# Antioxidant capacities of *Sideritis scardica* aerial parts' extracts and its essential oil composition


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***Abstract***

*Sideritis scardica* is an endemic species in the Balkan Peninsula, with a wide spectrum of application and an extensive range of traditional usage. The aim of this study was the evaluation of the antioxidant capacity of several plant extracts of this plant (DPPH· assay), as well as the analysis of its essential oil. One part of the plant material of the plant was extracted in a soxhlet apparatus with petroleum ether, dichloromethane, methanol and water, consecutively. Another part of the plant was exclusively extracted with water. Antioxidant results, according to the DPPH· test, showed that the methanolic fraction, after defatting, had the greatest antiradical efficiency in comparison to the standard used (trolox). The essential oil was obtained by hydrodistillation and was analyzed by GC/MS. B-caryophyllene (23%) was the main constituent of *S. scardica.* All this information may be useful for the promotion of this plant as a natural antioxidant in food and medicinal products, justifying also its traditional use as beneficial to health.


***Introduction***

*Sideritis scardica* is an endemic species in the Balkan Peninsula and it is widely used for the treatment of inflammation, gastrointestinal disorders, and coughs. In traditional medicine, it is utilized as a loosening agent in bronchitis and bronchial asthma, and as an active constituent of dietary supplements for the prevention of anemia. *S. scardica* is known as a healing aromatic herbal tea in traditional medicine of the Balkan countries. "Mountain tea" is made from the aerial parts of the plant by infusion or decoction. In the folk medicine the tea was used, along with other herbs, as antirheumatic and immune stimulating agent. Besides its uses as herbal tea, ethanolic extract of *S. scardica* is applied topically as antiseptic after tooth extraction and for oral sores (**1**).

Amongst the plethora of methods used for the evaluation of antioxidant activity, the DPPH· test is very useful in the micromolar range demanding minutes to hours for both lipophilic and hydrophilic samples. In the presence of an antioxidant, which can donate an electron to DPPH, the purple color being typical of the free DPPH radical decays and the change can be measured spectrophotometrically (515 nm). This interaction indicates its radical scavenging ability in an iron-free system. In cases where the structure of the electron donor is not known (e.g. plant extract), this method can afford data on the reduction potential of the sample and hence can be helpful in comparing the reduction potential of unknown materials.

The therapeutic benefit of medicinal plants is often attributed to their antioxidant properties. Plant infusions and decoctions from medicinal plants have been used for over 2000 years in traditional medicine. The beneficial properties of these herbal "teas" are often associated with the bioactive compounds of the medicinal plants, which are used for their preparation. *S. scardica* has also been used, on the basis of its antioxidant properties, to prepare herbal teas. Moreover, concerning the adverse effects of the crude extracts from medicinal plants on some organs, it is highly recommended that the crude extracts should be processed into different fractions, each to be used for a precise clinical disorder. The aim of this research was the study of the essential oil of *S. scardica,* to find bioactivities in this medicinal plant of the Greek flora, relating to ethnopharmacological uses as antioxidant and to evaluate the antioxidant capacity of the fractions obtained.

***Materials and Methods***

The aerial parts of *Sideritis scardica* were collected during flowering from selected plants, cultivated in experimental lines at IBPGR, Department of Aromatic & Medicinal Plants (Thessaloniki Greece). *Sideritis* germplasm was originated from wild grown populations of N. Greece. Voucher specimen is kept at the Department of Aromatic and Medicinal Plants.

One part of the plant material was extracted in a soxhlet apparatus with petroleum ether (*P.Ether extr.*), dichloromethane (*DM extr.*), methanol (*MeOH extr.*) and water ($H_2O$ *-1 extr.*), consecutively. Another part of the plant was exclusively extracted with water ($H_2O$*-2 extr.*) in the same Soxhlet apparatus.

The antioxidant potential of each extract was evaluated with the DPPH· method, according to *Parejo et al* (**2**)**.** A compound known for its good antioxidant potential, Trolox, was also evaluated for comparative reasons.

The essential oil was obtained by hydrodistillation of the aerial parts of the plant, using a Clevenger type distillation apparatus. The duration of distillation was 3 hours and the rate 3.0-3.5ml/min. The distilled essential oil was dried over anhydrous sodium sulphate and stored at 4-6°C until analyzed.

The essential oil was analyzed on a DB-5 Column [Length: 30m, I.d:0,25mm.], using a Gas Chromatograph Shimadzu GC-17A interfaced with a Mass Spectrometer Shimadzu QP-5050A supported by the GC-MS Solution software. GC conditions; temperature program: 55°C (1min), (1.5°C/min): 55°C→110°C, (3°C/min): 110°C→150°C, (8°C/min): 150°C→220°C, 220°C (10min), Inj. temp.: 260°C, Split ratio 1:30, (70 eV, ion source temp. 200°C). The relative content of each compound was calculated as percent of the total chromatographic area and the results are expressed as means of triplicate experiments. The identification of the components was based on comparison with their retention indices (RI) relative to *n*-alkanes ($C_7$-$C_{40}$), with corresponding literature data, and by matching their spectra with those from MS libraries.

***Results and Discussion***

Radical scavenging activity expressed as $EC_{50}$ (DPPH· test) ranged from 0.775 to 34.956μg dry extract / mg DPPH (**Figure 1**). The methanolic extract of the studied plant possessed, by far, the greatest antiradical activity. In comparison with the standard used, the methanolic extract of *S. scardica* was the strongest antioxidant, with the quarter of the value of the antioxidant capacity of the Trolox. The aquatic fractions showed also potent antiradical activity, except for the petroleum ether and the dichloromethane extracts, which were the weakest antioxidant fractions. The range of antioxidant capacity could be attributed to the physicochemical and structural characteristics of the components contained in each extract. For this reason, the low antioxidant property of the petroleum ether and the dichloromethane extracts could also be attributed to the low concentration of active chemical compounds. This is to be explored when the extensive phytochemical analysis of each fraction is accomplished.

The presence of 21 constituents with over 0.5% yield in the essential oil, was confirmed for the plant. β-Caryophyllene (23%), β-Pinene,(11%), β-Bisabolene (8%), 1-Octen-3-ol (8%) were the main constituents of *S. scardica* (**Table 1**).

This research work proves *in vitro* the good antioxidant capacity of *S. scardica* extracts. All this information may be useful for the promotion of extracts of this medicinal plant of Greek flora, as natural antioxidant in food and medicinal products. Moreover, the findings of antioxidant capacity validated the use of the plant for medicinal purposes and can be an answer to the continual demand for new natural antioxidants.

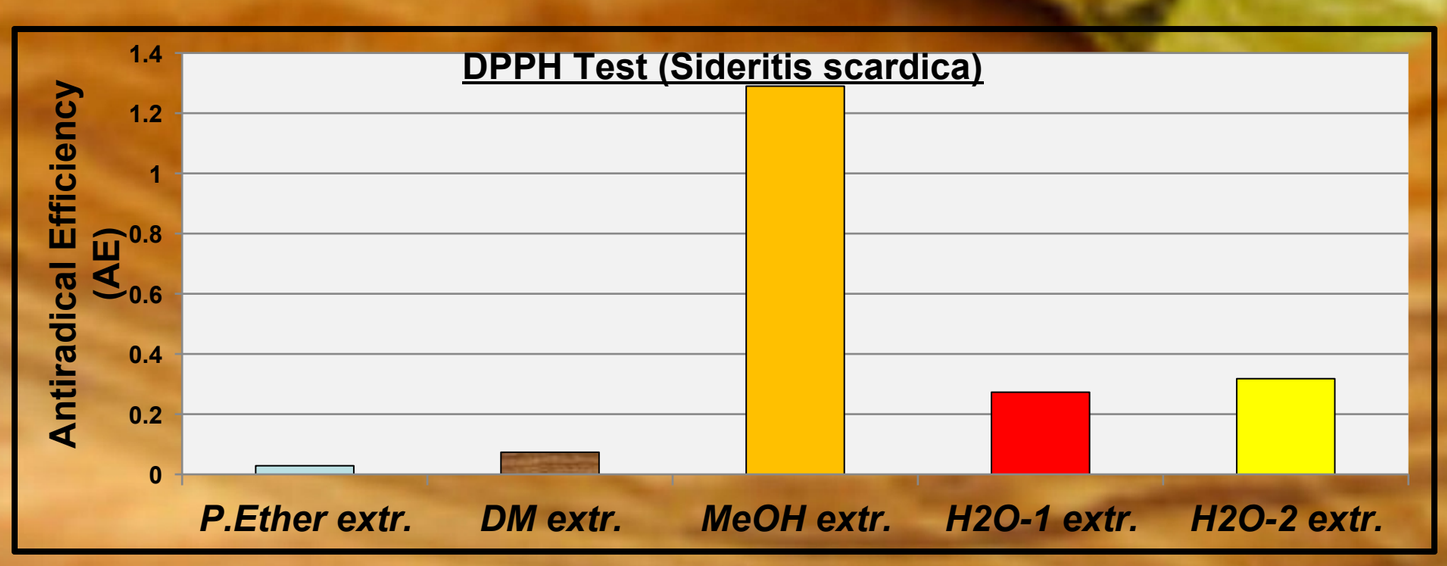
***Table 1***

The percentage yield of the main constituents of the essential oil of *S. scardica*

| | | ***Sideritis scardica*** |
|---|---|---|
| **R.I.** | **Constituents** | ***% yield*** |
| **935** | α-Pinene | 6,23 |
| **974** | Sabinene | 1,49 |
| **976** | β-Pinene | 10,93 |
| **980** | 1-Octen-3-ol | 7,53 |
| **991** | Myrcene | 0,73 |
| **1028** | Limonene | 5,14 |
| **1040** | cis β-Ocimene | 2,66 |
| **1099** | Linalool | 1,18 |
| **1175** | Terpinen 4-ol | 0,60 |
| **1295** | Thymol | 0,97 |
| **1302** | Carvacrol | 2,10 |
| **1374** | a-Copaene | 0,58 |
| **1417** | β-Caryophyllene | 22,75 |
| **1458** | trans β-Farnesene | 3,26 |
| **1480** | D-Germacrene | 1,91 |
| **1495** | Bicyclogermacrene | 6,28 |
| **1508** | β-Bisabolene | 7,91 |
| **1523** | δ-Cadinene | 0,79 |
| **1579** | Spathulenol | 1,49 |
| **1584** | Caryophyllene oxide | 2,77 |
| **1688** | α-bisabolol | 1,50 |

| | ***Antiradical Efficiency (AE)*** | | | | |
|---|---|---|---|---|---|
| | **P.Ether extr.** | **DM extr.** | **MeOH extr.** | **$H_2O$-1 extr.** | **$H_2O$-2 extr.** |
| ***S. scardica*** | 0.029 | 0.073 | 1.290 | 0.273 | 0.318 |

***Figure 1:*** Antiradical Efficiency (AE) of *S. scardica*




AE=1/$EC_{50}$ ; $EC_{50}$ : Efficient Concentration (mg antioxidant / mg DPPH·); amount of antioxidant needed to decrease the initial DPPH· concentration by 50%.

**Trolox AE = 5.59**


***References***

1) Todorova, M. et al. (2014), J. Ethnopharm. 152: 256-265.

2) Parejo, I. et al. (2000), J. Pharmac. Toxic. Methods 44: 507-512



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