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SCREENING OF TOTAL ANTIOXIDANT STATUS OF *RICOTIA* L. AND SOME *GRAMMOSCIADIUM* DC SPECIES FROM TURKEY

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Abstract

This study was aimed at screening the total antioxidant status (TAS) of genus *Ricotia* L. and some *Grammosciadium* L. from Turkey in order to find new potential sources of natural antioxidants. The TAS level of a total of 12 plant extracts from non-edible plant materials was examined. According to the results, hexanic extract of *R. sinuata* plant exhibited the lowest antioxidant status (2.41 mmol Trolox equivalent/l). The highest TAS level was determined in ethanolic extract of the *G. daucoides* (4.36 mmol Trolox equivalent/l). As a result, due to its antioxidative properties, ethanolic extract of *G. daucoides* can be utilized as a natural source of antioxidants.

Introduction

Free radicals are responsible for widespread and indiscriminate oxidation and peroxidation of lipids causing cell death or organ damage (Ansari 1993). Antioxidants are first line of defense against free radical damage, and are critical for maintaining health. The need for antioxidants becomes even more critical with increased exposure to free radicals (Yildirim *et al*. 2012, Rai *et al.* 2014, Hossain *et al.* 2015). Large number of medicinal plants have been investigated for their antioxidant properties. Natural antioxidants either in the form of raw extracts or their chemical constituents are very effective to prevent the destructive processes caused by oxidative stress (Zengin *et al.* 2011). Although the toxicity profile of most medicinal plants have not been thoroughly evaluated, it is generally accepted that medicines derived from plant products are safer than their synthetic counterparts (Vongtau *et al.* 2005, Oluyemi *et al.* 2007). Potential sources of antioxidant compounds have been searched in several types of plant materials such as vegetables, fruits, leaves, oilseeds, cereal crops, barks and roots, spices and herbs, and crude plant drugs (Ramarathnam *et al.* 1995). An organism's metabolism fights against oxidative effects with its own antioxidant defence systems. Elimination and neutralisation of reactive oxygen species is handled by both enzymatic and non-enzymatic antioxidant mechanisms. In practise, TAS represents all of these compounds (Halliwell 1994)

The genus *Ricotia* is represented by nine species which are distributed in the South East Europe, Eastern Mediterranean and adjacent Middle East (Burtt 1951, Appel and Shehbaz 2003). In the flora of Turkey, there are six species (*Ricotia tenuifolia* Sibth. & Sm., *R. sinuata* Boiss. & Heldr., *R. carnosula* Boiss. & Heldr., *R. davisiana* B.L. Burtt., *R. varians* B.L. Burtt. and *R. aucheri* (Boiss.) B.L. Burtt). *Ricotia* has five endemic species in Turkey. The rate of endemism of *Ricotia* species in the flora of Turkey is 83.3% (Davis 1985).

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The genus *Grammosciadium* DC. (Apiaceae) comprises nine taxa in Turkey [*G. daucoides* DC., *G. cornutum* (Nábělek) C.C.Towns., *G. macrodon* Boiss., *G. pterocarpum* Boiss., *G. platycarpum* Boiss. & Hausskn. ex Boiss., *G. confertum* Hub.-Mor. & Lamond, *G. schischkinii* (V.M.Vinogr. & Tamamsch.) V.M.Vinogr., and *G. haussknechtii* Boiss. *G. scabridum* Boiss.] (Hedge and Lamond 1972, Pimenov and Leonov 2004, Behçet *et al.* 2012) three of them - *G. schischkinii, G. haussknechtii* and *G. confertum* - are endemic to Turkey (Pimenov and Leonov 2004). All species of the genus have an Irano-Turanian element, except for *G. confertum* (Hedge and Lamond 1972).

The present study aims at investigating the total antioxidant status (TAS) of some plants from Turkey in order to find new potential sources of natural antioxidants.

Materials and Methods

Ricotia tenuifolia, *R. sinuata, R. carnosula, R. davisiana, R. varians, R. aucheri, Grammosciadium daucoides, G. macrodon, G. cornutum, G. platycarpum, G. pterocarpum, G. confertum* were collected from different regions of Turkey (Table 1). The plant material was identified by Dr. Mehmet Yavuz Paksoy, Department of Environmental Engineering, Tunceli University, Turkey. The fresh parts of the plant materials were cleaned and dried in the shadow for extraction.

Table 1. Location of the sampling area.

Plant species	Locations
Ricotia tenuifolia	Antalya; between Finike and Elmalı, Calcareus rocky, 390 m, 24.04.2010, Paksoy 1080
R. sinuata	Akseki; Çukurköy, 965 m, 26.04.2011, Paksoy 1135
R. carnosula	Antalya; Kemer, Goynuk canyon entrance, Goynuk stream vicinity, 10 m, 24.04.2010, Paksoy 1075
R. davisiana	Antalya; Kemer, Tahtalı mountain, Peynirlik located, 1600 m, 17.07.2010, Paksoy 1098
R. varians	Isparta; Aksu, Dedegol mountain, Obruk plateau, 1350 m, 01.08.2010, Paksoy 1104
R. aucheri (Boiss.)	Kahramanmaras; Caglayancerit, Oksuz mountain, Akdut located, 1200 m, 11.06.2010, Paksoy 1094
Grammosciadium daucoides	Tunceli; Ovacık,Around The Sahverdi village, 05.05.2012, Paksoy 1182
G. macrodon	Van; Bahçesaray, Karabel(Varikrapit) gateway-between Bahçesaray, 2400 m, 29.05.2012, Paksoy 1208
G. cornutum	Van; Çatak, Around Alacayer village, 2100 m, 29.05.2012, Paksoy 1207
G. platycarpum	Mus: Malazgirt, Kazgölü, 1800 m, 25.06.2012, Paksoy 1218
G. pterocarpum	Elazığ; Around Tekevler village, 1550 m, 27.05.2012, Paksoy 1203
G. confertum	Adana; Tufanbeyli, Güzelim village, larch forest, 1430 m, 14.07.2012, Paksoy 1244

Aqueous, hexanic, methanolic and ethanolic extracts were obtained from leaves. Fresh plant materials were washed with tap water, air dried and then chopped into small fragments, which were shade-dried and reduced to a coarse powder in a mortar and pestle. The aerial parts of the plant samples (2 g) were extracted with 20 ml methanol (MetOH), ethanol, water and hexane. The organic solvents were evaporated to dryness under vacuum at low temperature using a rotary evaporator (Yildirim *et al.* 2013). These extracts are: aquatic, methanolic, ethanolic and hexanic extracts of *G. macrodon, G. confertum, G. pterocarpum, G. cornutum, G. daucoides* and aquatic, methanolic, ethanolic and hexanic extracts of *R. carnosula, R. tenuifolia, R. davisiana, R. varians, R. sinuata, R. aucheri.*

Total antioxidant status was determined by using Rel assay diagnostics TAS assay kit (Lot. RL024) by Multiscan FC (Thermo). Antioxidants in the sample reduce dark blue-green colored ABTS radical to colorless reduced ABTS form.

The change of absorbance at 660 nm is related with total antioxidant levels of the sample. The assay is calibrated with a stable antioxidant standard solution which is traditionally named as Trolox equivalent that is a vitamine E analog.

Results and Discussion

TAS levels of genus *Ricotia* L. and some *Grammosciadium* L. from Turkey are shown in Table 2. The total antioxidant status of *Grammosciadium* ranged from 3.20 to 4.36 mmol Trolox equiv./l, (Table 2). The highest TAS value was found in ethanolic extract of *G. daucoides* (4.36 mmol Trolox equiv./l). Further, differences among the *Ricotia* L. species showed some variability, ranging from 4.29 to 2.41 mmol Trolox equiv./l. (Table 2). The hexanic extracts of *R. sinuata* exhibited lowest total antioxidant status among all samples evaluated in this study (2.41 mmol Trolox equiv./l.).

Table 2. The TAS levels of genus *Ricotia* and some *Grammosciadium* from Turkey.

Plant species	Extraction	TAS*	Plant species	Extraction	TAS*
G. macrodon	Aquatic	4,23	*R. carnosula*	Aquatic	3,98
	Methanolic	4,21		Methanolic	4,29
	Ethanolic	4,21		Ethanolic	4,14
	Hexanic	3,87		Hexanic	4,14
G. confertum	Aquatic	3,28	*R. tenuifolia*	Aquatic	4,13
	Methanolic	3,38		Methanolic	4,27
	Ethanolic	3,75		Ethanolic	4,12
	Hexanic	3,20		Hexanic	4,11
G. pterocarpum	Aquatic	4.00	*R. davisiana*	Aquatic	4.00
	Methanolic	4,19		Methanolic	4,08
	Ethanolic	4.32		Ethanolic	4,05
	Hexanic	3,90		Hexanic	4,27
G. platycarpum	Aquatic	4,23	*R. varians*	Aquatic	4,01
	Methanolic	4,21		Methanolic	4,14
	Ethanolic	4.15		Ethanolic	3,85
	Hexanic	3,85		Hexanic	3,98
G. cornutum	Aquatic	4,03	*R. sinuata*	Aquatic	4,24
	Methanolic	4,23		Methanolic	4,23
	Ethanolic	4,13		Ethanolic	4.21
	Hexanic	4,20		Hexanic	2,41
G. daucoides	Aquatic	4.20	*R. aucheri*	Aquatic	3,68
	Methanolic	4.15		Methanolic	3,95
	Ethanolic	4,36		Ethanolic	3,93
	Hexanic	4,21		Hexanic	4,05

*****TAS: Total antioxidant status (mmol Trolox equiv./l).

Exceptional advances in biomedical sciences since the past century give opportunities to understand the molecular basis of disease that could result in new strategies for treatment and for prevention of pathologies (Kusano and Ferrari 2008). Plants are important source of potentially useful structures for the development of new chemotherapeutic agents (Tona *et al.* 1998). Many reports are available on the antiviral, antioxidant, antibacterial, antifungal, anthelmintic, antimolluscal and anti-inflammatory properties of plants (Mahesh *et al.* 2008).

In our study, the total antioxidant status of *Grammosciadium* L. and *Ricotia* species varies depending on the selected solvents. The total antioxidant status of *Grammosciadium* ranged from 3.20 to 4.36 mmol Trolox equiv./l. The highest TAS value was found in ethanolic extract of *G. daucoides* (4.36 mmol Trolox equiv./l). Further, differences among the *Ricotia* species showed some variability, ranging from 4.29 to 2.41 mmol Trolox equiv./l. (Table 2). The hexanic extracts of *R. sinuata* exhibited lowest total antioxidant status among all samples evaluated in this study (2.41 mmol Trolox equiv./l). Ethanolic extract of *G. daucoides* can be utilized as an effective and safe source of antioxidants. Since the antioxidant compounds found in plants have different polarities, because different solvents are used to isolate antioxidants. Water, methanol, ethanol, and acetone were used in extraction processes. The antioxidant activity of the extract and the yield depend on the selected solvent (Gong *et al.* 2012).

The study was a part of a larger survey in which other functional properties of these extracts such as their antimicrobial, anti-inflammatory effects are also being evaluated. The results suggest that *Ricotia* L. and some *Grammosciadium* L. are potential source of antioxidant molecules. These plants can be used as natural antioxidants. However, further phytochemical analysis is required for the isolation of bioactive molecules from the plant that may show a broad spectra of pharmacological activities.

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