

Lipide-soluble vitamin contents of some *Astragalus* species in Turkey

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Summary. *Background:* It is believed that eight astragalus species of this study have rich vitamin contents. *Purpose:* To determine vitamin levels of eight *Astragalus* species in Turkey for the first time. *Methods:* 1 g seed was homogenised in solvent isopropanol/hexane/ (2:3 v/v) and was treated at 10.000 g along five minutes. Afterwards, at 40°C, extracts were treated on a rotary evaporator. Then, samples were prepared. All of analysis were conducted by HPLC. Seeds were dissolved in mobile phase (methanol/acetonitrile; 25/75 v/v) and were injected 50 µL. The temperature of analytical column was performed at 40 °C. Detection of retinol acetate and retinol were done at 320 nm, and the detection of D2, D3, α -tocopherol acetate, α -tocopherol, δ -tocopherol were done 215 nm for, detection of K1 was done 235 nm. Authentic external standard mixtures were used to detect the vitamins. The findings obtained from analysis were represented as µg/g. *Results:* beta carotene, gamma tocopherol, vitamin D3 and retinol amounts of the *Astragalus* species were found to be quite high. High vitamin amounts of astragalus species provide the use of these plants in the treatment of various diseases. However, alpha tocopherol and vitamin K1 values of astragalus species in the study were found to be lower than the other species belonging to other fabaceae. *Conclusion:* Vitamin contents of eight *Astragalus* species; *Astragalus asterias* Steven, *Astragalus christianus* L., *Astragalus suberosus* Banks & Sol., *Astragalus barbatus* subsp. *Nanus* Ponert, *Astragalus lagopoides* Lam., *Astragalus camptoceras* Bunge ve *Astragalus cretaceus* Boiss. & Kotschy were determined.

Key words: *Astragalus*, lipide-soluble vitamins, HPLC

Introduction

The genus *Astragalus* L. (Fabaceae) is the richest genus of *Angiospermae* in the world, distributed around semiarid steppic regions (1). *Astragalus* L. has two phylogenetic branches; one of them is Old World (Africa, Asia, and Europe) and the other one is New World (America). It has about 2000 taxa with 136 sections in the Old World (2). It is represented by 478 taxa in 63 sections and 202 (42%) taxa endemic to the Turkish flora (3). 6 new endemic taxa were added to the Turkish flora in 2012 and 2018 (4-9) and the number reached 484.

Astragalus species is as forage for livestock and wild animals, although some of them have been recognized as of use in foods, cosmetics, as substitutes

for tea or coffee, or as sources vegetable gums (10). The widespread use of legumes makes this food group an important source of lipid, fatty acids and protein in animal and human nutrition (11). Thus, species of Leguminosae have received considerable attention and their biochemical components (protein, fat, fatty acids, flavonoids) have been investigated (12). Although the fatty acid compositions of some Turkish *Astragalus* species were reported by different researchers (13-17) there was no studies about the lipide-soluble vitamin contents of eight *Astragalus* L. taxa. *Astragalus* species did not find enough studies about sources of vitamin content increased the importance of the research. Therefore, the aim of the present investigation is to determine the vitamin content of eight *Astragalus* L. taxa.

Materials and methods

Study area

Study area was located on the East of Anatolian diagonal, in the skirts of South-Eastern Taurus Mountains, in the Upper Euphrates Region of the Eastern Anatolia Region (18). Elazığ (Fig. 1) belongs to the Iran-Turan Plant Geography Region and falls within the B7 grid square according to the Grid classification system developed by Davis (19). Elazığ Province is bounded to the East by Bingöl, to the West by Malatya, to the South by Diyarbakır, and to the North by Tunceli. It is situated between longitudes 40–38° East and latitudes 38–39° North. The county is 1067 m above sea level (20).

Plant materials

Field study was carried out over a period of approximately one year. During this period, eight vascular *Astragalus* specimens were collected. The plants

were pressed in the field and prepared for identification. Plants were identified using the standard text, “Flora of Turkey and the East Aegean Islands” (19) and were compared with the specimens in Firat University Herbarium. The names of plant families were listed in alphabetic order. Scientific names of plant species were identified according to the International The Plant Name Index: <http://www.theplantlist.org>.

Extraction of plant materials

1 g seed was homogenised in solvent isopropanol/hexane/ (2:3 v/v) (21) and was treated at 10.000 g along five minutes. Afterwards, at 40°C, extracts were treated on a rotary evaporator. Then, samples were prepared based on the method of Sánchez-Machado (22).

HPLC analysis of vitamins

All of analysis were conducted by HPLC. Seeds were dissolved in mobile phase (methanol/acetonitrile; 25/75 v/v) and were injected 50 µL. The temperature of analytical column was performed at 40 °C. Detection of



Figure 1. Geographical location of the study area.

Table 1. Localities of studied *Astragalus* taxa

Plant No	Plant species	Voucher specimen	Locality and altitude
1	<i>Astragalus asterias</i> Steven	3020	Elazığ Firat University campus 1060 m
2	<i>Astragalus christianus</i> L.	3002	Elazığ Baskil, Belhan village 1520 m
3	<i>Astragalus suberosus</i> Banks & Sol.	3040	Elazığ Firat University campus 1060 m
4	<i>Astragalus barbatus</i> subsp. <i>nanus</i> (DC.) Ponert	4129	Elazığ Baskil, Bolucuk village 1600 m
5	<i>Astragalus lagopoides</i> Lam.	1917	Elazığ Baskil, Kayabeyli village 1430 m
6	<i>Astragalus camptoceras</i> Bunge	3030	Elazığ Firat University campus 1060 m
7	<i>Astragalus cretaceus</i> Boiss. & Kotschy	3234	Elazığ Baskil, Hacimustafa village 1800 m
8	<i>Astragalus aduncus</i> Willd.	3010	Elazığ Baskil, Kayabeyli village 1450 m

retinol acetate and retinol were done at 320 nm, and the detection of D2, D3, α -tocopherol acetate, α -tocopherol, δ -tocopherol were done 215 nm for, detection of K1 was done 235 nm. Authentic external standard mixtures were used to detect the vitamins (23). The findings obtained from analysis were represented as $\mu\text{g/g}$.

Results and discussion

In the study, vitamin contents of eight *Astragalus* species; *Astragalus asterias* Steven, *Astragalus christianus* L., *Astragalus suberosus* Banks & Sol., *Astragalus barbatus* subsp. *Nanus* (DC) Ponert, *Astragalus lagopoides* Lam., *Astragalus camptoceras* Bunge, *Astragalus cretaceus* Boiss. & Kotschy and *Astragalus aduncus* Willd. were determined. The lipide-soluble vitamin contents of studied *Astragalus* species were given table 2.

A large amount of beta carotene (β -carotene) was found in *Astragalus asterias* Steven species (1927,03 $\mu\text{g/g}$). Similarly, β -carotene was found in *Astragalus suberosus* Banks & Sol. (795,9 $\mu\text{g/g}$), *Astragalus christianus* L. (538,23 $\mu\text{g/g}$) and *Astragalus lagopoides* Lam. (360,57 $\mu\text{g/g}$). However in a study, 18 genotypes of the Fabaceae family, the highest amount of beta carotene was reported as 0,41 $\mu\text{g/g}$ (24). The amount of beta carotene in some species in the present study is significantly higher than this value.

When the amount of gamma tocopherol (γ -tocopherol) was examined, the highest amount was determined in *Astragalus aduncus* Willd. (1486,45 $\mu\text{g/g}$). Different amounts of gamma tocopherol were measured in the species such as *Astragalus suberosus* Banks & Sol. (1230,95 $\mu\text{g/g}$), *Astragalus lagopoides* Lam. (574,0 $\mu\text{g/g}$) and *Astragalus camptoceras* Bunge (495,39 $\mu\text{g/g}$). These values are higher than the maximum amount of gamma tocopherol 95.3 $\mu\text{g/g}$ in a previous study (25).

In the study, R-tocopherol and D2 were not found in eight *Astragalus* species, whereas vitamin D3 was found in all *Astragalus* species. The most biologically active form of vitamin D in humans is vitamin D3 (cholecalciferol), which is a fat-soluble steroid (26). In this study, the highest amount of vitamin D3 was found in *Astragalus christianus* L. (18,97 $\mu\text{g/g}$) while the lowest amount of *Astragalus asterias* Steven (1,61 $\mu\text{g/g}$) was measured. These values are higher than the maximum amount of vitamin D3 0,06 $\mu\text{g}/100\text{g}$ in a previous study (27).

The level of α -tocopherol acetate was measured at the highest level of *Astragalus christianus* L. (3,43 $\mu\text{g/g}$) but not found in *Astragalus camptoceras* Bunge and *Astragalus cretaceus* Boiss. & Kotschy species. Alpha tocopherol (α -tocopherol) was detected in different amounts of all *Astragalus* species. The amount of alpha tocopherol was highest in *Astragalus aduncus* Willd.

Table 2. Lipide-soluble vitamin amounts of *Astragalus* species

Taxa	β -carotene	γ -tocopherol	R-tocopherol	D2	D3	α -tocopherol	α -tocopherol acetate	K1	Retinol	Retinol acetate
<i>Astragalus asterias</i> Steven	1927,03	-	-	-	1,61	3,01	1,4	0,07	-	-
<i>Astragalus christianus</i> L.	538,23	-	-	-	18,97	5,95	3,43	0,07	0,42	0,3
<i>Astragalus suberosus</i> Banks & Sol.	795,9	1230,95	-	-	15,82	12,46	1,47	0,91	0,49	0,77
<i>Astragalus barbatus</i> subsp. <i>nanus</i> (DC) Ponert	-	-	-	-	4,48	6,16	0,63	-	0,07	0,01
<i>Astragalus lagopoides</i> Lam.	360,57	574	-	-	10,92	12,95	1,96	-	0,35	0,42
<i>Astragalus camptoceras</i> Bunge	-	495,39	-	-	13,23	2,38	-	-	0,07	0,07
<i>Astragalus cretaceus</i> Boiss. & Kotschy	-	-	-	-	3,85	4,13	-	-	0,14	0,07
<i>Astragalus aduncus</i> Willd.	-	1486,45	-	-	4,06	20,85	0,42	-	0,07	0,14

type (20,85 µg/g). However, in a study, the amount of alpha tocopherol was found to be approximately 89.4 µg/g in many species of Fabaceae, and it was significantly higher than the species in present study (25).

Small amounts of vitamin K1 were found in *Astragalus asterias* Steven, *Astragalus christianus* L. and *Astragalus suberosus* Banks & Sol. species, while vitamin K1 was not measured in other *Astragalus* species. In a study, vitamin K1 value in species of fabaceae found to be 1,839 µg/g (28). This value is higher than the vitamin K1 values of all *Astragalus* species in the present study.

Retinol (0,07-0,49 µg/g) and retinol acetate (0,07-0,77 µg/g) were found in small amounts in *Astragalus* species. The highest amounts of these vitamins were found in the *Astragalus suberosus* Banks & Sol. species (retinol 0,49 µg/g, retinol acetate 0,77 µg/g), while could not be detected in *Astragalus asterias* Steven species. In a study of 18 genotypes belonging to Fabaceae family, the highest amount of retinol was determined as 0,0441 µg/g (24). The amount of retinol in most species in the present study is significantly higher than this value.

In the study, vitamin levels of eight *Astragalus* species were determined for the first time. The plants belonging to eight *Astragalus* species in this study were compared with other plants belonging to Fabaceae family. Beta carotene, gamma tocopherol, vitamin D3 and retinol amounts of the species in the present study were found to be quite high. High vitamin amounts of *Astragalus* species in the study provide the use of these plants in the treatment of various diseases. However, alpha tocopherol and vitamin K1 values of *Astragalus* species in the study were found to be lower than the other species belonging to other Fabaceae.

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