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Phenolic Compounds, Organic Acid Profiles and Antioxidant Potential of *Salvia verticillata* L.

Züleyha ALMAZ[1*]

[1]Mus Alparslan University, Arts and Sciences Faculty, Molecular Biology and Genetics Department, Mus, Turkey
Züleyha ALMAZ ORCID No: 0000-0002-4532-4311

**Corresponding author: z.turkoglu@alparslan.edu.tr*

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Salvia verticillata L., Phenolic acid, Organic acid, Antioxidant

Abstract: *Salvia* genus, which is widely used in folk medicine and attracts great attention, is a rich source of polyphenols, which has been the subject of many chemical studies. Leaf and root ethanol extracts of *Salvia verticillata* L. plant sampled from Muş region were obtained by using the soxhlet extraction method. Antioxidant profiles of these extracts were defined by five different methods. Although *S. verticillata* L. leaf extract showed the best activity in all tests, it was lower than the antioxidants we used as standard. The presence of 17 phenolics, 13 organic acids, and sugars in these extracts was screened by HPLC and correlated with their antioxidant potential. In this context, the difference between the organs of the species examined and collected from a region was also revealed. Leaf extracts have been found to be rich in curcumin, which has anti-inflammatory, anti-cancer, and strong antioxidant capacity. It was determined that acetic acid, an organic acid that is also used as a food preservative, was found in very high amounts in root and leaf extracts. According to the results obtained, it can be thought that the phenolic and organic acid contents of *S. verticillata* L. may be among the parameters responsible for antioxidant activity, and they are also natural sources for pharmacological processes and the food industry.

Salvia verticillata L.'nin Fenolik Bileşikleri, Organik Asit Profilleri ve Antioksidan Potansiyeli

Anahtar Kelimeler
Salvia verticillata L, Fenolik asit, Organik asit, Antioksidan

Öz: Zengin bir polifenol kaynağı olan ve çok sayıda kimyasal araştırmaya konu olan ada çayı, büyük ilgi gören ve halk hekimliğinde yaygın olarak kullanılan bir cinstir. Muş yöresinden örneklenen *Salvia verticillata* L. bitkisinin yaprak etanol ve kök etanol ekstraktları soxhlet ekstraksiyon yöntemi kullanılarak elde edilmiştir Bu ekstraktların antioksidan profili, beş farklı metod kullanılarak belirlendi. *S. verticillata* L. yaprağı ekstresi tüm testlerde en iyi aktiviteyi göstermesine rağmen standart olarak kullandığımız antioksidanlardan daha düşüktü. Bu ekstraktlarda 17 fenolik, 13 organik asit ve şekerin varlığı HPLC ile taranmış ve antioksidan potansiyelleri ile ilişkilendirilmiştir. Bu bağlamda bir bölgeden toplanan ve incelenen türlerin organları arasındaki fark da ortaya konulmuştur. Yaprak özlerinin, anti-inflamatuar, anti-kanser ve güçlü antioksidan kapasiteye sahip kurkumin açısından zengin olduğu bulunmuştur. Gıda koruyucu olarak da kullanılan bir organik asit olan asetik asidin kök ve yaprak ekstraktlarında çok yüksek miktarlarda bulunduğu belirlendi. Elde edilen sonuçlara göre *S. verticillata* L.'nin fenolik ve organik asit içeriğinin antioksidan aktiviteden sorumlu parametreler arasında olabileceği, ayrıca farmakolojik süreçler ve gıda endüstrisi için doğal kaynaklar olduğu düşünülebilir.

1. INTRODUCTION

Since ancient times, people have used plants as a solution to nutrition and health problems. With the expansion of alternative medicine and the increasing demand for health treatment, the use of therapeutic plants has increased all over the world. Among the plants that are often the subject of research to determine their health benefits is the genus *Salvia*, which belongs to the mint family. *Salvia*, which contains about 900 species and belongs to the Lamiaceae

family, is an important genus used as a folk medicine in many diseases such as coronary heart disease, neuroasthenic insomnia, hepatitis, and chronic kidney failure [1]. It has been reported that different species of *Salvia* have biological activities such as antidiabetic, antioxidant, antimicrobial, anti-inflammatory, anticarcinogenic, anxiolytic, antiplasmodial, and produce various phenolic metabolites that attract attention with these properties [2, 3]. In addition, many types of *Salvia*, which are frequently used in the food industry due to herbal teas, flavoring and preservative properties, also attract great attention in the cosmetics, perfumery and pharmaceutical sectors [4, 5]. Although antioxidants such as BHT, BHA, and *ter*-butyl hydroperoxide (TBH) are widely used today to reduce the damage caused by free radicals, they are unsuitable for use in health and food fields as they can cause tumors and toxicities in the animal body [6]. *Salvia* species, one of the most common members of its family, and their isolated components have significant antioxidant activity [7]. Phenolics are the main compounds that make up the antioxidant effect of widely used sage [8]. Phenolic antioxidants scavenge free radicals by an electron transfer mechanism. The presence of the methoxy group in the phenolic structure of curcumin, which is one of them, causes its antioxidant and anti-inflammatory activity to be high. It is also a phenolic compound with strong antitumor, antiseptic, anti-carcinogenic activity [9]. In clinical studies, it has been reported that curcumin is not dangerous for humans even at high doses [10]. For these and other reasons, the desire for natural foods has led to the search for naturally occurring antioxidants.

In this study, root and leaf ethanol extracts of *S. verticillata* L. were prepared and their phenolic and organic acids profiles were investigated by HPLC. In addition, *in vitro* antioxidant tests such as DPPH and ABTS radical scavenging activities, CUPRAC and FRAP methods, reducing capacities of copper and iron ions and metal chelating activities of these extracts were performed. The results of the study and analysis were correlated with each other.

2. MATERIAL AND METHOD

2.1. The Extraction of Plant Samples

S. verticillata L. plant was collected in Mus province during the vegetation period in 2021 (38° 43' 33.0996" N; 41° 30' 44.4384" E). The identification of the collected plant samples according to the Flora of Turkey was done by Murat Kurşat (Bitlis, Turkey). The plant samples that was made into herbarium material was encoded (Z. Almaz: 4900). The leaves and roots of the plant were removed and left to dry in shade. *S. verticillata* L. leaf (*Sv*L) and root ethanol (*Sv*R) extracts were prepared by using the soxhlet extraction method.

2.2. Antioxidant Activity

2.2.1. Fe^{3+} reducing activity according to the FRAP method

The reduction of total Fe^{3+} power was performed according to the assay of Savci et al. [11]. *S. verticillata* L. extracts (15, 30 and 45 µg/mL) were added to test tubes at different concentrations and adjusted to 1 mL with deionized water. For each test tube, 500 µL of potassium ferricyanide [$K_3Fe(CN)_6$] and phosphate buffer (0.2 M, pH: 6.6) as well as plant extracts of different concentrations were added. The solution was incubated at 50°C for 20 minutes before the 500 µL of trichloroacetic acid (TCA) was added to the reaction mixture.and 500 µL of the phase was taken at the top of the mixture, 500 µL of deionized H_2O and 100 µL of $FeCl_3$ were added. The absorbances of the samples were measured by adjusting the UV visible spectrophotometer to 700 nm.

2.2.2. Cu^{2+} reducing activity according to the CUPRAC method

The copper ion (Cu^{2+}) reducing capacities of *S.verticillata* L. extracts and standard antioxidants were performed similar to the procedure used by Apak et al. [12]. Different concentrations *S. verticillata* L. extracts (15, 30, and 45 µg/mL) were added to test tubes and made up to 1 mL with deionized H_2O water. Firstly, 250 µL of $CuCl_2$ solution (0.01 M) and neocuproine solution ($7.5x10^{-3}$ M), 1 mL of CH_3COONH_4 (1 M) buffer were transferred to each tube. After half an hour of incubation to clearly determine their reducing capacity, absorbance at 450 nm was measured.

2.2.3. $ABTS^{\cdot+}$ scavenging activity

Re [13] method was used for calculation of 2,2′-Azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) radical scavenging activities of *S. verticillata* L. extracts. First of all, solutions of potassium persulphate (2.45 mM) and ABTS (7 mM) were mixed in a 1:1 ratio. It was incubated for 12 h at room temperature in the dark. Then the absorbance of the mixture was measured at 734 nm and diluted with ethanol until an absorbance of 0.700±0.02 was reached. After adding different concentrations of extracts to the test tubes, it was made up to 200 µL with ethanol. 800 µL of ABTS was added to them. It was incubated for up to two hours. The absorbance of the extracts was measured at 734 nm.

2.2.4. $DPPH^{\cdot}$ scavenging activity

1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity of the *S. verticillata* L. extracts and standards were done to an assay of Koçpınar et al. [14]. This method is based on the removal of DPPH free radicals by reacting with antioxidants. According to this assay, the extracts at different concentrations (15, 30, and 45 µg/mL) were placed in the test tubes, were made up to be adjusted to 600 µL with ethanol. 200 µL of 1 mM DPPH radical solution was added to each test tubes. The mixture was incubated for 30 minutes at room

temperature and in the dark and absorption spectra were calculated at 517 nm against the ethanol blank.

2.2.5. Fe^{2+} chelating activity

The Fe^{2+} chelating activity of *S. verticillata* L. extracts and standard antioxidants was studied, modifying the method performed by Alhafez et al. [15]. It was prepared from samples at concentrations of 15, 30 and 45 µg/mL. 2 mM solution containing 120 µL of $FeCl_2.4H_2O$ and 80 µL of distilled water was added to 60 µL of solution containing samples. After this step, the total volume was made up to 1000 µL with ethanol. To initiate the reaction, 60 µL of 5 mM ferrozine solution prepared earlier was added to the samples. The resulting solution was mixed by rapid vortexing and incubated for 10 minutes at room temperature. In the next step, the absorbance of the samples at 562 nm was measured by UV/VIS spectrophotometer.

2.3. Phenolic Substance Analysis by HPLC

Concentrations of 1 mg/mL were prepared by diluting the leaf (*Sv*L) and root (*Sv*R) extracts of *S. verticillata* L. for loading in HPLC. To determine the phenolic content by HPLC, the final concentrations of the standards were adjusted to 10 mg/mL. Then, 1% acetonitrile and acetic acid (1/9 ratio, respectively) were added to prepare the standards. Methanol was added at the same rate and stock standards were prepared. Different concentrations of 10, 25, 50, 75, and 100 µg/mL of the stock solution prepared for the standards were created to draw the standard graph [16]. The *S. verticillata* L. plant extracts we prepared earlier were diluted at 20 mg/mL concentration using standard solutions and filtered using a 0.45 µm membrane filter. HPLC analyses used for phenolic content determination were performed using 1260 Infinity II HPLC Agilent Tech. Standard chromatograms and curves generated by HPLC were used to determine the concentrations of 17 different phenolics. ACE 5 C18 (250 x 4.6 mm id) is the analytical column used for the analysis. The HPLC configuration used includes a 1260 DAD WR detector (272 nm, 280 nm, and 310 nm wavelength), a 1260 Quat Pump VL pump (flow rate 1.0 mL/min), a 1260 Vial sampler (20 µL injected), and a G7130A column furnace (28 °C).

2.4. Organic Acid and Sugar Content in The Extracts

The standards used to determine the sugar and organic acid content of *S. verticillata* L. extracts are fructose, glucose, rhamnose, tartaric acid, pyruvic acid, citric acid, maleic acid, malic acid, acetoin, fumaric acid, 2,3-butanediol, acetic acid, and succinic acid. The concentrations of the standards used were weighed to be 1 mg/mL. The standards that we will use as stocks were dissolved in falcon tubes with 0.03M H_2SO_4 (sulfuric acid). Stock standard solutions were prepared in 8 different dilutions (5, 10, 25, 50, 100, 200, 300 and 400 ppm). It was loaded on HPLC (Agilent Technologies 1260 Infinity II) and the calibration curve was calculated [17]. 0.2 ml was taken from the extracts. It was vortexed by adding 0.03M 1.8 mL H_2SO_4. The resulting mixture was centrifuged at 2000 rpm for 5 minutes and 100 µL was taken from the supernatant. It was added to ependorph containing 0.9 µL of 0.03 M H_2SO_4 and mixed. The resulting mixture was filtered through 0.45 µm pore diameter filters and transferred to approximately 0.5 mL bottles. The extracts we prepared to determine the amount of sugar and organic acid in HPLS were given to the device. 0.03 M H_2SO_4 was used as carrier phase.

2.5. Statistical Analysis of The Antioxidants

All antioxidant tests were repeated three times. Fe^{3+} and Cu^{2+} reducing activity, Fe^{2+} chelating activity results were calculated as µM Trolox equivalent in g extract (µM TE/g Extract). In other tests, the results of both the extract and the standards (DPPH· and $ABTS^{\cdot+}$ radical scavenging activities) were calculated as % radical removal, and the standards and extract results were compared. All comparisons were made using One-way ANOVA followed by Dunnett's multiple comparisons test, and values were given as mean±standard deviation (Mean±Standard Deviation). "●" symbol in comparison against BHA standard, "■" symbol in comparison against BHT standard, and "◆" symbol in comparison against ascorbic acid (AA) were used as degrees of significance, and comparison results that did not change significantly were expressed as "ns". Accordingly, one symbol (● or ■ or ◆) was expressed as meaningful, two symbols (●● or ■■ or ◆◆) were expressed as very meaningful, and three symbols (●●● or ■■■ or ◆◆◆) were expressed as highly meaningful. •/▪/◆P<0.05 (meaningful); ••/▪▪/◆◆ P<0.01 (very meaningful); •••/▪▪▪/◆◆◆P<0.001 (highly meaningful).

3. RESULTS AND DISCUSSION

3.1. Results of *In Vitro* Antioxidant Activity

Since various mechanisms of action may play a role in the antioxidant effects of studies on plant extracts, it is generally accepted that the application of a single antioxidant method is not sufficient to determine the free radical scavenging potential of the samples and is a limited approach. Therefore, it is considered useful to use simultaneous methods for a real test, that is, to understand the full antioxidant potential [18]. The current study has demonstrated the ability of leaf and root extracts from *S. verticillata* L. to capture DPPH· and $ABTS^{\cdot+}$ radicals, as well as reduce ferric to ferrous form (FRAP assays), to reduce Cu^{2+} into Cu^{+} (CUPRAC assay) and the analyzed antioxidant activity potentials such as the capacity to chelate (Fe^{2+} chelating activity). The antioxidant potential of the extracts is compared with standard antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), and ascorbic acid (AA) are given in Table 1. In the FRAP results obtained, it was determined that the leaf extract had better metal reducing power than the root extract, but it was weaker than BHT with the lowest activity. However, it was determined that the same extract showed better activity than AA in the CUPRAC results. ABTS radical scavenging activities were found to be similar to the antioxidants used as standard in leaf extract and even stronger than BHT. It

was also found that leaf extract showed better activity than root extract in DPPH scavenging activity. In the metal chelation results of the study, the extracts showed very low activity compared to the standard antioxidants, but the activity of the leaf extract was quite good compared to the root extract. Considering the results obtained, it was found that the leaf extract of the *S. verticillata* L. plant showed better antioxidant activity compared to the root extract and some standard antioxidants. *In vitro* antioxidant activity results of the highest concentrations of *Sv*L and *Sv*R extracts compared with standard antioxidants as µg TE/ml and percentages are presented in Figures 1 and 2.

Table 1. *In vitro* Antioxidant Activities of *S. verticillata* L. extracts.

Samples	FRAP (µg TE/ml)	CUPRAC (µg TE/ml)	DPPH scavenging (IC_{50})	ABTS scavenging (IC_{50})	Fe-chelating (IC_{50})
***Sv*L**	10.83±0.01	29.72±0.11	40.03±0.02	23.51±0.01	139.78±0.01
***Sv*R**	8.05±0.01	15.28±0.01	97.94±0.20	79.20±0.11	580.04±0.02
BHA	90.34±0.01	101.47±0.03	20.14±0.30	18.29±0.01	38.29±0.01
BHT	18.16±0.01	72.81±0.04	26.86±0.03	24.65±0.21	38.12±0.02
AA	38.71±0.02	21.33±0.01	19.40±0.01	20.19±0.01	32.41±0.01

*FRAP and CUPRAC results as µg TE / ml Extract, DPPH, ABTS, and Fe-chelating activity results as IC_{50}

In a study investigating the antioxidant activity of extracts of *S. verticillata* ssp. *verticillata* and *S. verticillata* spp. *amasiaca*, prepared using different solvents, it was reported that methanol extracts showed strong DPPH radical scavenging activity compared to other extracts and BHT [19]. In another study evaluating the biological activities of 7 *Salvia* species, they reported that all species had a strong DPPH clearance potential, and *S. verticillata* L. showed the strongest antioxidant activity. The ability of these *Salvia* species to chelate transition metal ions was determined by the iron-chelating activity assay, and *S. verticillata* L. had the weakest activity, consistent with our study. They reported that *S. verticillata* L. showed high activity in accordance with our study in the antioxidant activity experiment performed by reducing the power assay [4].

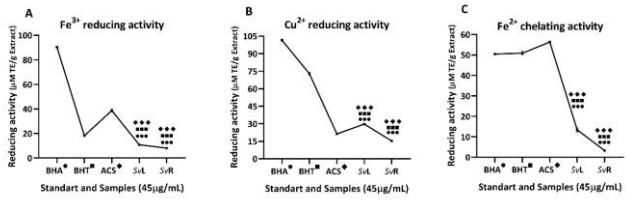

Figure 1. *In vitro* antioxidant activities of *S. verticillata* L. extracts as Trolox Equivalent (TE), BHA, BHT and ACS

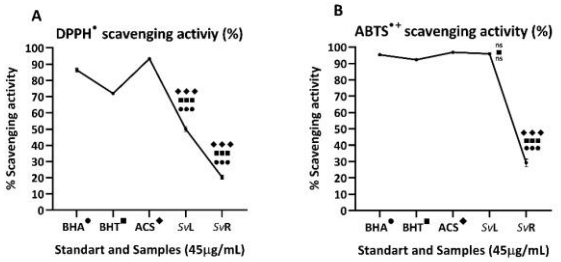

Figure 2. *In vitro* antioxidant activities of *S. verticillata* L. extracts as a percentage, BHA, BHT and ACS

3.2. Results of Phenolic Concentration

The phenolic concentration of root (*Sv*R) and leaf (*Sv*L) extracts of *S. verticillata* L. was determined using HPLC and the results are given in Table 2 in µg/mL. Leaf extract had the highest concentration of curcumin and root extract had the highest concentration of abscisic acid. The lowest concentrations for leaf and root extracts were seen in rosmarinic acid and catechol, respectively. The phenolic concentration of the leaf extract of *S. verticillata* L. appears to be significantly higher than the root extract. It is known that the pharmaceutical properties of *Salvia* plant species originate from phenolic compounds. Significant differences are observed in terms of the distribution and content of phenolic compounds, both within and between species of the plant. In addition, in our study, it was determined that there are more and different types of phenolic compounds in the leaves than in the roots, similar to the literature. In a previous study, it was determined that the leaves of *S. verticillata* are very rich in rosmarinic acid and salvianolic acid A [20]. Again, in a study evaluating the *S. verticillata* subsp. *amasiaca* of Turkey in the literature, it was reported that the leaves were quite rich in rosmarinic acid compared to the roots [3]. The part of our study, which is similar to the literature, is that the above-ground parts of *Salvia* species have high phenolic content. Differences with the current study may be due to the extraction method, the solvent used, and the differences in intra- and inter-species components. HPLC chromatogram results of leaf and root extracts are given in Figures 3 and 4, respectively.

Table 2. The concentrations (µg/mL) of phenolics in the *S. verticillata* L. extracts

Phenolic Compounds	Phenolic amounts (µg/mL)	
	*Sv*L	*Sv*R
Ascorbic acid	-	1.1197
Gallic acid	-	-
Myricetin	-	-
4-Hydroxybenzoic acid	1.2571	-
Trans-*p*-coumaric acid	-	-
3,4-Dihydroxybenzoic acid	-	-
Abscisic acid	1.1033	**1.4756**
Quercetin	-	-
Apigenin	-	-
Kaempferol	-	-
Curcumin	**39.2766**	-
Catechol	0.5635	0.1926
Vanillin	-	-
Caffeic acid	-	-
Cinnamic acid	0.7852	-
Rosmarinic acid	0.6375	0.7946
Salicylic acid	-	-
Total phenolic	**43.6232**	**3.5825**

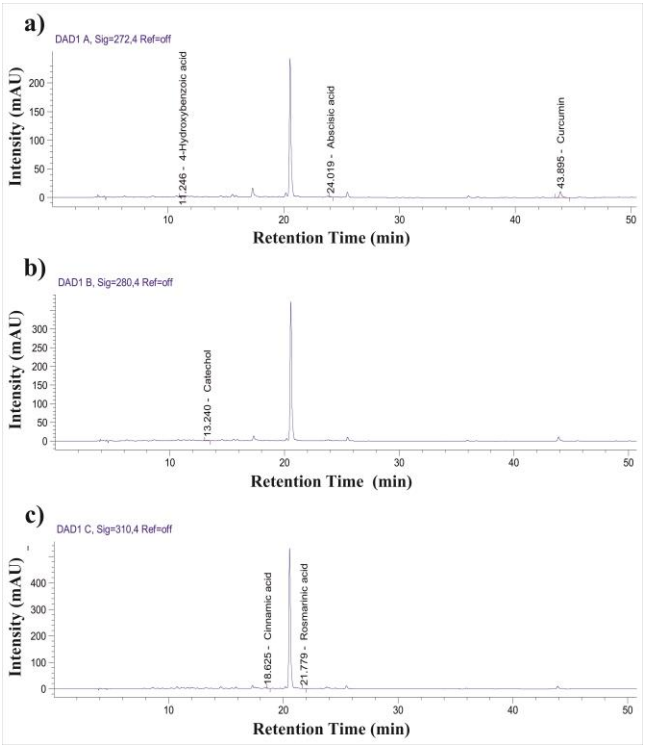

Figure 3. HPLC phenolic chromatogram of *Sv*L ethanol extracts

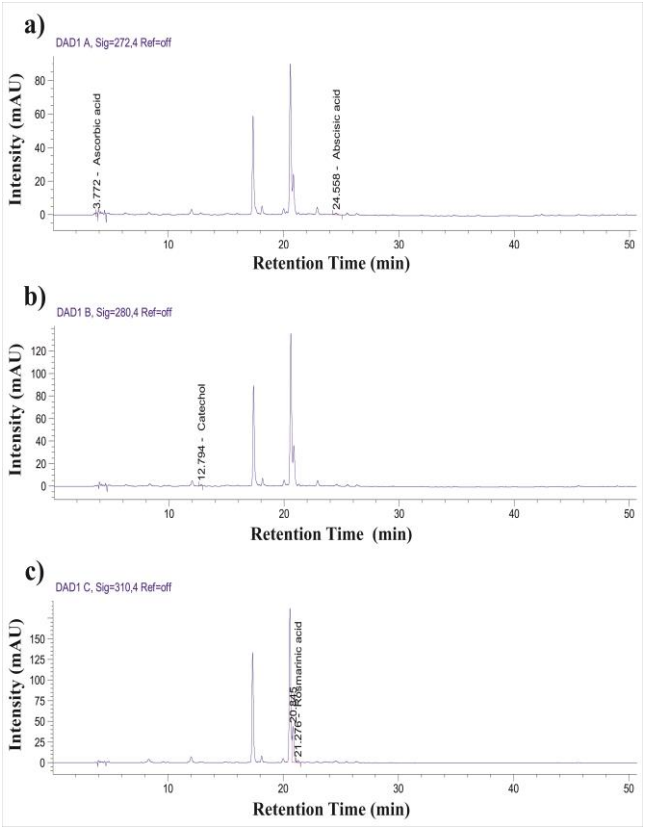

Figure 4. HPLC phenolic chromatogram of *Sv*R ethanol extracts

3.3. Results of Organic Acids and Sugars Concentration

Researchers often emphasize that organic acids and sugars are bioactive compounds, play important roles in antioxidant defense, and therefore have enormous therapeutic potential. They reported that organic acids reduce the pH, prevent food spoilage caused by organisms, and thus are widely used in the food industry, and they do not have negative effects on the body because they are rapidly oxidized in metabolism [21, 22]. In addition to organic acids, glucose and fructose are accepted as positive molecules in terms of antioxidant capacity [23]. In the current study, it was determined that the root and leaf ethanol extracts of *Salvia* contain a very high amount of acetic acid, followed by high amounts of maleic acid for both extracts. These two organic acids appear to be higher in root extract than in leaf extract. In addition to these, tartaric acid in leaf extract and fumaric acid in root extract were found at significant levels. According to the results, the ratios of glucose and fructose, which are free sugars, were found to be close to each other and moderately high in both extracts. In addition to organic acids, acetoin, a secondary metabolite with significant biological activity, was detected in leaf extract in lower amounts compared to other components. The results are given in Table 3 in µg/mL. In the literature review, no study was found on the organic acid and sugar content of the *Salvia* genus. HPLC chromatogram results of *Sv*L and *Sv*R ethanol extracts are given in Figure 5.

Table 3. The concentrations (µg/mL) of organic acids and sugars in the *S. verticillata* extracts

Organic acid and sugar compounds	Organic acid and sugar amounts (µg/mL)	
	***Sv*L**	***Sv*R**
Maleic acid	107.1578	139.0724
Citric acid	-	-
Tartaric acid	13.7790	-
Pyruvic acid	-	-
Glucose	51.6690	42.4277
Malic acid	-	-
Fructose	35.1951	52.1044
Rhamnose	-	-
Suksinic acid	-	-
Fumaric acid	-	-
Acetic acid	**1268.6395**	**1092.7171**
Acetoin	6.1567	23.8874
2-3 bütan	-	-
Total compounds	**1482.5975**	**1350.2090**

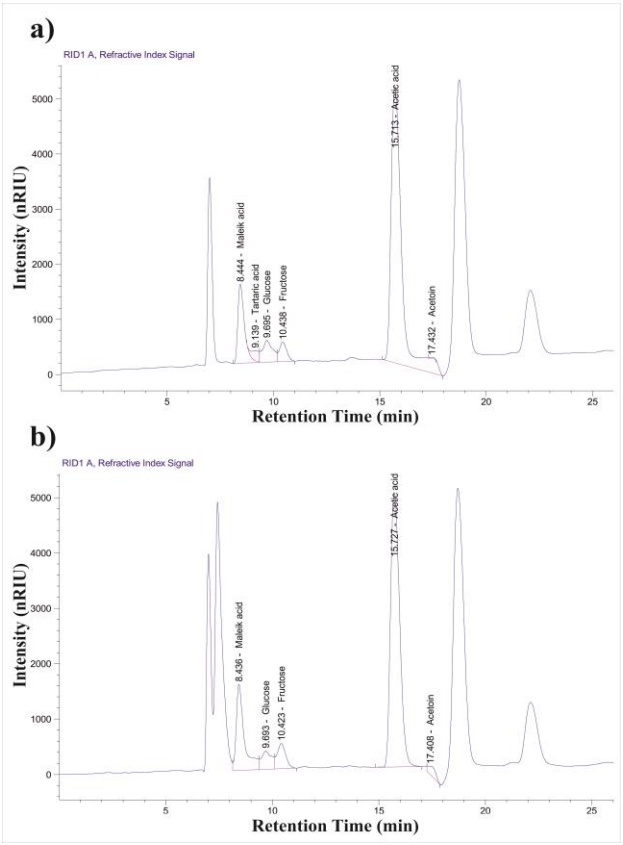

Figure 5. HPLC organic acid and sugar chromatogram results of *Sv*L (a) and *Sv*R (b) ethanol extracts

4. CONCLUSION

Research on the biological potential of plant phenolic compounds continues to be extremely interesting today. Determination of antioxidant properties in order to identify natural sources, including wild species, for use in functional foods, pharmaceuticals, and industry, is also one of the subjects that should be investigated. As a result, antioxidant activities, phenolic, organic acid, and sugar components of *S. Verticillata* L. were investigated. This strain exhibited good antioxidant activity. It has various phenolic and organic acids, including curcumin and acetic acid, as well as glucose content. It has been found to have reasonable and variable activities according to the antioxidant method used. It was also understood that *Salvia* species collected from different regions may have different phenolic contents and concentrations. This study provides a valuable reference for further biological activity research of *Salvia*, such as isolating and characterizing active compounds and conducting in vivo studies to better understand their activity.

REFERENCES

[1] Lu Y, Foo LY. Polyphenolics of Salvia—a review. Phytochemistry. 2002;59(2):117-40.

[2] Jeshvaghani ZA, Rahimmalek M, Talebi M, Goli SAH. Comparison of total phenolic content and antioxidant activity in different Salvia species using three model systems. Industrial Crops and Products. 2015;77:409-14.

[3] Zengin G, Llorent-Martínez EJ, Fernández-de Córdova ML, Bahadori MB, Mocan A, Locatelli M, et al. Chemical composition and biological activities of extracts from three Salvia species: S. blepharochlaena, S. euphratica var. leiocalycina, and S. verticillata subsp. amasiaca. Industrial Crops and Products. 2018;111:11-21.

[4] Mervić M, Bival Štefan M, Kindl M, Blažeković B, Marijan M, Vladimir-Knežević S. Comparative Antioxidant, Anti-Acetylcholinesterase and Anti-α-Glucosidase Activities of Mediterranean Salvia Species. Plants. 2022;11(5):625.

[5] Tosun M, Ercisli S, Sengul M, Ozer H, Polat T, Ozturk E. Antioxidant properties and total phenolic content of eight Salvia species from Turkey. Biological Research. 2009;42(2):175-81.

[6] Scott G. Antioxidants. Bulletin of the Chemical Society of Japan. 1988;61(1):165-70.

[7] Zupko I, Hohmann J, Rédei D, Falkay G, Janicsák G, Máthé I. Antioxidant activity of leaves of Salvia species in enzyme-dependent and enzyme-independent systems of lipid peroxidation and their phenolic constituents. Planta Medica. 2001;67(04):366-8.

[8] Schwarz K, Ternes W. Antioxidative constituents ofRosmarinus officinalis andSalvia officinalis. Zeitschrift für Lebensmittel-Untersuchung und Forschung. 1992;195(2):99-103.

[9] Raheem SMA. Evaluate the Efficiency of Sage (Salvia Officinalis) and Curcumin Mouthwash in the Treatment of Recurrent Aphthous Stomatitis (Comparative Study). Al-Rafidain University College For Sciences. 2021(48).

[10] Vitaglione P, Barone Lumaga R, Ferracane R, Radetsky I, Mennella I, Schettino R, et al. Curcumin bioavailability from enriched bread: the effect of microencapsulated ingredients. Journal of Agricultural and Food Chemistry. 2012;60(13):3357-66.

[11] Savcı A, Koçpınar E, Alan Y, Kurşat M. Antioxidant, antimicrobial, and DNA protection activities of some Tanacetum species and phenolic richness in their ethanolic extracts. International Food Research Journal. 2020;27(1).

[12] Apak R, Güçlü K, Özyürek M, Esin Karademir S, Erçağ E. The cupric ion reducing antioxidant capacity and polyphenolic content of some herbal teas. International journal of food sciences and nutrition. 2006;57(5-6):292-304.

[13] Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free radical biology and medicine. 1999;26(9-10):1231-7.

[14] Koçpınar EF, Kürşat M, Savcı A, Alan Y. Some biological properties of ethanol extract prepared from the aerial parts of Scutellaria albida L. subsp. condensata (Rech. f.) JR Edm. Bitlis Eren University Journal of Science and Technology. 2020;10(2):43-8.

[15] Alhafez A, Savci A, Alan Y, Söylemez R, Kilic A. Preparation of Cu (II), Ni (II), Ti (IV), VO (IV), and Zn (II) Metal Complexes Derived from Novel vic Dioxime and Investigation of Their Antioxidant and

Antibacterial Activities. Chemistry & Biodiversity. 2022:e202100768.
[16] Seal T. Quantitative HPLC analysis of phenolic acids, flavonoids and ascorbic acid in four different solvent extracts of two wild edible leaves, Sonchus arvensis and Oenanthe linearis of North-Eastern region in India. Journal of Applied Pharmaceutical Science. 2016;6(2):157-66.
[17] Ball S, Lloyd L. Agilent Hi-Plex columns for carbohydrates, alcohols, and acids. Application Note Pub. 2011;4:5990-8264.
[18] Tabart J, Kevers C, Pincemail J, Defraigne J-O, Dommes J. Evaluation of spectrophotometric methods for antioxidant compound measurement in relation to total antioxidant capacity in beverages. Food chemistry. 2010;120(2):607-14.
[19] Yumrutas O, Sokmen A, Ozturk N. Determination of in vitro antioxidant activities and phenolic compounds of different extracts of Salvia verticillata ssp. verticillata and spp. amasiaca from Turkey's flora. Journal of Applied Pharmaceutical Science. 2011;1(10):43.
[20] Fotovvat M, Radjabian T, Saboora A. HPLC fingerprint of important phenolic compounds in some Salvia L. species from Iran. Records of Natural Products. 2018;13(1).
[21] Lamani S, Anu-Appaiah KA, Murthy HN, Dewir YH, Rikisahedew JJ. Analysis of Free Sugars, Organic Acids, and Fatty Acids of Wood Apple (Limonia acidissima L.) Fruit Pulp. Horticulturae. 2022;8(1):67.
[22] Angonese M, Motta GE, de Farias NS, Mologni L, Daguer H, Brugnerotto P, et al. Organic dragon fruits (Hylocereus undatus and Hylocereus polyrhizus) grown at the same edaphoclimatic conditions: Comparison of phenolic and organic acids profiles and antioxidant activities. LWT. 2021;149:111924.
[23] Hu W, Sun DW, Pu H, Pan T. Recent developments in methods and techniques for rapid monitoring of sugar metabolism in fruits. Comprehensive reviews in food science and food safety. 2016;15(6):1067-79.