

## *Dorycnium vuralii* (Fabaceae), a new species from Türkiye

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**Abstract:** The present study describes a new species *Dorycnium vuralii* (Fabaceae) from Çankırı, Türkiye. The new species is compared morphologically to the taxa of sect. *Bonjeanea* and sect. *Dorycnium* along with pollen micromorphology, leaflet, and seed micromorphology. The geographical distribution of *Dorycnium vuralii* is mapped. The phylogenetic relationships between the new species and other closely related species in the genus are inferred based on DNA data from both cpDNA and nrDNA (rbcL1&ITS). Although phylogenetic implications are different in the two DNA barcoding regions, *D. vuralii* is shown to be a new species in both molecular data. The new species is assessed as critically endangered (CR) following the IUCN Red List Categories and Criteria.

**Key words:** *Dorycnium*, new species, pollen and seed morphology, phylogeny, Türkiye.

### 1. Introduction

The genus *Dorycnium* Miller belongs to the *Loteae* DC. tribe in the subfamily Papilionoideae within the family Leguminosae. *Lotus* L., *Tetragonolobus* Scop., and *Dorycnium* belong to the same tribe, *Loteae*, within the family Leguminosae. The genus *Dorycnium* comprises 12 species distributed mainly in the Mediterranean region and Macaronesia (Slavík, 1995). According to Rikli (1901), the genus is divided into three sections: sect. *Dorycnium*, sect. *Bonjeanea* (Rchb.) Taubert, and sect. *Canaria* Rikli. The first revision of the genus *Dorycnium* in Türkiye was conducted by Demiriz (1969).

According to Demiriz, 8 taxa grow in Türkiye. Later, Vural (1983) described a new species, named *Dorycnium sanguineum*, from Türkiye. Today, the genus has 7 species, 3 subspecies, and 2 varieties (12 taxa) in Türkiye (Kocabaş, 2014; Kocabaş and İlçim, 2020). *Dorycnium* is known as “gernevük” in Turkish.

Many *Dorycnium* species have similar morphological characteristics that make them difficult to distinguish. In such cases, macro- and micromorphological characteristics such as pollen, seed, and phylogeny are frequently used to distinguish taxa (Kocabaş and İlçim 2020; Kramina and Polevova, 2022; Kramina et al. 2021, 2022).

DNA barcoding techniques, or the analysis of standardized DNA markers, have proven useful for this

purpose (Zhang and Jiang, 2020). Kress et al. (2005), Chen et al. (2010), the China Plant BOL Group (2011), and Kress (2017) all detail the widespread usage of these markers in the context of species discrimination and the preservation of threatened species. In particular, the internal transcribed spacer (ITS) region of nuclear ribosomal DNA (nrDNA) has found widespread application in this context. Additionally, the large subunit of ribulose biphosphate carboxylase (rbcL) gene region on chloroplast DNA has also been used recently as a barcode in plants. Studying both cpDNA and nrDNA in one study provides reliable results for phylogenetic studies (China Plant BOL Group, 2011). There are many molecular studies on *Dorycnium* genus (Kramina et al. 2021, 2022; Kocabaş, 2014). Kocabaş (2014) made a revision study with both morphological and molecular data in his study.

During a field trip near Çankırı in June 2022 some interesting specimens of *Dorycnium* were collected by Şahin, an author of the present paper. A detailed morphological examination of the collected specimens revealed that they are close to *D. sect. Bonjeanea* and sect. *Dorycnium*, but they are distinct from both known species of these sections.

This study aims 1) to describe a new species, *Dorycnium vuralii*, 2) to distinguish micromorphological differences of the new species from sect. *Bonjeanea* and

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sect. *Dorycnium*), 3) to determine the phylogenetic relationships of the newly described species *Dorycnium vuralii* within the genus *Dorycnium* by using herbarium specimens and Gen Bank data.

## 2. Materials and methods

### 2.1 Plant materials

Flowering materials of the new species were collected twice during field trips in Çankırı in June and July 2022. All the specimens were identified using the relevant literature (Davis, 1970; Güner et al., 2000; Kocabaş, 2014). They were also compared with the type materials and other representative collections at GAZI and ANK herbaria, and the images of type specimens from E (Appendix 1). The authors followed Brummitt and Powell (1992) and IPNI 2020 (<www.ipni.org>) for the abbreviations of plant names.

Light and scanning electron microscopy were used to examine pollen for clues about its structure. The Wodehouse (1935) method was used to prepare pollen slides for LM. With the use of a Leica DM 500 digital imaging system LM, the following parameters of at least 30 pollen grains were measured and analysed; pollen shape, P: polar axis, E: equatorial axis, ratio P/E, Clg: colpus length; Clt: colpus width, Plg: porus length; Plt: porus width, Plg/Plt, aperture shape, and operculum membrane ornamentation, apocolpium and mezocolpium, intine and exine measurements and ornamentation type.

Gold particles were coated onto aluminum stubs using a sputter-coater (Çeter et al., 2013) to create SEM images of dried pollen grains, leaflets (adaxial and abaxial), and seeds. Kastamonu University's SEM Laboratory used a Jeol JSM 6490LV to make morphological observations. The shape classification of pollen grains is based on the P/E ratio proposed by Erdtman (1966) and the nomenclature is drawn from Faegri and Iversen (1992), Punt et al. (2007), Hesse et al. (2009), Punt and Hoen (2009), Çeter et al. (2013), and Halbritter et al. (2018).

### 2.2 Total DNA extractions, PCR amplifications, and phylogenetic analysis

*Dorycnium vuralii* leaf specimens were collected in the wild to analyze molecular data. Moreover, the herbarium samples of some *Dorycnium* species native to Türkiye were retrieved from the GAZI herbarium. The extraction of DNA from the complete genome was performed using a plant DNA extraction kit manufactured by MACHEREY-NAGEL (NucleoSpin Plant II, Minikit for DNA from plants). The amplification of the ITS region (ITS1+5.8S+ITS2) and *rbcl* (*rbcl1*) was conducted using the primer pairs specified by Hsiao et al. (1995) and Savolainen et al. (2000), respectively. In a total volume of 20 µL, PCR amplifications were performed with 2.5 µL of 5 Hot FirePol Blend PCR Mix (Solis Biotek) (12.5 mM MgCl<sub>2</sub>), 0.4 µL of each

primer pair, 1 µL of template DNA, and 15.7 µL of water. PCR reactions were performed using SuperCycler™ (Kyrattec) with the following optimized cycling parameters: 5 min at 95 °C for initial denaturation, followed by 30 cycles of 30 s at 95 °C for template denaturation, 30 s at 54 °C and 56 °C for the primers of ITS and *rbcl* regions, respectively, for annealing, 90 s at 72 °C for extension, and 10 min at 72 °C for final extension. BM Labosis Company (Ankara) purified and sequenced the materials after they were checked on an agarose gel (2%). Finch Tv software version 1.4.0 (Patterson et al., 2004), developed by the Geopiza Research Team, was used to validate the acquired sequences prior to analysis. The sequences were aligned using the multiple sequence comparison by log expectation (MUSCLE) tools in molecular evolutionary genetics analysis (MEGA) version 11 software (Tamura et al. 2021). In addition, the optimal replacement model for reconstructing the species' phylogeny was identified using MEGA. To analyze the sequences, the computer recommended the general time reversible (GTR) (Nei and Kumar 2000) model with Gamma distribution, and the phylogenetic tree was constructed using the maximum likelihood approach based on the GTR+G model and bootstrap test analysis. The Bayesian evolutionary analysis by sampling trees (BEAST) software package was employed in this study, utilizing the GTR+G substitution model with uniform rates for data partitions. The Yule tree prior was applied, and a starting tree was randomly generated with 10,000,000 Markov Chain Monte Carlo (MCMC) runs. Additionally, the raxmlGUI 2.0 program was utilized, implementing the GTR+fast bootstrap values method, to validate and ensure the reliability of the obtained results. The phylogenetic complex trees were then condensed and merged utilizing the Tree Annotator software, employing a posterior probability threshold of 1 (Drummond et al., 2012; Bouckaert et al., 2019). The visualization of the trees was accomplished using Fig Tree V 1.4.4. The phylogenetic trees generated by the MEGA and BEAST software separately with same model (GTR+G) and the bootstrap and posterior probability values were added on one tree manually for subjecting to a comparative and integrative analysis. The sequences of *Dorycnium* species, as well as selected species from allied genera such as *Lotus* and *Ornithopus* from the family Fabaceae, were obtained from the NCBI database. These sequences were included with the samples used in the study to offer an evolutionary context (Appendix 2).

Additionally, the Splits Tree 4 (v 4.19.0) program was used to indicate the clustal network analysis for *Dorycnium* species based on both ITS and *rbcl* regions. GTR parameters were used for the analysis of the clustal network, and the networks were drawn separately for each region.

### 3. Results

#### 3.1. Taxonomy

*Dorycnium vuralii* Karaman, B.Şahin & Aytaç **sp. nov.**  
(Figure 1,2&3)

#### Diagnosis

*Dorycnium vuralii* is similar to *D. hirsutum* (L.) Ser. but it is distinguished by having stem 10–25 cm long (versus 30–65 cm), leaves composed of 5 leaflets (versus 5–7); internodes 1–2 cm long (versus 1.5–5.7 cm); inflorescence with 3–6 flowers (versus 6–11 flowers); calyx 4–5 mm long (versus 6–12 mm); legume valves contorting at maturity (versus not contorting at maturity). Also, *D. vuralii* is similar to *D. rectum* (L.) Ser. but it is distinguished by having a stem 10–25 cm long (versus 35–100 cm long); leaves composed of 5 leaflets (versus composed of 5–7); internodes 1–2 cm long (versus 2–7 cm.); inflorescence with 3–6 flowers (versus 12–35), peduncle 1–1.4 cm long (versus 3–13 cm long); calyx teeth shorter than tube (versus teeth longer than tube), standard white and with purple lines, 4–5 mm long (versus white or pink, 6–7 mm.), legume ovate-oblong, 5–7 × 2–3 mm, glabrous, with a beak of 1.5–2 mm, and 1-seeded (versus linear-cylindrical, 8–14 × 1.5–3 mm, beak 3–4 mm, and 3–7 seeded).

On the other hand, it is similar to *D. graecum* (L.) Ser. distinguished from it by having stem 10–25 cm long (versus 25–90 cm), leaves composed of 5 leaflets (versus 5–7), rachis 2–5 mm long (versus 1–2.5 mm), leaflets 3–17 × 1–6 mm long (versus 10–40 × 3–15 mm long), internodes 1–2 cm long (versus 1.5–8 cm), inflorescence with 3–6 flowers (versus with 7–47), peduncle 1–1.4 cm long (versus 1.5–7 cm), calyx 4–5 mm long (versus 2–4.5 mm), standard 4–5 mm long (versus 7–7.5 mm), keel purple (versus white); style 2 mm long (versus ca.1 mm); legume ovate oblong, 1-seeded, opens at maturity (versus oblong-cylindrical to conic, 1–4-seeded, not open at maturity), seeds oblong-reniform (versus ovoid-orbicular).

#### Type

Türkiye, Çankırı: Bozkır mountain, Elmalıpınar village, 800–900 m, steppe, 08.06.2022, B. Şahin 8117 (holotype GAZI, isotypes: ANK, HUB).

#### Paratype

Türkiye, Çankırı: Bozkır mountain, Elmalıpınar village, 800–900 m, steppe, 10.07.2022, B. Şahin 8151 (GAZI).

#### Description

Glaucous, erect, suffrutescent, 10–25 cm high, with erecto-patent branches. Stems and branches with dense appressed to ascending sericeous hairs. Stipules minute, subulate, 1–2 mm, free. Leaves sessile, composed of 5 obovate-oblong leaflets; leaflets 3–17 × 1–6 mm, mucronate, long spreading to appressed-hairy, caduceus. Inflorescence a few-flowered subcapitate umbel, with 3–6 flowered. Peduncle 1–1.4 cm. Pedicels appressed to ascending sericeous, 8–16 mm long. Bract 1.5–4 mm. Calyx 4–5 mm long, densely

appressed-pubescent, green; teeth triangular, green, acute, equal, slightly shorter than the calyx tube. Standard white with purple lines, ovate, 4–5 × 2–3 mm, obtuse to slightly emarginate at the apex, glabrous; wings white with purple lines, 3.5–4 mm; keel purple, 3.5–4 mm. Stamen-tube 2–3 mm. Ovary sessile. Stylus 2 mm, glabrous. Legume ovate-oblong, 5–7 × 2–3 mm, glabrous, with a beak 1.5–2 mm, 1-seeded. Seeds light brown, spotted or not, oblong-reniform, 2–3 × 1–2 mm.

#### Phenology

Flowering is in June, and fruiting is in July.

#### Etymology

This species is named in honor of Prof. Dr. Mecit VURAL, who defined a new species belonging to the genus *Dorycnium* from Türkiye for the first time and contributed a lot to the flora of Türkiye.

#### Suggested Turkish name

*Dorycnium* is called “gernevük” in Turkish. We propose “Beygernevüğü” as a vernacular name (Kandemir et al., 2016).

#### Distribution and habitat

*Dorycnium vuralii* is currently known only from the type locality in Bozkır mountain, Elmalıpınar village, Çankırı, Türkiye (Figure 4). The area where *D. vuralii* is found in a place where evaporitic depositional rocks occurring in the tertiary and ophiolitic rocks come together and form a mixture. Clay soils are formed by the accumulation of soils formed on the rocks on the slopes by erosion. This species lives on a slope with heavy textured soil consisting of very dense clay compared to its environment. Microelement analysis was carried out from the soil on this slope since no plants were grown in some parts or sparsely at most. According to the analysis, boron is low in the vegetation part of these soils and high in the bare part, and salinity is the same. Potassium, magnesium, and phosphorus are all found to be high, along with a moderate level of iron, and little nitrogen. pH is neutral or slightly alkaline. In this case, salinity and boron levels are seen as prominent values. While there are no plants in salty and high-boron areas, there is a sparse variety of plants in some areas. Likewise, high clay saturation exists. Under the influence of these limiting factors, a special soil structure with dense clay and salt has developed and covered an area of about 10 ha, unlike the marly-gypsum-calcareous soils around it. *D. vuralii* is found at the base of about 1 ha of this area with invasive species in places. It has a habitat and ecological environment here in the form of an isolated island. Therefore, the living space is very limited. There are no ancestral species in close proximity with which it can hybridize, transfer, or share genes. Some processes which were created by extreme factors originating from the particular habitat may have probably been effective in the differentiation of this species. Therefore, future studies on the ecological environment would be useful.



Figure 1. Holotype of *Dorycnium vuralii* sp. nov. (scale bar: 1 cm).

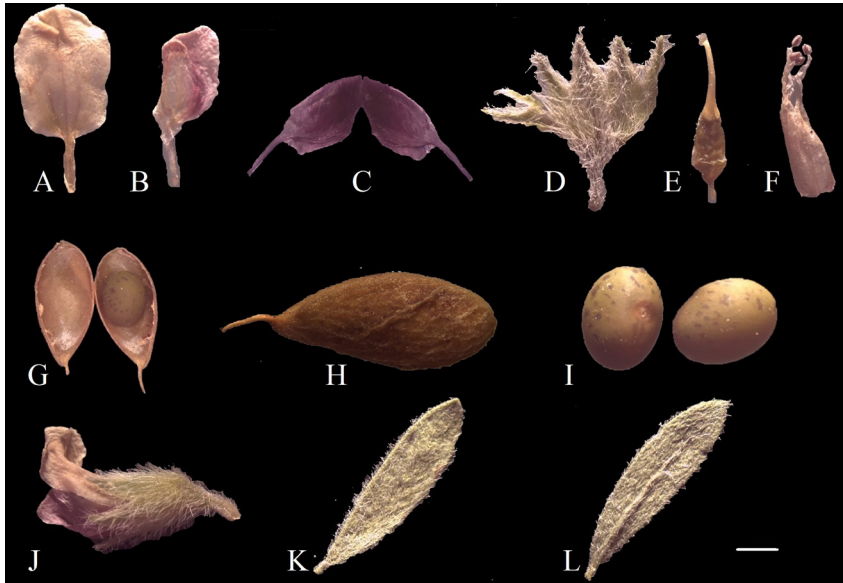
**Conservation status**

*Dorycnium vuralii* is known from a single location; its extent of occurrence (EOO) and area of occupancy (AOO) are approximately 1 km<sup>2</sup> (criterion B1, B2); number of mature individuals are less than 250 (D1); number of locations is only one (a), estimated continuing decline (b) in area of occupancy (ii), quality of habitat (iii), and number of mature individuals (v). Threats such as urbanization, overgrazing, habitat loss because of agricultural activities, and climate change exacerbate the situation. As a result, this new species is assessed as ‘Critically Endangered: CR B1ab (ii,iii,v) + B2ab (ii,iii,v)’ (IUCN, 2021).

**Distinguishing key of *Dorycnium* species in Türkiye**

1. Calyx with 5 equal teeth; corolla wings free at apex; legume 1–8 seeded, valves contorting or not contorting at maturity (Sect. *Bonjeanea*).  
 2. Leaves composed of 5–7 leaflets; inflorescence more than 6 flowered (6–47).

3. Flower 9–16 mm; legume valves not contorting at maturity.....*D. hirsutum*  
 3. Flower 4–5 mm; legume valves contorting or not contorting at maturity.  
 4. Rachis 5–11 mm; legume valves contorting at maturity; ..... *D. rectum*  
 4. Rachis 1–3(–5) mm; legume valves not contorting at maturity; .....*D. graecum*  
 2. Leaves composed of 5 leaflets; inflorescence 3–6 flowered .....*D. vuralii*  
 1. The calyx slightly 2-lipped, 2 upper calyx teeth wider compared to the other 3 lower teeth; corolla wings connected at apex and inflated; legume 1-seeded, valves not contorting at maturity.....  
 (Sect. *Dorycnium*)  
 4. Inflorescences almost sessile, 2–3 flower clusters per leaf axil.....*D. axilliflorum*  
 4. Inflorescences pedunculate and leaf axils with single flower clusters



**Figure 2.** *Dorycnium vuralii* A- standard, B- wing, C- keel, D- calyx, E- ovary, F- staminal tube, G- legume valves contorting at maturity, H- legume, I- seed, J- calyx and corolla, K- adaxial surface of leaflet, L- abaxial surface (scale bar: 1 mm).



**Figure 3.** Habit of *Dorycnium vuralii* in its natural habitat. A- general view, B- flower, C- legume (Photographed by B. Şahin).

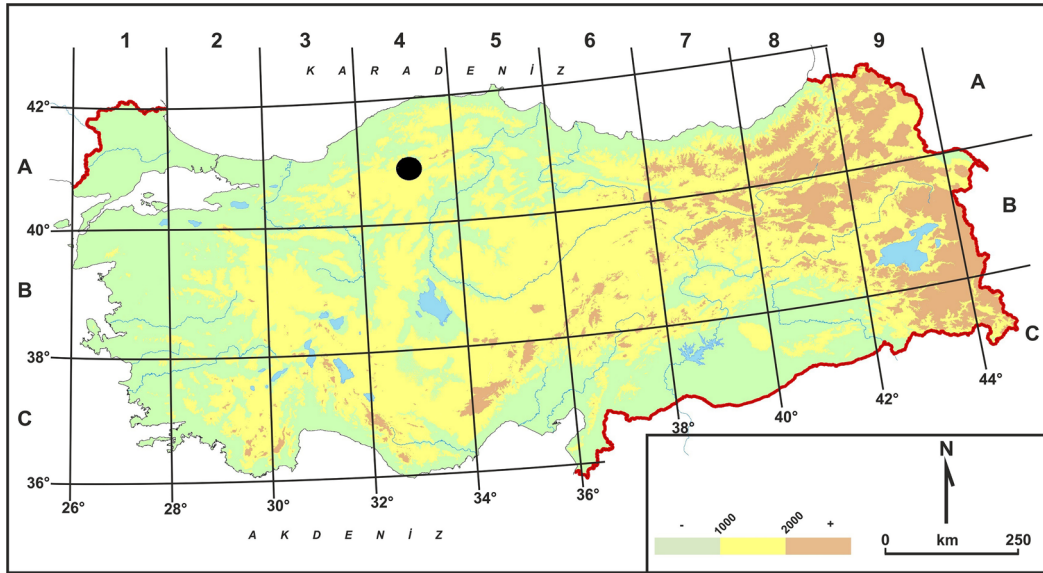


Figure 4. Distribution map of *Dorycnium vuralii* (●).

5. Leaflets up to 5 mm wide, lateral inflorescences long, pedunculate.....*D. pentaphyllum*  
 5. Leaflets 5–10 mm wide; lateral inflorescences short, pedunculate, or subsessile  
 6. Flowers white or bright yellow–white.....*D. amani*  
 6. Flowers blood red .....*D. sanguineum*

### 3.2. Pollen morphology

Pollen grains were observed to be trizonocolporate, radially symmetrical, and isopolar in *Dorycnium vuralii*. The pollen shape of *D. vuralii* is prolate–spheroidal, spheroidal, or rarely oblate–spheroidal (ratio P/E  $1.01 \pm 0.02$ ), polar view (P)  $22.72 \pm 0.93 \mu\text{m}$ , equatorial view (E)  $22.41 \pm 1.03 \mu\text{m}$ . Colpus length (Clg)  $14.93 \pm 1.15 \mu\text{m}$ , and colpus width (Clw)  $3.45 \pm 0.44 \mu\text{m}$ . Pore prolate–spheroidal (pore length/pore width (Plg/Plt)  $1.09 \pm 0.09$ ), pore length (Plg)  $7.43 \pm 0.77 \mu\text{m}$ , and pore width (Plt)  $6.73 \pm 1.08 \mu\text{m}$ . The aperture is operculate, and the operculum membrane has granulate ornamentation. Apocolpium  $11.27 \pm 3.58 \mu\text{m}$ , mezocolpium  $15.14 \pm 0.81 \mu\text{m}$ . Intine  $0.68 \pm 0.07 \mu\text{m}$  and exine  $0.69 \pm 0.07 \mu\text{m}$ . Ornamentation is psilate–perforate (Figure 5).

### 3.3. Leaflet morphology

The leaflets of *D. vuralii* have densely simple hairs at the upper and lower surfaces. Hairs are slender and spiral-like, showing striate–verrucate surface ornamentation. Leaflets are amphistomatic, and the stomata are mesomorphic (Figure 6).

### 3.4. Seed morphology

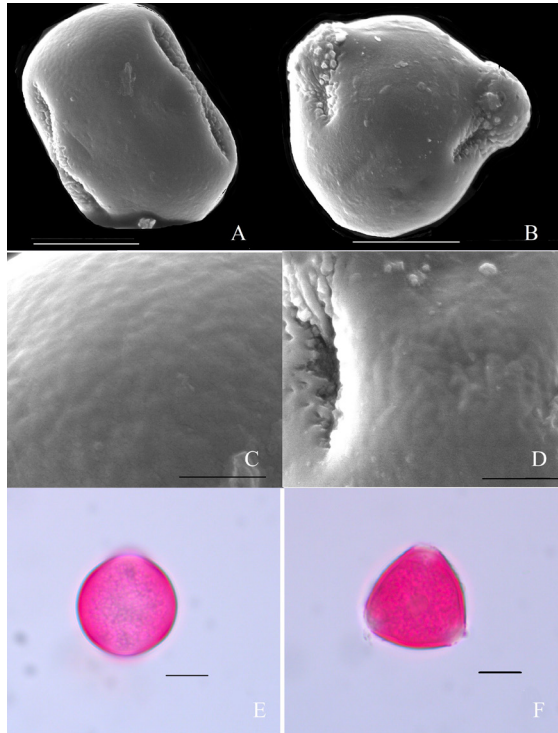
The seeds of *Dorycnium vuralii* are light brown, spotted or not, oblong–reniforme,  $3.22 \pm 0.32 \times 2.32 \pm 0.24 \text{ mm}$ . The surface ornamentation is psilate–verrucate near the hilum microreticulate. The hilum shape is spheroidal, and  $0.27 \pm 0.04 \times 0.27 \pm 0.03 \text{ mm}$  in size (Figure 7).

### 3.5. Phylogeny

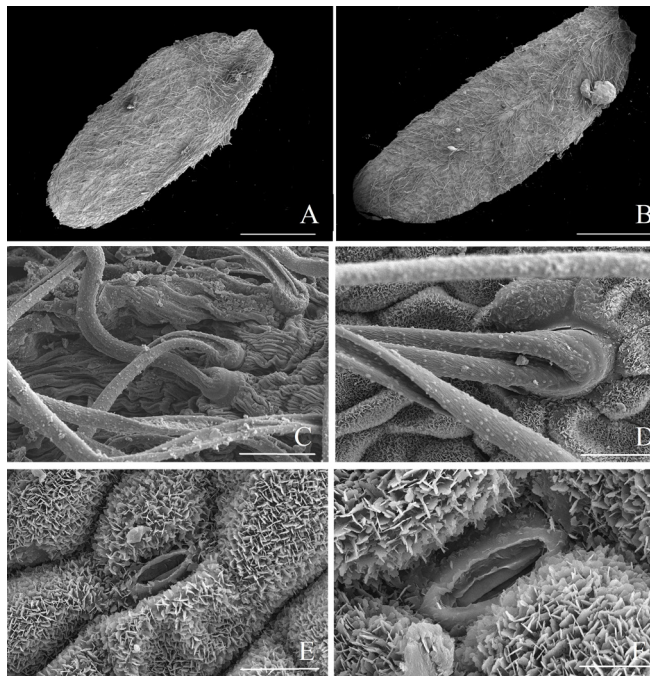
ITS sequences were obtained as a total of 587 bp in length (after exclusion of gap-rich and ambiguous bases) and 84 of them were variable. Moreover, rbcL1 sequences (after exclusion of gap-rich and ambiguous bases) were obtained partially as 570 bp in length, and 11 of them were variable. The overall mean genetic divergence among *Dorycnium* species based on ITS and rbcL1 regions were calculated as 0.034 and 0.007, respectively. According to the phylogenetic tree constructed using ITS region (Figure 8), there are 2 main clusters, and *D. vuralii* is positioned in the one with *D. sanguineum*, *D. rectum*, *D. graceum*, *D. amani*, and *D. axilliformum*. The other cluster is composed of *D. hirsutum*, *D. pentaphyllum* species, and their subspecies and varieties. Even if the overall mean genetic divergence is lower than the ITS region genetic divergence value, the phylogenetic tree of rbcL1 region (Figure 9) indicates sufficient phylogenetic separations supported with morphological data. *D. pentaphyllum* species and its subspecies formed a cluster, and the other species are positioned at another cluster. The new species, *D. vuralii* is positioned alone in the 2nd cluster and separated from *D. hirsutum* with high pp and bootstrap values. Moreover, both ITS and rbcL1 data were used to show network tree with the same parameters (GTR) and bootstrap analysis. The network indicates similar separations with phylogenetic trees (Figures 10 and 11).

### 4. Discussion and conclusion

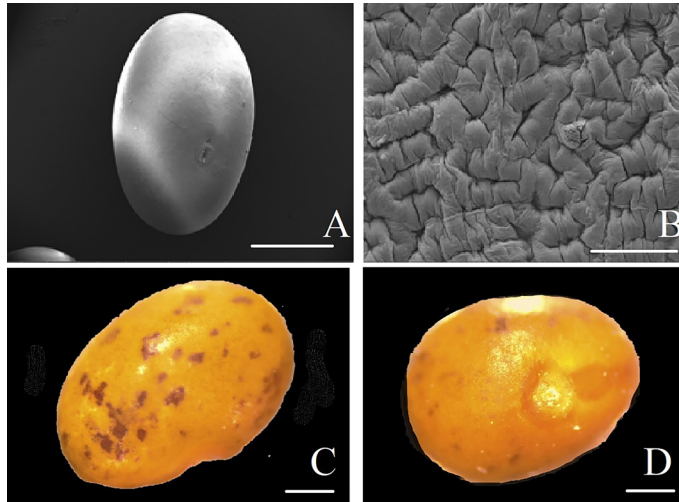
A detailed morphological examination of the collected specimens from *Dorycnium vuralii* revealed that it is close to *D. sect. Bonjeanea* and *sect. Dorycnium* and that



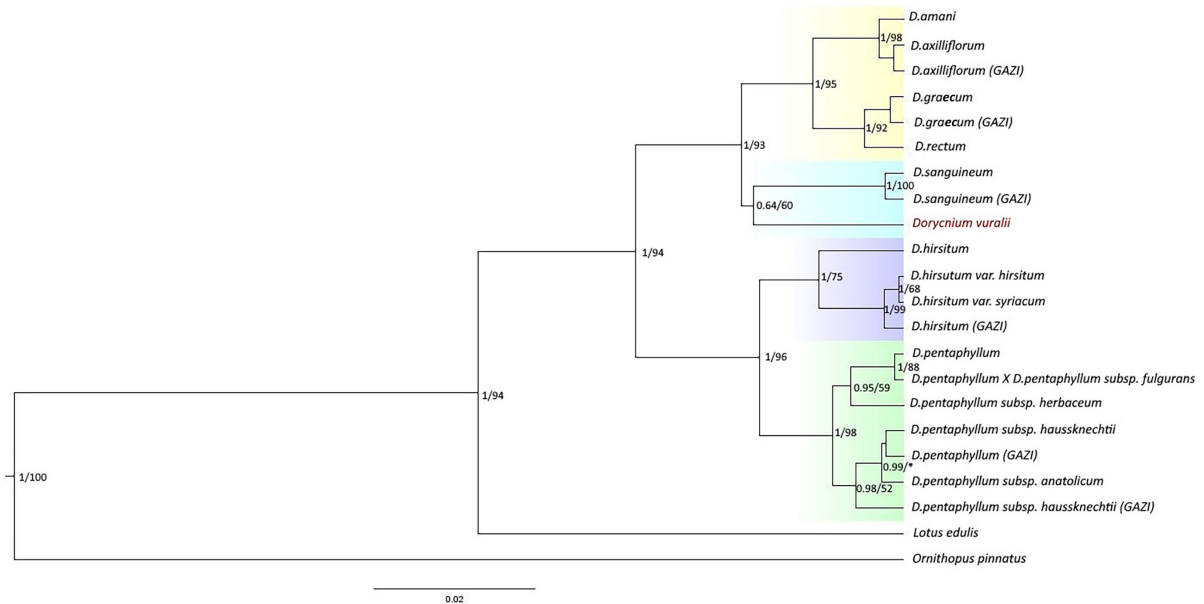
**Figure 5.** Pollen grain SEM microphotographs of *Dorycnium vuralii* A- equatorial view, B- polar view; ornamentation B- equatorial view, C- polar view; pollen grain light microphotographs, E- equatorial view, F- polar view. Scale bars: A, B: 10  $\mu$ m; C: 2  $\mu$ m; D: 4  $\mu$ m; E,F: 10  $\mu$ m.



**Figure 6.** Leaflet SEM microphotographs of *Dorycnium vuralii*. A,C,E- adaxial surface; B,D,F- abaxial surface. Scale bars: A,B: 2 mm; C, D: 50  $\mu$ m; E: 10  $\mu$ m; F: 5 mm.



**Figure 7.** Seed SEM microphotographs of *Dorycnium vuralii*. A-general view of hilum side, B-ornamentation of hilum side; light microphotographs, C- general view, D- hilum. Scale bars: A: 1 mm; B: 10 µm; C,D: 500 µm.



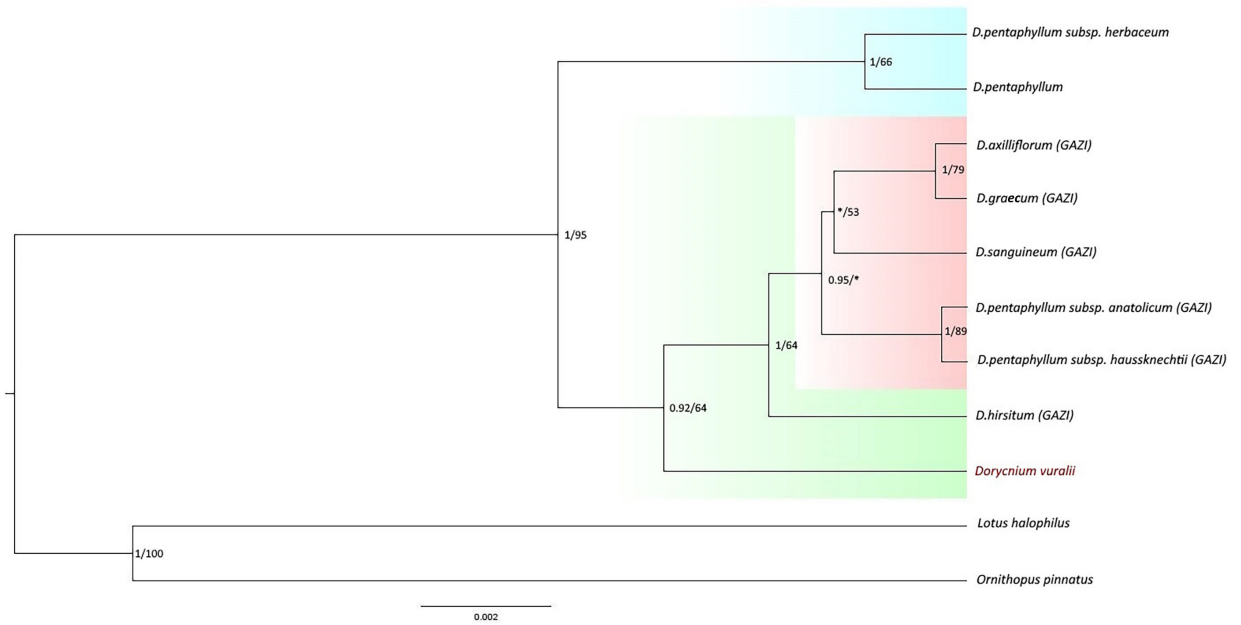
**Figure 8.** Bayesian consensus tree with GTR model with Gamma distribution of ITS gene region of species (FigTree v 1.4.4). The results of ML analysis with subsequent optimization (the posterior probabilities (PP) and bootstrap values with 1000 replicates) and values of over one are given next to the nodes and separated with slashes (the bootstrap values lower than 50 and posterior probability values lower than 0.90 are not shown).

it is distinct from both known species of the section. *D. vuralii* is similar to Sect. *Bonjeanea* by having calyx with 5 equal teeth corolla wings free at apex; valves contorting at maturity and Sect. *Dorycnium* by having calyx teeth shorter than calyx and 1-seeded legume. When the morphological features of the species were examined, it was deemed appropriate to be included in the sect. *Bonjeanea*. *D. vuralii*

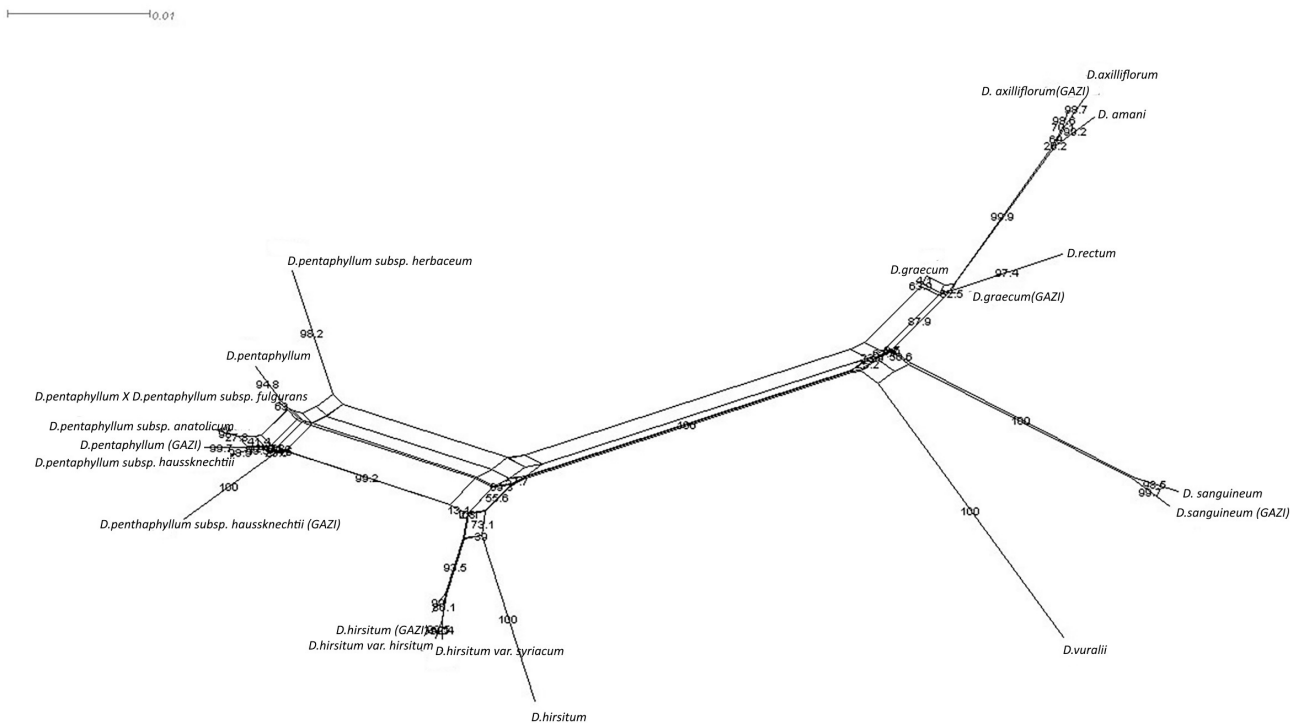
is similar to *D. hirsutum*, *D. graecum*, and *D. rectum*, but distinguished from them by having leaves composed of 3–5 leaflets and inflorescence more than 3–6 flowered. The comparison of diagnostic characteristics of species that belong to genus *Dorycnium* of Türkiye is given in Table 1.

*D. vuralii* grows on a slope with heavy textured soil consisting of very dense clay compared to its surroundings

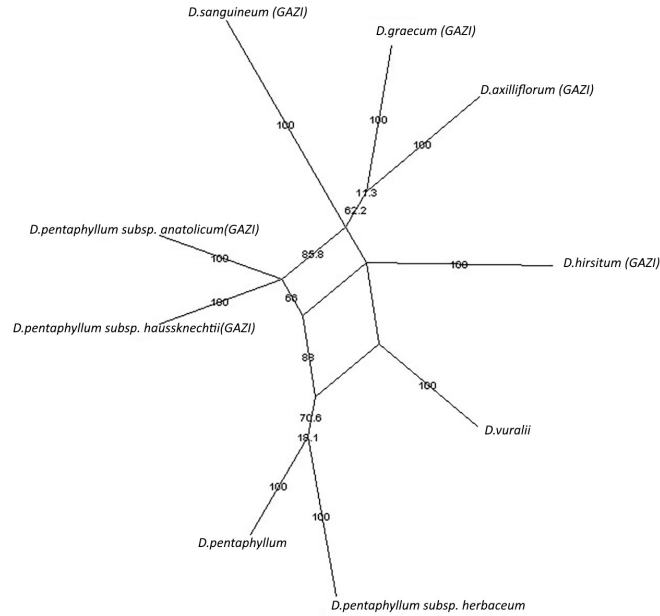




**Figure 9.** Bayesian consensus tree with GTR model with Gamma distribution of rbcL gene region of species (FigTree v 1.4.4). The results of ML analysis with subsequent optimization (the posterior probabilities (PP) and bootstrap values with 1000 replicates) values of over one are given next to the nodes and separated with slashes (the boot strap values lower than 50 and posterior probability values lower than 0.90 are not shown).



**Figure 10.** The clusteral network that is drawn using ITS data with GTR model at Splits Tree 4 (v 4.19.0) software (Bootstrap values are indicated on network branches).



**Figure 11.** The cluster network that is drawn by using rbcL data with GTR model at Splits Tree 4 (v 4.19.0) software (Bootstrap values are indicated on network branches).

at an altitude of 800–900 m. *D. hirsutum* var. *hirsutum* generally spreads in vegetations at flow altitude, high temperatures, and high humidity levels in the Aegean, Marmara, and Eastern Mediterranean regions close to the coast. *D. hirsutum*. var. *syriacum* is localized only in a narrow coastal area in Hatay Province. *D. rectum* grows in the Mediterranean and Marmara regions close to the sea but at different heights. *D. graecum* prefers temperate places at different altitudes but is partially humid in the western Black Sea, most of the Eastern Mediterranean, Central Anatolia, the northern Aegean, and the Marmara regions. *D. sanguineum* Vural is found in a narrow area in Karaman (Kocabaş, 2014).

When the pollen morphology of *D. vuralii* is examined, it differs from other species of *Dorycnium* taxa that grows in Türkiye with its spheroidal pollen shape. *D. sanguineum* is the closest species in terms of pollen shape and size (Kocabaş, 2014). In terms of seed morphology, *D. vuralii* differs from all species distributed in Türkiye with its psilate verrucate seed ornamentation. Seed shape of *D. vuralii* is closer to *D. pentaphyllum* Scop. and *D. hirsutum* (Kocabaş, 2014). Kramina and Polevova (2022) indicate that the seed size, shape, and color only partially allow distinguishing species in the studied group.

Internal transcribed spacer (ITS) regions in plants are frequently indicative of rapid concerted evolution with low levels of intragenomic sequence variation and a small number of polymorphic positions (Nieto-Feliner and Rosselló, 2007; Xu et al., 2017; Osuna-Mascaró

et al., 2022). The diversity of ITS sequences is often accompanied by hybridization events and concerted evolution, resulting in the homogenization of sequences. The process of sequence homogeneity is not fully achieved across ITS sequences, leading to a relatively high level of intragenomic variability (Kramina et al., 2021; Osuna-Mascaró et al., 2022). Conversely, nonconcerted evolution is expected to occur in recently produced hybrid species, when both parental ITS sequences are present (Álvarez and Wendel, 2003). The present study revealed variations in phylogenetic trees as a result of the ITS diversity, which is frequently associated with hybridization occurrences (Álvarez and Wendel, 2003; Osuna-Mascaró et al. 2022). This relatively slow evolutionary process, together with the recent emergence of the group, may not provide adequate differentiation between genetic lineages. On the other hand, the evolutionary trajectory of the nrITS in concert may have a significant impact on the evolution of taxa at lower taxonomic levels, functioning as a mechanism similar to lineage sorting (Mort et al., 2007).

Additionally, Kramina et al. (2022) studied morphology, biogeography, and molecular data of the genus *Lotus*. They found that nuclear ITS results conflicted with plastid phylogeny. They reported that although their plastid region has low variable percentage, the plastid tree has a well-supported clade that combines accessions of *L. dorycnium* s.l. and *L. hirsutus* from the eastern parts of their ranges (including Türkiye). Moreover, Kocabaş (2014), who studied Turkish *Dorycnium* taxa in

Table 1. Comparison of diagnostic characteristics of species which belong to genus *Dorycnium* of Türkiye (Kocataş, 2014).

Section	Sect. <i>Borjeanea</i>			Sect. <i>Dorycnium</i>				
	<i>D. vuralii</i>	<i>D. hirsutum</i>	<i>D. rectum</i>	<i>D. graecum</i>	<i>D. amani</i>	<i>D. pentaphyllum</i>	<i>D. axilliflorum</i>	<i>D. sanguineum</i>
Stem	10-25 cm, branches with dense appressed to ascending sericeous hairs	30-65 cm, sparsely short soft and subappressed hairs	35-100 cm, sparsely short soft and appressed hairs	25-90 cm, curly rough bristly hairs	25-45 cm long, sparsely short appressed hairs	17-65 cm, dense or not, short, soft and subappressed and erect long or short hairs	10-25 cm, sparsely short appressed hairs	20-25 cm, thin long hairs
Leaves	Composed of 3-5 leaflets, rachis 2-5 mm	Composed of 5-7 leaflets, rachis 3-11 mm	Composed of 5-7 leaflets, rachis 1-2.5 mm	Composed of 5-7 leaflets, rachis 1-2.5 mm	Composed of 5 leaflets, rachis 1.5-3 mm	Composed of 5 leaflets, rachis absent	Composed of 5 leaflets, rachis absent	Composed of 5 leaflets, rachis 1-4 mm
Leaflets	3-17 × 1-6 mm, obovate-oblong, long, spreading to appressed hairy, mucronate.	8-20 × 3-10 mm, oblong-obovate, dense soft hairy, mucronate.	12-35 × 5-21 mm, obovate-oblong, sparsely short and appressed-hairy, appressed-hairy, appressed-long hairy or glabrous mucronate	10-40 × 3-15 mm, obovate-oblong to oblanceolate, sparsely short and appressed-hairy, appressed-hairy, appressed-long hairy	10-18 × 5-12 mm, obovate-oblong, sparsely short and appressed-hairy, obtuse.	6-20 × 1-5 mm, obovate or linear, erect to spreading and long and appressed hair, acute to mucronate, obtuse	10-17 × 2-5 mm, oblong-linear, soft appressed hairy, acute.	5-20 × 3-10 mm, obovate-elliptic, dense thin long soft hairy, mucronate
Internodes	1-2 cm	1.5-5.7 cm	2-7 cm	1.5-8 cm	2.3- 7.2 cm	1-6 cm	1.6-4.3 cm	1-3 cm
Inflorescence	with 3-6 flowers, peduncle 1-1.4 cm; pedicel 1-1.5 mm long	with 6-11 flowers, peduncle 1.5-4 cm; pedicel 1-2 mm.	with 12-35 flowers, peduncle 3-13 cm; pedicel 1.5-2.5 mm.	with 7-47 flowers, peduncle 1.5-7 cm.; pedicel 1-2 mm.	with 5-12 flowers, peduncle 1.2-2 cm, pedicel 0.4-0.8 mm.	with 3-35 flowers, peduncle 1-6.5 cm., pedicel 0.7-2.2 mm.	with 2-15 flowers, peduncle 0.5-5 mm., pedicel absent or 1 mm.	with 4-8 flowers, peduncle 0.5-2 cm., pedicel absent or 1 mm.
Calyx	4-5 mm long, densely appressed-pubescent, green, teeth triangular, green, acute, equal, 1.5-2 mm long	6-12 mm long, densely soft long hairs, pinkish, teeth lanceolate-subulate, red to purple at the apex, 3.5-6.5 mm long	3-4.5 mm long, soft long hairs, base of tube pink, teeth subulate, 2-2.5 mm. long	2-4.5 mm long, thin soft long hairs, pinkish, teeth lanceolate-subulate, 1-2.5 mm. long	3-4 mm long, sparsely short appressed hairs, yellowish-green, teeth triangular-lanceolate, 1-2 mm long	1.2-4.5 mm long, densely or sparsely long or short hairs, yellowish or nearly pinkish, teeth dark green or purplish, 0.3-2.2 mm	2.5-3 mm long, soft appressed hairs, teeth triangular, lanceolate, 1.5-2 mm long	4.5-5 mm long, soft long hairs, yellowish-green, teeth narrow triangular-lanceolate, purplish-reddish

Table 1. (Continued.)

Standard	white with purple lines, ovate, 4-5 mm long	white or pink, 15-16 mm long	white or pink, 6-7 mm long	white, 5.7-7.5 mm long	white or yellow-white, 4-4.5 mm long	white, 4-7 mm long	white, 4-5 mm long	red, 5-7 mm, mm long
Wing	white with purple lines, 3.5-4 mm long				white or yellow-white, 4 mm long			
Keel	purple, 3.5-4 mm long				white or yellow-white, 3-3.5 mm.			
Style	2 mm. long	3 mm long	2-2.5 mm long	ca.1 mm long	ca. 1 mm long	0.5 mm long	ca. 1 mm long	ca.1 mm
Legume	ovate-oblong, 5-7 x 2-3 mm, glabrous, with a beak 1.5-2 mm, 1-seeded, opens at maturity	ovate-oblong 6-10 x 2-4.5 mm, with a beak 4 mm, 2-7-seeded, not open at maturity	linear- cylindrical, 8-14 x 1.5-3 mm, beak 3-4 mm, 3-7-seeded, opens at maturity	oblong- cylindrical to conic, 4-10 x 1.5-3 mm, beak 1-2 mm, 1-4-seeded, not open at maturity	ellipsoid, 4-6 x 2-5 mm, beak 1.4-1.8 mm long, 1-seeded, not open at maturity	ovoid, 2-8 x 1-3.5 mm, beak 1-2.2 mm long, 1-seeded, not open at maturity	ellipsoid, 4-6 x 2-5 mm, beak 1.4-1.8 mm, 1-seeded, not open at maturity	ovoid, 5-5.5 x 2.5-3 mm, beak 1.5 mm, 1-2-seeded, not open at maturity
Seeds	oblong-reinforced	oblong-reinforced	oblong-spheroidal	ovoid-orbicular	oblong	reniform or oblong	elliptic	oblong

his PhD thesis, concluded that ITS data cannot support morphological data but provides a different perspective for taxonomic studies. Therefore, these discrepancies, which previously suggested a taxonomic perspective on species differentiation within the phylogenetic trees, now provide new insights for discriminating species within the genus *Dorycnium*.

Allan et al. (2004) studied molecular phylogenetics in *Lotus* genus species. They used *Dorycnium hirsutum* and *Dorycnium pentaphyllum* species to compare the controversy of the *Dorycnium* species. In the phylogenetic tree, the two *Dorycnium* species were positioned close to the *Lotus* species but formed a separate clade. Again, Allan and Porter (2000) investigated the phylogenetic relationships of the families Loteae and Coronillae. In their study, the phylogenetic tree of the nrITS region indicated that *Dorycnium* species (*Dorycnium graecum*, *D. herbaceum*, *D. hirsutum*, *D. rectum*) were grouped with old-world *Lotus* species but formed a separate clade. Therefore, even if *Dorycnium* species were positioned with old-world *Lotus* species, they seemed to be a separated genus.

Moreover, due to the confictions and discussions regarding the systematic categories of certain *Lotus* and *Dorycnium* species, sequences of ITS gene region from Kramina et al. (2022) were obtained from NCBI gene bank and combined with the sequences studied in the current study. The phylogenetic tree supports the newly described species, *Dorycnium vuralii*, by placing it in a separate clade (Appendix 3).

However, it is evident that *D. vuralii*, distributed in Çankırı, represents a new species of the genus *Dorycnium* based on the evaluation of the morphological and molecular data. Therefore, the genus is now represented by 8 species in Türkiye with the inclusion of the species described in this study.

Based on the available morphological, phylogenetic, and palynological evidence, it has been determined that this particular taxon warrants assessment as a new species.

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**Appendix 1: Examined Specimens*****D. hirsutum* (L.) Ser. var. *hirsutum***

Balikesir: Akhisar-Sındırgı, 30 km S of Sındırgı, *Pinus brutia* forest, *Quercus infectoria*, *Cistus creticus*, *Paliurus spinachristi*, *Colutea cilicica*, serpentine soil, 276 m, 31.05.2003, Reeves & N. Adıgüzel 2647 (photo, E00248631!).

İstanbul: Büyükkada, 29.05.1931, H. Demiriz 3294 (ANK!).

İzmir: Near Urla, slopes, sea level, 26.04.1965, Davis 41850 (photo, E00354834!); Smyra, dans lersbroussaillers, 20.05.1854, Balansa 386 (photo, E00354832!).

Mersin: Kuzucubelen Adana: Karataş, nature reserve of Yumurtalık lagoon, dune, 1 m, 19.04.1998, H.Şağban 2058 (GAZI!). Kuzucu-Belen road, Kutuluk-Cemilli, *Pinus brutia* forest, 200 m, 08.05.1952, Demiriz 965 (photo, E00354835!).

Muğla: Marmaris, near the hill, 250 m, 27.06.1997, H.Şağban 1841 (GAZI!).

Osmaniye: Gavur mountain, 18.05.1956, Hub.-Mor. 47 (ANK!); Hatay: İskenderun, Nergislik, Amanos mountains, 700 m, 08.06.1967, Y. Akman 46 (ANK!).

***D. rectum* (L.) Ser.**

Gaziantep: 6 km west of Fevzipaşa, 1 km wordem Nurdağı, 15.07.1973, Holtz, Hanel&Kesercioğlu 00.687 (photo, E00339821!).

Muğla: Köyceğiz, Beyobası village, 50 m, A.Güner 9300 (GAZI!).

***D. graecum* (L.) Ser.**

Ankara: Çamkoru, 1300 m, 10.1958, Alpay 2544 (photo, E00339560!). Çamkoru-Çamlıdere, 1300 m, 10.1958, Alpay 2545 (photo, E00339646!).

Adana: Osmaniye, Zorkun plateau, Amanos mountains, *Pinus nigra* forest, 1300 m, 04.06.1968, Y. Akman 7772 (ANK!); Karsanti, Yapraklı province, bedrock: serpentine, *P. nigra* community, 100 m, 30.06.1973, E. Yurdakulol 1620 (ANK!).

Amasya: Karadağı, Moghuer mountain, 04.06.1889, Bornmueller 262 (photo, E00339543!).

Artvin: Prov. Çoruh, mountain above Artvin, 1450 m, banks, 19.06.1957, Davis&Hedge 29742 (ANK!, photo E00339540!).

Ankara: Kızılcahamam, Kargasekmezcol, 1000 m, 40°24 N, 32°41 E, 27.07.1989, H.M.Nesbitt& D.J. Samuel 2504 (GAZI!); Kızılcahamam, Soğuksu National Park, 1530 m, 31.07.1989, O. Eyuboğlu 1043 (GAZI!).

Bolu: Abant, abiesforest, 850 m, 07.07.1978, E.Yurdakulols.n. (ANK!). Yedigöller national park, Köyveri, 1100 m, 13.06.1977, R. İlarıslan s.n.(ANK!); Ilgazcol open *Abies* muntain, 29.06.1958, Markgraf & Birand 10594 (ANK!); Kale, Kırık plateau, 1550 m, 24.06.1990, İ.Kılınç 1126 (GAZI!); Akçakoca-Karasu, c, 1 km from Halk into Akçakoca, by the main road, c. 60 m, roadside in *Corylus maxima* plantation, Lampinen 7891 (photo, E00339571!).

Aladağ, Kartalköy 1750m, 12.07.1957, Bozakman 2549 (photo, E00339563!). Aladağ on Kartal kaya hill, 1700 m, rock igneous slope, 12.07.1962, Davis&Coode 37429 (photo E00339643!). 10 km sudl Yiğilca, Karadesetal, 400 m, 02.06.1958, macchie am Sudhang, Kühne 2626 (photo, E00339644!). Sunnice mountain nörlđ, Bolu, 1550 m (*Fagus orientalis*, *Abies bornmülleriana*), 12.09.1957, Wagenitz and Beng 92 (photo E00339667!).

Bursa: on road from Bursa to Uludağ, 10 km from hotel at Uludağ, 1000 m, sandy cliffs and scree along road, 16.05.1962, Dudley 34714 (photo, E00339648!). Uludağ, new Bursa, Garig vegetation, 09.06.1964, De Wilde 4049 (photo, E00339556!).

Çanakkale : Kara Biga province, scrub, 22.05.1976, Y.Akman and Quezel 9291 (ANK!).

Eskişehir: Namazlar, *Pinus nigra* forest area, ca. 1550 m, 04.07.1970, T. Ekim 1211 (photo, E00339649!) Above Mihalicçık, 1500 m, open *Pinus nigra* forest, Davis&Coode 37208 (photo, E00339570!).

Hatay: mountain Amanos, regiond'tlosan, 4000-5000 m, 06.1908, Haradjian 2244 (photo, E00339557!); Amanos mountain, Kusliji Mountain, 5000-6500 m, 08.1908, Haradjian 2516 (photo, E00339555!).

İstanbul: Aydos Mountain, S slope, 360m, 26.08.1964, Demiriz 3768 (photo, E00339576!); Yakacık-Aydos road, roadside, 23.04.1966, Demiriz&Enberker 5603 (photo, E00339577!); Alemdağ, near summit, macchie and dry meadow, 29.05.1962, Demiriz 4921 (photo, E00339578!); Alemdağ- Ömerli, 10 km, roadside, 04.06.1967, Demiriz 25498 (photo, E00339579!); Anadolu Hisarı, Elmalibendi (Yeni bend), roadside, 02.07.1966, Demiriz5662 (photo, E00339580!); Sarıyer, around Hünkarsuyu, slopes, 18.06.1967, Demiriz 25543 (photo, E00339581!); Derbent karayolu-İstiridye, macchie, 26.05.1966, Demiriz 5605 (photo, E00339582!); Fatihormanı-Bahçeköy road, 3 km from Bahçeköy, roadside and edge of forest, 08.06.1966, Demiriz&Enberker 5625 (photo, E00339583!); Büyükkada, S of Dilburnu, 02.05.1965, Demiriz 5211 (photo, E00339573!); Kartal, Aydos Mountain, around Altınpinar, Aytuğ&Yaltırık 3333 (photo, E00339651!); Aydos Mountain, S slopers, stony macquis, 19.05.1967, ISTF 25470 (photo, E00339652!); 4 km E Beykoz, roadside, 23.06.1973, Edmondson 422 (photo, E00339655!); Terkos, 3 km offer Arnavutköy, between Cinilihan, ruins and Tabyabayırı, 200 m, macchie, maquis, 06.06.1958, H. Demiriz 3878 (photo, E00339641!); Kilyos, 27 km N of İstanbul, hill, 2 km E of village, clearings in low oak scrub, 40 m, 21.06.1971, Edmondson 394 (photo, E00339647!).

Kastamonu: Ilgaz Mountain, *Picea* forest, south slope, 2000 m, 20.07.1969, J.Darrah 114 (photo, E00038185!); Daday, Sarioluk, oaky, 1000 m, 19.06.1978, O.Ketenöglu



1218 (ANK!); north slope of Ilgaz Mountain, 1600 m, 08.09.1954, Davis & O. Polunin 25049 (ANK!). İnebolu-Abana exit, scrub community, 20 m, 12.06.1990, E. Yurdakulol 3193 (ANK!); Tosya, Çukurhan, Kazdağ province, deduction field, 1600 m, 12.09.1976, 6245 (ANK!); Ayancık, down slope of beech-fir mixed forest, 1480 m, 10.08.1949, H. Demiriz 1967 (ANK!). Küre-İnebolu, ledges of metamorphic rock, 07.06.1954, Davis 21612 (photo, E00339642!). N. side of Ilgaz Mountain, 1600 m, banks in Arboretum, 08.09.1954, Davis & Polunin 25049 (photo, E00339544!); Ilgaz Mountain (from Ilgaz pass, *Abies* forest, NW near top of ridge glades and dry clearings, 17.10.1967, Watson (photo, E00339549!); İnebolu-Küre, 2000 ft, 08.06.1954, Davis 21664 (photo, E00339545!); 150 m, 05.07.1934, Balls 1571 (ANK!).

Kırklareli: Istranca forest, Karamanbayırı, 21.07.1965, Demiriz 5550 (photo, E00339584!). S.E. of Demirköy, *Fagus orientalis* forest, 07.1978, Polunin 15141 (photo, E00339550!); Istranca forest, Valika köprüsü-Balaban deciduous forest and roadside, 21.07.1965, Demiriz 5568 (photo, E00339658).

Kütahya: Dumlupınar, Gökdağ, Akdere, *P. nigra* ormanı, 1350 m, 22.07.1983, M. Vural & H. Malyer 2459 (GAZI!, E00339546!); Simav, Kiçir to Akdağ, on forest road, *Pinus nigra* forest, 19.06.1965, 1600 m, Coode and Jones 2686 (photo, E00339553!).

Manisa: Soma, up to middle station between 200 m., to 400 m, 12.05.1977, Ö. Seçmen 961 (ANK!).

Samsun: New Ankara road, 5 miles inland, 08.06.1963, Tobay 288 (photo, E00339554!).

Sinop: Samsun-Sinop border (Gerze), 250 m, growelly clay roadside bank, 10.06.1965, C. Tobey (photo, E00339548!).

Trabzon: Sürmene, 10 m, road side, 12.07.1966, Demiriz, H. 569 (photo, E00339572!); Çukurçayır, 1 ml S of USAF radar station, 240 m, 25.05.1966, Denton 26 (photo, E00339645!) Sürmene, Kale üstü, 450 m, 20.06.1984, M. Vural (GAZI!).

Uşak: c. 13 miles Dumlupınar to Banaz, 13.06.1965, open *Pinus nigra*, c. 1000 m, woodland, Coode & Jones 2368 (photo, E00339558!).

Yozgat: Akdağmadeni, İşletme müdürlüğü 6. kısım, 04.07.1979, T. Ekim 3930 (ANK!); Akdağmadeni, Çulhalı bölgesi, Aktaş Deresi, 1700 m, 30.08.1965, H. Kayacık, G. Eliçin 3859 (photo, E00339552!).

Zonguldak: E outskirts of town, 70 m, E and E-SE of the hill Fenerburnu hill, 26.08.1964, Demiriz 2473 (photo, E00339574!); North east of Zonguldak, between Kazanyeri and Kapuz, 120 m, edge maquis, 05.05.1951, Demiriz 337 (photo, E00339575!). Mezarlıksırtları-İhsaniye arası tepelerin güney yamaçları, 123 (ANK!); Cide, maki, 100 m, 22.06.1978, O. Ketenoğlu 1219 (ANK!).

***D. pentaphyllum* subsp. *herbaceum* (Vill.) Rouy**

Ankara: Beynam forest, 28.06.1948, H. Bağdas.n. (ANK!).

Artvin: Çoruh, Borçka, 21.06.1957, 150 m, Davis & Hedge 29995 (ANK!); Azdavur, 26.06.1957, 1100 m, Davis & Hedge 30084 (ANK!); Ardanus-Kardevandağı, 1100 m, eroded sholey banks, 26.06.1957, Davis & Hedge 30084 (photo, E00339767!); Borçka, 150 m, igneous banks, 24.06.1957, Davis & Hedge 29995 (photo, E00339768!).

Balıkesir: Edremit, Kazdağı, 1000 m, 11.08.1970, R. Çetik 6 (ANK!); Edremit, Kazdağı, 17.06.1955, K. Karamanoğlu (ANK!).

Bursa: Çekirge, 24.06.1948, H. Bağda (ANK!); Çekirge, 02.07.1936, Gassner 586 (ANK!), İnegöl to Tahtaköprü, 300 m, bozkır, 2.7.1962, Davis & Coode 36633 (photo, E00339772!).

Çanakkale: Bayramiç, between Bayramiç-Evciler, 600 m, 07.1966, H. Uçar 7725 (photo, E00038184!).

Eskişehir: Sündiken mountain, Mihaliçcik, 1200 m, 27.07.1976, T. Ekim 152 (ANK!).

İstanbul: Dragos, roadside sand maguis, 04.06.1966, Demiriz S611 (photo, E00339774!); Şile, Seashore, limestone slopes, 10-15 m, 08.07.1952, Demiriz 1188 (photo, E00339775!); Side, Ağlayan kaya, macchie, 0-10 m, 04.06.1967, Demiriz 25515 (photo, E00339776!) Kilyos, 4 km W, 3,5 km W of Hisarkaya, 08.06.1966, sandy slopes, Demiriz & Enberker S627 (photo, E00339778!); Yakacık, S slopes, macchie, 05.06.1966, Demiriz 5617 (photo, E00339779!); Sarıyer, Belgrad forest (near Kartal hill), 18.07.1960, Yaltırık 1394 (photo, E00339763!).

Kocaeli: İstanbul-Kocaeli road, c. 55 km from İstanbul, 100 m, damp clay slope by road side, 20.07.1956, McNeill 228 (photo, E00339764!).

Kütahya: Kütahya to Tavşanlı 10 km from Tavşanlı, 23.06.1962, Dudley 36116 (photo, E00339773!).

Ordu: southern part of Ordu at 20 km, 600 m, 05.07.1958, H. Birand 10754 (ANK!). Fatsa-Aybastı, 450 m, clay over volcanic rock, 21.07.1965, Tobay 1347 (photo, E00339770!).

Samsun: Kızılırmak oter Asarvillage, 250 m, limestone, slopes, 08.07.1967, Tobay 2241 (photo, E00339765!).

Sinop: Sinop-Bayabat, 18.08.1973, F. Holtz 01.228 (photo, E00339771!).

Trabzon: Meryemana-Maçka, 2 km, roadside, 13.7.1966, Demiriz 5741 (photo, E00339777!).

Zonguldak: Zonguldak to Çaycuma, 450 m, scrubby slopes, 17.07.1962, Davis, Coode & Yaltırık 37634 (photo, E00339759!). 15 km sudl. Ereğli, macchie, Kühne 3293, 09.07.1958 (photo, E00339762!).

Zonguldak: Yeltepe, above Sorgun plateau, 1600 m, limestone, 20.07.1962, D.37881 (photo, E00339766!).

***D. pentaphyllum* subsp. *anatolicum* (Boiss.) Gams**

Adapazarı: nr. Geyve, 100 m, eroded banks, 01.07.1962, Davis 36308 (photo, E00339799!).

Amasya: Yaylacık village, open forest, 1250 m, 19.06.2003, E.Yücel 1043; Yenice-Direkliası, 950 m, 30.05.1987, S.Peker (GAZI!)

Kastamonu: Tosya, Yapraklıyolu, 1350-1400 m, 03.07.1976, M.Kılınç 6228 (ANK!); Devrenkani, İnceğizcivarı, 1150 m, 23.07.1990, E.Yurdakulol 3146 (ANK!).

Ankara: Beynam, 55 km south west of Ankara, 12.07.1965, Ledingham&Ekim 4315 (photo, E00339786!). Beypazarı, *P.nigra* forest, H.Demiriz 8851 (ANK!); Ayaş, H. Karamanoğlu (ANK!); Kalecik, Kalkan province, 17.06.1971, M.Kılınç 90 (ANK!); Beynam mountain, 08.06.1968, Y.Akman 6908 (ANK!); Çubuk, Ovacık-Saraycık village, 1250-1380 m, 27.08.1992, E. DüNDAR (GAZI!); Kızılcahamam, 1600 m, 01.08.1991, M.Vural 5885 (GAZI!); Kızılcahamam, Soğuksu National Park, 1150 m, 10.06.1990, O. Eyüboğlu 1578 (GAZI!); Ayaşbeli, 1150 m, 27.06.1986, M. Vural 4208 (GAZI!); Çubuk II barajı, 110-1200 m, 28.06.1982, F. Demircioğlu 1127 (GAZI!).

Bolu: Pelitçik road junction, 900 m, 07.07.1978, E.Yurdakulol (ANK!)

Burdur: Dirmil-Armutlu, 2-3 km N., scattered, *Quercus sp* forest, 1200m, 02.06.1965, H. Deviriz, G. Attila, T. Aslanerler& I Dölek 5406. (photo, E00339795!); Armutlu-Dirmil, N of Dirmil; N of Dirmilgeçidi (pass), *Pinus brutia-Quercus* forest, 1150 m, 02.06.1965, H. Demiriz, G. Attila, T. Aslanerler& I Dölek 5386 (photo, E00339797!)

Çankırı: Atkaracalar, Dumanlı mountain, 1400-1500 m, 05.07.1992, A. Duran 1591 (GAZI!) , Eskişehir: 7 km NE of Mihalıccık, 1430 m, serpentine oil, , R.D. Reeven 2030, A.R. Kruckeberg, N. Adıgüzel, 28.7.1998 (photo, E00281535!)

Elazığ: Yayılım mountain, 1250 m, 23.06.1983, H.Evren 1564 (ANK!)

Erzincan: Eğin, Kodschodur dagh, 18.06.1890, Sintenis 2656 (photo, E00339789!)

Eskişehir: 7 km NE of Mihalıccık, 1430m, serpentine soil, in pine forest, 18.07.1998, Reeves, Kruckeberg&Adıgüzel 2030 (photo, E00281535!); Sündiken Mountain, 1400 m, 22.06.1974, T.Ekim 953 (ANK!); Türkmen Mountain, Hizanderesi, 1500 m, T.Ekim 2485 (ANK!); Afyon: Sandıklı, Kemer village, 1300-1400 m, 25.06.1985, Y.Akman 13970 (ANK!).

Kastamonu: between Kastamonu and Aras, 06.1967, 1200 m, T. Baytop 911373 (photo, E00339800!)

Kayseri: between Yahyalı-Kuzoluk, Habib bridge province, 1450 m, 09.06.1995, Y. Bağcı 603; Kıranardı, 1560 m, 17.07.1973, R.Çetik (ANK!);

Tokat: Artova, Arabacı Musa village, 1200-1300 m, 11.06.1981, R. İlarıslan 1259 (ANK!).

Kayseri: Bakırdağ above Kısge, 1400 m, 28.06.1952, Davis, Dodds&Çeltik 19306 (photo, E00339787!).

Kırıkkale: Delice, Büyükkavşar village, 1150 m,

10.07.1990, C.Birden 1172 (GAZI!); Koçubaba kasabası, bozkır, 1150 m, 16.06.1990, A.Dönmez 1966 (GAZI!).

Konya: in collibus agricus planiti, Von Heldreich 1845 s.n. (photo, E00339788!); Ağaçyurdu village, 1400 m, 21.06.1979, M.Vural 1637 (ANK!); Ermenek, Damlaçalı, 1800 m, 08.07.1978, M.Vural 967 (GAZI!).

Kütahya: Tavşanlı to Emet, 800 m, calcareous banks, Davis&Coode 36578, 03.07.1962 (photo, E00339790!); Murat Mountain (above Gediz) at Hamam, 1400 m, *Pinus nigra* forest, on slopes, Davis&Coode, 03.07.1962 (photo, E00339732!); N above Şaphane, S foot of Şaphane mountain, stay pasture, 1050-1150m, 24.06.1954, H. Deniz 2099 (photo, E00339796!).

Mersin: 1240 m, 23.06.1971, T.Uslu 891 (ANK!); Kavaklıpınar, 1240 m, 23.06.1971, T.Uslu 891 (ANK!).

Sivas: Karayün, Kızılca kıslavillage province, K. yamaç, 1430 m., 13.96.2009, Ö. Tan 1231; Yıldızeli, Bayat village, 1700 m, 18.07.1979, T.Ekim 3929 (ANK!); next to Ulaş to 25 km, 1500 m, 10.07.1956, H.Demiriz 145 (ANK!).

Yozgat: national park, 1500 m, 26.06.1979, B.Sayın 72 (ANK!); Kayseri: Pınarbaşı, Hınzır mountain, 1850 m, 17.07.1981, N.Çelik 2045 (ANK!); next to ulaş up to 25 km sonra, mountain steppe, 1500 m, 10.7.1956, Birand&Karamanoğlu 145 (ANK!).

#### ***D. pentaphyllum* subsp. *haussknechtii*** (Boiss.) Gams

Isparta: Eğridir, near 5 km to Yılanlıvillage, P.nigramountain, 1200 m, 29.06.1974, H.Peşmen 1365 (ANK!); Antalya: between Manavgat-Akseki 10. km., 300-350 m, 28.03.1993, H.Duman 4980 (GAZI!); Akseki, Geyranplateau, old field, 1150-1250 m, 10.06.1995, A.Duran 2627 (GAZI!).

Adana: Pozantı, Nanelik province, 1570 m, 17.07.1995, Z.Aytaç 707 (GAZI!); Feke GöksunGoryebelowHimmetli, 700-800 m, 09.07.1952, Davis 19837, Dadds, Çetik(photo, E00339806!) Seyhan: above Osmaniye on Fevzipaşa road, 250 m, banks with *Myrtus*, 01.05.1966, Davis 42252 (photo, E00339803!).

Antalya: Akseki to Beyşehir, 10 miles from Akseki, 1400-1500 m, limestone scree, *Abies* and *Cedrus*, Dudley 14.06.1962, D. 35822 (photo, E00339810!); Antalya: Korkuteli, Barutlu Kahu, Edge of *Pinus brutia* forest, 800 m, 03.06.1965, H. Demiriz, P. Enberker, I Dölek& S. Ümit 5449 (photo, E00339813!); Bükormanı, 500 m, 13.06.1959, Alpay 2457 (photo, E00339814!); Manavgat to Akseki, 8 miles from main road junction, macchie with *Erica*, 14.06.1962, Dudley 35758 (photo, E00339817!);

Osmaniye: Eastern part of Osmaniye, 08.09.1884, Postn (BM!).

Hatay: Dört Yol, Kuzuculu to Bülke on forest road, open *Pinus brutia* forest with *Erica*, *Cistus* etc., c. 3000 m, 04.05.1965, CoodeM&Jones, B. S10. (photo, E00339815!); İskenderun, down to Fatırlı village, Amanos mountains, 315 m, 08.06.1967, Y.Akman 7776 (ANK!).

Kahramanmaraş: Engizek mountain, Aksu, 1100 m, 05.07.1986, H. Duman 2232 (GAZI!); Maraş-Göksun road, 24 km from Maraş, alt. 1300 m, in follow field, 14.06.1960, Stainton&Henderoon 5534 (photo, E00339804!)

Mersin: Mut, 5 miles from Mut to Karaman, 300 m, slope under *Pinus brutia*, Coode, M.&Jones, 91813 (photo, E00339807); Anamur: Orman bakımevi ve jandarma yakını, Çam ormanı altı, 27.05.1966, A. Baytop, T. Baytop, B. Çubukçu 9732 (photo, E00339808!, E00339809!); West of Gülnar: East of Bulkari province, İbili suyu, limestone slopes, 930 m, 25.05.1951, H. Demiriz 427 (photo, E00339812!).

**D. amani Zohary**

C6 Hatay: Amanus: AşağıZorkun, 30.6.1932, Eig&Zohary 38187 (photo, E00038187!)

**D. axilliflorum Hub.-Mor.**

Burdur: Burdur-Salda Gölü edges, 1100 m, 05.06.1996, Duman 4796 (GAZI!); Yesilova, south of Salda Lake, 1150

m, 11.07.1983, *Quercus* forest, H. Duman 5089, Z. Aytaç, A.Dönmez (photo, E00077736!).

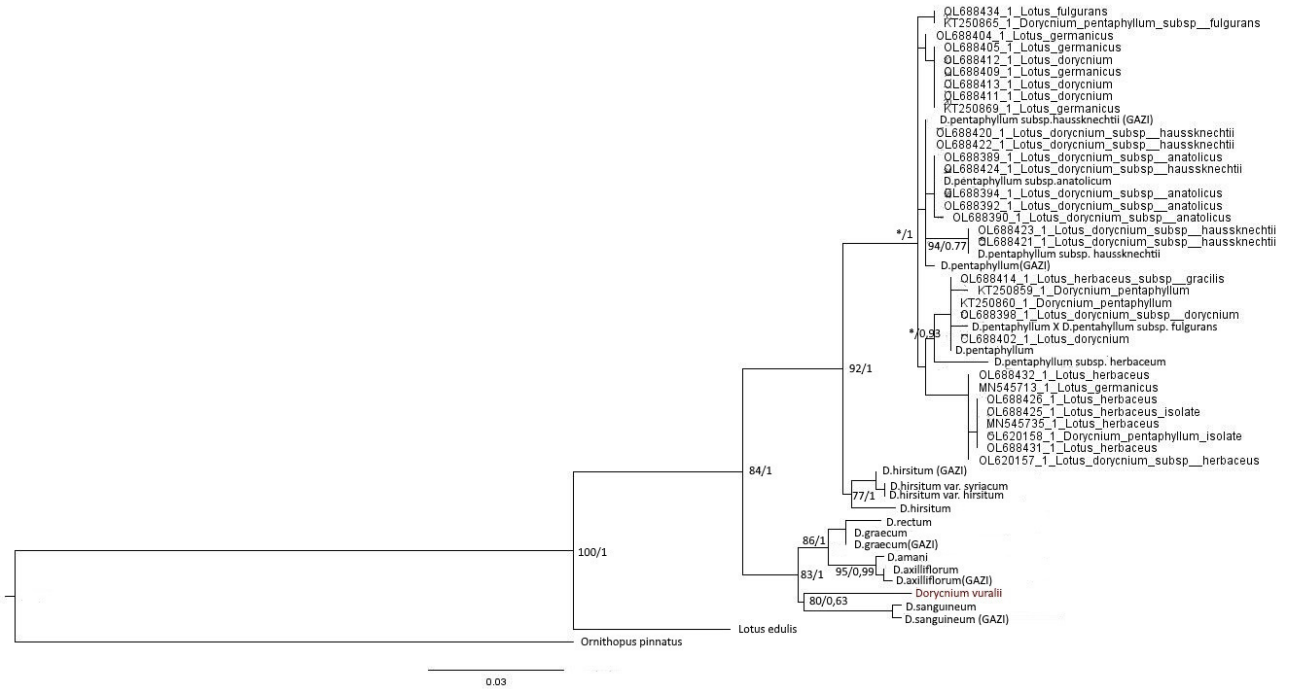
Denizli: Kazıkbeli, 27 km sudlich Denizli steinige Trockenwiese, 02.007.1973, Holtz, F., Hänel, P. &Kesercioglu, T. 470 (photo, E00038186!); Yesilova, south part of Salda lake, 1150 m, 11.07.1993, oakly aperture, H. Duman, Z. Aytaç, A. Durmaz 5086 (photo, E00077736!); Tefenni-Dirmil, 9 km südlichTefenni, *Quercus macchie*, Huber-Morath 8510. 26.06.1948 (photo, E00339537!); Tefenni-Çavdır road, 9 km NNE of Karakuzu pass, stony meadow in deciduous *Quercus* scrub, 1200 m, 02.06.1965, H. Demiriz, G. Attila, T. Aslaner& I Dölek 5362 (photo, E00339538!); Dirmil-Armutlu, 2-3 km N., H. Devris, G. Attila, T. Aslaner& I Dölek 5407 (photo, E00339539!).

**D. sanguineum Vural**

Karaman: Bucakkuşla, eastern slope of Çevlik Mountain, *Pinus brutia* mountain aperture, 550 m, M. Vural 1976 (GAZI, E00038189)

**Appendix 2:** Table indicates the accession numbers and references of the sequence of *Dorycnium* species and outgroups that were obtained from NCBI databank. Additionally, some sequences were obtained from the thesis of Kocabaş (2014) manually.

Species name	NCBI accession number (ITS)	NCBI accession number (rbcL1)	References
<i>Dorycnium pentaphyllum</i>	GQ483311.1	MN019712.1	Conesa et al. (2010) /Jaén-Molina et al. (2021)
<i>Dorycnium herbaceum</i>	-	KF724310.1	Little (2014)
<i>Dorycnium pentaphyllum x Dorycnium pentaphyllum</i> subsp. <i>fulgurans</i>	GQ483313.1	-	Conesa et al. (2010)
<i>Dorycnium hirsutum</i>	AY294292.1	-	Allan and Porter (2000)
<i>Dorycnium rectum</i>	-	-	Kocabaş (2014)
<i>Dorycnium graecum</i>	-	-	
<i>Dorycnium axilliflorum</i>	-	-	
<i>Dorycnium amani</i>	-	-	
<i>Dorycnium sanguineum</i>	-	-	
<i>Dorycnium hirsutum</i> var. <i>hirsutum</i>	-	-	
<i>Dorycnium hirsutum</i> -var. <i>syriacum</i>	-	-	
<i>Dorycnium pentaphyllum</i> subsp. <i>haussknechtii</i>	-	-	
<i>Dorycnium pentaphyllum</i> -subsp. <i>anatolicum</i>	-	-	
<i>Dorycnium pentaphyllum</i> subsp. <i>herbaceum</i>	-	-	
<i>Lotus edulis</i>	KT250863.1	-	Kramina et al. (2016)
<i>Lotus halophilus</i>		KX282862.1	Abdullah (2017)
<i>Ornithopus pinnatus</i>	AY325278.1	MK924798.1	Degtjareva et al. (2003)/ de Vere et al. (2012)



**Appendix 3:** Figure indicates the comparison of *Lotus* and *Dorycnium* species, which have contradictions. ITS gene region sequences were used for constructing phylogenetic tree. The sequences of some *Lotus* and *Dorycnium* species, obtained from the NCBI database using the accession numbers provided from the table in the article by Kramina et al. (2022), represent the origins in various countries. All sequences were combined and were aligned by using MEGA 11 program. In order to examine the sequences, the computer suggested employing the GTR (general time reversible) model proposed by Nei and Kumar (2000), which incorporates a Gamma distribution. Subsequently, the phylogenetic tree was generated using the maximum likelihood method, utilizing the GTR+G model and doing a bootstrap test study with raxmlGUI2.0 program (1000 replicates). The BEAST (Bayesian Evolutionary Analysis by Sampling Trees) software tool was used to analyze the data with posterior probability value of 1. The GTR+G substitution model with uniform rates was employed for the data partitions. The Yule tree prior was employed, and an initial tree was stochastically constructed using 10,000,000 iterations of the Markov Chain Monte Carlo (MCMC) algorithm. The phylogenetic complex trees were subsequently condensed and combined using the Tree Annotator software, with a posterior probability threshold of 1. The trees were indicated using Fig Tree V 1.4.4. For a comparative and integrative analysis, the trees were combined and shown. Bootstrap values lower than 50 and posterior probability values lower than 0.60 were not shown in the figure.