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A new species from Türkiye, *Centaurea turhanii* (Cardueae, Asteraceae)

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Abstract: In this article, a new cornflower, *Centaurea turhanii* Uysal & Hamzaoğlu (Asteraceae) from Türkiye was described and reported. Morphological, karyological, and molecular analyses were used to reveal the taxonomic status of the new species with the species thought to be related to taxa. A modified determination key was submitted and its ecology and phenology were given.

Key words: Endemic, karyomorphology, molecular, SEM, Türkiye

1. Introduction

An important and relatively large genus for The Flora of Turkey (Wagenitz, 1975), *Centaurea* L. (Asteraceae) shows a broad distribution across Eurasia, especially in Irano-Turanian and Mediterranean Regions (Susanna and García-Jacas, 2007). Even the genus was taxonomically problematic, the number of species in the genus is increasing day by day, especially in Asia minor. This shows that the genus is still evolving in terms of speciation. Various methods and approaches have been applied by different researchers for a long time to solve taxonomic problems within the genus *Centaurea* (Romaschenko et al., 2004; Garcia-Jacas et al., 2000; 2001; 2006; Uysal et al., 2009, 2010, 2015, 2016, 2017a; Hilpold et al., 2014; Negaresh and Rahiminejad 2018). Although many species have been transferred to different genera (Wagenitz and Hellwig, 2000; Greuter, 2003; Greuter and Raus, 2009; Negaresh, 2019), Türkiye is still the main diversity center of the genus *Centaurea* (Wagenitz, 1986). The discovery of many new species in the time is strong evidence to support this (Yıldırımlı, 2012; 2018; Köse and Alan, 2013; Bancheva et al., 2014; Bancheva and Kaya, 2015; Bona, 2015, 2016; Negaresh et al., 2015; Yüzbaşıoğlu et al., 2015; Kültür et al., 2016; Pınar, 2016, Uysal et al., 2016, 2017b; Uysal and Hamzaoğlu, 2017; Behçet et al., 2017; Armağan and Uysal, 2018; Negaresh and Rahiminejad, 2018; Şirin et al., 2019, 2020, 2022; Hamzaoğlu and Koç 2020; Duman et al., 2021; Özbek, 2021; Uysal et al., 2022). According to the List of Plants of Türkiye (Uysal, 2012), while 194 taxa in Türkiye represent the genus *Centaurea*, this number has currently reached 221 with the latest additions and new records (Uysal, 2012; Hamzaoğlu and Koç, 2020; Duman et al., 2021; Özbek, 2021; Şirin et al., 2022; Uysal et al., 2022). Of these taxa, 134 are endemic and the endemism rate of the genus for Türkiye is approximately 60%. This study introduced a new species of *Centaurea* to the scientific world as based on morphological, karyological, and molecular data.

2. Materials and methods

Morphology: Interesting *Centaurea* specimens were collected around Kayseri in 2019. Interesting specimens of the genus were examined using the Flora of Turkey and the East Aegean Islands and the neighboring country floras (Wagenitz, 1975, 1980; Dostál J (1976); Tzvelev, 2001). They were compared with the specimens found in KNYA, GOET, E and K herbariums. In the morphologic works, fifteen specimens were used and they were deposited KNYA and GAZI herbarias. Acronyms are according to Thiers (2023).

SEM micromorphology: For achene micromorphology, achenes of new and closely related species (at least 10) were first passed through a series of different concentrations of alcohol (70%, 80%, 96%, and 100%) (Kashi et al., 2014). Then, after the surfaces of the achenes were coated with gold, images of the achene surface at different magnifications were taken under a high vacuum in a ZEISS EVO LS-10 scanning electron microscope (SEM).

Karyology: Mature seeds of the new species and related taxa were selected and germinated for karyomorphological analyses. Root meristems from germinating seeds were

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pretreated with 0.002 M 8-hydroxyquinoline at 4 °C for 8 h. Then, the material was fixed with Carnoy for 24 h in the fridge at 4 °C. Before staining, the material was hydrolyzed with 5 M HCl for 1 h at room temperature, stained with 1% aceto-orcein and mounted in 45% acetic acid. At least 10 metaphases were examined per taxa; the best metaphase plates were photographed (100×) with a digital camera (Olympus DP-72), mounted on an Olympus BX53 microscope. Karyotype asymmetry, ideograms, and karyograms of these taxa were made using the KAMERAM analysis system. While chromosome terminology was made according to the design proposed by Levan et al. (1964), karyotype asymmetries were calculated according to the indices given in the literature (Romero Zarco 1986; Paszko, 2006; Peruzzi and Eroğlu 2013).

DNA isolation, PCR amplification, and data analysis: DNA isolation of the new species and some taxa (Table 1) were performed on leaves from herbarium specimen (Doyle and Doyle, 1987; Soltis et al., 1991; Cullings, 1992). The ITS region was amplified according to the method given by Garcia-Jacas et al. (2006). For ITS region amplification, ITS1-ITS4 primers were used for PCR amplifications (White et al., 1990). ITS sequences of outgroups and new species-related taxa were downloaded from the gene bank (Table 1). All sequences obtained were aligned with the Bioedit 7.2.6.1 program (Hall, 1999). The PAUP 4.0b10 program was used for parsimony analysis and TrNef+I was found to be the most suitable model in AIC (Swofford 2002). Moreover, retention (RI), consistency (CI), and homoplasy (HI) indices were calculated. We

Table 1. Locality and Genebank numbers of *Centaurea* and its related taxa.

Section	Taxa	Locality	Genebank Number ITS
Pseudoseridia	*Centaurea cheirolopha* (Fenzl) Wagenitz		DQ319101
Pseudoseridia	*Centaurea lycopifolia* Boiss. & Kotschy ex Boiss. & Kotschy		DQ319130
Pseudoseridia	*Centaurea hermanni* F.Herm.		DQ319118
	Centaurea odyssei Wagenitz		DQ319140
	Centaurea xylobasis Rech.f.		DQ319176
	Centaurea kirmacii Uysal & Armağan	Türkiye; Muğla: Kavaklıdere, Menteşe village, 3.5 km Southern of Gökçukurplateu, above limestones, 1620 m, 27 vii 2017, Armağan 7797	OQ746369 (In this study)
Cheirolepis	*Centaurea drabifolia* Sm.		DQ319111
Cheirolepis	*Centaurea deflexa* Wagenitz		DQ319105
Microlophus	*Centaurea thracica* (Janka) Janka ex Gugler		DQ319171
	Centaurea kotschyi (Boiss. & Heldr.) Hayek		DQ319127
	Centaurea cheirolepidoides Wagenitz		DQ319100
	Centaurea ensiformis P.H.Davis		DQ319112
	Centaurea paphlagonica (Bornm.) Wagenitz		DQ319142
	Centaurea nallihanense Uysal & Hamzaoğlu		KX158188
	Centaurea isaurica Hub.-Mor.		DQ319124
	Centaurea derderiifolia Wagenitz		Q319106
	Centaurea nivea (Bornm.) Wagenitz		DQ319139
	Centaurea cankiriensis A.Duran & H.Duman		DQ319096
Ptosimopappus	*Centaurea ptosimopappa* Hayek		DQ319152
Ptosimopappus	*Centaurea ptosimopappoides* Wagenitz		DQ319153
	Centaurea turhanii Uysal & Hamzaoğlu	Türkiye, Kayseri, Akkışla, between Ortaköy and Seydinali villages, 1460 m a.s.l., gypseous steppe slopes, 28 vi 2019, Hamzaoğlu 7600 (KNYA!).	OQ746370 (In this study)
Cheirolepis	*Centaurea saligna* (K.Koch) Wagenitz		DQ319159

Table 1. (Continued).

	Centaurea leptophylla (K.Koch) Tchich.		AM087154 AM087155
Microlophus	*Centaurea behen* L.		AY826250
Microlophus	*Centaurea rigida* Banks & Sol.		DQ319158
Microlophus	*Centaurea polypodiifolia* Boiss.		DQ319148
	Centaurea antitauri Hayek		DQ319084
	Centaurea kirikkalensis Özbek	Türkiye; Kırıkkale: Delice, above Baraklı village, *Quercus pubescens*Willd. clearings and steppe, 1100–1150 m, a.s.l., 03 vi 2016, Hamzaoğlu 7239	OQ778863 (In this study)
Rhizocalathium	*Centaurea rhizantha* C.A.Mey.		DQ319157
Rhizocalathium	*Centaurea rhizocalathium* (K.Koch) Tchich.	Türkiye; Artvin: Yusufeli-Çevreliköyüarası, İspir yol ayrımından 1 km, yol kenarı eğimli taşlık kayalık alanlar, 06 vi 2020, T.Uysal 4062 (KNYA!).	OQ778864 (In this study)
Rhizocalathium	*Centaurea armena* Boiss.		DQ319085
Outgroup	*Rhaponticoides pythiae* (Azn. & Bornm.) M.V.Agab. & Greuter		ON694102

used MrBayes 3.2 (Ronquist et al., 2012) to perform the Bayesian phylogenetic analyses. In the Bayesian analyses, random starting trees were used, which were run for 7.1×10^4 generations, comprising 2 independent runs that consisted of four metropolis-coupled chains. Tracer version 1.7 software was used to analyze the trace files created by the Bayesian Markov chain Monte Carlo studies (Rambaut et al., 2018) and, after checking them for convergence, the first 1000 samples (20%) were discarded as burn-in. FigTree v1.4.0 software (Rambaut, 2006) was used as the graphic viewer of the phylogenetic tree.

3. Results

Centaurea turhanii Uysal & Hamzaoğlu **sp. nova** (Figure 1; Table 2)

Type:— Türkiye, Kayseri, Akkışla, between Ortaköy and Seydinali villages, 1460 m a.s.l., gypseous steppe slopes, 28 vi 2019, Hamzaoğlu 7600 (**holo**. GAZI, **iso**. ANK, GAZI, HUB, KNYA).

Diagnosis: Although the new species is not morphologically closely related to any known species in the genus, diagnosis will be made in the focus of these two species, since *C. ptosimopappa* Hayek is located in the closest position in the phylogenetic tree. In addition, the species *C. leptophylla* (K.Koch) Tchich., and C. malatyensis which were thought to show a similarity at first glance in terms of habitat and habitat form as well as appendage structure, were also compared with the new species. Therefore, the new species could be related to *C. leptophylla, C. malatyensis*, and *C. ptosimopappa* but it is different from them by having a shorter stem (not more than 8 cm long), floccose-tomentose leaves (not glabrous or tomentose to villous), ovate-oblong involucre (not narrowly ovoid or obovoid-oblong), alone with ciliated middle phyllaries (not all ciliated), shorter flowers (not more than 12 mm long), more large with oblong-elliptic achenes (not ovate-oblong and not less than 5 mm long) and nondeciduous simple scabrous pappus (not deciduous, biserial and plumose or barbellate).

Description:

Perennial herbs with woody rootstock, sterile shoots usually present. Stem decumbent, 3–8 cm long, densely grey floccose-tomentose, terete, simple or with a few branches in upper part, 0.9–1.5 mm diameter at base. Leaves densely grey floccose-tomentose, elliptic-oblanceolate, attenuate at base, long petiolate, apex acute, with a cartilaginous mucro; lower and median entire, denticulate or with few coarse teeth or lobes, 5–9 ′ 0.5–1.5 cm; upper linear-elliptic to linear-oblanceolate, usually entire. Capitula solitary at end of branches. Involucre oblong-ovate, 14–18 × 8–12 mm. Phyllaries with 5–6 series, sparsely floccose-tomentose; outer and median ones ovate-triangular; inner ones linear-lanceolate. Appendages small, usually a simple spine, mostly very deciduous, spinule simple or lanceolate, rarely ciliate, sparsely floccose-tomentose, patent or reflexed; outer linear, simple, without cilia, only with a terminal spinule; median ones linear-lanceolate, 1–2 cilia (0.5–1.5 mm) on each side, ending in a 2–3.5 mm spinule. Flowers bright yellow in vivo, pale yellow in sicco; central florets hermaphroditic, 23–25 mm long, corolla 17–19 mm long, 5-lobed, lobes 4–6 mm long, with brownish nerves, anther tube straw-colored; peripheral florets sterile, finely

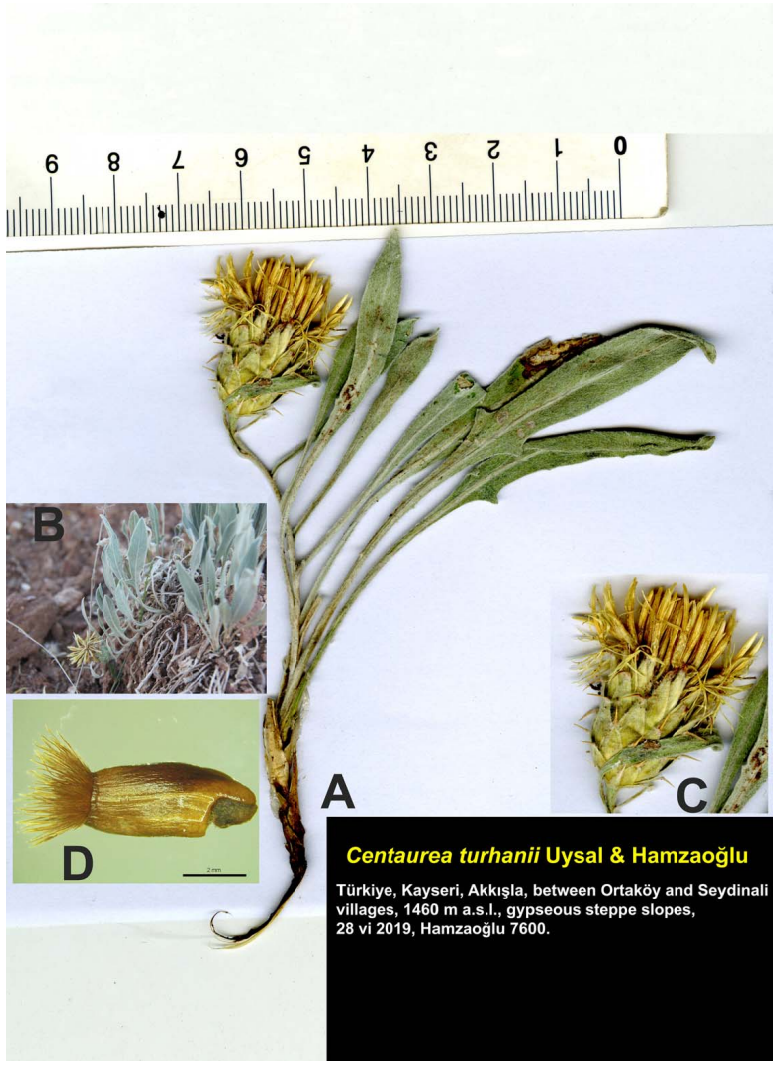

Figure 1. Holotype of *Centaurea turhanii* (A); Habit (B); Capitula (C); Achene (D).

Table 2. Diagnostic characters of *Centaurea turhanii* sp. nov. and related species, *C. leptophylla*, *C.ptosimopappa, C. malatyensis* and *C. cheirolopha*.

→**Taxa** ↓**Character**s	*Centaurea turhanii*	*C. leptophylla* (the measurements base on both our samples (O.Tugay 5172 & T.Uysal) and the type specimen (GOET; Barcoding number 001277)	*C. ptosimopappa*	*C. malatyensis*	*C. cheirolopha*
Stem	**3-8 cm long**, decumbent, densely grey floccose-tomentose, terete, simple or with a few branches in upper part, 0.9–1.5 mm diameter at base	up to **25 cm**, erect or ascending, simple	up to **180 cm**, erect and branched from the base	7–15 cm, decumbent, simple or with few branches in the lower part and with 1(–2) capitula	Up to **25** cm, erect or ascending, simple or with few branches.

Table 2. (Continued).

Leaves	Basal leaves soft, densely **grey floccose-tomentose,** elliptic-oblanceolate, long petiolate; lower and median entire, denticulate or with few coarse teeth or lobes, 5–15 mm broad; upper similar, usually entire.	soft, **glabrous**, linear-lanceolate, shortly petiolate, 2–4 mm broad; upper similar, entire.	firm and leathery, lanceolate-spathulate to obovate, upper similar, entire.	Basal leaves short petiolate (20–25 mm), oblanceolate, 5–6 mm wide, entire, tomentose to villous, lateral nerves indistinct, median and upper leaves sessile, linear to lanceolate; median leaves 4–6 × 0.5–0.7 cm; upper leaves 3–4.5 × 0.4–0.6 cm with entire margin	Leaves bicolored, green, and glabrescent above, grey-tomentose below, of very variable form, basal and lower usually lyrate with large lanceolate or hastate terminal segment and 1–3 pairs of lateral segments or undivided, lanceolate, entire or denticulate; median and upper leaves lanceolate to linear-lanceolate, sometimes narrowly decurrent.
Involucre	**Ovate-oblong**, 14–18 × 8–12 mm; Phyllaries with 5–6 series, sparsely floccose-tomentose; outer and median ovate-triangular; inner linear-lanceolate.	narrowly obovoid-oblong, 12–15 × 9–12 mm; Phyllaries with 5–6 series, glabrous; outer and median ovate-triangular; inner linear-lanceolate.	narrowly ovoid 18–25 × 8–16 mm; Phyllaries very numerous, glabrous.	Ovoid, 37–47 × 12–17 mm; villous in upper parts, greenish-yellow	Ovoid, 15–20 × 9–12(–15) mm,
Appendage	small, usually a simple spine, mostly very deciduous, spinule simple or lanceolate, rarely ciliate, sparsely floccose-tomentose, patent or reflexed; outer linear, simple, without cilia, only with a terminal spinule; median linear-lanceolate, **1–2 cilia (0.5–1.5 mm) on each side,** ending in a 2–3.5 mm spinule	small, with a simple spine, mostly very deciduous, spinule simple or lanceolate, without cilia, glabrous, patent or reflexed; outer linear, simple, without cilia, only with a terminal spinule; median linear-lanceolate, **without cilia**, ending in a 2.5–3.5 mm spinule	a minute, deciduous, spinule simple and too short, **without cilia**, glabrous, patent, or reflexed; all similar, ending in a 0.3–0.5 mm spinule.	triangular, not concealing basal part of phyllaries, not decurrent, entire in the outer and the median phyllaries with a spinule 2–3 mm long, irregularly lacerate in the inner phyllaries ending in a spinule 0.5–1.5 mm long.	Appendages small, brown, broadly triangular, spreading or reflexed, with 5–7 digitate 1–5 mm spinules.
Flowers	yellow, not radiant, **9–12 mm** long, with 5 linear-filiform 4–6 mm lobes; anther tubes yellow	yellow, not radiant, **18-20 mm** long, with 5 linear-filiform 5–6 mm lobes; anther tubes yellow.	yellow, not radiant, **20-25 mm long**, with 5 linear-filiform 5–6 mm lobes; anther tubes yellow.	yellow, marginal not radiant, ca. 35 mm long, corolla tube glabrous, 18–22 mm long, lobes 6–8 mm, linear with brown stripes along corolla	Flowers yellow

Table 2. (Continued).

Achene	**5–6(–7) × 1.8–2.3 mm, oblong-elliptic**, brownish, glabrous, notched and rounded at base.	4.5–5 × 2.2–2.5 mm, narrowly ovoid-**oblong**, creamish-brown in mature, superficially with sparsely pilose hairy.	4–5 × 2 mm, narrowly ovoid to oblong, brown, with sparsely pilose hairy.	6–6.5 × 3.5–4.5 mm, obovoid with striations, light brown, covered with adpressed whitish hairs;	4-5.5 mm;
Pappus	**simple**, scabrous, brownish to purplish, 2-3(–4) mm long, **not deciduous**	**double**, scabrous, outer series whitish-cream, 8–10 mm, barbellate, inner series 0.5 mm, similar to ascale, **deciduous**.	**Simple**, scabrous, creamish-brown, c. 4 mm, deciduous.	**simple**, 18–22 mm, plumose, brownish	0.5-1.5 mm, inner row not distinctly different.

dissected, not radiant, nearly equal to central ones, 15–17 mm long, 4–5-lobed, lobes 4–6 mm long. Achenes 5–6 × 1.8–2.3 mm, oblong-elliptic, brownish, glabrous, notched and rounded at base. Pappus scabrous, brownish to purplish, simple, 2-3(–4) mm long, persistent.

Phenology:—The new species blooms in June and starts fruiting in mid-July.

Group B (Volume V, Flora of Turkey (Wagenitz, 1975)

1. Flowers pink or purple
1. Flowers yellow
3. All leaves linear-lanceolate to linear or elliptic-oblanceolate, entire
4. Leaves linear lanceolate, glabrous 172. ***leptophylla***
4. Leaves elliptic-oblanceolate, densely tomentose or grey floccose-tomentose
5. Pappus plumose, 18–22 mm 172.a. ***malatyensis***
5. Pappus scabrous, 2–4 mm 172.b. ***turhanii***
3. All leaves pinnatifid to lyrate

Etymology: The species is named in honor of our precious friend and scientist Prof. Dr. Turhan ÇETİN (Ankara, Türkiye), who has participated in many field studies with us and made geographical observations and created maps. He is very fruitful a scientist who has studies on Türkiye's natural environment and botanical tourism.

Distribution, habitat, and ecology: *Centaurea turhanii* grows on gypseous steppes between Ortaköy and Seydinali villages in the Akkışla district of Kayseri Province at altitudes of 1370–1590 m a.s.l. (Figure 2), and it is associated with *Gypsophila turcica* Hamzaoğlu, *Thesium stelleroides* Jaub. & Spach, *Thymus cappadocicus* Boiss., *Isatis glauca* Willd. ex DC. subsp. *sivasica* (P.H.Davis) Yıld., *Matthiola anchoniifolia* Hub.-Mor., *Ferula szowitsiana* DC., and *Paracaryum racemosum* Britten. The area where the species grows is not threatened other than intensive grazing, and it is part of the Irano-Turanian floristic region.

Due to the lack of detailed observations and studies on the distribution area of the species and the number of individuals for several years, no suggestions were made regarding the threat category and protection status.

Achene structure: Achenes of *Centaurea turhanii* are glabrous, oblong-ellipical, 5.9–6.3 × 2.7–2.9 mm, with ornamentation irregular sulcate. Cells are distinct and cell boundaries are thin (Figure 3). Achenes of *C. ptosimopappa* are sparsely pilose, ovoid to oblong, 5.24 × 2.05 mm, with ornamentation irregular sulcate. Ripe achene lacks the pappus (deciduous). Cells and cell walls are distinct (Figure 3).

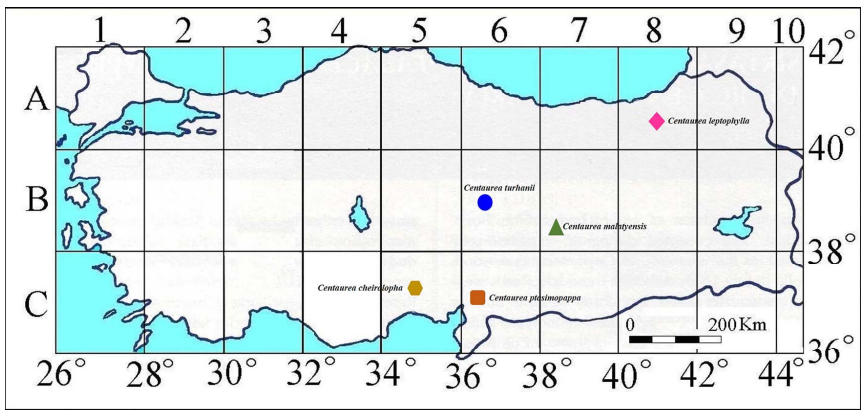

Figure 2. Distribution map of *C. turhanii* and related taxa.

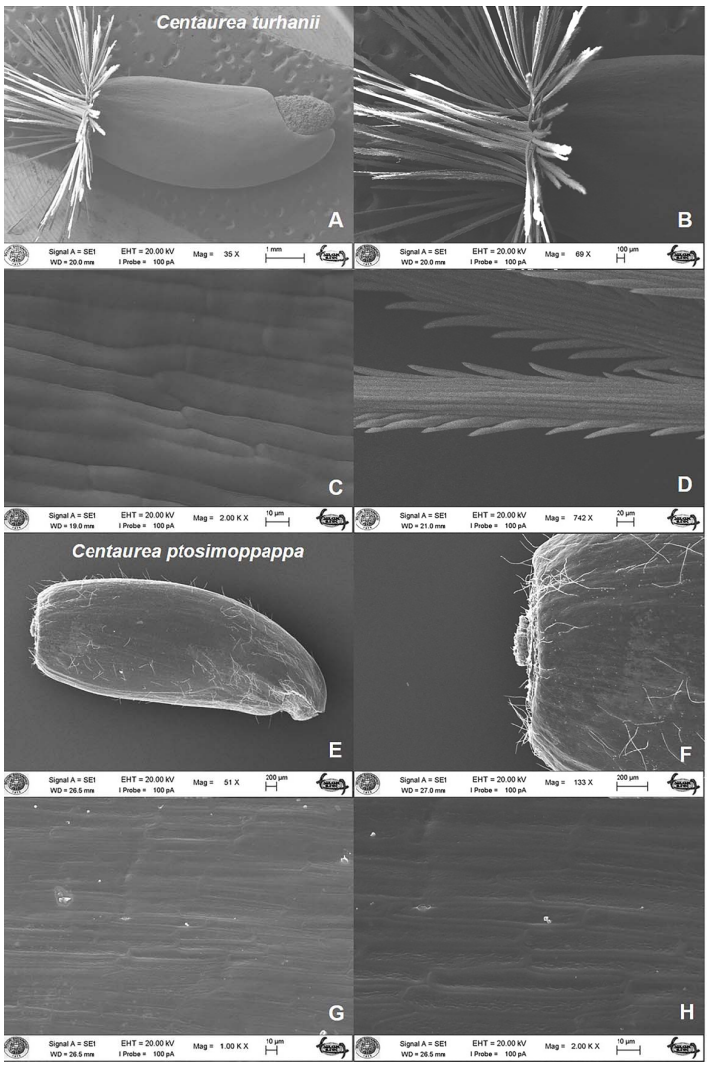

Figure 3. Scanning electron micrographs of achene in *Centaurea turhanii* (A–D) and *C. ptosimoppappa* (E–H).

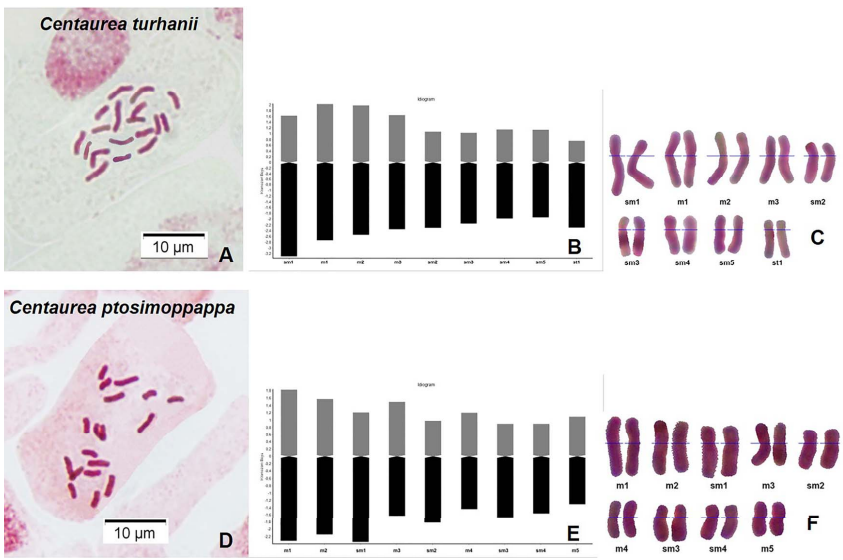

Figure 4. Mitotic metaphase chromosomes, idiograms and karyograms of *Centaurea turhanii* (A–C) and *C. ptosimopappa* (D–F).

Table 3. The chromosome features of *Centaurea* taxa.

Taxa	2n	R SC-LC (µm)	R- LC / SC	p (µm) mean (±SD)	q (µm) mean (±SD)	CL (µm) mean (±SD)	TCL (µm)	CI mean (±SD)	KF
C. turhanii	18	3.05– 4.92	1.613	1.38 (±0.43)	2.40 (±0.39)	3.78 (±0.73)	33.995	36 (±0.06)	6m + 10sm + 2st
C. ptosimopappa	18	2.40– 4.14	1.725	1.24 (±0.30)	1.80 (±0.36)	3.05 (±0.59)	27.415	41 (±0.05)	10m + 8sm
***Centaurea ptosimopappa*(a)** (Koçyiğit& Bona 2013)	18								3m+2msm+4sm
***Centaurea ptosimopappa*(b)** (Koçyiğit& Bona 2013)	18								4m+5msm
C. *leptophylla* (Uysal et al. 2015)	36	1.36– 2.43	1.78	0.67 (±0.15)	1.07 (±0.22)	1.75 (±0.33)	31.5	38 (±0.05)	2m+7sm

R: Range, SC: Shortest chromosome length, LC: Longest chromosome length, p: Mean length of the long short arm, q: Mean length of the long arm, CL: Mean chromosome length, TCL: Total haploid complement length, CI: Mean centromeric index, SD: Standard deviation, KF: Karyotype formula, m: Metacentric, sm: Submetacentric, st: Subtelocentric.

Table 4. The karyotype indices of *Centaurea* taxa

Taxa	A_1	A_2	CV_{CL}	CV_{CI}	AI	M_{CA}
C. turhanii	0.429	0.194	19.418	15.968	3.101	26.98
C. ptosimopappa	0.302	0.193	19.269	12.58	2.424	18.42
C. *leptophylla* (Uysal et al 2015)	0.36	0.19	18.95	12.99	2.46	22.99

A_1: Intrachromosomal asymmetry index, A_2: Interchromosomal asymmetry index, CV_{CL}: Coefficient of variation of chromosome length, CV_{CI}: Coefficient of variation of centromeric index, AI: Karyotype asymmetry index, M_{CA}: Mean centromeric asymmetry.

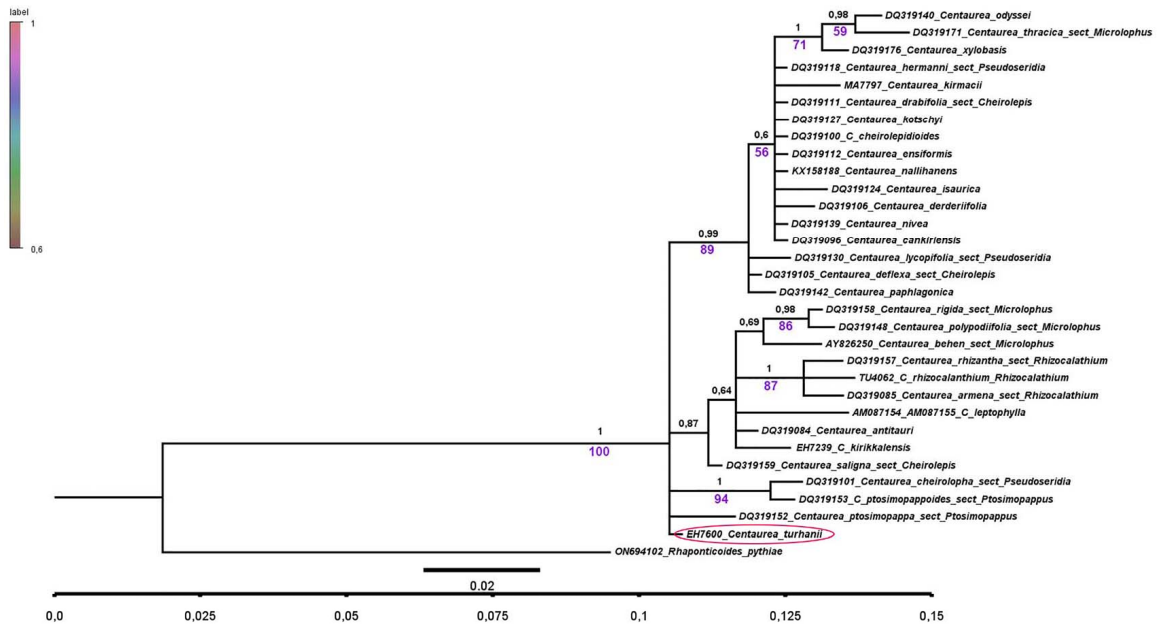

Figure 5. Majority rule consensus tree obtained from Parsimony and Bayesian analysis of the ITS data for *C. turhanii* and related taxa (tree length: 608, CI: 0.732, RI: 0.762, HI: 0.268). Values in the branches are shown as maximum Bayesian posterior probabilities/Parsimony bootstrap.

Karyology: According to our counts, *Centaurea turhanii* is diploid with chromosome number $2n = 18$. The karyotype formula of the species is $2n = 6m + 10sm + 2st$ (Figure 4). According to the classification based on the length of the chromosomes (Lima De Faria 1980), the new species has four pairs of medium and five pairs of small chromosomes and chromosomes get shorter gradually from medium to small (Table 3). When we compare our results with the information from the literature, although there is agreement in terms of the chromosome number of *C. ptosimopappa*, there are significant differences in terms of karyomorphology. In the study conducted by Koçyiğit and Bona (2013), different karyotype formulas of two different populations of *C. ptosimopappa* were specified. Interestingly, there is no match between the related study and the karyotype formulas determined in our study. (Figure 4 and Table 3). Chromosomal features of the new species and related taxa are given in Tables 3 and 4.

Phylogeny: Phylogenetic analyses included 32 taxa, one of which was an outgroup. The aligned data matrix consists of 608 base pairs long and 533 of them are constant. While the informative character number for parsimony was 28 and the uninformative was 47. The tree created by the PAUP program has reliable tree scores [consistency index (CI: 0.732), retention index (RI: 0.762), and homoplasy (HI: 0.268)]. Parsimony analysis was executed with the TrNef+I model according to chosen AIC criteria. Trees revealed by parsimony and Bayesian analysis formed similar topologies for the studied *Centaurea* taxa (Figure 5). Therefore, a common tree was created by combining the analysis. In the combined tree based on ITS regions, the new species is in a different clade from all studied *Centaurea* taxa in terms of its phylogenetic history. Indeed, we concluded that the new species is located phylogenetically differently from the taxa belonging to the sections analyzed.

4. Discussion

Centaurea turhanii has been characterized by achene surface micromorphology, karyomorphology, nrDNA ITS, and morphological studies. *Centaurea turhanii* which was published as a new species to science is a unique species both morphologically and phylogenetically, it does not have any close relatives in the genus in terms of the characteristics given. Previously, the involucrum bract and appendages have played a vital role morphologically in the taxonomy of the genus (Wagenitz, 1975), and the subgroups were classified according to these characters. However, in the ongoing studies, it has been pointed out that the classification based on phyllaries and appendages is unreliable in *Centaurea*. In particular, the analyses based on molecular studies suggested that there is a mismatch between morphology and molecular data and that the reason for this is rapid evolution due to hybridization (Garcia-Jacas et al., 2006; Hilpold et al., 2014). Similarly, it is declared that sectional classification had difficulties in establishing natural and clear division based on bract appendices as depended on the entanglement of sections, as previously suggested by Ertuğrul et al. (2004) and Hilpold et al. (2014). The virtual absence of congruence between morphology and molecular data suggests that the morphological characters that have been used taxonomically are not reliable with respect to the true relationships. Some morphological traits may have developed convergently in several cases– a high plasticity of morphological characters is also likely. The development of equal or similar morphological character states may have partly been due to an adaptive response to similar ecological conditions, for example, the development of small divided tomentose leaves as a response to dry conditions (cf. Larcher, 2003), or of spiny bract appendages as a response to herbivores (cf. Cooper and Owen-Smith, 1986; Hilpold et al., 2014). Because of these problems, we will focus on the groups and species to which the new species is relative according to the information from the molecular and karyological data, rather than morphology.

In terms of involucrum bracts, the new species exhibits an intermediate state on the point of main types of appendages. While the outer phyllaries are in the form of a simple spinule, the appendages of some middle phyllaries irregularly contain one or two pairs of cilia at the base or mostly simple spinule. The inner phyllaries are in the form of a narrowly triangular simple membranous spine. Based on this morphology, especially in group B, in which the appendage has been reduced to a terminal spiny or spinule (Wagenitz, 1975). One of the sections defined in Group B is *Microlophus* and the section is represented by six species in the Flora of Turkey. Another section thought to be related according to phylogenetical analyses is *Ptosimopappus* and this section includes only two endemic species. The known species of these sections in the mentioned groups consist of taller plants with long branches and erect stems. Even if it is not completely morphologically, the new species can be more associated with the taxa of the *Ptosimopappus* too, considering the mainly achene and pappus structures. However, a very short and creeping stem is not a usual for the species of neither A nor B groups. Besides with this general comparison, the new species display morphologically an interesting similarity with *C. leptophylla* which are a local endemic species spreading in Çoruh Valley of Artvin Province. Although the two species are similar in terms of growing on rocky slopes, creeping habitat form and especially appendages, they differ in terms of leaf structure, indumentum and pappus characteristics. In addition, the new species is similar to *Centaurea malatyensis*. However, it is easily distinguished

from the *C. malatyensis* by its smaller capitula and very short and scabrous pappus. Moreover, the two species (new species and *C. malatyensis*) are different in their ecology; the new species is distributed on gypseous steppes while the other species (*C. malatyensis*) prefers serpentine cliffs. The new species clearly looks different when compared to *C. cheirolopha*. Not only *C. turhanii* is different in terms of leaf hairy and color (not bicolor) but also it has short creeping stem (not longer than 10 cm and not erect-ascending stem). Additionally, while *C. turhanii* is rarely including irregularly one or two pair cilia at the base of medium phyllaries, *C. cheirolopha* has 5–7 pair digitate cilias in all phyllaries (not all phyllaries have several pairs of cilia). Lastly, it includes longer and scabrous pappus hairs (not 0.5–1.5 mm and not plumouse) (Table 2).

So far, it has been reported that achene surface morphology is effective in the differentiation of taxa belonging to the genus *Centaurea* (Uysal et al., 2016; Güzel, 2022). The cell boundaries of *C. malatyensis* are relatively expanded. The cell boundaries of *C. leptophylla, C. ptosimopappa*, and *C. cheirolopha* species are thinner than *C. malatyensis*. However, even the boundaries of these species are thicker compared to the new species. It is seen that the new species differs from *C. ptosimopappa, C.malatyensis, C. leptophylla*, and *C. cheirolopha* in terms of achene shape, surface ornamentation and hairy status (Kültür et al., 2016; Aydın et al., 2019; Güzel 2022).

The chromosome number determined had been reported before for many *Centaurea* species in several studies (Georgiadis and Phitos, 1976; Phitos and Damboldt, 1976; Romaschenko et al., 2004; Uysal, 2008, 2009; Uysal et al., 2015, 2017). Additionally, the basic chromosome numbers of some taxa belonging to *Microlophus* were reported as $x = 8$ and $x = 17$ (Garcia-Jacas et al., 1998; Romashchenko et al., 2004; Uysal et al., 2009; Tasar et al., 2018). These results disagree with number of the new species. The basic chromosome number of the *C. thracica* (Janka) Janka ex Guglerwas reported as $x = 9$ (Damboldt and Matthäs, 1975; Constantinidis et al., 2002). However, the taxonomic position of this species is controversial according to phylogeny studies (Garcia-Jacas et al., 2006). Based on the information we obtained from the literature, with exceptions, we reconfirmed the $x = 9$ basic chromosome number commonly seen in the genus *Centaurea*. According to our karyological comparisons among *C. turhanii*, *C. ptosimopappa*, and *C. leptophylla*, the new species has distinctive karyomorphological features (Karyotype formula and asymmetry indices; Tables 3 and 4) and is clearly different karyologically from its relatives. Unlike of *C. leptophylla*, the new species is a diploid like *C. ptosimopappa*. The karyotype formula is unique and consist of mostly submetacentric chromosomes as well as subtelocentric. Subtelocentric chromosomes are not seen in related taxa.

Romaschenko et al. (2004) reported the chromosome number of *C. cheirolopha* as $2n = 2x = 18$ (diploid). *C. cheirolopha* is the mostly similar species in terms of karyomorphology with the new species. However, the sixth and ninth chromosome pairs of *C. cheirolopha* are metacentric. In particular, *C. cheirolopha* differs from the new species because it has metacentric chromosome pairs.

As related to chromosomal counts and analyses of *C. ptosimopappa*, there is a minor conflict among present and previous reports (Tables 3 and 4) Even so, the karyotype formulas of this taxon are not hold related to the new species.

According to the ITS based phylogenetic tree; the new species is taken place relatively far from the better resolved clade containing *Microlophus* species (PP: 0.69) and in the immediate near of the clade containing *Ptosimopappus* species (PP: 1/BS: 94), which can be considered as a polytomic clade. For this reason, it can be said that the species does not display high closeness to any members of mentioned groups but partly more closely related to species of the sections *Microlophus* and *Ptosimopappus*. However, based on the current data, it is thought that it would not be correct to evaluate the species in one of these two sections, and it may be considered to determine a common new section for *C. leptophylla*, *C. malatyensis* and *C. turhanii* species in a future study.

Additional specimens examined

Centaurea ptosimopappa Hayek

C6 Osmaniye, Zorkumroad, 1050 m, roadside, 05 July 2003, A. Duran 6306 & M. Sağıroğlu (KNYA!); C6 Osmaniye, between Zorkum, 300–400 m, roadside, 12 September 2003, A. Duran 6344 & M. Sağıroğlu (KNYA!).

Centaurea leptophylla (K.Koch) Tchich.

Artvin; Yusufeli, Sarıgöl girişi 790 m, 08 viii 2007, O. Tugay 5172 & T.Uysal (KNYA!).

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