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Notes on *Campanula argaea* group (Campanulaceae) in Turkey and related species

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Campanula pamphylica (Contandr., Quézel & Pamukç.) E. Akçiçek & Vural *comb. & stat. nov.*, previously regarded as a synonym, is shown to be a distinct species. *Campanula pamphylica* subsp. *afyonica* E. Akçiçek & Vural *subsp. nova* is described as a new subspecies from Anatolia, Turkey. *Campanula pamphylica* subsp. *tokuri* (A. Ocak) E. Akçiçek & Vural *comb. & stat. nov.* is proposed as a new combination. Diagnostic, morphological and molecular data are discussed for the taxa.

Key words: *Campanula*, nomenclature, RAPD, taxonomy

Introduction

The genus *Campanula* occurs mainly in the northern hemisphere and Mediterranean region. It is represented by ca. 300 species (Rosatti 1986). It is also a large genus in Turkey, where it has 114 species. Since *Campanula* was revised by Damboldt (1978) for the *Flora of Turkey*, 15 new species and one subspecies have been described from Turkey. One species and one subspecies were also added as new records for Turkey (Davis *et al.* 1988, Güner *et al.* 2000).

This paper recognizes *C. pamphylica* as a distinct species.

Campanula specimens were collected during 2000–2003 while we were working on the flora of Kumalar Dağı, in Afyon province. One of them superficially resembles *C. argaea*, but does not have decurrent stem leaves. *Campanula argaea* subsp. *pamphylica* was not distinguished by Damboldt (1978). Specimens in Hacettepe

University (HUB), the specimens cited in the original publication, and further specimens collected from the type locality were examined, and it was observed that subsp. *pamphylica* does not have decurrent stem leaves, while subsp. *argaea* does. It is indicated in the original publication that *C. argaea* subsp. *pamphylica* was observed only when in bud; as such, its classification has yet to be established and so it was temporarily included in *C. argaea*. The species *C. tokuri*, which is also known only from its type locality (B3 Eskişehir: Türkmen Dağları), resembles the specimens collected from Afyon and Burdur in that it is lacking decurrent stem leaves (Ocak 2003). Therefore, we studied and compared morphological and molecular aspects of *C. argaea* subsp. *argaea*, *C. argaea* subsp. *pamphylica*, *C. tokuri*, and other specimens collected from Afyon province. We conclude that *C. argaea* subsp. *pamphylica*, which was previously treated as a synonym of *C. argaea*, is best treated as a

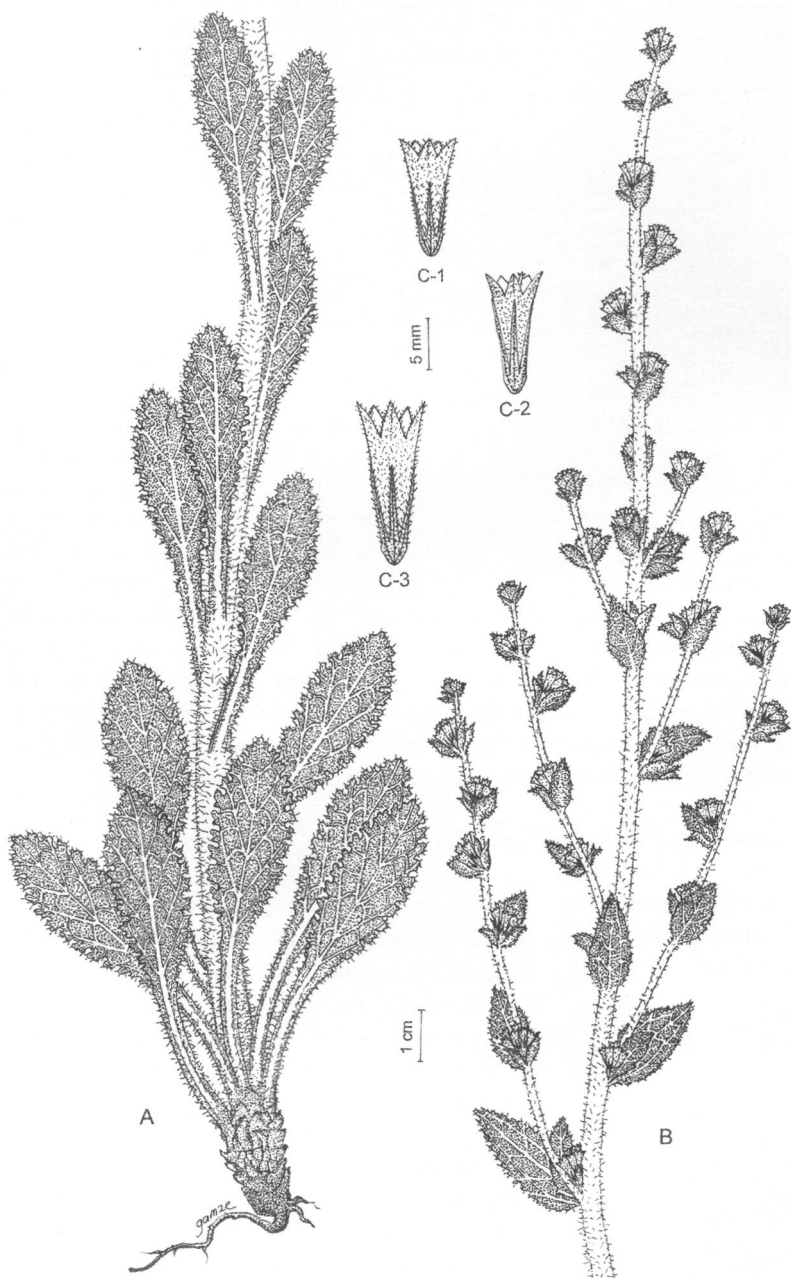


Fig. 1. — **A** and **B**: Habit of *Campanula pamphylica* subsps. *afyonica* (from holotype, E. Akçiçek 3758). — **C-1–C-3**: Flowers. **C-1**: *C. pamphylica* subsp. *afyonica* (from holotype, E. Akçiçek 3758). **C-2**: *C. pamphylica* subsp. *tokuri* (from isotype, Ocak 9051). **C-3**: *C. pamphylica* subsp. *pamphylica* (from A. Pamukçuoğlu s.n., HUB).

distinct species and that the specimens collected from Eskişehir and Afyon provinces are best recognized as subspecies of *C. pamphylica*.

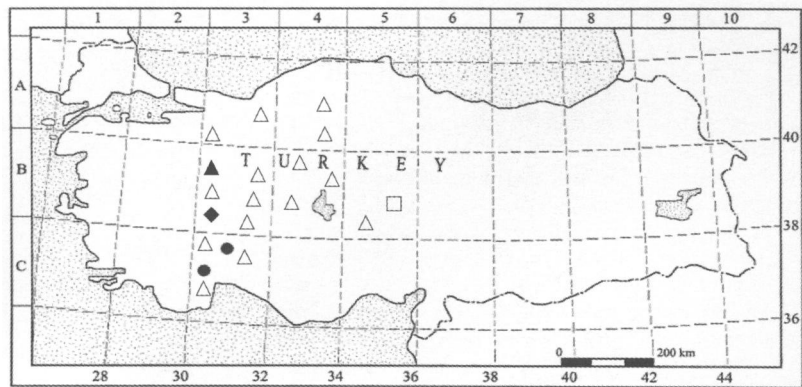
Material and methods

The plants were collected from Kumalar Dağı (Afyon) and near Burdur. The specimens were

compared with similar materials at GAZI, ANK, and HUB, and with records in the literature (Damboldt 1978, Davis *et al.* 1988, Güner *et al.* 2000, Ocak 2003). The authors of plant names follow Brummitt and Powell (1992).

A set of 40 oligonucleotides from Operon Technologies were used for RAPD amplification. Of 40 primers, 10 primers were chosen for further amplification of the DNA. For the

Fig. 2. Geographic distribution of (●) *Campanula pamphylica* ssp. *pamphylica*, (▲) *C. pamphylica* ssp. *tokuri*, (◆) *C. pamphylica* ssp. *afyonica*, (△) *C. argaea*, and (□) its type locality.



morphological analysis, 17 different numbers of characters were used in order to calculate genetic distances between *Campanula* species. The conditions reported by Williams *et al.* (1990) and Welsh (1992) were used for RAPD-PCR. Amplification was achieved in a Techne (UK) Progene thermocycler. After the cycling was completed, 15 μ l of the reaction products were analyzed alongside small molecular weight markers on 2% agarose gels in the presence of ethidium bromide, and gels were photographed under UV light (Maniatis 1982).

Bands on RAPD gels were scored as present (1) or absent (0), for all taxa studied. Common band analysis was conducted using the computer programme POPGEN to determine values of genetic distance among them (Nei 1978). The figures for genetic distance were then used as input data for cluster analysis to generate dendrograms.

Results

Campanula pamphylica (Contandr., Quézel & Pamukç.) E. Akçiçek & Vural, *comb. & stat. nov.* (Figs. 1 and 2)

Campanula argaea Boiss. & Bal. subsp. *pamphylica* Contandr., Quézel & Pamukç., Ann. Univ. Provence Sci. 46: 54. 1972. — TYPE: Turkey. C3 Burdur: 20 km S of Burdur towards Antalya, 700 m, 1970 Quézel *et al.* s.n. (MARSSJ, not seen).

Tomentose-hispid or setulose-hirsute, biennial herbs. Stem thick, ascending-erect, usually many-stemmed, sometimes single, 20–50(–70) cm, unwinged, longitudinally ridged, densely

leafy in lower part. All leaves acute to obtuse with strongly undulate to plane and entire to deeply dentate margins, more densely hairy beneath. Basal leaves oblanceolate, obovate, or oblong-spathulate, 2–4.5(–8) \times 0.7–1.5 cm, attenuate into a short broad petiole or petiole up to 5.5 cm. Cauline leaves gradually decreasing or \pm same in size. Median cauline leaves oblong-spathulate, obovate-spathulate, oblanceolate or ovate-lanceolate; sessile, not decurrent on stem. Upper cauline leaves bract-like, ovate, acute. Flowers many, sessile or subsessile, axillary, solitary or in clusters of 2–7 in a long interrupted spike. Bracts ovate, ovate-lanceolate, 5–10 \times 2.5–4 mm. Bracteoles navicular or lanceolate, 4–8 \times 1–3 mm. Calyx lobes spreading to erect or incurved, triangular or lanceolate, acute, 4–8 mm, shorter than or nearly as long as corolla tube; appendages inconspicuous. Corolla narrowly campanulate or narrowly infundibular, 8–17 mm, tomentose or setulose outside, lavender blue or light blue, divided for 1/5–1/3 its length into oblong-lanceolate or lanceolate, acute or mucronate, erect lobes. Stamens 5; filaments free, ca. 2 mm, ciliate, flattened and dilated at base; anthers free, linear-oblong or oblong-elliptic, ca. 4 mm, light yellow. Ovary 3-locular; style included in corolla, elongate, hairy, 7–10 mm; stigmas 3, recurved, 1–2 mm. Capsule subglobose or ovoid, ribbed, opening by 3 basal pores. Seeds numerous, ellipsoid, oblong-oblancheolate, flattened, 0.6–1 \times 0.15–0.4 mm, brown, shiny or dark brown with narrow black margin.

In the key of the *Flora of Turkey* (Güner *et al.* 2000), *Campanula pamphylica* can be inserted as follows:

17. Corolla broadly campanulate. 25–35(–60) mm long *C. incurva*
17. Corolla cylindrical-campanulate to narrowly campanulate or narrowly infundibular, (8–)10–20 mm 18
18. Plant scabrid to hirsute; calyx appendages as long as ovary *C. sibirica*
18. Plant tomentose-hispid or setulose-hirsute; calyx appendages much shorter than ovary 19
19. Median cauline leaves decurrent on stem; stem winged by decurrent leaf bases; basal leaves 3–7 × 1.5–3 cm; corolla tomentose-hispid outside *C. argaea*
19. Median cauline leaves not decurrent on stem; stem unwinged; basal leaves 2–4.5(–8) × 0.7–1.5 cm; corolla tomentose or setulose outside *C. pamphylica*

Key to the subspecies of *Campanula pamphylica*

1. Basal leaves with flat entire margins; calyx lobes 7–8 mm long; style 10 mm long *C. pamphylica* subsp. *pamphylica*
1. Basal leaves with strongly undulate and deeply dentate margins; calyx lobes 4–7 mm long; style 7–7.5 mm long 2.
2. Plant tomentose-hispid; bracts 7–8 × 4 mm; bracteoles navicular, 6–7 × 2–3 mm *C. pamphylica* subsp. *afyonica*
2. Plant setulose-hirsute; bracts 5 × 2.5 mm; bracteoles lanceolate, ca. 4 × 1 mm ... *C. pamphylica* subsp. *tokuri*

Campanula pamphylica subsp. *pamphylica*

Tomentose-hispid herbs. Stem 20–35 cm. Cauline leaves ± same in size. Basal leaves oblanceolate, 3–4.5 × 1–1.5 cm, with flat entire margins. Median cauline leaves oblong-spathulate, ovate-lanceolate; with flat entire margins. Flowers 3–7 in clusters. Bracts ovate-lanceolate, ca. 10 × 4 mm. Bracteoles lanceolate, 7–8 × 2 mm. Calyx lobes 7–8 mm. Corolla up to 17 mm. Anthers linear-oblong. Style 10 mm. Stigmas 1.5–2 mm. Endemic, Irano-Turanian element.

SPECIMENS EXAMINED: **Turkey.** C3 Isparta: Eğridir, Kovada Gölü çevresi, kalker kayalığı, 27.VI.1971 A. Pamukçuoğlu s.n. (HUB!); C3 Isparta, Kovada Gölü çevresi, kalker kayalığı, 27.VI.1971 Ö. Inceoğlu s.n. (HUB!).

Campanula pamphylica subsp. *tokuri* (A. Ocak) E. Akçiçek & Vural, *comb. & stat. nov.*

Campanula tokuri A. Ocak, Israel J. Pl. Sci. 51: 321. 2003.

TYPE: Turkey. B3 Eskişehir: Türkmen Dağları, Sandıközü Köyü çevresi, 1450 m, rocky slopes, 18.VI.2001 Ocak 9051 (holotype E; isotypes E, K, GAZI!).

Setulose-hirsute herbs. Stem (20–)30–40 cm. Cauline leaves gradually decreasing in size. Basal leaves spathulate, obovate, 2–4 × 1–1.5 cm, with strongly undulate and deeply dentate margins. Median cauline leaves obovate-spathulate, with strongly undulate and deeply dentate margins. Flowers solitary or 2–5 in clusters. Bracts ovate, ca. 5 × 2.5 mm. Bracteoles lanceolate, ca. 4 × 1 mm. Calyx lobes 4–7 mm. Corolla 9–10(–12) mm. Anthers oblong-elliptic. Style 7–7.5 mm. Stigmas 1 mm.

Campanula pamphylica subsp. *afyonica* E. Akçiçek & Vural, *subsp. nova*

Affinis C. pamphylicae subsp. *tokuri* et *C. pamphylicae* subsp. *pamphylicae*, sed ab *C. pamphylicae* subsp. *tokuri* planta tomentosohispido, 30–70 cm alto (non setuloso-hirsuto, 20–40 cm), bracteis 7–8 × 4 mm (non ca. 5 × 2.5 mm), bracteolis navicularibus, 6–7 × 2–3 mm (non lanceolatis, ca. 4 × 1 mm) et stigmatibus 1.5–2 mm (non 1 mm) longis differt. Ab *C. pamphylicae* subsp. *pamphylicae* caule 30–70 cm (non 20–35 cm) alto, foliis basalibus valde crispis undulatis et profunde dentatis marginatis (non haud undulatis et dentatis marginatis), bracteis ovatis, 7–8 × 4 mm (non ovatis-lanceolatis, ca. 10 × 4 mm), lobis calycis 5–6 mm (non 7–8 mm) longis, corolla 8–12 mm (non usque ad 17 mm) longo, stylo 7–7.5 mm (non 10 mm) longo differt.

TYPE: Turkey. B3 Afyon: Şuhut, Akyatak mahallesi, 1300–1350 m, crevices of volcanic rocks, 16.VII.2002 E. Akçiçek 3758 (holotype GAZI; isotypes ANK, ISTF).

Tomentose-hispid herbs. Stem 30–50(–70) cm. Cauline leaves gradually decreasing in size. Basal leaves oblanceolate or oblong-spathulate, 2.5–4.5(–8) × 0.7–1.3 cm, with strongly crisped undulate and deeply dentate margins. Median cauline leaves oblanceolate or oblong-spathulate, with strongly undulate and deeply dentate margins. Flowers solitary or 2–5(–7) in clusters. Bracts ovate, 7–8 × 4 mm. Bracteoles navicular, 6–7 × 2–3 mm. Calyx lobes 5–6 mm. Corolla 8–12 mm. Anthers linear-oblong. Style 7–7.5 mm. Stigmas 1.5–2 mm.

Flowering in June and July.

ADDITIONAL SPECIMENS EXAMINED (paratypes): Turkey. B3 Afyon: Şuhut, Akyatak mahallesi, 1300–1350 m, crevices of volcanic rocks, 23.VI.2001 *E. Akçiçek 3694* & *M. Sağıroğlu* (GAZI); same locality, 26.VII.2003 *E. Akçiçek 3762* (GAZI); B3 Afyon: 10 km from Sandıklı to Şuhut, Başkaya tepesi, 1300 m, rock crevices, 25.VII.2000 *E. Akçiçek 3107* (GAZI); B3 Afyon: Şuhut, Aydın köyü, 1100 m, on rock, 26.VII.2003 *E. Akçiçek 3761* (GAZI).

DISTRIBUTION: Endemic to Turkey (Afyon province). Irano-Turanian element.

HABITAT ECOLOGY: *Campanula pamphylica* subsp. *afyonica* grows in crevices of volcanic rocks of steppe at 1100–1350 m with *Noaea mucronata* subsp. *mucronata*, *Dianthus balansae*, *Poa bulbosa*, *Scabiosa argentea*, *Eryngium campestre*, *Echinophora tournefortii*, *Bromus scoparius*, *Phleum boissieri*, *Centaurea virgata*, *Aegilops cylindrica* and *Cruciata* sp.

For RAPD analysis, of 40 primers, 10 were chosen for further amplification of the DNA. RAPD bands ranged from 200 bp to 1500 bp in size. Some of the bands were monomorphic, while some of them showed at least one polymorphism.

Genetic similarity and distance between species of *Campanula* species calculated based on morphological and RAPD data is summarised in Table 1. Dendrograms were constructed using POPGENE computer program. Topology obtained from RAPD electrophoretic analysis of four species of *Campanula* is presented in Fig. 3. The dendrogram allows two groups to be distinguished. The upper group contains only *C. argaea*, which has a genetic distance between 56% and 61% from the others. The lower cluster contains *C. pamphylica* subsp. *afyonica*, *C. pamphylica* subsp. *tokuri*, and *C. pamphylica* subsp. *pamphylica*. Of these cluster members, *C. pamphylica* subsp. *afyonica* differs from *C. argaea*, *C. pamphylica* subsp. *tokuri* and *C. pamphylica* subsp. *pamphylica* with the genetic distance of 44%, 26%, and 37%, respectively.

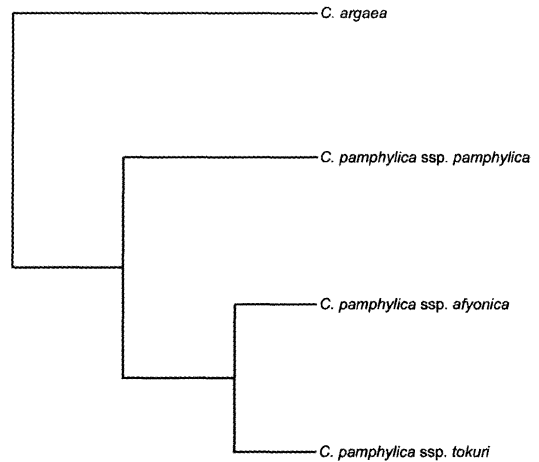


Fig 3. Dendrogram of the *Campanula* taxa based on morphological and RAPD data.

The taxon described as *Campanula argaea* subsp. *pamphylica* is not close to *C. argaea*. Specimens were examined in the herbaria of GAZI, ANK, HUB and compared with specimens of *C. argaea*. The differences are indicated in Table 2.

Campanula pamphylica shows additional variation that is best recognized at subspecific level.

These subspecies resemble each other in their unwinged stems, inconspicuous calyx appendages, and that the style is included in the corolla. However, they differ in their leaf and floral features. Specifically, *C. pamphylica* subsp. *afyonica* differs from *C. pamphylica* subsp. *tokuri* in its tomentose-hispid indumentum, taller stem 30–50(–70) cm, larger bracts (7–8 × 4 mm) and bracteoles (6–7 × 2–3) mm. On the other hand, *C. pamphylica* subsp. *tokuri* has setulose-hirsute indumentum, shorter stems (20–)30–40 cm, shorter bracts (ca. 5 × 2.5 mm) and shorter bracteoles (ca. 4 × 1 mm). *Campanula pamphylica* subsp. *tokuri* differs from *C. pamphylica*

Table 1. Nei's genetic distance (below diagonal) values based on morphological and RAPD data.

	<i>C. argaea</i>	<i>C. pamphylica</i> ssp. <i>afyonica</i>	<i>C. pamphylica</i> ssp. <i>tokuri</i>	<i>C. pamphylica</i> ssp. <i>pamphylica</i>
<i>C. argaea</i>	0			
<i>C. pamphylica</i> ssp. <i>afyonica</i>	56	0		
<i>C. pamphylica</i> ssp. <i>tokuri</i>	68	26	0	
<i>C. pamphylica</i> ssp. <i>pamphylica</i>	61	37	35	0

Table 2. A morphological comparison of *Campanula pamphylica* with *C. argaea*.

Character	<i>Campanula argaea</i>	<i>Campanula pamphylica</i>
Plant	tomentose-hispid, 10–35 cm	setulose-hirsute or tomentose-hispid, 20–70 cm
Stem	winged by decurrent leaf bases	unwinged
Basal leaves	oblong-spathulate, 1.5–3 cm wide	oblanceolate, obovate or oblong-spathulate; 0.7–1.5 cm wide
Median cauline leaves	oblong, decurrent on stem	oblong-spathulate, obovate-spathulate, oblanceolate or ovate-lanceolate; not decurrent on stem
Flowers	3–7 in clusters	solitary or 2–7 in clusters
Calyx lobes	6–8 mm, nearly as long as corolla tube	4–8 mm, shorter than or nearly as long as corolla tube
Corolla	up to 12 mm, densely tomentose-hispid outside	up to 17 mm, setulose or tomentose outside
Capsule	globose	ovoid or subglobose
Seeds	brown, shiny	brown, shiny or dark brown with narrow black margin

subsp. *pamphylica* in its setulose-hirsute indumentum; spathulate, obovate, strongly undulate and deeply dentate margined basal leaves; bracts ovate, ca. 5 × 2.5 mm; bracteoles ca. 4 × 1 mm; corolla 9–10(–12) mm; and style 7–7.5 mm. *Campanula pamphylica* subsp. *pamphylica* and *C. pamphylica* subsp. *afyonica* have long hispid hairs. This feature easily differentiates them from *C. pamphylica* subsp. *tokuri*, which has short hirsute hairs.

These subspecies are confined in SW Anatolia, in squares B3 and C3 (Fig. 2).

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