E. Liveri, E. Katopodi & G. Kamari

Karyosystematic study of some taxa from the Ionian floristic region (Greece). II

Abstract

Liveri, E., Katopodi, E. & Kamari, G.: Karyosystematic study of some taxa from the Ionian floristic region (Greece). II. [In Kamari, G., Blanché, C. & Siljak-Yakovlev, S. (eds), Mediterranean plant karyological data - 29]. – Fl. Medit. 29: 308-320. http://dx.doi.org/10.7320/FlMedit29.308

A karyosystematic study of six interesting taxa from the Ionian Islands (Greece) is presented. The chromosome number (2*n* = 16) and karyotype analysis for two ecologically different populations of the Greek endemic *Allium callimischon* subsp. *callimischon* are given and its karyotype variation is also confirmed. The chromosome number (2*n* = 16) and karyotype morphology of *Allium flavum* subsp. *tauricum* is reported for the first time in material from Ionian Islands. Karyotype analysis of *Ficaria calthifolia* (2*n* = 16) is given for first time in Greek material. Chromosome data (2*n* = 22) for *Freesia leichtlinii* subsp. *alba* are presented for the first time for this taxon. Previous karyological references of *Paeonia mascula* subsp. *russoi* (2*n* = 10) confirm our findings from a new population from north Kefallinia isl. The first karyological attempt at *Romulea bulbocodium* in Greece is performed here, resulting in two ploidy levels (2*n* = 3x = 33 and 2*n* = 4x = 44) for, ecologically, different populations and additionally the basic chromosome number of the genus is discussed.

Keywords: *Allium callimischon*, *A. flavum*, *Ficaria calthifolia*, *Freesia leichtlinii*, *Paeonia mascula*, *Romulea bulbocodium*, karyology, distribution, taxonomy.

Introduction

The present article is the second contribution to the flora of the Ionian floristic region, under the same title, also published in Flora Mediterranea journal by the research team of D. Phitos & G. Kamari. The reasons of this effort are mentioned in the introduction of the first article (Bareka & al. 2018).We hope that this effort will be continued in a follow-up project.

1962. ***Allium callimischon*** Link subsp. ***callimischon*** — 2*n* = 2x = 16 (Figs 1A, 1B & 3A, 3B).

Gr: Ins. Lefkada: Regio insulae borealis, Gyra dicta; in stagnosis, alt. 0-1 m, 20º 40΄ 54.77΄΄ E, 38º 50΄ 20.70΄΄ N, 05.XI.2017, leg.: *D. Phitos, G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29383*, UPA). – Figs 1A & 3A.

— Ins. Lefkada: Mons Stavrotas, ad cacumen Elati, in petrosis calc., alt. ca. 1100 m, 20°

38′ 47.38′′ E, 38° 42′ 27.14′′ N, 27.IX.2017, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29428*, UPA). – Figs 1B & 3B.

Allium callimischon (*Amaryllidaceae*) consists of two subspecies, according to Stearn (1978):

(a) subsp. *callimischon* (Figs 1A, 1B & 1C), endemic to Greece, described from NE Peloponnisos, has a wide distribution in the whole Peloponnisos, especially in the eastern (Constantinidis & Kalpoutzakis 2016) and southern part (Alibertis pers. com.); it also occurs on Kithira isl., W Ipiros, Ionian Islands (Tzanoudakis & al. 1991) and W Sterea Ellas (Bareka & al. 2000).

(b) subsp. *haemostictum* Stearn (Fig. 1D), restricted in Crete (Stearn 1978) and probably in SW Anatolia (Özhatay & Özhatay 1981).

The examined material of *Allium callimischon* subsp. *callimischon* was collected from Lefkada isl. The distribution of this subspecies in Lefkada is rather wide, extending from the sea level up to 1100 m. One of the observed populations occurs at the northern part of the island, specifically at the area of Gyra, in alt. 0-1 m (Fig. 1A). On Mt. Stavrotas, we observed three subpopulations: on the summit Elati (Fig. 1B), at the area of Agios Donatos and above the village Eglouvi. Moreover, in the eastern part of Lefkada isl. we also noticed two more subpopulations; above the village Nikiana (alt. 10-20 m) and in the clearing of the forest Skari (alt. ca. 600 m).

The cytotaxonomical study, hereby, includes plants from the two different habitats of the island. The first population of *A. callimischon* subsp. *callimischon* was collected at the area Gyra, on stagnate places (Fig. 1A), whereas the second population grows on the summit Elati of Mt. Stavrotas, on rocky limestone places (Fig. 1B). The above mentioned different ecological conditions of the two examined populations (i.e. altitude and substrate) have resulted in a different flowering time and size of the plants. More specifically, the plants of Elati are flowering from the end of August until the end of September and their stems size does not exceed 15 cm, whereas the plants of Gyra area are flowering from the end of October until mid-November and the stem height can reach up to 40 cm.

It is noteworthy that in the taxonomical keys for the two subspecies, given by Stearn (1978), only the difference in the shape of the spots on the upper part of the tepals is mentioned. As we have observed in a great amount of material from Lefkada, but also from all the distributional range of the taxon, the two subspecies differ additionally in the tepal shape and mainly in the ovary colour (mentioned here for the first time); green to yellow-green in the typical one (Figs 1A, 1B & 1C), whereas always dark reddish (colour of blood) in subsp. *haemostictum* (Fig. 1D).

The two karyologically examined populations of *A. callimischon* subsp. *callimischon* from Lefkada isl. are found to be diploids, with $2n = 16$ chromosomes (Figs 3A & 3B). The same chromosome number for the typical subspecies has been also given by Stearn (1978), Johnson (1982) and Iatrou (1986) in material from Peloponnisos, Tzanoudakis (1983a) and Tzanoudakis & al. (1991) in material from Peloponnisos, Lefkada, Ipiros and Kithira island and Bareka & al. (2000) in material from Sterea Ellas.

Concerning the chromosome morphology of *A. callimischon* subsp. *callimischon*, Johnson (1982), Tzanoudakis (1983a) and Bareka & al. (2000) give figures and more details. Tzanoudakis (1983a) highlights the great karyotype variation of all the examined populations. The karyotype formula given by Bareka & al. (2000) is 2*n* = 12m + 2m/sm + 2sm = 16 chromosomes.

The karyotypes of both examined populations are symmetrical, consisting mostly of metacentric chromosomes. The karyotype of Gyra population includes two pairs of marker-chromosomes: one pair is metacentric with a clear secondary restriction close to the centromere and one satellited pair with an additional secondary restriction close to the centromere (not always visible on both chromosomes) (Fig. 3A). On the contrary, the karyotype of the population from Elati has 4 marker-chromosomes with a strong secondary restriction close to the centromere, lacking, however, the small spherical satellites (Fig. 3B). Moreover, the karyotype of Elati population consists of chromosomes varying in size from 10.23 to 15.35 μm, while the chromosomes of Gyra population's karyotype vary from 12.22 to 19.44 μm.

1963. *Allium flavum* subsp. ***tauricum*** (Besser ex Rchb.) K. Richt — 2*n* = 2x = 16 (Figs 1E & 3C).

Gr: Ins. Lefkada: Mons Mega Oros, supra pagum Eglouvi in regione Striftouna; in petrosis, alt. ca. 880 m, 20° 37′ 38.45′′ E, 38° 43′ 35.15′′ N, 18.VI.2016, *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29116*, UPA).

Stearn (1978) recognizes under *Allium flavum* (*Amaryllidaceae*) two subspecies:

(a) subsp. *flavum*, distributed in S Europe and C Europe, from France to Turkey (Euro+Med 2006-).

(b) subsp. *tauricum*, distributed in SE Europe from Greece to European Russia (Euro+Med 2006-).

The geographical distribution of *Allium flavum* s.l. in Greece is notably unclear. Especially, for the Ionian floristic region, Dimopoulos & al. (2013) give only the presence of the typical subspecies, whereas the Flora Ionica Working Group (2016) refers to subsp. *tauricum* in Lefkada isl. and particularly from Mt. Stavrotas.

The taxonomical examination of our studied material from Mt. Stavrotas attributes it with certainty to subsp. *tauricum* (Fig. 1E).

The karyological studies of *Allium flavum* s.l. remain similarly unresolved, like their taxonomical relationships within the species. Many chromosome data with 2*n* = 16 and 2*n* = 32 are referred to *A. flavum* s.l., without further discrimination into a subspecies. Tzanoudakis & Vosa (1988) report both diploid and tetraploid populations from N and S Greece. Populations of *A. flavum* s.l. from Serbia are given as tetraploids (2*n* = 32) with B-chromosomes observed in some individuals (Vujosěvić & al. 2013). In total, 26 reports for chromosome numbers of *A. flavum* s.l. have been given mainly for European populations showing both diploids (2*n* = 16+0-2B) and tetraploids (2*n* = 32) (for more references see the Missouri Botanical Garden Database - Tropicos.org & PhytoKaryon - Kamari & al. 2017).

Concerning the subsp. *tauricum*, Baltisberger & Baltisberger (1995) report in material from adjacent Albania the chromosome number 2*n* = 16+1B and give a karyotype, consisting of meta- to submetacentric chromosomes, four of which are satellited. Also,

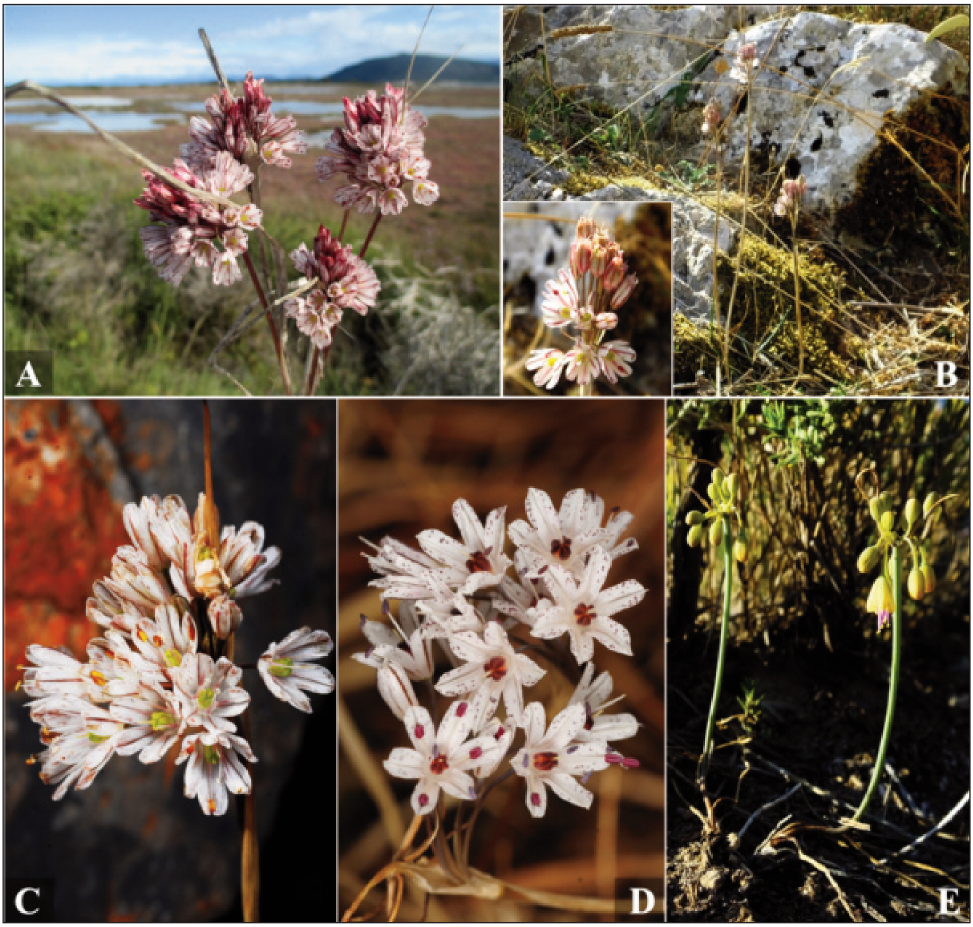

Fig. 1. *Allium* taxa studied: **A**, *Allium callimischon* subsp. *callimischon* on stagnate place (Gyra) and **B**, on rocky limestone (Mt. Stavrotas) from Lefkada island; Inflorescences of *A. callimischon*: **C**, subsp. *callimischon* from Lakonia (Peloponnese) and **D**, subsp. *haemostictum*from Crete; **E**, Individuals of *Allium flavum* subsp. *tauricum* from Mt. Mega Oros, Lefkada. – Photos C & D by A. Alibertis.

Karavokyrou (1994) reports 2*n* = 16 for populations of subsp. *tauricum* from Lesvos isl. (East Aegean). However, Özhatay (1990) describes a tetraploid karyotype (2*n* = 4x = 32), in material from Turkey, consisting of five sets of metacentrics, one set of submetacentric and two set of acrocentrics, with one of them bearing a satellite on the short arm.

The karyological study of *A. flavum* subsp. *tauricum* from Lefkada is the first report for the Ionian floristic region. The examined material is diploid (2*n* = 16), has always a symmetrical karyotype with all chromosomes being metacentric, varying in size from 9.44 to 17.78 μm and at least two pairs bearing small spherical satellites (one of them occurring on the longer arm) not always visible. The karyotype formula is: 2*n* = 2x = 12m + 4m-SAT = 16 chromosomes (Fig. 3C). No B-chromosomes have been observed in our material, but several reports mention one or two of them (Cheshmedzhirev 1971; Loidl 1979; Baltisberger & Baltisberger 1995; Krahulcová 2003; Vujosěvić & al. 2013).

1964. ***Ficaria calthifolia*** Rchb. — 2*n* = 2x = 16 (Figs 2A & 3D).

Gr: Ins. Kephallinia (Cephalonia vet.): Mons Aenos: ad cacumen Roudi, in loco Thea dicto; in apertis silvae Abietis cephalonicae, solo petroso, alt. 1055 m, 20° 36´ 59.68´´ E, 38° 11´ 24.03´´ N, 15.II.2017, leg.: *D. Phitos & D. Spanou* (Herb. Phitos & Kamari *29382*, UPA).

The genus *Ficaria* Guett. (*Ranunculaceae*) is known for its great morphological polymorphism and the subsequent confusion created by the use of different synonymies (*Ficaria* or *Ranunculus* L. etc.). Molecular data support the discrimination of *Ficaria* and *Ranunculus* into two different genera (Paun & al. 2005).

The *Ficaria verna* complex in Ionian Islands (e.g. Corfu, Lefkada, Kefallinia, etc.) is the case of a very variable group within the genus. According to Gutermann & al. (2014) and Flora Ionica Working Group (2016), at least three taxa of *Ficaria verna* agg. can be identified as being present on Ionian Islands: *F. calthifolia* as diploid, *F. verna* Huds. as tetraploid and *F. chrysocephala* (P. D. Sell) Galasso, Banfi & Soldano as tri/tetraploid.

However, the karyological reports for *F. calthifolia* in its wider distribution give several levels of polyploidy: 2*n* = 16 in material from Bosnia & Herzegovina, Croatia, Slovenia, 2*n* = 16+0-9B, 24, 40 in material from Croatia and 2*n* = 32 in material from Croatia and Slovenia (Papes & Trinajstic 1981, Druskovic & Lovka 1995, Lovka 1995). More chromosome counts are reported under the synonyms of the species e.g. 2*n* = 16 and 2*n* = 16+1-8B as *Ranunculus ficaria* subsp. *calthifolius* Arcang. in material from Armenia, Poland, Hungary, Bulgaria, Yugoslavia (Pogan & Wcisslo 1973, 1974, 1975, 1981, 1986) and Italy (Miceli & Garbari 1976, Capineri & al. 1978).

The studied population of *F. calthifolia* from the summit Roudi of Mt. Aenos shows diploid chromosome number 2*n* = 16. The karyotype is rather asymmetrical varying in size from 5.00-10.36 µm, consisting of 2*n* = 2x = 4m + 6sm + 4st + 2t-SAT= 16 chromosomes (Fig. 3D). The karyotype includes a SAT-chromosome pair, bearing large satellites that often are separated from the arm and, thus may seen as B-chromosomes. This chromosome count is the first for Greek material.

1965. ***Freesia leichtlinii*** subsp. ***alba*** (G. L. Mey.) J. C. Manning & Goldblatt — 2*n* = 2x = 22 (Figs 2B & 3E).

Gr: Ins. Kephallinia (Cephalonia vet.): Pars occidentalis insulae, supra sinu Lagadakia; in nanofruticetosis, terramroseam, alt. ca. 20 m, 20° 38´ 47.38´´ E, 38° 42´ 27.14´´ N, 16.II.2017, leg.: *D. Phitos& P. Minetos* (Herb. Phitos & Kamari *29385*, UPA).

The genus *Freesia* (*Iridaceae*) includes about 16 species the eastern tropical and southern Africa as their center of distribution (Goldblatt & al. 2006). *F. leichtlinii* is native to

South Africa, but it is found to grow naturalized in the wild also in other continents (Manning & Goldblatt 2001, 2010; Kim 2014). In Greece, it is also found as naturalized, usually growing at low altitude at open shiny stony places. Its occurrence in the Greek area cannot be considered frequent or at least it is not often collected. In Ionian Islands it is known from the islands Kefallinia, Lefkada, Paxi etc.

Freesia leichtlinii subsp. *alba* (Fig. 2B) is a geophyte, producing underground fleshy buds and arising from an underground corm. It grows from autumn to winter, flowering in early spring and dying back to the ground and remaining dormant in summer (Kim 2014). More information for *F. leichtlinii*, but also generally for the genus *Freesia* are offered in the exhaustive article of Manning & Goldblatt (2010).

As mentioned by previous researchers, the basic chromosome number of this genus is x = 11 (Taylor 1926, Goldblatt & Takei 1977, Wongchaochant & al. 2002, Goldblatt & al. 2006 etc.). Only one chromosome count (2*n* = 22) has been reported for *F. leichtlinii*, but no more details for its karyotype were provided then (Goldblatt 1982).

The chromosome number of the examined material is 2*n* = 22 with a diploid, symmetrical karyotype, consisting of metacentric and submetacentric chromosomes; the largest pair bears satellites (Fig. 3E). The size of the chromosomes varies from 1.47 to 2.06 μm. The given chromosome data comprise the first report not only for *F. leichtlinii* subsp. *alba*, but for the whole genus in the Greek area. Probably, this happens because *Freesia* does not attract particular interest in Greece, as it is considered naturalized.

1966. *Paeonia mascula* subsp. ***russoi*** (Biv.) Cullen & Heywood — 2*n* = 2x = 10 (Figs 2C & 3F).

Gr: Ins. Kephallinia (Cephalonia vet.): Inter pagum Neochori et sinum Giagana, alt. ca. 70 m, 20° 37΄ 02.30΄΄ E, 38° 20΄ 43.50΄΄ N, 27.IV.2015, leg.: *D. Phitos, N. Katsouni & V. Karagianni* (Herb. Phitos & Kamari *29441*, UPA).

The genus *Paeonia* L. (*Paeoniaceae*) includes ca. 35 species distributed widely in the northern hemisphere: eastern & central Asia, the western Himalayas, the Mediterranean region and the Pacific North America (Stern 1946, Tzanoudakis 1983b, Sang & al. 1997). In the Mediterranean area almost 22 species have been recorded so far (Aghababian 2011).

Paeonia mascula comprises five subspecies: (1) subsp. *mascula*, (2) subsp. *bodurii* N. Özhatay, (3) subsp. *hellenica* Tzanoud., (4) subsp. *icarica* Tzanoud. and (5) subsp. *russoi* (Aghababian 2011). All the above mentioned subspecies of *P. mascula* occur in Greece except for the Turkish endemic subsp. *bodurii*. However, the taxonomic status of the Greek subspecies is not clear. Tzanoudakis (1977, 1983b) highlights the variability within the species and recognizes the above mentioned four subspecies in the Greek area.

The plants of *Paeonia mascula* that occur on the islands of the Ionian floristic region Zakinthos, Kefallinia and Lefkada, as well as on the Akarnanika mountain range at the opposite side (W Sterea Ellada), were recognized as subsp. *russoi* (Cullen & Heywood 1964; Tzanoudakis 1977; 1983b). However, Hong & Wang (2006) and Hong (2010), based on some morphological similarities, as well as on the same chromosome number (2*n* = 10), assume that the Greek populations of subsp. *russoi* belong to *P. corsica* Sieber. The same

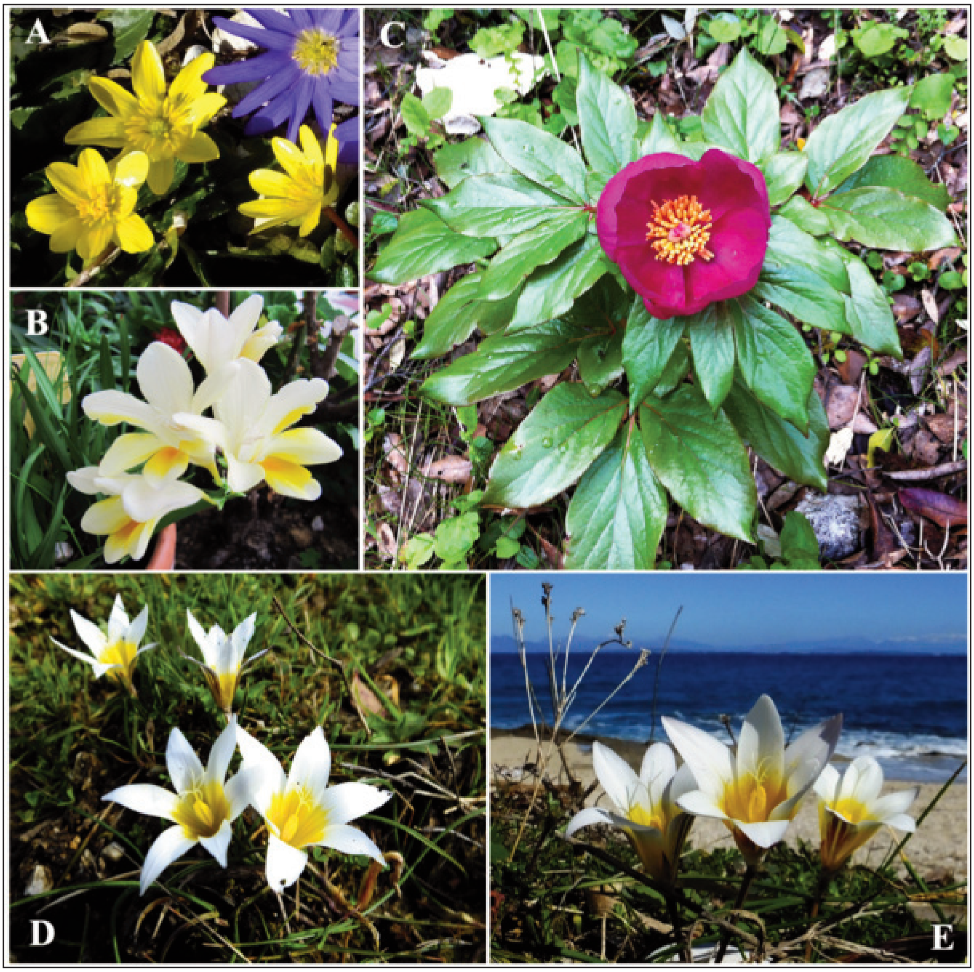

Fig. 2. Studied taxa from Kefallinia and Lefkada islands: **A**, *Ficaria calthifolia* from Mt. Aenos, summit of Mt. Roudi; **B**, Cultivated material of *Freesia leichtlinii* subsp. *alba*; **C**, individual of *Paeonia mascula* subsp. *russoi* from Giagana, N Kefallinia; *Romulea bulbocodium* from Lefkada: **D**, from Mt. Stavrotas and E, from the sand dunes of cape Gyrapetra.

name is used by Stearn & al. (2012), Dimopoulos & al. (2013) and Strid (2016). However, Phitos & al. (2015) consider the above facts insufficient to rename the plants of the Greek populations to *P. corsica*. Even less convincing is the phytogeographical distribution of *P. corsica* (Stearn & al. 2012). It is evident, mostly because of the numerous synonyms of *P. mascula* at the southern tip of the Italian Peninsula, Sicily, Corsica and Sardinia that the distinction of the representative taxa of the species has been ambiguous. For the above reasons we preserve here the previous name *P. mascula* subsp. *russoi* with its distribution in Ionian Islands and W Sterea Ellas. Moreover, molecular analysis of *P. mascula* subsp. *russoi* and

P. corsica from the whole distribution areas is under progress in order to elucidate their taxonomic relationships.

It is noteworthy that *P. mascula* subsp. *russoi* is included in the Red Data Book of Rare and Threatened Plants of Greece (Phitos & al. 2009) categorized as Near Threatened (NT), according to IUCN criteria (2001).

The evolutionary history of the genus *Paeonia* in the Mediterranean area has been closely related to polyploidy events. Concerning the ploidy levels within *P. mascula* subspecies, subsp. *russoi*, is diploid ($2n = 2x = 10$) and the remaining four are tetraploids ($2n = 4x = 20$) (Tzanoudakis 1977, 1983b; Özhatay & Özhatay 1995).

The chromosome number $2n = 10$ has been given for *P. mascula* subsp. *russoi* by Tzanoudakis (1977, 1983b) in material from the Ionian Islands (Lefkada, Kefallinia, Zakinthos) and W Sterea Ellas (Mt. Boumistos). Additionally, Passalacqua & Bernardo (2004) reported the tetraploid chromosome number $2n = 4x = 20$ for Italian populations under the taxonomic level of variety as *P. mascula* subsp. *mascula* var. *russoi* (Biv.) N. G. Passal. & Bernardo.

The chromosome number and the karyotype morphology, which is presented here from a new population in northern Kefallinia isl. (Fig. 3F), is in accordance with that given by Tzanoudakis (1977). The karyotype is diploid and symmetrical, consisting of $2n$ = 4m + 2m-SAT + 2sm-SAT + 2st-SAT = 10 chromosomes. The chromosome size varies from 9.30-13.95µm.

1967. *Romulea bulbocodium* (L.) Sebast. & Mauri — $2n = 3x = 33$ (Figs 2D & 3G) & $2n = 4x = 44$ (Figs 2E & 3H).

Gr: Ins. Lefkada: Mt. Stavrotas, ad cacumen Elati, alt. ca. 1100 m, 20° 38´ 47.38´´ E, 38° 42´ 27.14´´ N, 31.III.2016, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29442*, UPA). – Figs 2D & 3G.

— Ins. Lefkada: Regio borealis insulae, ad promunturium Gyrapetra dictum; in semiarenosis et in solo humidulo, alt. 2-5 m, 20° 40´ 54.77´´ E, 38° 50´ 20.70´´ N, 02.III.2017, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari*29210*, UPA). – Figs 2E & 3H.

The genus *Romulea* Maratti (*Iridaceae*) comprises ca. 95 taxa (Goldblatt & al. 2008). The distribution centre of the genus is found in South Africa and Arabian Peninsula, where 80 taxa occur (Manning & Goldblatt 2001, 2004, 2008). The remaining 15 taxa are endemic to the Mediterranean area (Peruzzi & al. 2011a).

Romulea is represented in Greece by five species: 1) *R. bulbocodium*, 2) *R. columnae* Sebast. & Mauri, 3) *R. linaresii* Parl., 4) *R. ramiflora* Ten. And 5) *R. tempskyana* Freyn (Dimopoulos & al. 2013). The first four species have been reported in some Ionian Islands, whereas *R. bulbocodium* occurs in most of them (see Flora Ionica Working Group 2016).

Despite the relatively wide distribution of *Romulea* species in Greece, the only taxon that has been studied karyologically is *R.* c.f. *linaresii* ($2n$ = ca. 39) from Athens (Goldblatt & Takei 1997).

Concerning the ancestral basic chromosome number in *Romulea*, the opinions of the authors are not concordant. For example, de Vos (1972) suggested the number $x = 12$, whereas Goldblatt & Takei (1997) $x = 13$. Additionally, several secondary basic chromo-

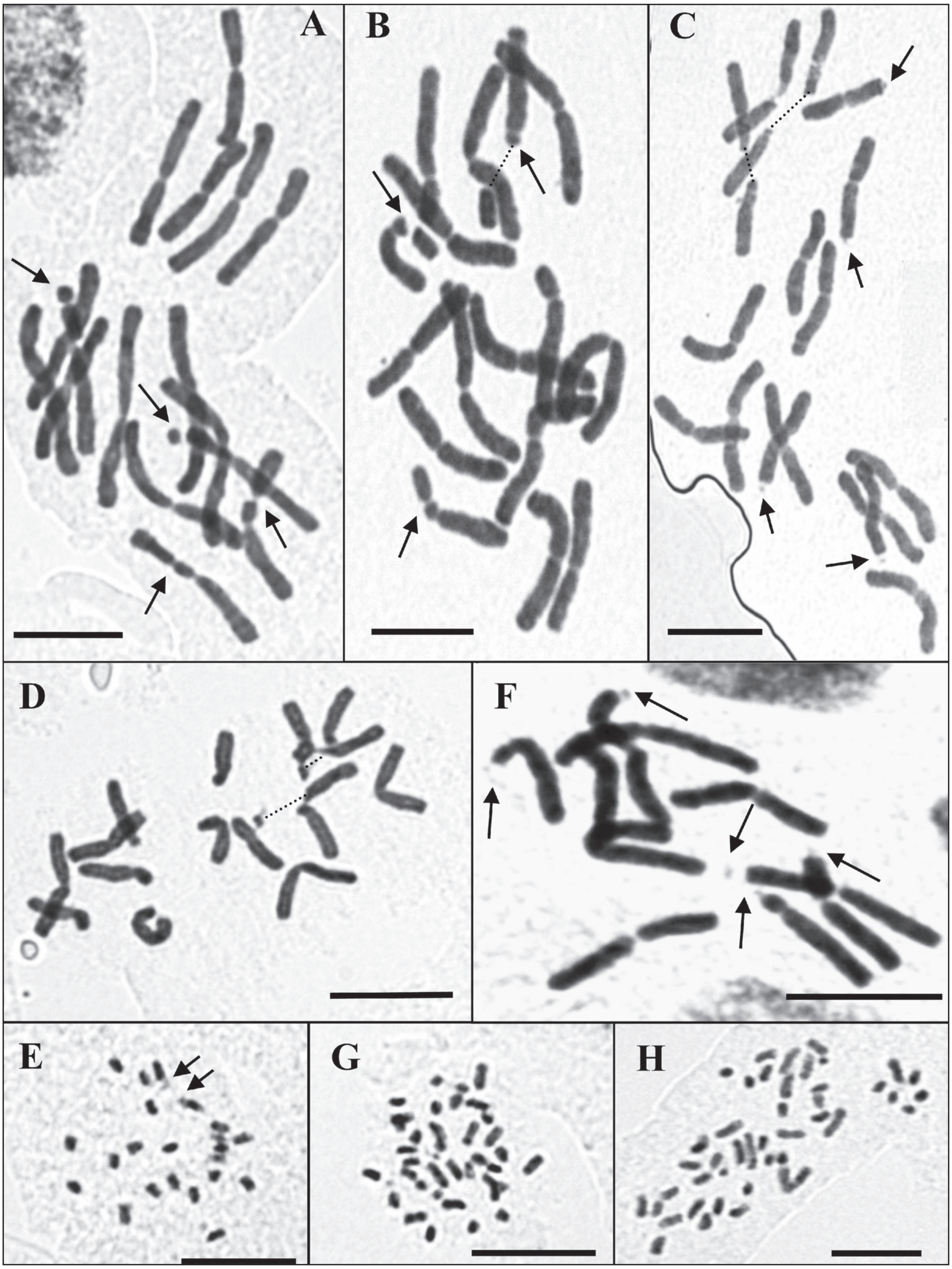

Fig. 3. Microphotographs of mitotic metaphase plates of the studied taxa: **A**, *Allium callimischon* subsp. *callimischon* from Gyra, 2*n* = 16 and **B**, from Mt. Stavrotas, 2*n* = 16; **C**, *Allium flavum* subsp. *tauricum*, 2*n* = 16; **D**, *Ficaria calthifolia*, 2*n* = 16; **E**, *Freesia leichtlinii* subsp. *alb*a, 2*n* = 22; **F**, *Paeoniamascula* subsp. *russoi*, 2*n* = 10; **G**, *Romulea bulbocodium* from Mt. Stavrotas, 2*n* = 3x = 33 and **H**, from Gyra, 2*n* = 4x = 44. – Arrows indicate satellites and/or secondary restrictions. Scale bars = 10 μm.

some numbers are reported, such as x = 9, 10, 11, 14 that are justified, due to dysploidy (de Vos 1972; Goldblatt & Takei 1997; Castro & Roselló 2005; Peruzzi & al. 2011a). Peruzzi & al. (2011a) suggest x = 9 as a shared basic chromosome number for all investigated taxa, opposing x = 14, favored by Castro & Rosello (2005). The discordant chromosome numbers given in literature are often the case for several Mediterranean *Romulea* species and could be interpreted as miscounting, due to the small chromosome size and their reluctance to stain, or as dysploid chromosome sets, realized through descending (i.e., 2*n* = 34) or ascending (i.e., 2*n* = 56) aneuploidy, or finally, as a possible effect of B-chromosomes (Peruzzi & al. 2011a).

The reported chromosome numbers for *Romulea bulbocodium* are: 2*n* = 28 in material from Spain (Castroviejo 1984), 2*n* = 34 in material from Iberian Peninsula (recognized as var. *rectifolia*; Fernandes & al. 1948) and in material from eastern Mediterranean of unknown origin (Darlington & Wylie 1955), 2*n* = 36 in material from Italy, Sicily and Sardinia (Peruzzi & al. 2011a, b) and 2*n* = 42 in material from Croatia (Susnik & Lovka 1973).

Our results confirm the intense karyotype variability of the species and the genus in general. One of the examined populations from Mt. Stavrotas (Fig. 2D) shows the chromosome number 2*n* = 3x = 33 with chromosome size varying from 1.07-2.86 μm (Fig. 3G), whereas the second population from the sand dunes of cape Gyrapetra (Fig. 2E) shows 2*n* = 4x = 44 with chromosome size from 0.88-3.53 μm (Fig. 3H). The karyological findings of the studied, here, populations support that the basic chromosome number for the species is x = 11. According to that, one population is triploid and the other one is tetraploid (Figs 3G & 3H). We present the chromosome number and a karyotype microphotograph because of the difficulty in counting and staining the material, as also mentioned by previous authors (e.g. Goldblatt & Takei 1997; Peruzzi & al. 2011a). The chromosome data given here constitute the first karyological contribution for *R. bulbocodium* from Greece.

Acknowledgements

We would like to thank Mr. A. Alibertis who provided us with photos of *A. callimischon* and also the staff of the Management Body of Mt. Aenos National Park (V. Karagianni, P. Minetos and D. Spanou) for their company and help during field work.

References

Aghababian, M. 2011+: *Paeoniaceae*. In: Euro+MedPlantbase - the information resource for Euro-Mediterranean plant diversity. – http://ww2.bgbm.org/EuroPlusMed/ [Last Accessed May 2019]

Baltisberger, M. & Baltisberger, E. 1995: Cytological data of Albanian plants. – Candollea **50(2):** 457-493.

Bareka, E. P., Koutoula, M. & Kamari, G. 2000: Reports (1106-1109). [In Kamari, G., Felber, F. & Garbari, F. (eds), Mediterranean chromosome number reports - 10]. – Fl. Medit. **10:** 381-430.

—, Katopodi, E., Kamari, G. & Phitos, D. 2018: Karyosystematic study of some taxa from the Ionian floristic region (Greece). I. – Fl. Medit. **28:** 85-97. https://doi.org/10.7320/flmedit28.085

Capineri, R., D' Amato, G. & Marchi, P. 1978: Numeri cromosomici per la Flora Italiana: 534-583. – Inform. Bot. Ital. **10(3):** 421-465.

Castro, M. & Rosselló, J. A. 2005: Chromosome numbers in plant taxa endemic to the Balearic islands. – Bot. J. Linn. Soc. **148:** 219-228. https://doi.org/10.1111/j.1095-8339.2005.00397.x

Castroviejo, S. 1984: Numeros cromosomaticos de plantas occidentales, 280-289. – Anales Jard. Bot. Madrid **40:** 457-462.

Cheshmedzhiev, I. V. 1971: A contribution to the cytotaxonomy of certain species of *Allium* L. – Bot. J. **56:** 662-670.

Constantinidis, Th. & Kalpoutzakis, E. 2016: Plant Guide to Mount Parnon and Moustos wetland protected area. Endemic, rare and threatened species. – Astros.

Cullen, J. & Heywood, V. H. 1964: Notes on the European species of *Paeonia*. – Feddes Repert. Spec. Nov. Regni Veg. **69:** 32-35.

Darlington, C. D. & Wylie, A. P. 1955: Chromosome Atlas of Flowering Plants. – London.

Dimopoulos, P., Raus, T., Bergmeier, E., Constantinidis, Th., Iatrou, G., Kokkini, S., Strid, A. & Tzanoudakis, D. 2013: Vascular plants of Greece. An annotated checklist. – Englera **31:** 1-372.

Druskovic, B. & Lovka, M. 1995: Reports. [In Stace, C. A. (ed.), IOPB chromosome data 9]. – Int. Organ. Pl. Biosyst. Newslett. (Zurich) **24:** 15-19.

Euro+Med (2006-): Euro+Med PlantBase - the information resource for Euro-Mediterranean plant diversity. – http://ww2.bgbm.org/EuroPlusMed/ [Last Accessed May 2019].

Fernandes, A., Gareia, J.& Fernandes, R. 1948: Herborizaçoesnos dominos da Fundaçao da Casa de Bragança. – Mem. Soc. Brot. **4:** 5-87.

Flora Ionica Working Group (2016 onwards): Flora Ionica- An inventory of ferns and flowering plants of the Ionian Islands (Greece). – https://floraionica.univie.ac.at/ [Last Accessed January 2018]

Goldblatt, P. 1982: Systematics of *Freesia* Klatt (*Iridaceae*). – J. S. African Bot. **48:** 39-91.

— & Takei, M. 1997: Chromosome Cytology of *Iridaceae* - Patterns of Variation, Determination of Ancestral Base Numbers, and Modes of Karyotype Change. – Ann. Missouri Bot. Gard. **84:** 285-304. https://doi.org/10.2307/2400005

—, Davies, J., Manning, J. C., van der Bank, M. & Savolainen, V. 2006: Phylogeny of *Iridaceae* subfamily *Crocoideae* based on a combined multigene plastid DNA analysis. – Aliso **22:** 399-411. https://doi.org/10.5642/aliso.20062201.32

—, Rodriguez, A., Powell, M. P., Davies, T. J., Manning, J. C., van der Bank, M. & Savolainen, V. 2008: *Iridaceae* "Out of Australasia"? Phylogeny, biogeography, and divergence time based on plastid DNA sequences. – Syst. Bot. **33:** 495-508. https://doi.org/10.1600/036364408785679806

Gutermann, W., Schneeweiss, G. M., Schönswetter, P., Staudinger, M. & Wallinger, C. 2014: The vascular plants of Kálamos (Ionian Islands, Greece): a catalogue. – Phyton **54(2):** 161-196.

Hong, D.-Y. 2010: Peonies of the world, taxonomy and phytogeography. – Kew.

— & Wang, X.-Q. 2006: The identity of *Paeonia Corsica* Sieber ex Tausch (*Paeoniaceae*), with special reference to its relationship with *P. mascula* (L.) Mill. – FeddesRepert. **117(1-2):** 65-84. https://doi.org/10.1002/fedr.200511081

Iatrou, G. 1986: Contribution to the study of endemism of the flora of Peloponnisos. – Ph.D. Thesis, University of Patras, Patras, Greece [In Greek with English summary].

IUCN 2001: IUCN Red List Categories and Criteria: version 3.1. − Gland & Cambridge.

Johnson, M. A. T. 1982: Karyotypes of some Greek species of *Allium*. – Ann. Mus. Goulandris **5:** 107-119.

Kamari, G., Bareka, P. & Stavropoulos, P. (eds) 2017 onwards: Phytokaryon – A karyological database for the Flora Hellenica. – http://www.phytokaryon.gr [Last Accessed April 2019]

Karavokyrou, E. 1994: Study of the plant genus *Allium* in Eastern Greece. – Ph.D. Thesis, University of Patras, Patras, Greece [In Greek with English summary].

Kim, T. K. 2014: Edible Medicinal and Non Medicinal Plants, **8.** – Dordrecht, Heidelberg, New York, London

Krahulcová, A. 2003: Chromosome numbers in selected monocotyledons (Czech Republic, Hungary, and Slovakia). – Preslia**75:** 97-113.

Loidl, J. 1979: C-band proximity of chiasmata and absence of terminalisation in *Allium flavum* (*Liliaceae*). – Chromosoma**73:** 45-51. https://doi.org/10.1007/bf00294843

Lovka, M. 1995: Reports. [In Stace, C. A. (ed.), IOPB chromosome data 9].– Int. Organ. Pl. Biosyst.Newslett. (Zurich) **24:** 21-23.

Manning, J. C. & Goldblatt, P. 2001: A synoptic review of *Romulea* (*Iridaceae: Crocoideae*) in sub-saharian Africa, the Arabian Peninsula and Socotra including new species, biological notes and a new infrageneric classification. – Adansonia **23:** 59-108. https://doi.org/10.4102/abc.v34i1.401

— & — 2004: Two new species of *Romulea* (*Iridaceae*, *Crocoideae*), from the western Karoo, northen Cape and notes on infrageneric classification and range extensions. – Bothalia **34:** 17-22. https://doi.org/10.4102/abc.v34i1.401

— & — 2008: *Iridaceae-Romulealitea* and *R. tubulosa* (*Crocoideae*), two new species from Namaqualand, South Africa. – Bothalia **38:** 78-82.

— & — 2010: Botany and horticulture of the genus *Freesia* (*Iridaceae*). – Sterlitzia **27:** 1-114.

Miceli, P. & Garbari, F. 1976: Numeri cromosomici per la flora italiana: 255-262. – Inform. Bot. Ital. **8(2):** 207-216.

Özhatay, N. 1990: The genus *Allium* in European Turkey and around Istanbul. – Ann. Mus. Goulandris **8:** 115-128.

— & Özhatay, E. 1981: A new record for the Turkish flora: *Allium callimischon* Link ssp. *Haemostictum* Stearn. – J. Fac. Pharm. Istanbul **17:** 103-106.

— & — 1995: A new white *Paeonia* L. from north-western Turkey: *P. mascula* Miller subsp. *bodurii* N. Özhatay. – Karaca Arbor. Mag. **3(1):** 17-26.

Papes, D. & Trinajstic, I. 1981: Reports. [In Löve, Á. (ed.), IOPB Chromosome number reports LXX]. – Taxon **30:** 70.

Paun, O., Lehnebach, C., Johansson, J. T., Lockhart, P. & Hörandl, E. 2005: Phylogenetic relationships and biogeography of *Ranunculus* and allied genera (*Ranunculaceae*) in the Mediterranean region and in the European Alpine System. – Taxon **54(4):** 911-930. https://doi.org/10.2307/25065478

Passalacqua, N.G. & Bernardo, L. 2004: The genus *Paeonia* L. in Italy: taxonomic survey and revision. – Webbia **59(2):** 215-268. https://doi.org/10.1080/00837792.2004.10670771

Peruzzi, L., Iiriti, G. & Frigani, F. 2011a: Contribution to the karyological knowledge of Mediterranean *Romulea* species (*Iridaceae*). – Folia Geobot. **46:** 87-94. https://doi.org/10.1007/s12224-010-9094-1

—, — & — 2011b: Reports. [In Marhold, K. (ed.), IAPT/IOPB chromosome data 11]. – Taxon **60(4):** 1220-1223.

Phitos, D., Constantinidis, Th. & Kamari, G. 2009: Red Data Book of Rare and Threatened Plants of Greece, **2(E-Z).** – Patras.

—, Kamari, G., Katsouni, N. & Mitsainas, G. 2015: The Mt. Aenos of Cephalonia island. History-Physiography-Biodiversity. – Cephalonia.

Pogan, E. & Wcisslo, H. 1973: Studies in *Ranunculus ficaria* L. I. Karyological analysis of *R. ficaria* L. ssp. *bulbifer* (Marsden-Jones) Lawalrée and *R. ficaria* L. ssp. *calthifolius* (Rchb.) Arc. – Acta Biol. Cracov., Ser. Bot. **16:** 135-143. https://doi.org/10.5586/asbp.1981.040

— & — 1974: Studies in *Ranunculus ficaria* L. II. Further karyological studies. – Acta Biol. Cracov., Ser. Bot. **17:** 165-173.

— & — 1975: Studies in *Ranunculus ficaria* L. III. Karyotype analysis. – Acta Biol. Cracov., Ser. Bot. **18:** 79-99.

— & — 1981: Studies in *Ranunculus ficaria* L. IV. Cytoembryological studies. – Acta Biol. Cracov., Ser. Bot. ,**23:** 37-54.
— & — 1986: Studiesin *Ranunculus ficaria* L. VII. Additions to chromosome numbers. – Acta Biol. Cracov., Ser. Bot. **28:** 87-92.
Sang, T., Crawford, D. J. & Stuessy, T. F. 1997: Chloroplast DNA phylogeny, reticulate evolution, and biogeography of *Paeonia* (*Paeoniaceae*). – Amer. J. Bot. **84(9):** 1120-1136. https://doi.org/10.2307/2446155
Stearn, W. T. 1978: European species of *Allium* and allied genera of *Alliaceae*: A synonymic enumeration. – Ann. Musei Goulandris **4:** 83-198.
—, Davis, P. & Tan, K. 2012: Peonies stin Ellada. – Athens.
Stern, F. C. 1946: A study of the genus *Paeonia*. – London.
Strid, A. 2016: Atlas of the Aegean Flora. Part 1: Text & Plates. Part 2: Maps. – Berlin.
Susnik, F. & Lovka, E. H. 1973: Reports. [In Löve, Á. (ed.), IOPB Chromosome numbers reports XLI]. – Taxon **22:** 462-463. https://doi.org/10.1002/j.1996-8175.1973.tb03387.x
Taylor, W. R. 1926: Chromosome morphology in *Fritillaria*, *Alstromeria*, *Silphium* and other genera. – Amer. J. bot. **13:** 179-193. https://doi.org/10.1002/j.1537-2197.1926.tb05877.x
Tropicos.org. Missouri Botanical Garden. – http://www.tropicos.org [Last Accessed April 2019].
Tzanoudakis, D. 1977: Cytotaxonomic study of the genus *Paeonia* in Greece. – Ph.D. Thesis, University of Patras, Patras, Greece [In Greek with English summary].
— 1983a: Karyotypes of ten taxa of *Allium* sect. *Scorodon* from Greece. – Caryologia **36(3):** 259-284. https://doi.org/10.1080/00087114.1983.10797667
— 1983b: Karyotypes of four wild *Paeonia* species from Greece. – Nord. J. Bot. **3:** 307-318
— & Vosa, C. G. 1988: The cytogeographical distribution pattern of *Allium* (*Alliaceae*) in the Greek peninsula and islands. – Pl. Syst. Evol. **159:** 193-215. https://doi.org/10.1007/bf00935972
—, Iatrou, G., Kypriotakis, Z. & Christodoulakis, D. 1991: Cytogeographical studies in some Aegean *Liliaceae*. – Bot. Chron. **10:** 761-775.
Vos de, M. P. 1972: The genus *Romulea*in South Africa. – J. South Afr. Bot.**9:** 1-307.
Vujosěvić, M., Jovanović, V. & Blagojević, J. 2013: Polyploidy and B chromosomes in *Allium flavum* from Serbia. – Arch. Biol. Sci., Belgrade **65(1):** 23-32.
Wongchaochant, S., Doi, M. & Imanishi, H. 2002: Phylogenetic Classification of *Freesia* spp. by Morphological and Physiological Characteristics and RAPD Markers. – J. Japan. Soc. Hort. Sci. **71(6):** 758-764. https://doi.org/10.2503/jjshs.71.758

Addresses of the authors:
Eleni Liveri[1], Eleni Katopodi[2] & Georgia Kamari[1],
[1]Botanical Institute, Section of Plant Biology, Department of Biology, University of Patras, GR-26500 Patras, Greece. E-mails: eleniliveri@upatras.gr; kamari@upatras.gr
[2]Laboratory of Systematic Botany, Faculty of Crop Science, Agricultural University of Athens, IeraOdos 75, GR-11855 Athens, Greece. E-mail: elkatopodi@hotmail.com