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CHROMOSOME NUMBERS IN *HIERACIUM* (ASTERACEAE) FROM CENTRAL AND SOUTHEASTERN EUROPE V

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Chromosome numbers for 15 taxa of *Hieracium* s.lat. (including two taxa of *Pilosella* Vaill.) from Bosnia and Herzegovina, Bulgaria, Greece, North Macedonia, Poland, Romania and Slovakia are given and their metaphase plates are illustrated. Chromosome numbers are published for the first time for *H. pannosum* subsp. *parnassi* Nägeli & Peter from Greece (3x and 4x), and for an undescribed species of *H.* sect. *Cernua* from North Macedonia (4x). A new, diploid chromosome number was found in *H. bracteolatum* s.lat. from Greece.

Keywords: Asteraceae, chromosome number, Europe, *Hieracium*, karyotype

INTRODUCTION

This paper continues a series on the karyology of *Hieracium* L. in Central and Southeastern Europe conducted at the Department of Plant Cytology and Embryology of the Jagiellonian University (Musiał and Szelag, 2015; Musiał et al., 2016, 2017, 2018). Here we present the chromosome numbers for 13 taxa of *Hieracium* s.str. and two species of *Pilosella* Vaill. from 20 populations in Bosnia and Herzegovina, Bulgaria, Greece, North Macedonia, Poland, Romania and Slovakia, including an undescribed species of *H.* sect. *Cernua* R. Uechtr. from North Macedonia. It is noteworthy that the diploid chromosome number for *H. bracteolatum* s.lat. from Greece was determined for the first time; this Aegean relict was hitherto known from tri- and tetraploid populations.

MATERIAL AND METHODS

The seeds for karyological investigations were collected from plants in nature or in an experimental garden. Then they were germinated on moistened filter paper in Petri dishes. The 3- or 4-day-old seedlings were incubated in saturated aqueous solution of 8-hydroxychinoline for 4 h at room temperature. They were subsequently fixed in a mixture of absolute ethanol and glacial acetic acid (3:1, v/v) for 24 h. The fixed material was stained in 2% acetic orcein for 4 days at room temperature. The stained seedlings were transferred to 45% acetic acid and heated to boiling over a flame. For slide preparation, root tip meristems were cut off and squashed in a drop of 45% acetic acid. The coverslip was removed after freezing in liquid nitrogen and the slide was thoroughly air-dried, and mounted in Entellan. The metaphase chromosomes were counted and photographed using a Nikon Eclipse E400 microscope equipped with a CCD camera. At least 10 seedlings were analyzed for each taxon and depending on the species, the somatic chromosome number was established on 9–23 well-spread mitotic metaphase plates in the meristematic cells of roots.

RESULTS AND DISCUSSION

Hieracium alpinum L.; 2n = 3x = 27 (Fig. 1a)
Bosnia and Herzegovina, Vranica Mts., Prokoško Jezero lake, 1750 m a.s.l., grassy places in *Pinus mugo* communities on silicate bedrock.

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Our chromosome count well matches that previously published from the Vranica Mts. (Mráz et al., 2009; Ilnicki and Szeląg, 2011). The Prokoško Jezero lake was the first locality of *H. alpinum* on the Balkan Peninsula discovered by Erich Brandis in 1888 (Szeląg, 2015).

Hieracium atratum var. *subnigrescens* (Fr.) R. Uechtr.; 2n = 4x = 36 (Fig. 1b)
Poland, Karkonosze Mts., Kocioł Łomniczki glacial cirque, 1280 m a.s.l., grassy slope on granite.

This is the first chromosome number report for this taxon from the Polish part of the Karkonosze Mts. which confirms the numbers published from the Czech part of the mountains (Chrtek, 1994).

Hieracium bifidum s.lat.; 2n = 3x = 27 and 2n = 4x = 36 (Fig. 1c)
1. Poland, Wyżyna Krakowsko-Częstochowska upland, Góra Zborów hill, 440 m a.s.l., calcareous rocks with *Valeriana tripteris*, 2n = 3x = 27.
2. Poland, Wyżyna Krakowsko-Częstochowska upland, Okiennik Wielki rock, 330 m a.s.l., calcareous rocks with *Festuca pallens*, 2n = 3x = 27.
3. Poland, Karkonosze Mts. Mały Śnieżny Kocioł glacial cirque, 1320 m a.s.l., basalt rock crevices, 2n = 3x = 27.
4. Poland, Wyżyna Krakowsko-Częstochowska upland, Wilcza Skała rock near Podlesice village, 360 m a.s.l., calcareous rocks with *Valeriana tripteris*, 2n = 4x = 36 (Fig. 1c).

Our data confirm the chromosome number published for *H. bifidum* s.lat. from other parts of Poland (Musiał et al., 2018).

Hieracium bracteolatum s.lat.; 2n = 2x = 18 (Fig. 1d)
1. Greece, Island of Evia, Mt. Xeroboúni (Ξεροβούνι) massive, SW of the Chapel of Agia Marina (Αγία Μαρίνα), 920 m a.s.l., an eroded slope in the *Abies cephalonica-Castanea sativa* forest on calcareous bedrock.
2. Greece, Island of Thasos, Mt. Toumpa (Τούμπα), 800 m a.s.l., in the *Pinus nigra* forest on the southern slope on silicate bedrock.

This is the first diploid chromosome number for *H. bracteolatum* s.lat., the Circum-Aegean endemic, previously known only from tri- and tetraploid populations (Chrtek et al., 2009; Merxmüller, 1975; Strid and Franzén, 1981; Schuhwerk and Lippert, 1998).

The analyzed plants are morphologically similar to *H. scamandris* Zahn from western Turkey (Sell and West, 1975) and will be the subject of ongoing studies to be presented separately (Vladimirov and Szeląg, in prep.).

Hieracium fritzei F.W. Schultz; 2n = 3x = 27 (Fig. 1e)
Poland, Karkonosze Mts., Łabski Kocioł cirque, 1300 m a.s.l., grassy slope along a turist path to Mt. Szrenica.

This is the first chromosome number report for this taxon from the Polish part of the Karkonosze Mts. which confirms the numbers published from the Czech part of the mountains (Chrtek, 1994).

Hieracium pannosum subsp. *parnassi* Nägeli & Peter; 2n = 3x = 27 and 2n = 4x = 36 (Fig. 1f and g)
1. Greece, Parnassus Mts. (Παρνασσός), along a road from Kalivia Arachovas (Καλύβια Αράχοβας) to the Parnassos ski center (Χιονοδρομικό Κέντρο Παρνασσού), 1250 m a.s.l., calcareous scree in the *Abies cephalonica* forest. 2n = 3x = 27 (Fig. 1f)
2. Greece, Parnassus Mts. (Παρνασσός), along a road from Kalivia Arachovas (Καλύβια Αράχοβας) to the Parnassos ski center (Χιονοδρομικό Κέντρο Παρνασσού), 1650 m a.s.l., calcareous rocks in the *Abies cephalonica* forest. 2n = 4x = 36 (Fig. 1g)

These are the first chromosome numbers for this taxon described from the Parnassus Mts.

Hieracium schmidtii Tausch; 2n = 4x = 36 (Fig. 1h)
Bulgaria, Stara Planina Mts., Čiprovska Planina Mts., above the village of Kopilovci along a tourist path to Mt. Kopren, 1150 m a.s.l., andesite rocks in the *Fagus sylvatica* forest.

This is the first chromosome number report for this taxon from Bulgaria. The tetraploid cytotype of *H. schmidtii* is rare; it was previously found in Poland (Musiał et al., 2018).

Hieracium sparsum Friv.; 2n = 2x = 18 (Fig. 2a)
Bulgaria, Rila Mts., between the village of Sestrimo and the Beklemen dam, 1500 m a.s.l., eroded slopes along a road on silicate bedrock.

This Balkan endemic is a diploid (Vladimirov and Szeląg, 2001; Szeląg et al., 2007). The analyzed plants are characterized by black achenes not observed in *H. sparsum* to date.

Hieracium tommasinianum K. Malý; 2n = 4x = 36 (Fig. 2b)
Bulgaria, Struma valley, along a road from the town of Boboshevo (Бобошево) to the Church of St. Demetrius (Бобощевски манастир Свети Димитър), 650 m a.s.l., eroded siliceous slope on *Carpinus orientalis* thickets margin.

This is the first chromosome number report for this taxon from Bulgaria. The tetraploid chromosome number was reported from Montenegro (Schuhwerk and Lippert, 1998).

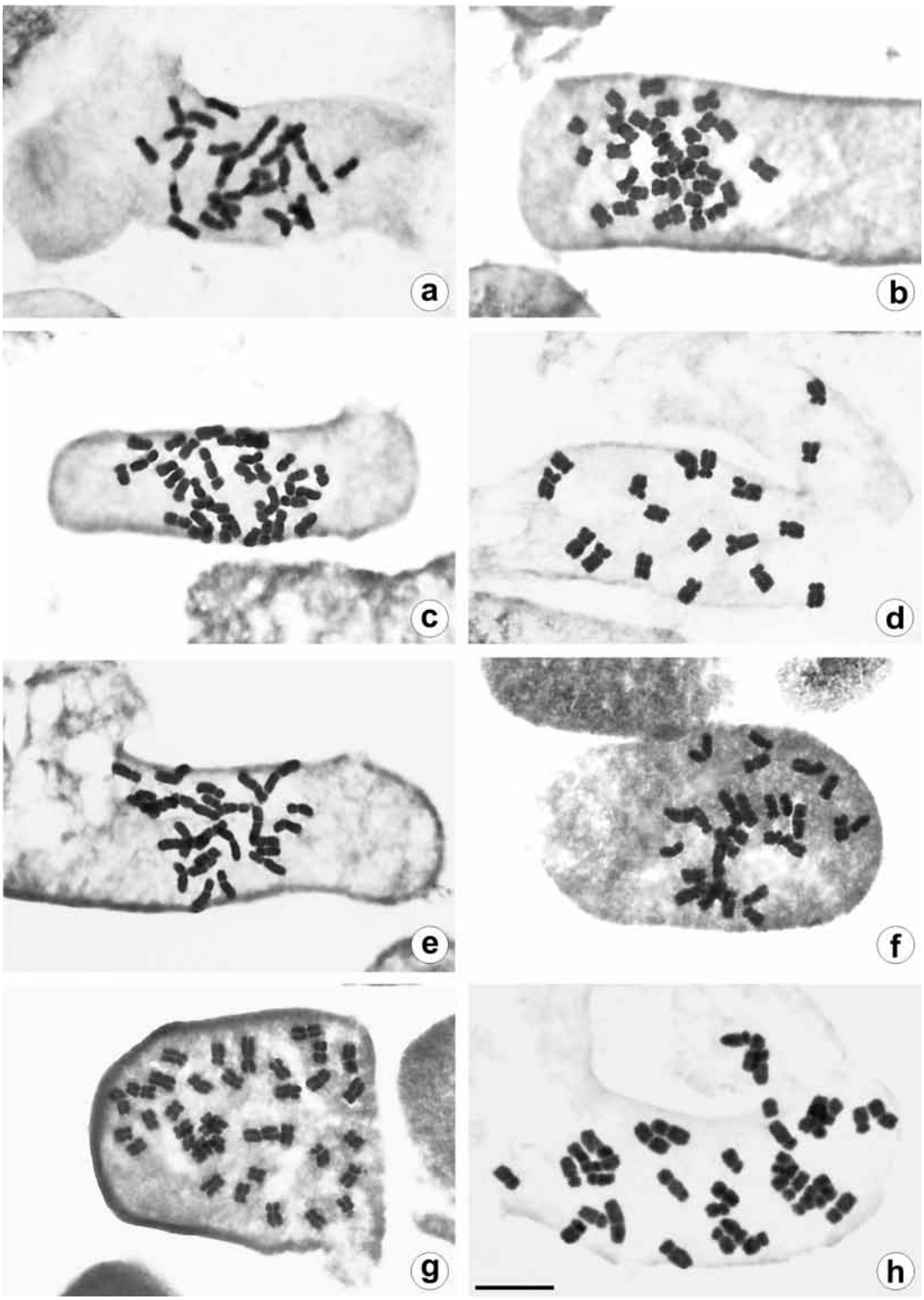

Fig. 1. Metaphase plates of: (**a**) *Hieracium alpinum* 2n=3x=27, (**b**) *H. atratum* var. *subnigrescens* 2n=4x=36, (**c**) *H. bifidum* s.lat. 2n=4x=36, (**d**) *H. bracteolatum* s.lat. 2n=2x=18, (**e**) *H. fritzei* 2n=3x=27, (**f**) *H. pannosum* subsp. *parnassi* 2n=3x=27, (**g**) *H. pannosum* subsp. *parnassi* 2n=4x=36 (**h**) *H. schmidtii* s.lat. 2n=4x=36. Scale bar in Fig. 1h = 10 μm and refers to all figures.

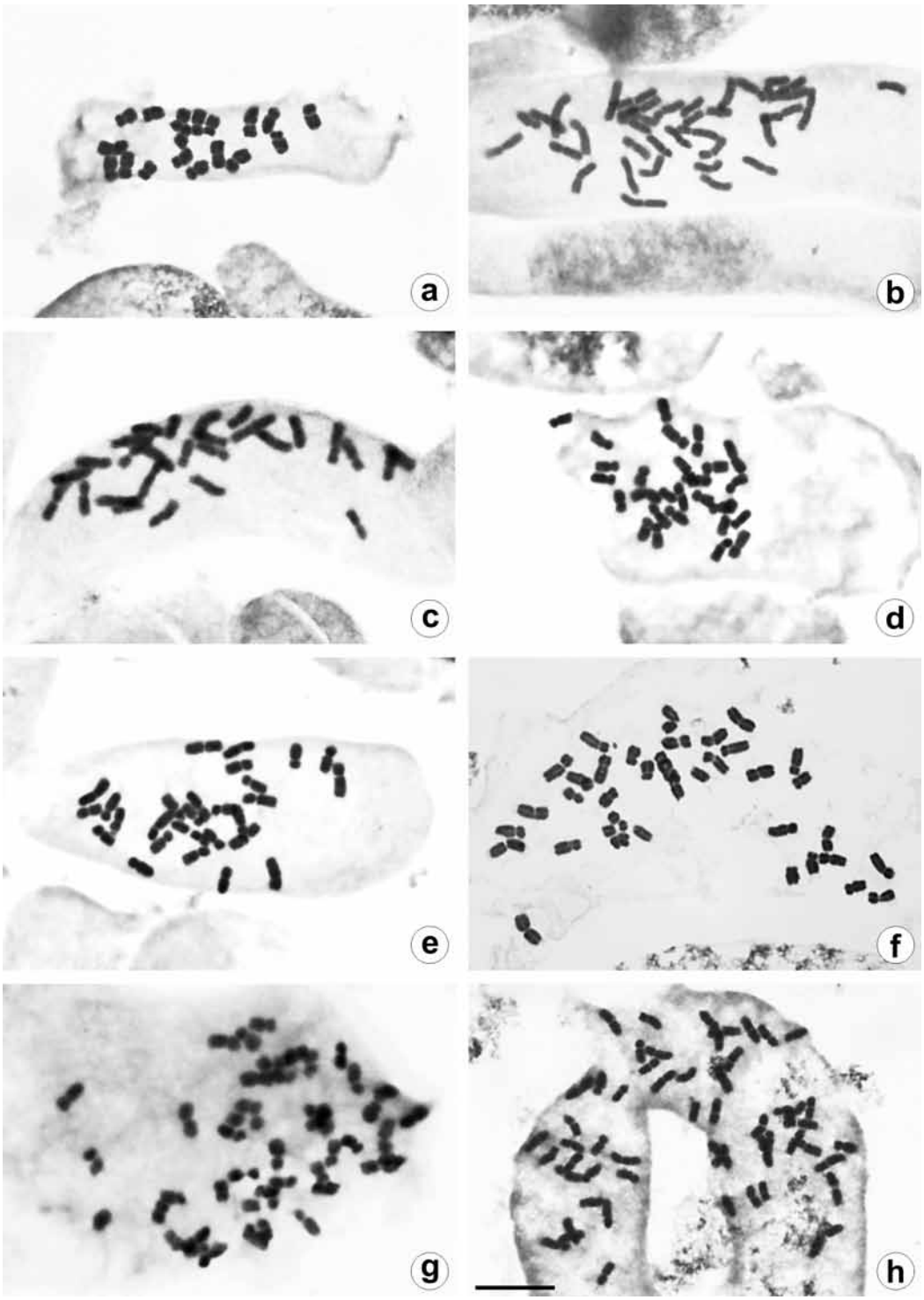

Fig. 2. Metaphase plates of: (**a**) *Hieracium sparsum* 2n=2x=18, (**b**) *H. tommasinianum* 2n=4x=36, (**c**) *H. virgicaule* 2n=3x=27, (**d**) *H. vulgatum* 2n=3x=27, (**e**) *H. wiesbaurianum* subsp. *kelainephes*, 2n=3x=27, (**f**) *H.* sp. 'Krivul' 2n=4x=36, (**g**) *Pilosella aurantiaca* 2n=5x=45 (**h**) *P. cymosa* 2n=6x=54. Scale bar in Fig. 2h = 10 μm and refers to all figures.

Hieracium virgicaule Nägeli & Peter; 2n = 3x = 27 (Fig. 2c)
Slovakia, Western Carpathians, Slovenský raj Mts., 250 m north-east of the road junction No. 66 and No. 67, 930 m a.s.l., on calcareous rocks along a road.

The same chromosome number was given by Chrtek et al. (2004).

Hieracium vulgatum Fries; 2n = 3x = 27 (Fig. 2d)
Poland, Western Carpathians, Gorce Mts. Mt. Wdżar near Kluszkowce, 700 m a.s.l., eroded calcareous slope in an old quarry.

The triploid chromosome number was reported in plants from Great Britain (Marton, 1974) and from Poland (Musiał and Szeląg, 2015)

Hieracium wiesbaurianum subsp. *kelainephes* Nyár. & Zahn; 2n = 3x = 27 (Fig. 2e)
Romania, Southern Carpathians, Mehedinți Mts, Cheile Țăsnei gorge, 650 m. a.s.l., calcareous scree.

A new locality of this taxon, hitherto known only from the Cheile Turzii gorge. Our data confirm the chromosome number published in plants from the *locus classicus* (Musiał et al., 2018).

Hieracium sp. 'Krivul'; 2n = 4x = 36 (Fig. 2f)
North Macedonia, Jakupica Mts. Mt. Krivul, 1900 m a.s.l., grassy places in *Pinus mugo* communities on silicate.

Most probably a new, undescribed species of *Hieracium* sect. *Cernua* R. Uechtr. similar to *H. sparsum* subsp. *ipekanum* Rech. fil. & Zahn from Montenegro.

Pilosella aurantiaca (L.) F.W. Schultz & Schultz-Bip.; 2n = 5x = 45 (Fig. 2g)
Romania, Southern Carpathians, Ciucaş Mts. Şaua Gropşoarele pass, 1650 m a.s.l., subalpine grasslands on silicate bedrock.

The pentaploid cytotypes of *P. aurantiaca* are rare; our data confirm those published from Czechia (Krahulcová et al., 2001).

Pilosella cymosa (L.) F.W. Schultz & Schultz-Bip;. 2n = 6x = 54 (Fig. 2h)
North Macedonia, Jakupica Mts., Begovo Pole, 2050 m a.s.l., grasslands on calcareous slope.

The same chromosome number was published from the French Alps (Gadella and Kliphuis, 1970).

AUTHORS' CONTRIBUTION

KM – karyological analysis, preparation of figures and interpretation of results; ZS – sampling and drafting of manuscript. The authors have declared that there is no conflict of interest.

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