUĞLA Turkey