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Research Article

Antioxidant and oxidant status, DPPH activity, total phenolic and flavonoid contents of mountain tea (*Sideritis libanotica* subsp. *kurdica* (Bornm.) Hub.-Mor)

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ABSTRACT

Samples of *Sideritis libanotica* subsp. *kurdica* (Bornm.) Hub.-Mor, often known as mountain tea, were gathered in Iraq and analyzed for their antioxidant and oxidant status, DPPH activity, total phenolic and total flavonoid contents. The plant's aerial parts were utilized for this purpose. The levels of antioxidants and free radicals were measured by Rel assay kits. The ability to scavenge free radicals was evaluated using the DPPH technique. Folin-Ciocalteu and aluminum chloride reagent are used. As a result TAS values of 7.934±0.179 mmol/L, TOS values of 10.626±0.275 µmol/L, and OSI values of 0.134±0.001. The total phenolic and flavonoid contents were found to be 129.75±2.37 and 111.47±3.15 mg/g, respectively. Furthermore, DPPH activity at 2 mg/mL was calculated to be 75.15±1.45 *S. libanotica* subsp. *kurdica* has been found to have antioxidant activity and is thus a viable natural antioxidant source in this context.

Keywords: Antioxidant, phenolic, flavonoid, ironwort, mountain tea.

Dağ çayının (Sideritis libanotica subsp. kurdica (Bornm.) Hub.-Mor) antioksidan ve oksidan durumu, DPPH aktivitesi, toplam fenolik ve flavonoid içerikleri

ÖZ

Genellikle dağ çayı olarak bilinen *Sideritis libanotica* subsp. *kurdica* (Bornm.) Hub.-Mor Irak'tan toplanmıştır. Toplam antioksidan ve oksidan durumu, DPPH aktivitesi, toplam fenolik ve toplam flavonoid içerikleri açısından analiz edilmiştir. Bu amaçla bitkinin toprak üstü kısımlarından yararlanılmıştır. Toplam antioksidan ve oksidan durumları, Rel assay kitleri ile ölçülmüştür. Serbest radikalleri temizleme yeteneği, DPPH tekniği kullanılarak değerlendirilmiştir. Toplam fenolik ve flavonoid içeriği için sırasıyla Folin-Ciocalteu ve aliminyum klorür reaktifi kullanılmıştır. TAS değeri 7.934±0.179 mmol/L, TOS değeri 10.626±0.275 µmol/L ve OSI değeri 0.134±0.001 olduğu belirlenmiştir. Toplam fenolik ve flavonoid içerikleri sırasıyla 129.75±2.37 ve 111.47±3.15 mg/g olarak belirlenmiştir. Ayrıca 2 mg/mL'de DPPH aktivitesi 75.15±1.45 olarak hesaplanmıştır. *S. libanotica* subsp. *kurdica*'nın antioksidan aktiviteye sahip olduğu ve bu bağlamda doğal antioksidan kaynağı olduğu bulunmuştur.

Anahtar Kelimeler: Antioksidan, fenolik, flavonoid, demirotu, dağ çayı.

1. INTRODUCTION

Plants are one of the constant elements of human life. The human race has found several uses for various plants. [1] Plants have been utilized by people all across the world for anything from building materials to food to medicine.[2] Plants are one of the few permanent aspects in human medicine, especially in the treatment of

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illnesses. The antioxidant, anticancer, anti-inflammatory, anti-allergic, anti-aging, DNA-protective, antibacterial, and hepatoprotective properties of plants have been the subject of many research. [3–7] Determining the biological activity of plants is, therefore, crucial for their application as supplementary medicines in this setting. They have further pharmacological use as natural materials.[8]

The bioactive chemicals found in many plants provide them unique biological characteristics.[9] These bioactive compounds are not nutritional but are also very important medicinally. Compounds having antioxidant activity are produced by many different plant species.[10,11] They can serve as a natural antioxidant source if sufficient quantities of antioxidant molecules are produced.[12] The purpose of this research was to evaluate the antioxidant properties of *Sideritis libanotica* subsp. *kurdica* (Bornm.) Hub.-Mor. In addition, Total flavonoid and phenolic content was also determined.

Ironwort, often called mountain tea or shepherd's tea, is a species of *Sideritis* (Lamiaceae). It is a herb that is commonly used to make tea. It thrives in high altitude regions with scant soil and often directly on the rocks itself. They are 8-50 cm in height and are xerophytic plants. Because of its pleasant scent, it is a popular herbal tea ingredient in many cultures. The plant's stem, leaves, and flowers are employed in a boiling lemon or honey concoction.[13,14]

In this work, we analyzed samples of *S. libanotica* subsp. *kurdica* from Iraq (Duhok) to assess its antioxidant, oxidant potential, DPPH activity, total phenolic, and total flavonoid content.

2.MATERIALS AND METHODS

2.1. Materials

Specimens of *S. libanotica* subsp. *kurdica* were obtained from the city of Duhok (Iraq). Shaded and well-ventilated space was used for the drying operations of the plant samples. After that, we cartridged 30 g of the plant sample and extracted it with 250 mL of ethanol at 50 ^{0}C for around 6 hours. The solvents of the obtained extracts were removed in the concentrator. (Heidolph Laborota 4000 Rotary Evaporator).

2.2. Methods

2.2.1. Antioxidant tests

The plant's antioxidant (TAS) and oxidant (TOS) levels were measured using Rel Assay kits. Calibrators included trolox for the antioxidant test and hydrogen peroxide for the oxidant test.[15,16] The oxidative stress index was determined by dividing the TOS value with the TAS value. [17]

The ability to scavenge free radicals was measured using the DPPH assay. Plant extracts were dissolved in DMSO to make 1 mg/mL stock solutions. To 50 L of this solution, 160 L of a 0.039% DPPH solution was added. After a 30-minute room-temperature incubation, the sample's absorbance was measured at 517 nm. All plant preparation concentrations were tested again. The antioxidant ascorbic acid served as a standard.[18] DPPH free radical scavenging percentages were calculated using the following formula: % inhibition = [(Abs control-Abs sample)\Abs control]x100.

2.2.2. Total phenolic and flavonoid tests

Plant extract (originally 0.1 mL) was diluted with 1 mL of distilled water. After that, we added 1 mL of Folin-Ciocalteu reagent (1:9, v/v) and gave it a good whirl. The finished product added 0.75 mL of 1% Na2CO3. The mixture was then allowed to incubate at room temperature for 2 hours. The incubation period was followed by a 760 nm measurement. From the gallic acid standard solution calibration curve, the total phenolic content was calculated and represented as mg.GAE/g.[19]

Measurement of flavonoids was performed using aluminum chloride.[20] Quercetin (0.5 mL), plant extract (0.5 mL), methanol (4.3 mL), 10% Al(NO3)3, and 1 M NH4CH3COO (0.1 mL) were mixed together. After a 40-minute incubation period, the absorbance was measured at 415 nm. Flavonoids were reported as mg.QE/g.

3. RESULTS AND DISCUSSION

3.1. Antioxidant activity

Oxidant molecules are byproducts of metabolic processes in all living things.[21] When these oxidant molecules accumulate, the body's antioxidant defense mechanism steps in to neutralize them. When the body's antioxidant defenses against oxidizing substances are deficient, a condition known as oxidative stress can develop.[22,23] Many illnesses, including cancer, Alzheimer's, Parkinson's, and cardiovascular disease, may develop in humans as a result of oxidative stress.[24] The consequences of oxidative stress can be mitigated or even prevented with the use of antioxidant supplements.[25] We investigated the antioxidant capacity of mountain tea in this work. The results are shown in Table 1 and Table 2.

Table 1. TAS, TOS, OSI, TPC and TFC values of *S. libanotica* subsp. *Kurdica*.

	TAS mmol/L	*TOS* µmol/L	*OSI*	*TPC mg/g*	*TFC mg/g*
Mountain tea	7.934±0.179	10.626±0.275	0.134±0.001	129.75±2.37	111.47±3.15

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Table 2. DPPH activity of *S. libanotica* subsp. *Kurdica*.

	0.25 mg/mL	**0.5** mg/mL	**1** mg/mL	**2** mg/mL
Mountain tea	47.45±1.39	58.15±0.89	62.93±1.46	75.15±1.45
Ascorbic acid	76.91±0.86	88.07±0.90	92.90±0.85	96.77±0.42

Antioxidant chemicals are produced by the bodies of many plants. Because of these qualities, they are used into alternative medicine as antioxidant agents.[26] . We investigated the antioxidant capacity of mountain tea in this work. The maximum DPPH activity was seen at a concentration of 2 mg/mL (75.15±1.45). The value of 2 mg/mL ascorbic acid, which was employed as a standard, was found to be 96.77±0.42. We observed that mountain tea has less DPPH activity than ascorbic acid. Antioxidant activity in many species of *Sideritis* has been described in the scientific literature.[27–29] In our experiment, the DPPH activity of the mountain tea was found to be high.

The total antioxidant capacity (TAS) of a product is a measure of all of its antioxidant components.[30] TAS, TOS and OSI values of *Sideritis libanotica* subsp. *kurdica* have not been reported in the literature. It was determined for the first time in our study. Numerous plant species have had their TAS, TOS, and OSI published in the scientific literature. Among these studies, TAS values of *Mentha longifolia* ssp. *longifolia*, *Rhus coriaria* var. *zebaria*, *Alcea kurdica*, *Rumex scutatus*, *Helianthemum salicifolium* and *Scorzonera papposa* were reported as 3.628, 7.342, 3.298, 8.656, 9.490 and 6.328, respectively. TOS values were reported as 4.046, 5.170, 8.312, 4.951, 14,839 and 11,525, respectively. OSI values are reported as 0.112, 0.071, 0.252, 0.057, 0.157 and 0.182, respectively.[1,29,31–33] Compared to this study, the TAS value of mountain tea used in our study was determined to be higher than *M. longifolia* ssp. *longifolia*, *R. coriaria* var. *zebaria*, *A. kurdica* and *S. papposa*, and lower than *R. scutatus* and *H. salicifolium*. In this context, it has been determined that mountain tea has an important antioxidant potential.

The total oxidant status (TOS) measures the total amount of oxidant components in an organic product.[30] Our research showed that the mountain tea had a lower TOS value than both *H. salicifolia* and *S. papposa*, and a higher TOS value than *M. longifolia* ssp. *longifolia*, *R. coriaria* var. *zebaria*, *A. kurdica*, and *R. scutatus*. The oxidant components in the mountain tea we tested in this investigation were found to be within normal ranges.

When the TOS value is divided by the TAS value, the OSI value emerges. The higher the OSI number, the more likely it is that the product contains harmful levels of oxidant chemicals and should not be consumed.[30] In this study, we found that mountain tea had a lower OSI than *A. kurdica*, *H. salicifolium*, and *S. papposa*, but a higher OSI than *M. longifolia* ssp. *longifolia*, *R. coriaria* var. *zebaria*, and *R. scutatus*. Our findings suggest that the mountain tea utilised in this study has significant promise as a means of reducing oxidant chemicals.

3.2. Total phenolic and flavonoid contents

Secondary metabolites include the phenolic and flavonoid chemicals found in plants.[34] The health advantages of these non-nutritional substances are comparable to those of natural goods.[35] Our research looked on the overall phenolic and flavonoid content of mountain tea. Total phenolic contents as 35.5-366.9 mg/g and total flavonoid contents as 14.2-155.7 mg/g of different *Sideritis* species (*S. rubriflora*, *S. libanotica* subsp. *violascens*, *S. erythrantha* var. *cedretorum*, *S. congesta*, *S. brevidens* and *S. viralli*) have been reported in the literatüre.[36]

Total phenolic contents of *S. lycia* and *S. libanotica* subsp. *linearis* have been reported as 16.52 and 10.33 g/kg, and total flavonoid contents as 14.30 and 9.68 g/kg.[28] Instead of using these species, we assessed the total phenolic and flavonoid contents of *S. libanotica* subsp. *kurdica*. Our research indicates that the mountain tea we utilised can be a natural supply of phenolics and flavonoids in this setting.

4. CONCLUSION

The antioxidant capacity of mountain tea was investigated in this study. Total phenolic and flavonoid concentrations in the plant were also calculated. The research indicated that the plant has potential as a useful natural antioxidant source. In addition, it is thought to be a source of phenolics and flavonoids. Therefore, mountain tea has been identified as a potential supplementary antioxidant source for reducing oxidant chemicals.

Conflict of interest

Authors declare that there is no a conflict of interest with any person, institute, company,

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