



## Report on the chromosome numbers of four *Carex* taxa in Korea (Cyperaceae)

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**ABSTRACT:** We report the meiotic chromosome numbers of four *Carex* taxa from Korean populations. Three are the first reports made on taxa from Korean populations: *Carex appendiculata* (Trautv. & C. A. Mey.) Kük. ( $n = 27_{II}$ ), *C. fernaldiana* H. Lév. & Vaniot ( $n = 33_{II}$ ), and *C. metallica* H. Lév. ( $n = 15_{II}$ ). Reports on the other species expand the range of variation in the chromosome number within a taxon, *C. miyabei* Franch. ( $n = 43_{II}, 44_{II}, 45_{II}$ ). *Carex* L. (Cyperaceae) consists of more than 2,000 species worldwide and is the most species-rich genus in Korea. The species diversity in the genus has been hypothesized to be associated with the chromosome variation, but chromosome information pertaining to Korean *Carex* taxa is not well known. This report updates the chromosome number inventory on Korean *Carex* to 24 out of 180 taxa.

**Keywords:** *Carex*, chromosome number, Cyperaceae

Chromosome information has long been an interesting research topic to numerous natural scientists. Chromosome contains hereditary genomic information allowing us to understand functional, structural, genetic, and evolutionary features of organisms. Chromosome number information detects major genomic events such as aneuploidy and polyploidy and has been consistently investigated (Guerra, 2008; Rice et al., 2015). In plant systematics, cytotaxonomic, phylogenetic and speciation inferences of chromosome numbers have been appreciated throughout the vascular plant lineages (de Azkue and Martínez, 1990; Chung and Kim, 1997; McArthur and Sanderson, 1999; Windham and Yatskievych, 2003; Peruzzi et al., 2009; Rockinger et al., 2016).

In *Carex* L. diversity research, chromosomes have provided critical information in various levels of taxonomic and phylogenetic groups, dealing with taxonomic delimitation, gene flow, lineage divergence, and evolution rate related questions (Tanaka, 1948; Davies, 1956; Rothrock and Reznicek, 1996; Hoshino, 1981; Rothrock et al., 2009; Hipp et al., 2010; Chung et al., 2012; Escudero et al., 2012). The genus is the most species-rich group in the temperate zones with more than 2,000 species worldwide (Global *Carex* Group, 2015). As a holocentric chromosome group, chromosome number increase

and/or decrease without DNA duplication and/or deletion events (agmatoploidy and symploidy) (Luceño and Guerra, 1996; Hipp et al., 2013) might cause relatively rapid speciation (Hipp et al., 2010; Chung et al., 2012; Escudero et al., 2012). Although chromosome information has been considered critical features for *Carex* diversity, chromosome numbers of about only 20 taxa out of 180 Korean taxa have been documented (Kim, 2006; Lee and Kim, 2008; Chung et al., 2013; Chung et al., 2016; Chung et al., 2017; Chung et al., 2018; Chung and Im, 2018).

In this study, we report meiotic chromosome numbers for four *Carex* species from Korean populations and discuss their taxonomic and cytological significances: *C. miyabei* Franch., *C. metallica* H. Lév., *C. fernaldiana* H. Lév. & Vaniot, and *C. appendiculata* (Trautv. & C. A. Mey.) Kük.

### Materials and Methods

Spikes with unopened staminate flowers were fixed in March and/or April, and voucher specimens with fully mature perigynia were collected in May and/or June, either 2018 or 2019. The methods for chromosome observation were applied as described in Rothrock and Reznicek (1996) and Chung et

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**Table 1.** *Carex* taxa investigated with voucher specimens and chromosome numbers.

Taxon (locality, voucher specimen)		Chromosome numbers counted, $2n$ ( $n$ )	Previous counts, $2n$
Sect. <i>Carex</i>	<i>Carex miyabei</i> Franch. Songdeok-ri, Goesan-gun, Chungcheongbuk-do, South Korea ( <i>Chung</i> 6077, 7 Jun 2019, KH)	86, 88, 90 ( $n = 43_{II}, 44_{II}, 45_{II}$ )	90 (Tanaka, 1948) 84 (Chung et al., 2018) 66, 72 (Chung and Im, 2018)
Sect. <i>Hymenochlaenae</i>	<i>Carex metallica</i> H. Lév. Yongsu-ri, Hangeong-myeon, Jeju-si, South Korea ( <i>Chung</i> 5150, 1 Jun 2018, KH)	30 ( $n = 15_{II}$ )	30 (Hoshino et al., 2011)
Sect. <i>Mitratae</i>	<i>Carex fernaldiana</i> H. Lév. & Vaniot Surisan Mt., Anyang-si, Gyeonggi-do, South Korea ( <i>Chung</i> 6048-I, 20 May 2019, KH)	66 ( $n = 33_{II}$ )	67 (Tanaka, 1948) 66–74 (Hoshino et al., 2011)
Sect. <i>Phacocystis</i>	<i>Carex appendiculata</i> (Trautv. & C. A. Mey.) Kük. Yupyeong-ri, Nam-myeon, Jeongseon-gun, Gangwon-do, South Korea ( <i>Chung</i> 6070, 19 May 2019, KH)	54 ( $n = 27_{II}$ )	c.80 (Krogulevich, 1976) 76 (Zhukova and Petrovsky, 1976)

al. (2016). Fixed anthers in a fixing mixture of methanol, chloroform, and propionic acid (6:3:2) were squashed in 1% acetic-orcein and then observed at 1,000 $\times$  magnification (Nikon Eclipse 50i, Nikon, Tokyo, Japan). To have solid meiotic chromosome numbers, each figure observed was drawn and photographed. In addition, variations in meiotic chromosome numbers ( $n$ ) were determined after observations of at least three pollen mother cells per sample. All voucher specimens were identified following Hoshino et al. (2011) and Park et al. (2016), and deposited at the Korea National Arboretum Herbarium (KH) (Table 1).

## Results and Discussion

Observed meiotic chromosome numbers are presented with previous records in Table 1, and representative meiotic chromosome figures are given in Fig. 1. All chromosomes are very short, less than 1  $\mu$ m long, and distinct primary constriction cannot be pointed, which is one of characteristics of holocentric chromosomes (Hipp et al., 2013; Cuacos et al., 2015). Furthermore, bivalents are only observed in all the meiotic chromosomes in the present study.

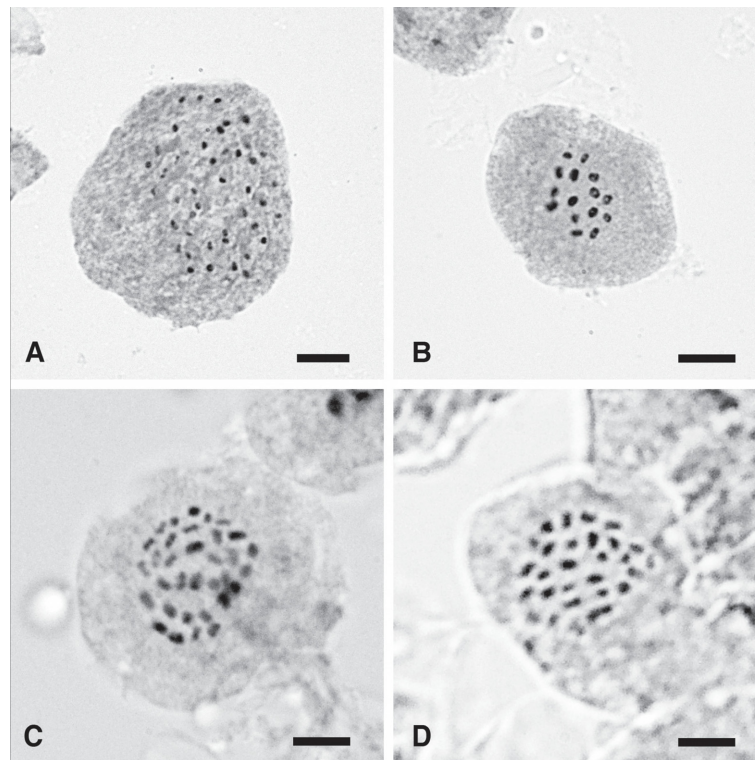
### ***Carex miyabei* ( $n = 43_{II}, 44_{II}, 45_{II}$ ) (Fig. 1A) – Sect. *Carex***

The meiotic chromosome numbers of the species vary,  $n = 43_{II}, 44_{II}, 45_{II}$ . The counts are incongruent with previous reports from Korean populations,  $2n = 66, 72$  (Chung and Im, 2018),  $2n = 84$  (Chung et al., 2018); and a Japanese population,  $2n = 90$  (Tanaka, 1948). The species has been considered as a Japanese endemic, but recently distributions of the species in Japan and Korea have been known (Im et al., 2008; Hoshino

et al., 2011; Park et al., 2016). Although *C. miyabei* is distinctly characterized pubescent perigynia with long beaks, and pistillate scales with aristate apices, the species has been often confused with *C. glabrescens* (Kük) Ohwi for similarity on female spike numbers (more than two), and sharing ecological habitats (Im et al., 2008). These species occur throughout South Korea, but the latter grows mainly middle parts of South Korea (Park et al., 2016). Because both favor sunny, sandy places along rivers, it is not hard to find populations of the two taxa close each other (Im et al., 2008; Park et al., 2016). Variations in morphological characters such as perigynium shapes and surface features have made it hard to distinguish them from each other. Chromosome numbers of *C. glabrescens* have not been reported. As proposed in the previous studies (Chung and Im, 2018; Chung et al., 2018), taxonomic and cytological research should be conducted to understand the two morphologically challenging taxa in the section with taxon sampling covering the distribution areas of the taxa.

### ***Carex metallica* ( $n = 30_{II}$ ) (Fig. 1B) – Sect. *Hymenochlaenae***

The meiotic chromosomes of *C. metallica* from a Korean population is observed for the first time,  $n = 30_{II}$ , which is identical to a previous report from Japan (Hoshino et al., 2011). The species only occurs in Jeju Island, South Korea and is mainly distributed in southern parts of China and Japan (Park et al., 2016; Chen and Zhang, 2018; Masaki, 2018). Dai et al. (2010) reported distributions of the species in Fujian (China) and Taiwan. The *Hymenochlaenae* comprised of about 60 species in Africa, Asia, and Central and North America, and Europe, and many Asian species exhibit narrow distribution patterns (Dai et al., 2010). Thirteen taxa out of fifteen Chinese



**Fig. 1.** Photomicrographs of *Carex* meiotic chromosomes. **A.** *C. miyabei* ( $n = 43_{II}$ , Chung 6077). **B.** *C. metallica* ( $n = 15_{II}$ , Chung 5150). **C.** *C. fernaldiana* ( $n = 33_{II}$ , Chung 6048-1). **D.** *C. appendiculata* ( $n = 27_{II}$ , Chung 6070). Scale bars = 5  $\mu$ m.

species in the section are endemic to China, and *C. hondoensis* Ohwi occurs only in Japan (Dai et al., 2010; Hoshino et al., 2011).

#### ***Carex fernaldiana* ( $n = 33_{II}$ ) (Fig. 1C) – Sect. *Mitratae***

Chromosome number of  $n = 33_{II}$  for *C. fernaldiana* is identified, which is the first report on the taxon from a Korean population. In Japanese populations, the species exhibits a range of chromosome number variations,  $2n = 66-74$  (Tanaka, 1948; Hoshino et al., 2011). Although the species is distinguished from closely related taxa such as *C. stenostachys* Franch. et Sav. and *C. polyschoena* by long rhizomes and glabrous perigynia (Park et al., 2016; Nam, 2017), *C. fernaldiana* is considered as a synonym of *C. pisiformis* Bott (Govaerts, 2019). Govaerts (2019) lists homotypic names of *C. fernaldiana*: *C. sachalinensis* var. *fernalidiana* (H. Lév. & Vaniot) T. Koyama, *C. pisiformis* var. *fernalidiana* (H. Lév. & Vaniot) T. Koyama, and *C. sachalinensis* subsp. *fernalidiana* (H. Lév. & Vaniot) T. Koyama. Nomenclatural synonyms need to be reevaluated. The evaluation should be conducted along with examinations on taxonomic synonyms covering the distribution areas of *C. fernaldiana* and *C. pisiformis* complex.

#### ***Carex appendiculata* ( $n = 27_{II}$ ) (Fig. 1D) - Sect. *Phacocystis***

For the first time, chromosome number of *C. appendiculata* from a Korean population is revealed,  $n = 27_{II}$ . Previously,  $2n = 76$  and c. 80 were reported from Russia (Krogulevich, 1976; Zhukova and Petrovsky, 1976). The species is mainly distributed in northern areas of South Korea and is characterized with 1–2 terminal female spikes and broad perigynia (Park et al., 2016). Although taxonomic status and delimitation are not congruent among distributed countries, the species occurs in China (north east), Japan (north), Russia (east), and Korea (north) (Dai et al., 2010; Hoshino et al., 2011, Ji, 2013; Park et al., 2016; Chen and Zhang, 2018; Masaki, 2018).

Counts on three taxa are the first reports from Korean populations: *Carex appendiculata* (Trautv. & C. A. Mey.) Kük. ( $n = 27_{II}$ ), *C. fernaldiana* H. Lév. & Vaniot ( $n = 33_{II}$ ), and *C. metallica* H.Lév. ( $n = 15_{II}$ ). The reports on the other species increase a range of variation in chromosome number within a taxon: *C. miyabei* Franch. ( $n = 43_{II}$ ,  $44_{II}$ ,  $45_{II}$ ). The counts confirm that chromosome number variations within and/or among taxa resulted from aneuploidy/agmatoploidy, and connote high species diversity in *Carex*. The report updates

chromosome number inventory on Korean *Carex* to 24 taxa out of 180 taxa. Accumulating chromosome number data in *Carex* contributes to understanding species diversity of the genus.

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## Conflict of Interest

The authors declare that there are no conflicts of interests.

## Literature Cited

- de Azkue, D. and A. Martínez. 1990. Chromosome number of the *Oxalis tuberosa* alliance (Oxalidaceae). *Plant Systematics and Evolution* 169: 25–29.
- Chen, W. and S. Zhang. 2018. *Species Catalogue of China*. Vol. 1. Plants. Spermatophytes (II). Angiosperms (Arecaceae-Poaceae). Science Press, Beijing. Pp. 50, 72. (in Chinese)
- Chung, G. Y. and Y. S. Kim. 1997. Study on the somatic chromosome numbers of Korean *Aster* L. and its allied taxa. *Korean Journal of Plant Resources* 10: 292–299.
- Chung, K.-S. and H.-T. Im. 2018. Meiotic chromosome numbers of five *Carex* taxa in Korea (Cyperaceae). *Korean Journal of Plant Taxonomy* 48: 201–205.
- Chung, K.-S., A. L. Hipp and E. H. Roalson. 2012. Chromosome number evolves independently of genome size in a clade with nonlocalized centromeres (*Carex*: Cyperaceae). *Evolution* 66: 2708–2722.
- Chung, K.-S., T. Hoshino, T. Masaki and H.-T. Im. 2017. Cytological investigations on eight *Carex* species in Korea (Cyperaceae). *Cytologia* 82: 329–334.
- Chung, K.-S., T. Hoshino, T. Masaki, H.-T. Im and S.-J. Ji. 2018. Chromosome counts of six Korean *Carex* species (Cyperaceae). *Cytologia* 83: 229–233.
- Chung, K.-S., T. Hoshino, T. Masaki, J. C. Yang and H.-T. Im. 2016. Cytological studies on seven species of Korean *Carex* (Cyperaceae). *Cytologia* 81: 143–147.
- Chung, K.-S., J. C. Yang and Y.-M. Lee. 2013. Chromosome numbers of *Carex* section *Siderostictae* from Korea populations (Cyperaceae). *Korean Journal of Plant Taxonomy* 43: 22–26.
- Cuacos, M., F. C. H. Franklin and S. Heckmann. 2015. Atypical centromeres in plants: what they can tell us. *Frontiers in Plant Science* 6: 913.
- Dai, L., S. Liang, S. Zhang, Y. Tang, T. Koyama and G. C. Tucker. 2010. *Carex* Linneaus. In *Flora of China: Acoraceae through Cyperaceae*. Zhengyi, W., P. H. Raven and H. Deyuan (eds.), Missouri Botanical Garden Press, St. Louis, MO. Pp. 285–461.
- Davies, E. W. 1956. Cytology, evolution and origin of the aneuploid series in the genus *Carex*. *Hereditas* 42: 349–365.
- Escudero, M., A. L. Hipp, M. J. Waterway and L. M. Valente. 2012. Diversification rates and chromosome evolution in the most diverse angiosperm genus of the temperate zone (*Carex*, Cyperaceae). *Molecular Phylogenetics and Evolution* 63: 650–655.
- Global *Carex* Group. 2015. Making *Carex* monophyletic (Cyperaceae, tribe Careiceae): a new broader circumscription. *Botanical Journal of the Linnean Society* 179: 1–42.
- Govaerts, R. 2019. World Checklist of Cyperaceae. Facilitated by the Royal Botanic Gardens, Kew. Published on the Internet. Retrieved Sep 23, 2019, available from <http://wccsp.science.kew.org/>.
- Guerra, M. 2008. Chromosome numbers in plant cytotaxonomy: concepts and implications. *Cytogenetic and Genome Research* 120: 339–350.
- Hipp, A. L., M. Escudero and K.-S. Chung. 2013. Holocentric chromosomes. In *Brenner's Encyclopedia of Genetics*. 2nd ed. Vol. 3. Maloy, S. and K. Hughes (eds.), Elsevier, New York. Pp. 499–501.
- Hipp, A. L., P. E. Rothrock, R. Whitkus and J. A. Weber. 2010. Chromosomes tell half of the story: the correlation between karyotype rearrangements and genetic diversity in sedges, a group with holocentric chromosomes. *Molecular Ecology* 19: 3124–3138.
- Hoshino, T. 1981. Karyomorphological and cytogenetical studies on aneuploidy in *Carex*. *Journal of Science of the Hiroshima University, Division 2, Series B* 17: 155–238.
- Hoshino, T., T. Masaki and M. Nishimoto. 2011. *Illustrated Sedges of Japan*. Heibonsha Ltd., Publishers, Tokyo. Pp. 324, 326, 340, 486, 512, 778.
- Im, H.-T., K.-S. Kim and B.-U. Oh. 2008. *Carex miyabei* Franchet. (Cyperaceae) and its distribution in Korea. *Korean Journal of Plant Taxonomy* 38: 539–545. (in Korean)
- Ji, S. J. 2013. A systematic study of the genus *Carex* sect. *Phacocystis* (Cyperaceae) in Korea. PhD dissertation, Chungbuk National University, Cheongju, 197 pp. (in Korean)
- Kim, S.-Y. 2006. Establishment of chromosome D/B and molecular cytogenetic analysis of Korean native plants. PhD disser-

- tation, Chungnam National University, Cheongju, 169 pp. (in Korean)
- Krogulevich, R. E. 1976. Rol poliploidii v genesise flory Putorana. In 1976 Flora Putorana: Materiali k Poznaniiu Osobnostei Sostava i Genezisia Gornykh Subarkticheskikh Flor Sibir. Novosibirsk. Malyshev, L. I. (ed.), Nauka, Novosibirsk. Pp. 217–235.
- Lee, J. and S. Y. Kim. 2008. Chromosomes of Endemic Plants in Korea 2008. Korea Research Institute of Bioscience and Biotechnology, Daejeon, 75 pp.
- Luceño, M. and M. Guerra. 1996. Numerical variations in species exhibiting holocentric chromosomes: a nomenclatural proposal. *Caryologia* 49: 301–309.
- Masaki, T. 2018. Distribution Maps of *Carex* (Cyperaceae) in Japan. The Japanese Society of Cyperology, Okayama. Pp. 426, 427, 451, 724, 754. (in Japanese)
- McArthur, E. D. and S. C. Sanderson. 1999. Cytogeography and chromosome evolution of subgenus *Tridentatae* of *Artemisia* (Asteraceae). *American Journal of Botany* 86: 1754–1775.
- Nam, G.-H. 2017. A systematic study of the genus *Carex* sect. *Mitratae* (Cyperaceae) in Korea. PhD dissertation, Andong National University, Andong. Pp. 144–145. (in Korean)
- Park, S.-H., Y.-M. Lee, H.-J. Kim, J.-C. Yang, C.-S. Jang, K.-H. Lee, J.-S. Lee, J.-S. Han, H.-J. Kim, K.-S. Jeong, D.-C. Son, D.-H. Lee, M.-J. Joo, E.-M. Sun, C.-H. Shin, K. Choi, S.-H. Oh, K. S. Chang, S.-Y. Jung and S.-J. Ji. 2016. Illustrated Cyperaceae of Korea. Munyoungsa, Seoul. Pp. 10, 23–38, 96, 180, 182, 220, 292 3, 102, 220, 284, 282, 334.
- Peruzzi, L., I. J. Leitch and K. F. Caparelli. 2009. Chromosome diversity and evolution in Liliaceae. *Annals of Botany* 103: 459–475.
- Rice, A., L. Glick, S. Abadi, M. Einhorn, N. M. Kopelman, A. Salman-Minkov, J. Mayzel, O. Chay and I. Mayrose. 2015. The Chromosome Counts Database (CCDB): a community resource of plant chromosome numbers. *New Phytologist* 206: 19–26.
- Rockinger, A., A. Sousa, F. A. Carvalho and S. S. Renner. 2016. Chromosome number reduction in the sister clade of *Carica papaya* with concomitant genome size doubling. *American Journal of Botany* 103: 1082–1088.
- Rothrock, P. E and A. A. Reznicek. 1996. Documented chromosome numbers 1996:1. Chromosome numbers in *Carex* section *Ovales* (Cyperaceae) from Eastern North America. *Sida* 17: 251–258.
- Rothrock, P. E., A. A. Reznicek and A. L. Hipp. 2009. Taxonomic study of the *Carex tenera* group (Cyperaceae). *Systematic Botany* 34: 297–311.
- Tanaka, N. 1948. The problem of aneuploidy (Chromosome studies in Cyperaceae, with special reference to the problem of aneuploidy). *Biological Contribution in Japan* 4: 1–327. (in Japanese)
- Windham, M. D. and G. Yatskievych. 2003. Chromosome studies of Cheilantheid ferns (Pteridaceae: Cheilanthoideae) from the western United States and Mexico. *American Journal of Botany* 90: 1788–1800.
- Zhukova, P. G. and V. V. Petrovsky. 1976. Chromosome numbers of some Western Chukotka plant species, II. *Botanicheskii Zhurnal (Moscow & Leningrad)* 61: 963–969.