

A new subspecies of *Cardamine amara* (Brassicaceae) from Bulgaria and Greece

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Received 11 January 1996, accepted 20 March 1996

Karyological and distributional data about *Cardamine amara* L. (Brassicaceae) in Bulgaria and Greece, together with the description of a new subspecies and a short review of the history of taxonomic treatment of this species in the area studied is presented. *Cardamine amara* subsp. *balcanica* Marhold, Ančev & Kit Tan, distributed in SW Bulgaria and NE Greece, is characterised by a densely hairy stem and (15–) 17–34 (–37) cauline leaves with (2–) 3–4 (–5) pairs of lateral leaflets.

Key words: Brassicaceae, Bulgaria, *Cardamine*, Cruciferae, Greece, karyology, new subspecies, taxonomy

INTRODUCTION

Cardamine amara L. is a species (or species group, according to some authors, e.g. Jones 1964) which exhibits an interesting pattern of morphological and karyological variation. On the diploid level only two subspecies have been known until now, namely *C. amara* subsp. *amara* and *C. amara* subsp. *opicii* (J. Presl & C. Presl) Čelak. (Marhold 1992, 1993a, 1995). *Cardamine amara* subsp. *amara* is widespread throughout most of Europe except its most northern parts. Southwards it extends to the Pyrenees, Italy and Stara planina in Bulgaria. *Cardamine amara* subsp. *opicii* is restricted to the upper montane to alpine belt of the Sudety Mts. and the Carpathians. During the study of the Bulgarian and Greek populations it was revealed that they are diploid and

can be classified as a separate subspecies. The aim of this article is to present karyological and distributional data about *C. amara* in Bulgaria and Greece, together with the description of the new subspecies and a short review of the history of taxonomic treatment of *C. amara* in the area studied. Detailed morphometric comparison of the diploid populations of *C. amara*, currently classified as *C. amara* subsp. *amara*, *C. amara* subsp. *opicii*, and *C. amara* subsp. *balcanica* Marhold *et al.* will be presented in the second part of this study (Marhold, in prep.).

MATERIAL AND METHODS

The study is based on the herbarium material deposited in C, K, PR, SAV, SO, SOA, SOM, and the private herbarium of E.

Eleftheriadou (herb. Eleft.), and on the plants collected in the field in 1993–1995 and cultivated in Bratislava and Sofia. The karyotypes were studied from mitotic metaphase plates obtained from root tips taken from cultivated plants or in the field, fixed in ethanol:acetic acid (3:1), hydrolysed in 1N HCl for 10–12 min, and stained with haematoxylin after Gomori (Sharma & Sharma 1965). Localities and voucher specimens are marked by an asterisk (*) in the list of localities.

RESULTS AND DISCUSSION

Taxonomy and phytogeography

As shown by Marhold (1992), diploid populations of *Cardamine amara* vary considerably with respect to number of leaves, congestion of leaves below the inflorescence, number of leaflets, indumentum, width and branching of stem. According to these characters, separation of the montane to alpine populations from the Sudety Mts. and the Carpathians at subspecific level (*C. amara* subsp. *opicii*) from the populations of the lower altitudes in the area (subsp. *amara*) was clearly demonstrated. Further multivariate morphometric studies (Marhold, in prep.) were carried out on the material from the Sudety Mts. and the Carpathians (*C. amara* subsp. *amara* and *C. amara* subsp. *opicii*) together with the 14 population samples (altogether 427 plants) from the Bulgarian mountains. These studies have shown that Bulgarian populations from the Rhodope, Rila and Pirin Mts., and from Mt. Vitoša are close to subsp. *opicii* with respect to the number of leaves, width of stem and indumentum, but they are sufficiently distinct from this subspecies with respect to number of leaflets of the stem leaves and the branching of stem (see following identification key). At the same time the populations from the above mentioned Bulgarian mountains were shown to be clearly different from subsp. *amara* with respect to the number of leaves and the indumentum of stem. These results, together with those from karyological studies (see below), led us to segregate the Bulgarian populations and morphologically similar plants from NE Greece as a separate subspecies, *C. amara* subsp. *balcanica*.

It is clear from the following historical review that most Bulgarian authors who attempted to classify *C. amara* in a more detailed manner have included *C. amara* subsp. *amara* and what we presently recognise as *C. amara* subsp. *balcanica* in both

C. amara and *C. barbaraeoides* Halácsy (or alternatively, as *C. amara* subsp. *amara* and *C. amara* subsp. *barbaraeoides* (Halácsy) Stojanov & Stefanov).

The first record of *Cardamine amara* for the area of Bulgaria and Greece was that of Velenovský (1886), who mentioned it as occurring “Unter den Balkansabhänge stellenweise”. Later on Velenovský mentioned this species in his *Flora Bulgarica* and its *Supplementum* (1891: 29, 1898: 17) from several localities in the Stara planina and Rila Mts., and from Mt. Vitoša. He did not provide any information on the infraspecific status of these plants. Later it was reported by Urumov (1910: 17, 1913: 15–16, 1917: 24, 1926: 11, 1928: 9) from the Rila and Rhodope Mts. and from the Stara planina. Stojanov and Stefanov (1924: 507) in the first edition of their Bulgarian flora reported *C. amara* from the following mountains: Stara planina, Rila, Rhodope, Vitoša, Osogovo and Pirin. Apart from the glabrous “typical form” they classified the plants with scarce hairs from Rila, collected by Georgiev, as var. *hirta* Koch (Koch 1892: 39). Var. *hirta* is, however, an illegitimate name, now treated as synonymous with *C. amara* subsp. *amara*. The only corresponding specimen (collection of Georgiev) in Bulgarian herbaria is that from Kostenecki balkan, cited below, which has densely hairy stems. The account of *C. amara* is accompanied by the note that “according to Velenovský, the occurrence of *C. barbaraeoides* Halácsy in Bulgaria is quite probable”. Stojanov and Stefanov (1933: 463–464) recognised two subspecies of *C. amara* for Bulgaria. Apart from “subsp. *typicum*”, they published a new combination *C. amara* subsp. *barbaraeoides* (Halácsy) Stojanov & Stefanov [“*barbareoides*”], based on the species name, published by Halácsy (1894). *Cardamine barbaraeoides* is now treated as *C. raphanifolia* subsp. *barbaraeoides* (Halácsy) Strid and as pointed out by Strid (1986: 258) it is clearly different from *C. amara* (e.g. in having yellow anthers) and its known distribution area is restricted to the chain of mountains from Timfristos to Smolikas and Vasilitsa in Greece. This means that Stojanov and Stefanov confused this taxon with part of the Bulgarian populations of *C. amara* (namely from the Central Stara planina and Rhodope). In the third edition of the Flora of Bulgaria Stojanov and Stefanov (1948: 509) slightly changed the classification of *C. amara* and they recognised “subsp. *typica*” [sic!] with two formae (glabrous “forma *genuina*”, commonly occurring in

Bulgaria, sparsely hairy “forma *hirta* (Wimm. et Grab.)”, reported from Central Rhodope) and subsp. *barbaraeoides* [“*barbareoides*”], reported for Central Stara planina, Rhodope and Vitoša. Hayek (1925: 392–393) reported *C. barbaraeoides* for Bulgaria and Greece and *C. amara* for Macedonia (in his sense including parts of Bulgaria, Greece and the former Yugoslav Republic of Macedonia) and Bulgaria. Assenov (1970) reported *C. amara* and *C. barbaraeoides* for Bulgaria. According to his treatment *C. amara* occurs in the West Stara planina, Vitoša and Rila Mts., while *C. barbaraeoides* in the West and Central Stara planina, Osogovska planina, Vitoša, Pirin, Rila and the West and Central Rhodope.

Karyology

For *Cardamine amara* diploid ($2n = 16$) and tetraploid ($2n = 32$) chromosome numbers have so far been reported. *Cardamine amara* subsp. *amara* and *C. amara* subsp. *opicii* represent diploid taxa, while tetraploid populations widespread in the Eastern Alps probably form a separate subspecies (Marhold 1993b, 1994). *Cardamine amara* subsp. *olotensis* O. Bolós, an endemic subspecies described from Catalonia, is probably tetraploid as well (E. Rico, pers. comm.). We now report that chromosome number $2n = 2 \times = 16$ was found in 12 populations of *C. amara* subsp. *balcanica* from the Rila, Pirin, Rhodope Mts. and from Mt. Vitoša and one population of *C. amara* subsp. *amara* from the Stara planina. The karyotype of *C. amara* subsp. *balcanica* is symmetrical with 8 chromosome pairs similar in length. The chromosomes are most probably of m- and Sm-type as the primary constriction is visible in most of them, but not well expressed. In populations nos. A9326, A9328, A9329 and A9470 a pair of SAT-chromosomes with very small satellites were found (Fig. 1).

Description of new subspecies

Cardamine amara subsp. *balcanica* Marhold, Ančev & Kit Tan, *subspecies nova* (Fig. 2)

Rhizoma crassum, 5–12 mm latum. *Caulis erectus simplex aut in parte superiore ramosus, dense hirsutus (non nisi cum exceptione subglabratus)*. *Folia caulina numero (15–)17–34(–37); folia cum (2–)3–4(–5) jugis, aut paulum aut nullo modo congesta sub inflorescentia; foliola hirta aut*

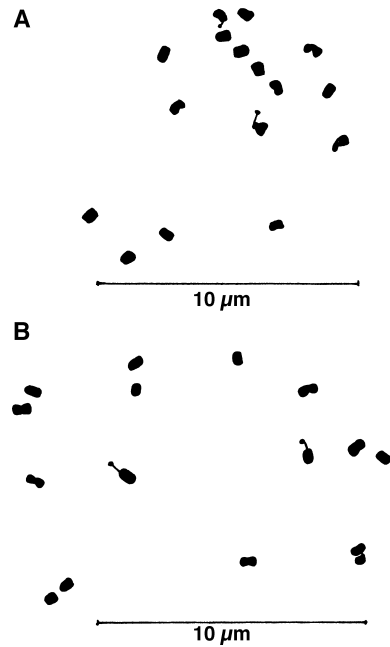


Fig. 1. Chromosomes (root tip mitoses) of *Cardamine amara* L. subsp. *balcanica* Marhold *et al.* (a) Bulgaria, Mt. Vitoša, “Aleko”, A9326, (b) Bulgaria, Rila Mts., Borovec, A9328.

subglabrata; foliolum apicale expressim majus quam foliola lateralialia.

Type: Bulgaria. Pirin Mts., valley of the river Demjanica, near Demjanica chalet, 1 850 m, 29.VI.1995, *M. Ančev & K. Marhold* (holotype SOM, isotypes B, C, SAV).

Perennial herb, 20–90 cm tall. Rhizome long, prostrate to ascending, thick, 5–12 mm in diam., clad with slender fibrous roots. Stem erect, simple or branched above, slightly sulcate, densely hairy (only very exceptionally subglabrous). Leaves not forming a basal rosette, cauline leaves (15–)17–34(–37), slightly or not congested below the inflorescence, with (2–)3–4(–5) pairs of lateral leaflets; leaflets sessile or shortly petiolulate, elliptic, oblong to ovate, seldom suborbiculate, shallowly lobed or slightly crenate, bright green, slightly flaccid, ciliate, puberulent to subglabrous, terminal leaflet much larger than lateral ones. Inflorescence racemose (corymbose at anthesis), simple or compound, 3–10 cm, with 6–40 flowers in the main inflorescence. Peduncles pubescent. Sepals ovate-oblong to lanceolate, 2.7–4.7 mm, glabrous, greenish membranous at margins. Petals white, obovate to oblanceolate, (5.9–)6.4–9.0(–9.4) × (2.3–)2.6–3.8(–4.2) mm, with short claw, apex

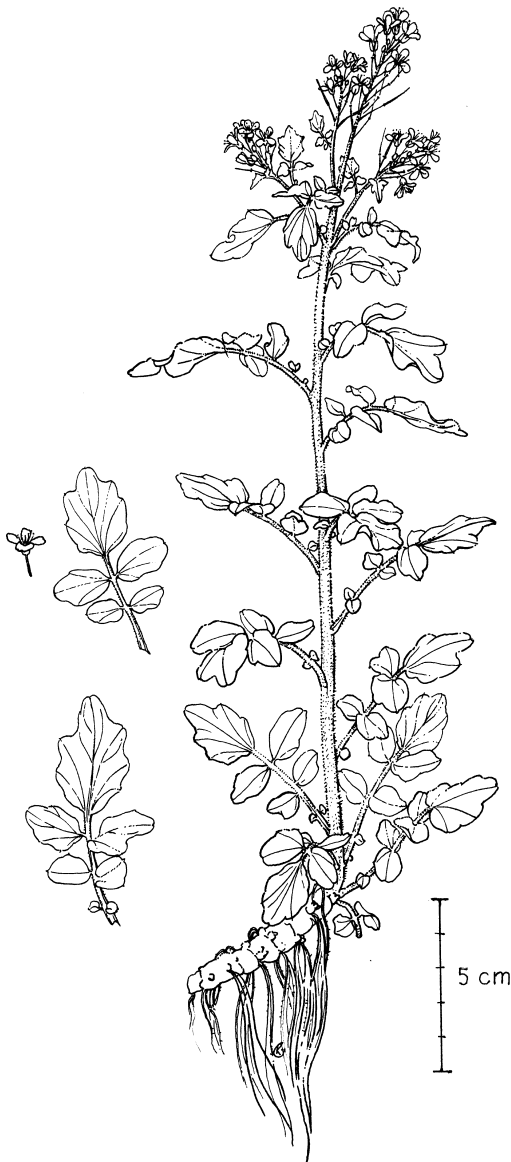


Fig. 2. Habit of *Cardamine amara* L. subsp. *balcanica* Marhold et al. from the *locus classicus* (drawn by K. Cigánová).

truncate to emarginate, glabrous. Stamens 6, indistinctly tetradynamous, equalling to slightly exceeding petals; filaments (3.5–)4.9–7.3(–7.8) mm, anthers reddish- to blackish-violet before anthesis, stigma inconspicuous. Pedicels ascending-erect to patent elongating to 15 mm in fruit, \pm glabrous. Siliquae diverging at the same angle or erect, 15–28 \times 1–2 mm glabrous, style 1.5–2.1 mm, seeds 0.8–1.5 \times 0.9–1.1 mm, pale brown.

Cardamine amara subsp. *balcanica* has the most southerly distribution of all the subspecies of *C. amara*. In SW Bulgaria it occurs by springs, streams and other wet places above 900 m, in mainly coniferous forest. In higher mountains it rarely extends above the timberline to the subalpine belt, up to 2 200 m in the Rila Mts. In NE Greece this subspecies occurs in damp shady places, along streams, tracks and openings in mixed forest dominated by *Picea abies* and *Fagus sylvatica*, on schist, at 1 450–1 800 m. *Cardamine amara* subsp. *balcanica* flowers from June to mid-August.

Identification key to the diploid taxa of *Cardamine amara*

- 1 Stem glabrous or sparsely hairy 2
- Stem densely hairy 3
- 2 Stem simple or branched; stem leaves (2–)3–14(–24), not congested beneath inflorescence; lower stem leaves with 2–6(–7) pairs of lateral leaflets subsp. *amara*
- Stem simple; stem leaves (10–)13–46(–53), congested beneath inflorescence; lower stem leaves with (4–)5–9(–11) pairs of lateral leaflets subsp. *opicii*
- 3 Stem simple; lower stem leaves with (4–)5–9(–11) pairs of lateral leaflets subsp. *opicii*
- Stem simple or branched; lower stem leaves with (2–)3–4(–5) pairs of lateral leaflets subsp. *balcanica*

Distribution of *Cardamine amara* in Bulgaria and Greece

Karyologically investigated populations and voucher specimens are marked with an asterisk (*).

Cardamine amara subsp. *amara*

Bulgaria. Stara planina: Vitinja, 880 m, 1952, *Železova* (SOM 31995). Below the pass Vitinja, Žerkovo, 750 m, 1995, *Marhold & Ančev* (SAV, SOM). Čurek, 840 m, 1995*, *Marhold & Ančev* (SAV, SOM A95124). Gabrovski Balkan, Arnautcki grobišta, 1902, *Nejčev* (SOM 32012). (?) Armankaja rivulet, 1903, *Nejčev* (SO 28089).

Cardamine amara subsp. *balcanica* (paratypes)

Bulgaria. Stara planina: Trevnenski Balkan, 1 900, *Urumov* (SOM 32008, 32014, 32015). — Vitoša region: Mt. Vitoša, 1903, *Mrkvička* (SOM 32011, 32013); 1915, *Střibný* (SOM 32019, 32022); 1915 *Stefanov* (SOA 4570). Mt. Vitoša, blatata,

1932, *Drjanovski* (SOM 32021). Mt. Vitoša, Reznjovete, 1 800 m, 1938, *Kitanov* (SO 28076, 28077). Mt. Vitoša, "Aleko", 1 800 m, 1993*, 1994*, *Marhold & Ančev* (SAV, SOM A9326*, A9466*). — Belasica Mts.: Above Petrič, 1922, *D. Jordanov* (SOA 15576). — Pirin Mts.: Pirin, 1915, *Urumov* (SOM 32017). Bajuvi dupki, 1977, *Andreev* (SOM 137834). Mutorok, 1969, *Kuzmanov* (SOM 124323, 124324). Mutorok, Dälgiya Čučur, 1983, *D. Stojanov* (SO 92141). Demjanica, 1929, "S. S. G." (SOA 4574); 1936, *Klásterský & Deyl* (PR). Demjaniška poljana, 1 620 m, 1995*, *Marhold & Ančev* (SAV, SOM A95102*). Near Demjanica chalet, 1 850 m, 1995*, *Marhold & Ančev* (SAV, SOM A95110*). BANSKO, near rivulet Glazne, 1 050 m, 1995, *Marhold & Ančev* (SAV, SOM). — Rila Mts.: Borovec, 1978, *Dočeva, Todorova & Peev* (SOM 137244); 1971, *Markova & Černeva* (SOM 137934); 1 300 m, 1993*, 1994, *Marhold & Ančev* (SAV, SOM A9328*). Above Borovec, 1993, *Strid & Kit Tan*, obs. Between Borovec and Sitnjakovo, 1 600 m, 1994*, *Marhold & Ančev* (SAV, SOM A9470*). BISTRICA, 1 600 m, 1993*, *Marhold & Ančev* (SAV, SOM A9329*). Carska Rila, Sokolec, 1 400 m, 1910, *Davidov* (SOM 32007). Carska Rila, "Šupalata", 2 000 m, 1911, *Davidov* (SOM 32001, 32002, 32004, 32005). Lagodaš, 1 200 m, 1931, *Fedčenko* (SOM 32016). Iskär river, Samokovsky Lag, 1 000 m, 1909, *s. coll.* (SOM 31994). Musala, 1901, *Tošev* (SOM 32018). Kostenecki Balkan, near Čavča river, 1889, *Georgiev* (SO 28090, 28091). Kosteneck, Jurukova poljana, Sokolovec, Inčov most, 1 500 m, 1994*, *Marhold & Vassilev* (SAV, SOM*). Kosteneck, Jurukova poljana, Sokolovec, 1 500 m, 1994*, *Marhold & Vassilev* (SAV, SOM*). Predela, 1961, *Simeonovski* (SO 28095). Samokov, 900 m, 1911, *Davidov* (SO 83216), 1 000 m, 1915, *Davidov* (SOM 32009). Maljovica, Meča poljana, 1 900 m, 1970, *Vichodceviski* (SO 28096). Komplex Maljovica, 1995*, *Marhold & Vassilev* (SAV, SOM*). Between Komplex Maljovica and Maljovica chalet, 1995, *Marhold & Vassilev* (SAV). Between Maljovica and Ovnarsko, 1995, *Marhold & Vassilev* (SAV). Komplex Maljovica, below Jončevo ezero, 2 100 m, 1995, *Marhold & Vassilev* (SAV). Bela Rila, Dolna Leva Reka, 2 000 m, 1911, *Davidov* (SOM 31997, 31998, 32006). Bela Rila, Mala Čerkva, 1 400 m, 1911, *Davidov* (SOM 32000). Bela Rila, Beli Iskär river, 1 400 m, 1915, *Davidov* (SOM 31996). Bela Rila, Beli Iskär river, 1 500 m, 1915, *Davidov* (SOM 31999). Belerečki izvori, 1897, *Stříbrný* (SOA 15575). Zelena Rila, Kriva Reka, 1893, *Davidov* (SOM 32003). Below Ribni ezera, along Rilska reka, 2 200 m, 1995, *Ančev* (SAV, SOM). Parangalica, Chajduvka river, 1975, *Andreev* (SOM 132166). Partizanska poljana, 1957, *Bondev* (SOM 109090, 109131). Parangalica, Kartala, 1 500 m, 1975, *Bondev, Andreev & Slavova* (Pl. Bulg. Exs. no. 921, as *C. barbaraeoides* Halácsy) (SOM 134237). — Rhodope Mts.: "Kaljač-burun", *s. a.*, *Urumov* (SOA 4572). Central Rhodope, between Kesten and Arda, 1938, *D. Jordanov* (SO 28093). Central Rhodope, Karläk, 1936, *Klásterský & Deyl* (PR); 1962, *s. coll.* (SOA 29523, 29524); 1962, *Dimitrov & Delipavlov* (SOA 22402); 1962, *Delipavlov* (SOA 08047-08049). Central Rhodope, Izgrev, 1984, *Delipavlov* (SOA 40450). West Rhodope, Pamporovo, 1 150 m, 1946, *N. Stojanov* (SO 28094). West Rhodope, Trigrad,

1963, *s. coll.* (SOA 08051-08054). West Rhodope, Beglika, 1935, *D. Antonov* (SOA 19097); 1956, *s. coll.* (SOA 08045, 08046). West Rhodope, Beglika, Toškovo dere, 1994*, *Marhold & Vassilev* (SAV, SOM*). West Rhodope, Beglika, Semsiza river, near Kolarova poljana, 1994*, *Marhold & Vassilev* (SAV, SOM*). West Rhodope, Beglika, natural reserve, near the right tributary of Semsiza river, 1994*, *Marhold & Vassilev* (SAV, SOM*). Greece. Nomos & Eparchia Dramas: Rodopi Mts., Eripiia Elatias, 1 450 m, 1988, *Eleftheriadou 2004* (herb. Eleft.). Tsakalos, 1 620 m, 1988, *Eleftheriadou 2067* (herb. Eleft.). Likolakka, Frakto forest reserve, 1 680 m, 1993, *Eleftheriadou 2388* (herb. Eleft.); 1 700 m, 1994, *Eleftheriadou 2785* (herb. Eleft.). Rema Likolakka, 1 600 m, 1993, *Eleftheriadou 2616*, Kanalies, 1 680 m, 1994, *Eleftheriadou 2888* (herb. Eleft.). Elatia forest, 13 km from Elatia to Plakia, 1 470–1 500 m, 1990, *Kit Tan*, obs. Along forest track to the right from Elatia-Plakia and Elatia-Stavrorema crossroads, ca. 1 500 m, 1990, *Kit Tan & G. Vold 8751* (C). Zagradenia area, Partheno Dasos, 1 600–1 800 m, 1988, *Strid & Kit Tan 27219* (C). — Nomos & Eparchia Serron: Vrontous Mts., Lailias, 1 650 m, 1935, *Tedd 1593* (K); *Voliotis* (ATHU) (*Voliotis* 1976: 36, 49).

Acknowledgements. The authors are indebted to Mrs. Katarína Cigánová (Bratislava) for illustration, to Mrs. Valja Goranova (Sofia) for technical assistance, and to Mr. Rossen Vassilev (Sofia) for his kind help in the field. This study was supported by the Grant Agency for Science (Bratislava) (Grant no. 2/2023/95) and by the National Science Fund of the Ministry of Science and Education (Sofia) (Grant no. B-410) which is gratefully acknowledged.

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