

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

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Chromosome numbers for 16 *Hieracium* s.str. species from Bosnia and Herzegovina, Bulgaria, Macedonia, Montenegro, Poland, Romania and Serbia are given and their metaphase plates are illustrated. Chromosome numbers are published for the first time for *H. filarszkyi* Jav. & Zahn $2n = 3x = 27$, *H. fritschianum* Hayek & Zahn $2n = 3x = 27$, *H. fritzeiforme* Zahn $2n = 3x = 27$, *H. hercegovinicum* (Freyn & Vandas) Szelag $2n=3x=27$, *H. nivimontis* (Oborny & Zahn) Chrtk fil. $2n = 4x = 36$, *H. vagneri* Pax $2n = 4x = 36$, as well as three undescribed species of hybrid origin between *H. olympicum* Boiss. and *H. sparsum* Friv. $2n = 4x = 36$, *H. naegelianum* Panč. and *H. scardicum* Bornm. & Zahn $2n = 3x = 27$, and *H. transylvanicum* Heuff. and *H. umbellatum* L. $2n = 4x = 36$.

Key words: Asteraceae, chromosome number, Europe, *Hieracium*, karyotype.

INTRODUCTION

During several years of studies on *Hieracium* L. in Central and Southeastern Europe, the second author collected many living plants and transplanted them to an experimental garden for future taxonomic investigations. To date, many of them have never been analysed karyologically. Knowledge of ploidy level, which especially in *Hieracium* s.str. indicates the mode of reproduction, is of particular interest in understanding taxonomic and phylogenetic relationships within the genus.

The present paper gives chromosome numbers for 16 species of *Hieracium* s.str. including three undescribed species of hybrid origin. The ploidy levels of 9 species are published for the first time.

This study is a continuation of karyological investigations of *Hieracium* L. in Europe conducted at the Department of Plant Cytology and Embryology of the Jagiellonian University (Szelag et al., 2007; Ilnicki et al., 2010; Ilnicki and Szelag, 2011; Szelag and Ilnicki, 2011).

MATERIAL AND METHODS

For cytological studies, seeds of the investigated species were collected from natural populations. Then they were germinated on moistened filter paper in Petri dishes. Karyological analysis was performed as described by Marciniuk et al. (2012) with some modifications. Briefly, 3- or 4-day-old seedlings were incubated in saturated aqueous solution of 8-hydroxychinoline for 4 h at room temperature. Subsequently they were fixed in a mixture of absolute ethanol and glacial acetic acid (3:1, v/v) for 24 h. Fixed material was stained in 2% acetic orcein for 4 days at room temperature. 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 cover-slip 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 Nikon Eclipse E400 microscope equipped with CCD camera.

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RESULTS AND DISCUSSION

Hieracium atratum s.l.; $2n = 4x = 36$ (Fig. 1a)
 Poland, Eastern Carpathians, Bieszczady Mts, Mt. Tarnica 1330 m a.s.l., rocky places with *Empetrum hermaphroditum*.

This is the first chromosome number report for this species from the Eastern Carpathians and from Poland which confirms the number previously published by Chrtek et al. (2004) from the Western Carpathians. Chrtek (1994) also published tri- and tetraploid chromosome numbers for two species belonging to the *Hieracium atratum* aggregate from the Western Sudetes.

Hieracium chrysostyloides (Zahn) Chrtek fil.; $2n = 5x = 45$ (Fig. 1b)

Poland, Eastern Sudetes, Mt. Śnieżnik Kłodzki, 1410 m a.s.l., alpine grassy places on western slope.

This is the first chromosome number report for this species from Poland which confirms the number previously published from the Hrubý Jeseník Mts, Eastern Sudetes (Chrtek, 1996). *Hieracium chrysostyloides* is endemic to the Eastern Sudetes and is one of three pentaploid *Hieracium* s.str. species known so far. The second one, *H. pentaploideum* P.D. Sell & D.J. Tennant, grows in Scotland and also belongs to the *H. nigrescens* agg. (Sell and Murrell, 2006; Tennant and Rich, 2008). The third one is *H. virosum* Pall. from Siberia in Russia (Pulkina and Tupitsyna, 2000).

Hieracium filarszkyi Jáv. & Zahn; $2n = 3x = 27$ (Fig. 1c)

Romania, Southern Carpathians, Retezat Mts, Mt. Butea, 1820 m a.s.l., grassy slope with *Pinus mugo* communities on granite.

This is the first chromosome number report for this endemic to the Retezat Mts species.

Hieracium fritschianum Hayek & Zahn; $2n = 3x = 27$ (Fig. 1d)

Bosnia and Herzegovina, Prenj Mts, Mt. Otiš, 1900 m a.s.l., calcareous rocks on eastern slope.

This is the first chromosome number report for this species known until now only from Albania.

Hieracium fritziforme Zahn; $2n = 3x = 27$ (Fig. 1e)
 Romania, Southern Carpathians, Retezat Mts, Saua ciurila pass, 1800 m a.s.l., grassy slope with *Pinus mugo* communities on granite.

This is the first chromosome number report for this endemic to the Retezat Mts species.

Hieracium heldreichii s.l. sensu Zahn; $2n = 3x = 27$ (Fig. 1f)

Serbia, Mt. Besna Kobila, 1840 m a.s.l., eroded slope along road on the summit, surrounded by pastures with *Festuca valida*, *Festuca nigrescens* and *Deschampsia caespitosa*, on schist.

It is quite a frequent on the Balkan Peninsula collective species of the morphological formula *H. pannosum* < *H. racemosum*. The same chromosome number was earlier reported in plants of unknown origin (most probably from Bulgaria) by Gentscheff (1938). This is the first chromosome number report for this species from Serbia.

Hieracium hercegovinicum (Freyn & Vandas) Szelag; $2n = 3x = 27$ (Fig. 1g)

Montenegro, Prokletije Mts, Mt. Maja Rosave, below Zastan Koliba military checkpoint, 1430 m a.s.l., *Fagus sylvatica* forest margin on calcareous bedrock.

This is the first karyological data for this species of presumably hybrid origin between *H. prenanthoides* Vill. and *H. gymnocephalum* Griseb. ex Pant., morphologically closer to the former species (Szelag, 2002). The plants analysed here were collected in the area in which diploid, sexual *H. gymnocephalum* grows (Szelag and Ilnicki, 2011). The same chromosome number was reported by Niketić et al. (2003) in plants referred to as *H. calophyllum* aggregate (morphological formula *H. gymnocephalum* – *H. prenanthoides*).

Hieracium nigritum R. Uechtr.; $2n = 4x = 36$ (Fig. 1h)

Poland, Eastern Sudetes, Mt. Śnieżnik Kłodzki, Hala pod Śnieżnikiem glade, 1260 m a.s.l., subalpine meadows with *Festuca supina*.

This is the first chromosome number report for this species from Poland which confirms the number previously published by Chrtek (1996) from the Hrubý Jeseník Mts, Eastern Sudetes, and from the Western Carpathians (Chrtek et al., 2004).

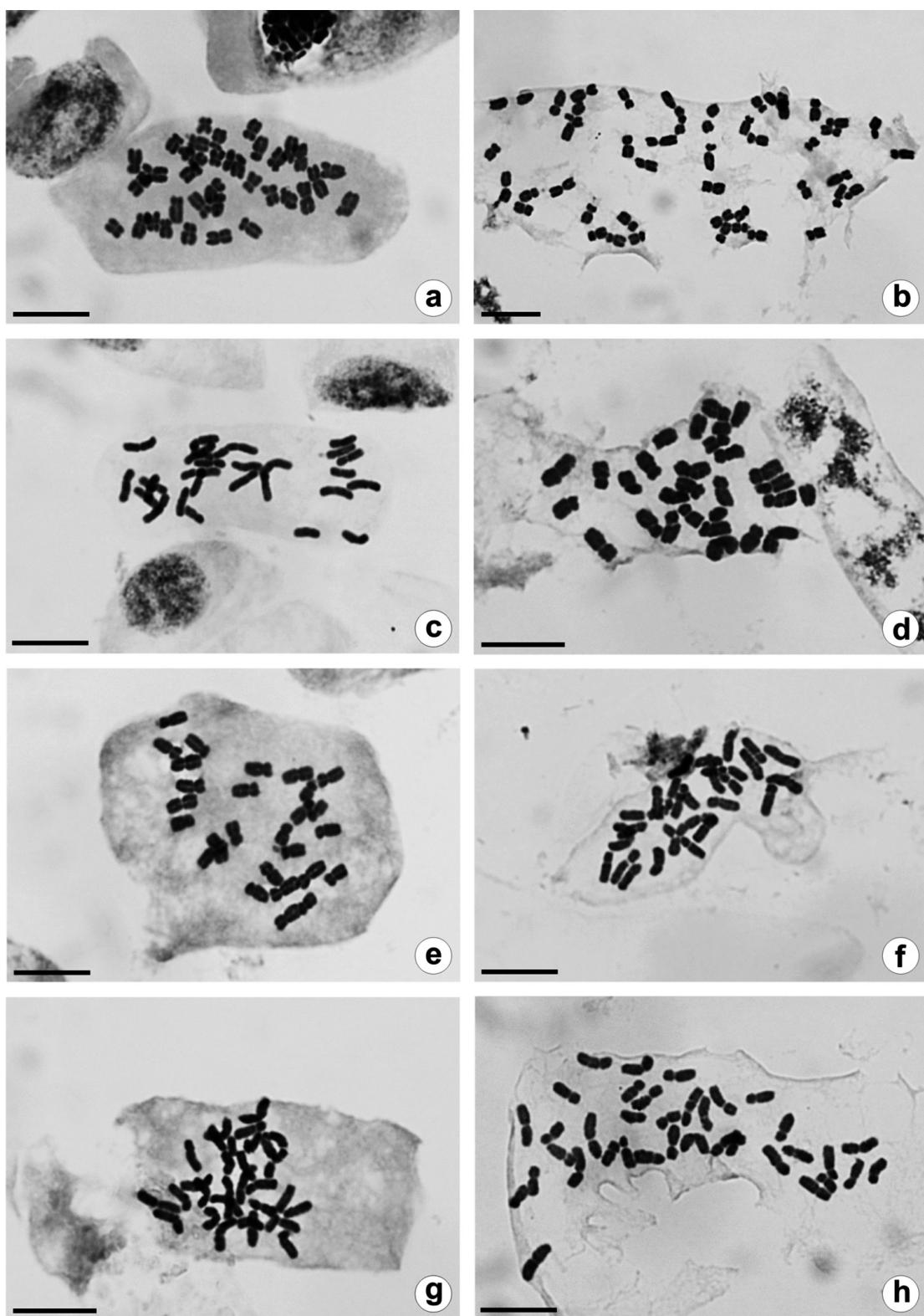


Fig. 1. Metaphase plates of: (a) *H. atratum*, $2n = 36$. (b) *H. chrysostyloides*, $2n = 45$. (c) *H. filarszkyi*, $2n = 27$. (d) *H. fritschianum*, $2n = 27$. (e) *H. fritzeiforme*, $2n = 27$. (f) *H. heldreichii*, $2n = 27$. (g) *H. hercegovinicum*, $2n = 27$. (h) *H. nigritum*, $2n = 36$. Bar = 10 μm .

Hieracium nivimontis (Oborny & Zahn) Chrtek fil.; $2n = 4x = 36$ (Fig. 2a)
 Poland, Eastern Sudetes, Mt. Śnieżnik Kłodzki, 1380 m a.s.l., alpine grassy places on western slope.

This is the first chromosome number report for this species known only from Mt. Śnieżnik Kłodzki.

Hieracium schustleri Zlatník; $2n = 4x = 36$ (Fig. 2b)

1. Poland, Eastern Sudetes, Mt. Śnieżnik Kłodzki, 1400 m a.s.l., alpine grassland on western slope (population with dark styles).
2. Poland, Eastern Sudetes, Mt. Śnieżnik Kłodzki, 1410 m a.s.l., rocky places on north-western slope (population with yellow styles) (Fig. 2b).

The same chromosome number was reported in plants from the Karkonosze/Krkonoše Mts (Chrtek, 1994).

Hieracium sudetotubulosum Szelag; $2n = 4x = 36$ (Fig. 2c)

Poland, Central Sudetes, Góry Stołowe Mts, upper verge of Mt. Szczeliniec Wielki, NW cliff next to the lookout, 900 m a.s.l.

Newly validated species (Szelag, 2014) known from the Western Sudetes and recently discovered also in the Góry Stołowe Mts (Szelag and Wójcik, 2014).

The same chromosome number was reported in plants from the Karkonosze/Krkonoše Mts, Western Sudetes by Chrtek (1994).

Hieracium vagneri Pax; $2n = 4x = 36$ (Fig. 2d)
 Poland, Eastern Carpathians, Bieszczady Mts, Mt. Bukowska Kopa, 1310 m a.s.l., rocky places on sandstone with calcicolous plants.

This is the first chromosome number report for this species of morphological formula *H. alpinum* – *H. caesium* which comprises a few taxa at the subspecies rank occurring in the Carpathians and Sudetes (Zahn, 1938).

Hieracium vulgatum Fries; $2n = 3x = 27$ (Fig. 2e)
 Poland, Wyżyna Miechowska upland, Kalina Lisiniec, 330 m a.s.l., termophilous chalk grassland *Inuletum ensifoliae*.

This is the first chromosome number report for this species from Poland. Triploid chromosome number was reported in plants from Great Britain (Morton, 1974). Information from Belarus (Dmitrieva, 1987) refers to *H. lachenalii* Suter.

Hieracium sp. 'Botev'; $2n = 4x = 36$ (Fig. 2f)
 Bulgaria, Stara Planina Mts, Kamenlivica on the southern slope of Mt. Botev, 1200 m a.s.l., eroded, siliceous rocks along touristic road from Panicite to the chalet Raj.

A new, undescribed species of presumably hybrid origin between *H. olympicum* Boiss. and *H. sparsum* Friv.

Hieracium sp. 'Ljuboten'; $2n = 3x = 27$ (Fig. 2g)
 Macedonia, Šarplanina Mts, Mt. Ljuboten, 2350 m a.s.l., calcareous rock crevices.

A new, undescribed species of presumably hybrid origin between *H. naegelianum* Panc. and *H. scardicum* Bornm. & Zahn. The hybrid grows together with diploid *H. naegelianum* (Szelag and Ilnicki, 2011) and tetraploid *H. scardicum* (Ilnicki and Szelag, 2011).

Hieracium sp. 'Raušor'; $2n = 4x = 36$ (Fig. 2h)
 Romania, Southern Carpathians, Retezat Mts, above Raušor ski centre, 1520 m a.s.l., grassy slope on *Picea abies* forest margin.

A new, undescribed species of presumably hybrid origin between sexual *H. transylvanicum* Heuff. and sexual *H. umbellatum* L. An emasculation experiment in the garden-cultivated plants shows that the new species reproduces apomictically. Detailed information on the experimental method followed here is given in Szelag (2004).

The diploid chromosome number was stated in the artificial F1 hybrid received in experimental crosses between both diploid, parental species (Mráz and Paule, 2006).

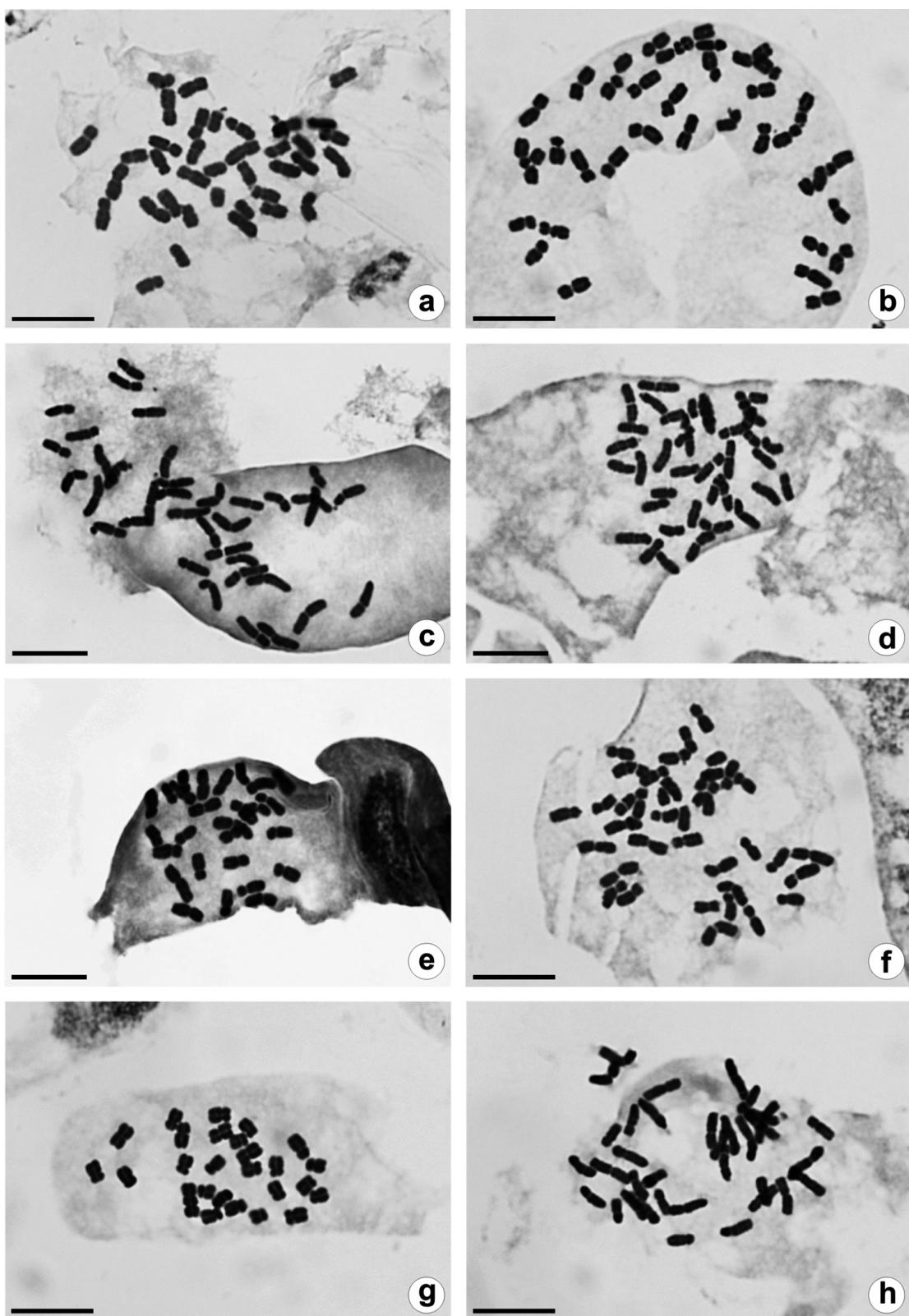
The three above-enumerated undescribed species are the subject of ongoing studies to be presented separately (Szelag, in prep.).

AUTHORS' CONTRIBUTION

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

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Figs. 2. Metaphase plates of: (a) *H. nivimontis* $2n = 36$. (b) *H. schustleri* $2n = 36$. (c) *H. sudetotubulosum* $2n = 36$. (d) *H. vagneri* $2n = 36$. (e) *H. vulgatum* $2n = 27$. (f) *Hieracium* sp. 'Botev'; $2n = 36$. (g) *Hieracium* sp. 'Ljuboten', $2n = 27$. (h) *Hieracium* sp. 'Raušor', $2n = 36$. Bar = $10 \mu\text{m}$.

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REFERENCES

- CHRTEK J. 1994. Chromosome numbers in selected *Hieracium* species in the Krkonoše Mts. (the West Sudeten). *Folia Geobotanica et Phytotaxonomica* 29: 91–100.
- CHRTEK J. 1996. Chromosome number in selected species of *Hieracium* (Compositae) in the Sudeten Mts. and the Western and Ukrainian Eastern Carpathians. *Fragmenta Floristica et Geobotanica* 41: 783–790.
- CHRTEK J, MRÁZ P, AND SEVERA M. 2004. Chromosome numbers in selected species of *Hieracium* s. str. (*Hieracium* subg. *Hieracium*) in the Western Carpathians. *Preslia* 76: 119–139.
- DMITRIEVA SA. 1987. Kariologicheskaja kharakteristika nekotorykh predstaviteley sem. slozhnovenetnykh (Asteraceae Dumort.) flory Belorussii. *Botanika (Minsk)* 28: 23–33.
- GENTSCHOFF G. 1938. Zytologische und embryologische Studien über einige *Hieracium*-Arten. *Planta* 27: 165–195.
- ILNICKI T, HASTEROK R, AND SZELAG Z. 2010. Cytogenetic analysis of *Hieracium transylvanicum* (Asteraceae). *Caryologia* 63: 192–196.
- ILNICKI T, AND SZELAG Z. 2011. Chromosome numbers in *Hieracium* and *Pilosella* (Asteraceae) from Central and Southeastern Europe. *Acta Biologica Cracoviensia Series Botanica* 53/1: 102–110.
- MARCINIUK P, MUSIAL K, JOACHIMIAK AJ, MARCINIUK J, OKLEJEWICZ K, AND WOLANIN M. 2012. *Taraxacum zajacii* (Asteraceae), a new species from Poland. *Annales Botanici Fennici* 49: 387–390.
- MORTON JK. 1974. Chromosome numbers of British plants, 3. *Watsonia* 10: 169.
- MRÁZ P, and PAULE J. 2006. Experimental hybridization in the genus *Hieracium* s. str.: crosses between diploid taxa. *Preslia* 78: 1–26.
- NIKETIĆ M, BAREKA P, AND KAMARI G. 2003. Karyosystematic study of selected *Hieracium* taxa (Compositae) from Mt Durmitor (Montenegro). *Botanika Chronica* 16: 23–45.
- PULKINA SV, AND TUPITSYNA NN. 2000. Poliploidnye kompleksy v rode *Hieracium* (Asteraceae). *Turczaninowia* 3: 79–81.
- SELL P, AND MURRELL G. 2006. *Flora of Great Britain and Ireland. Volume 4. Campanulaceae – Asteraceae.* Cambridge University Press, Cambridge.
- SZELAG Z. 2002. Hieracia balcanica II. Taxonomic and nomenclatural notes on *Hieracium calophyllum* and its relatives (Asteraceae). *Feddes Repertorium* 113: 503–510.
- SZELAG Z. 2004. Taxonomic and nomenclatural notes on *Hieracium* sect. *Cernua* (Asteraceae) in the Alps. *Polish Botanical Journal* 49: 111–115.
- SZELAG Z. 2014. *Hieracium sudetotubulosum* (Asteraceae), a new name for the illegitimate *H. tubulosum* (Tausch) Tausch. *Phytotaxa* 156(4): 250.
- SZELAG Z, ILNICKI T, NIKETIĆ M, AND TOMOVIĆ G. 2007. Diploid chromosome numbers in five *Hieracium* species from Serbia and Montenegro. *Acta Biologica Cracoviensia Series Botanica* 49(1): 119–121.
- SZELAG Z, AND ILNICKI T. 2011. Diploid chromosome numbers in *Hieracium* and *Pilosella* (Asteraceae) from Macedonia and Montenegro. *Acta Biologica Cracoviensia Series Botanica* 53(2): 124–126.
- SZELAG Z, AND WÓJCIK G. 2014. *Hieracium sudetotubulosum* (Asteraceae) rediscovered outside the Karkonosze Mts. *Polish Botanical Journal* 59: 117–119.
- TENNANT DJ, AND RICH TCG. 2008. *British alpine hawkweeds. A monograph of British Hieracium section Alpina.* Botanical Society of the British Isles, London.
- ZAHN KH. 1938. *Hieracium* L. In: Graebner P. fil. (ed.), *Synopsis der Mitteleuropäischen Flora.* 12(3): 1–708 Borntraeger, Berlin.