Youji SHIGENOBU*: **Karyomorphological studies on *Gentiana jamesii*, *G. nipponica* and *G. nipponica* var. *robusta***

重信陽二*: リシリリンドウ，ミヤマリンドウおよびイイデリンドウの核形態学的研究

Among the members of *Gentiana*, *G. jamesii* has a wide distribution in the alpine zones of Hokkaido, and *G. nipponica* in the same zones of central and northern Honshu and Hokkaido in Japan (Hara 1948, Ohwi 1978). In contrast, *G. nipponica* var. *robusta* is restricted to Mt. Iide in northern Honshu. Some taxonomical confusions have been reported on the treatment of *G. nipponica* var. *robusta* in connection with the other two taxa (Makino 1903, Takeda 1935, Hara 1947, Ohwi 1965, Ohba 1973). The present paper describes the results of karyotype analysis in the three taxa.

Materials and methods The plants materials investigated were *Gentiana jamesii* Hemsl., *G. nipponica* Maxim. and *G. nipponica* Maxim. var. *robusta* Hara. Foliage leaves and flowers of these plants were quite usual morphologically. The sources of the materials are shown in Tab. 1. *G. jamesii* from Mt. Rishiri was supplied by Mr. Ishinari Ohgiya, Principal of Higashi-rishiri choritsu Oshidomari Junior High School and Mr. Mitsuru Yoshida of the same town. The author expresses his cordial thanks for supply of the material.

These plants were grown in pots in the gardens of the Botanical Institute of Hiroshima University and the Biological Institute, College of Child Development, Kochi Women's University. The preparation and observation were carried out by the same method used in the previous report of *Nymphoides* (Shigenobu & Tanaka 1980). That is, root tips were pretreated with 0.002 mol 8-hydroxyquinoline for 3 hr at about 10°C and fixed in modified Carnoy's solution (99% ethanol: chloroform: glacial acetic acid=2:1:1) for over 24 hr at about 10°C. The fixed root tips were treated with 45% acetic acid for 15 min at about 10°C, hydrolyzed in a mixture of 1N HCl and 45% acetic acid (2:1) for 15 sec at 60°C, stained with 2% aceto-orcein and squashed. Voucher specimens of the studied plants were deposited in the Herbarium of the Botanical Institute of

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Tab. 1. Sources and chromosome numbers of three taxa of *Gentiana*.

Taxa	Sources	No. of plants studied	Chromosome number (2n)
G. jamesii	Mt. Rishiri, Higashi-rishiri, Hokkaido	2	36
G. nipponica	Mt. Iide, Yamato, Fukushima Pref.	6	36
	Unknown, obtained from Chugai-shokubutsuen	1	36
G. nipponica var. *robusta*	Mt. Iide, Yamato, Fukushima Pref.	8	96
		1	97
		1	98

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Observations

1) **Gentiana jamesii** Hemsl. Chromosomes at resting stage were observed as chromomeric granules and several spherical chromocentral blocks in the nucleus. The chromocentral blocks were more rounded than those of *G. nipponica* Maxim. The blocks varied in number from 20 to 25 per nucleus and ranged from approximately 0.3 μm to 0.7 μm in diameter (Fig. 1A). The karyotype at resting chromosomes was considered to belong to the category of the simple chromocenter type proposed by Tanaka (1971).

At mitotic metaphase 2n=36 chromosomes were counted in two samples (Tab. 1, Fig. 1B). The present paper is the first report of the 2n=36 chromosome number for this species. The chromosomes showed gradual variation of length ranging from approximately 3.0 μm to 1.5 μm, and had the centromere situated in the median or submedian position. Minute satellites were found in nine chromosomes (Fig. 1B). They were observed at the distal end of the long arm in seven chromosomes and the short arm in two chromosomes.

2) **Gentiana nipponica** Maxim. Chromosomes at resting stage were observed as many chromomeric granules scattered in the nucleus. Some of the chromomeric granules formed spherical chromocentral blocks with chromatin threads irregularly sticking out from the surface. The blocks varied in number from 35 to 45 per nucleus and ranged from approximately 0.3 μm to 1.0 μm in diameter (Fig. 1C). Thus, the karyotype at resting chromosomes was considered to be of the complex chromocenter type (Tanaka 1971), which was

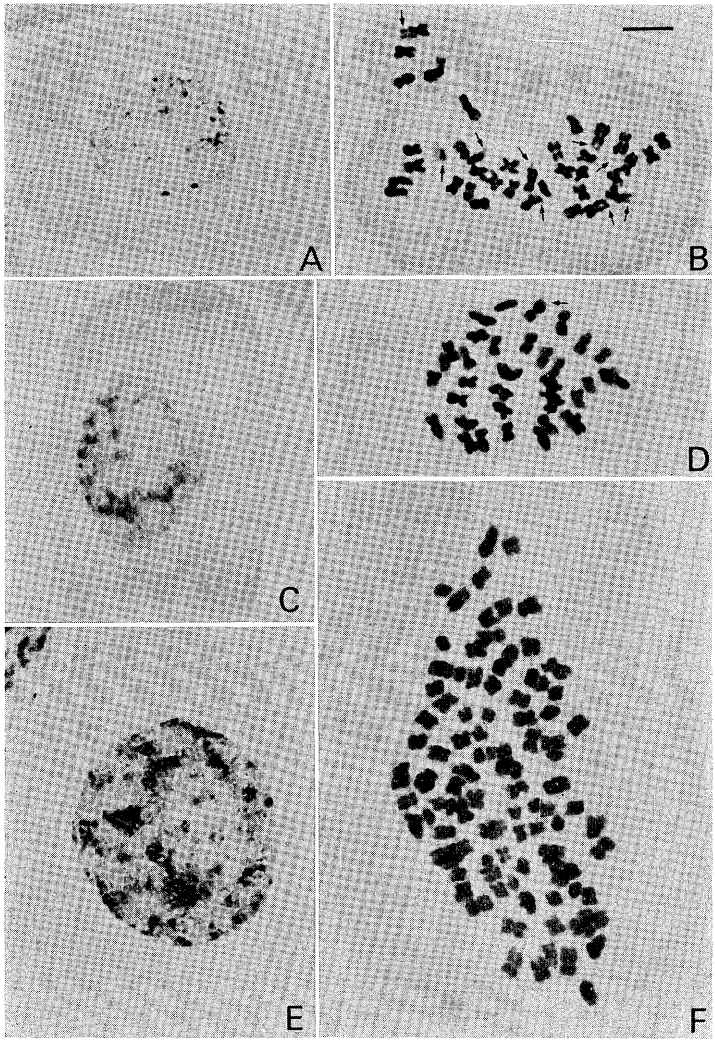

Fig. 1. Photomicrographs of somatic chromosomes in three taxa of *Gentiana*. A, B: *G. jamesii*, 2n=36. C, D: *G. nipponica*, 2n=36. E, F: *G. nipponica* var. *robusta*, 2n=96. A, C and E are at resting stages. Bar indicates 5 μm. Arrows show satellites.

very different from that of the previous species, *G. jamesii*.

At metaphase 2n=36 chromosomes were counted in all of the seven samples examined (Tab. 1). The present paper is the first report on the chromosome number of this taxon. A satellite was found in one chromosome (Fig. 1D). It was minute and located at the distal end of the long arm in the chromosome.

3) **Gentiana nipponica** Maxim. var. **robusta** Hara Chromosomes at resting stage were observed as many chromomeric granules dispersed in the nucleus. Some of the chromomeric granules formed large spherical chromocentral blocks and stained densely. Compared with the typical variety, *G. nipponica* var. *nipponica*, the present variety has many chromocentral blocks, which varied in number from 100 to 130 per nucleus (Fig. 1E). These blocks were mostly round in shape and were approximately 0.3 μm to 1.0 μm in diameter. Thus, the morphology of chromosomes at resting stage was considered to be of the complex chromocenter type (Tanaka 1971), which was also found in the previous taxon, *G. nipponica* var. *nipponica*.

At metaphase 2n=96 chromosomes were counted in eight out of ten plants studied (Fig. 1F), while in other two 2n=97 and 98 chromosomes were counted respectively (Tab. 1). The 2n=97 and 98 chromosome numbers were considered to be aneuploidy in this species. In the present investigation the chromosome number of this taxon was newly counted. The 2n=96 chromosomes showed gradual variation of length ranging from 4 μm to 1.5 μm, and had the centromeres situated in the median or submedian positions. Satellites were not observed.

Discussion The basic numbers of chromosome in *Gentiana* have been reported to be x=5, 6 and 7 (Darlington 1955, Fedorov 1974). On the other hand, chromosome numbers of 36 and 96 along with exceptions of 97 and 98 were found in the three taxa investigated in the present study. Therefore, the basic chromosome number may be considered to be x=6 in these three taxa. Thus, *G. jamesii* and *G. nipponica* can be considered to be hexaploids and *G. nipponica* var. *robusta* is a high polyploid of 16x. Whereas the chromosome at resting stage in *G. jamesii* was of the simple chromocenter type, that of *G. nipponica* and *G. nipponica* var. *robusta* was of the complex chromocenter type. Although many satellites were observed on the chromosomes at metaphase of *G. jamesii*, only one satellite was found on those of *G. nipponica* and no one on those of *G. nipponica* var. *robusta*. With regard to the karyological features men-

tioned above, *G. nipponica* and *G. nipponica* var. *robusta* belong to a polyploid series and have the same chromosome type at resting stage. Although *G. jamesii* has the same chromosome number as in *G. nipponica*, in contrast, The chromosomes at resting stage are quite different in these two species, having different types, the simple and complex chromocenter types.

The results of the present karyological study seem to support the taxonomical treatment of *G. jamesii* as a species distinct from *G. nipponica* var. *robusta* by Hara (1947) and Ohba (1973).

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Literature cited

Darlington, C.D. & Wylie, A.P. 1955. Chromosome atlas of flowering plants. 519 pp. George Allen & Unwin LTD. Fedorov, A. 1974. Chromosome numbers of flowering plants. 926 pp. Otto Koeltz Science Publisher, Western Germany. Hara, H. 1947. Annotationes miscellaneae ad plantas orientalis (II). Journ. Jap. Bot. **21**: 14-21. —— 1948. Enumeratio Spermatophytarum Japonicarum, 1. 300 pp. Iwanami Shoten (Tokyo). Makino, T. 1903. Observations on the flora of Japan. Bot. Mag. Tokyo **17**: 207-217. Ohba, H. 1973. A taxonomic note on *Gentiana nipponica* Maxim. var. *robusta* Hara. Journ. Jap. Bot. **48**: 373. Ohwi, J. 1965. Flora of Japan. Revised Ed. 1560 pp. Shibundo (Tokyo). —— 1978. Flora of Japan. 1584 pp. Shibundo (Tokyo). Shigenobu, Y. & Tanaka, R. 1980. Karyomorphological studies on three species of *Nymphoides* in Japan. Journ. Jap. Bot. **55**: 244-248. Takeda, H. 1935. Nippon no Kozan-shokubutsu (XII). Shokubutsu oyobi Dobutsu **3**: 2205-2213 (in Japanese). Tanaka, R. 1971. Types of resting nuclei in Orchidaceae. Bot. Mag. Tokyo **84**: 118-122.

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日本産リンドウ属2種，1変種の核形態学的研究を行った。*Gentiana jamesii* リシリリンドウと *G. nipponica* ミヤマリンドウはともに 2n=36 で，*G. nipponica* var. *robusta* イイデリンドウは 2n=96 であった。染色体の基本数が x=6 であるとすれば，前者は 6x に，後者は 16x になる。静止期染色体は，リシリリンドウが単純染色中央粒

型で，他の１種，１変種がともに複雑染色中央粒型であった。分裂期染色体は，すべて勾配的染色体長および対称的腕比で類似していたが，付随体染色体の数において相違していた。すなわち，リシリリンドウは９個の，ミヤマリンドウは１個の，イイデリンドウは０個の付随体染色体が観察された。これらの事実から，ミヤマリンドウとイイデリンドウとは染色体数において倍数関係が，核形態学的には近縁な関係があるのに対し，リシリリンドウは大きく離れた位置にあることがわかった。

□原　寛ほか：**Ozegahara, scientific research of the highmoor in central Japan.** 456 pp., 11 pls. 1982，日本学術振興会，東京，￥18,000，取次店 丸善．“尾瀬ヶ原”（1934）の出版から 26 年振りで陣容を新にして本書出版のはこびとなった。第一回では昭和25年度から３カ年計画で調査が進められたが，今回は1977年から同じく３カ年計画で文部省の科研費によって遂行された。この間には自然も人も非常な変化を遂げた。そしてその変化した自然を新しい目で見直すために行われたのである。本書に盛られた41篇の論文名は省略するが，地学，生態関係14，動物18，植物９篇である。参加した研究者は64名で前回よりも10名増えている。前回に比べて新しいことは，生態関係が新しい手法によって活溌に業績を挙げたことである。また前回殆ど手がけられて居らなかった水生植物資料が委しくまとめられたこと，コウモリ類やトリコマイセスなどが新しく加えられたことである。反対に植物のリスト，浮島関係は変化が少ないために省かれた。藻類の資料が一つもないのは残念である。

この４半世紀の間に尾瀬ヶ原の自然は大きく変えられた。否，悪化して仕舞った。原の周辺は乾燥して周囲から尾瀬ザサなどがどっと侵入して来つつあり，帰化植物も目立つ。弥四郎小屋から滝へ至る途にあった池などは跡かたも無くなった。道路には木材や板が敷かれ，原を原形に回復させるために生態学者の努力が行われているが，その成果はどんなものであろうか。以前は弥四郎小屋が只一軒あったあたりには多くの山小屋や売店が並んで居り，山の鼻あたりも同様である。これらから排泄された汚水，汚物は目の届かぬところに山積している。聞くところによれば東京都では南側の山をぶち抜いて，只見川の水を頂戴する計画があるそうである。飛んでもないことで，それほど欲しければ原に訪れた者達に１升瓶１本宛の水を分けてやっても宜しかろうとも思う。

前回活躍された研究者のうち他界された方々は次の如くである。小倉 謙，多田文男，堀川芳雄，水島正美，朝比奈泰彦，山崎文男（炭素同位元素利用法を導入した），久野久，猪熊泰三，倉田 悟の諸氏。なお父子二代に亘って活躍した息子さんの方は朝比奈正二郎，上野俊一。

尾瀬ヶ原の今後の成行は誰にも判らない。出来るならばこのかけがえのない自然に対して末永く弔辞を述べなくてすむことを望む。　　（小林義雄）