Junko NOGUCHI* & Shoichi KAWANO**: **Brief notes on the chromosomes of some Japanese plants** (3)***

野口順子*・河野昭一**: 日本植物の染色体短報 (3)

This is the third report of this series (Kawano, 1961, 1963). In the present study the following six species were karyologically investigated (Table 1). All the materials were collected in the field, transplanted into pots, and cultivated in the Biology Department greenhouse of Toyama University until examined cytologically. All preparations were made by using a modification of the acetic-orcein method of Tjio & Levan (1950) (cf. Kawano, 1965).

Observations

1. **Allium monanthum** Maxim. (Figs. 1 and 2).

Only asexual and female individuals were found in the populations in Akitsu, Higashi-Murayama City, Tokyo. The reproductive method of this species is exceedingly complex (Nagai, 1972).

Previously, both diploid and tetraploid chromosome numbers of 2n=16 and 32 were known from this species (Brat, 1965; Sokolovskaja, 1966). However, all the materials examined in the present study proved to be triploid with 2n=24 somatic chromosomes, which is a new chromosome number to this species.

Of the 24 chromosomes of the complement, fifteen chromosomes, each group consisting of three chromosomes, are large with a constriction at the median or submedian position, six chromosomes are somewhat shorter and possess a subterminal constriction, thus designated as *j*. The remaining three chromosomes are typical telocentric and have a terminal constriction.

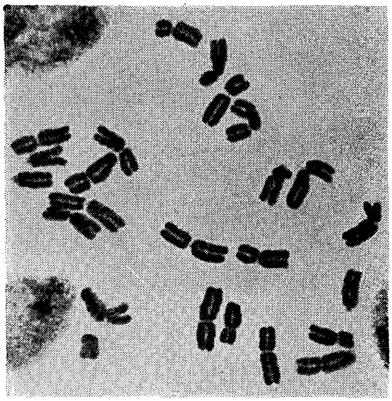
The karyotype of this plant may be expressed as follows:

$$K(2n)=24=3V_1+6V_2+6V_3+6j+3h$$

* Department of Botany, Hiroshima University, Hiroshima 730. 広島大学理学部植物学教室.
** Department of Biology, Toyama University, Toyama 930. 富山大学教養部生物学教室.
*** The second report of this series was published in Journ. Jap. Bot. 38: 47. 1963.

Table 1. A list of the plants studied in the present investigation.

Taxa	Previous report 2n	n	Authors	Present report 2n	Localities
Liliaceae					
Allium monanthum	16	8	Brat, 1965	24	Akitsu, Higashi-Murayama, Tokyo
	32		Sokolovskaja, 1966		
Amana edulis	48		Sato, 1942	48	Akitsu, Higashi-Murayama, Tokyo
		30	Oikawa, 1961		
Cardiocrinum cordatum var. *cordatum*	24		Takamine, 1915	24	Akitsu, Higashi-Murayama, Tokyo
var. *glehni*				24	Ohoiwa, Kamiichi, Toyama
Disporum sessile	16		Hasegawa, 1932, 1933 Washiashi, 1935 Matsuura & Suto, 1935 Therman, 1956 Kayano, 1960, 1961 Arano & Nakamura, 1967 Lee, 1967 Fujishima & Kurita, 1973	16	Tochiori, Toyama
Podophyllaceae					
Diphylleia grayi	12		Matsuura & Suto, 1935 Soeda, 1942 Kuroki, 1967 Noda & Fujimura, 1970	12	Nikko, Tochigi
	16		Lee, 1967		
Adoxaceae					
Adoxa moschatellina	36		Sokolovskaja, 1966 Zhukova, 1967 Johnson & Packer, 1968 Packer, 1968	54	Akitsu, Higashi-Murayama, Tokyo
		18	Lepper, 1970		
	45, 54		Hara, 1956		



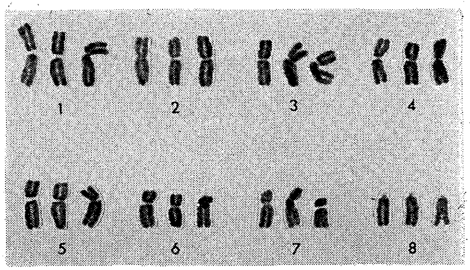


Fig. 2. Somatic chromosome complement of *Allium monanthum*. Reproduced from Fig. 1.

Fig. 1. Somatic chromosomes of *Allium monanthum* from Akitsu, Higashi-Murayama, Tokyo.

Since the sexuality of this species is very complex, further critical karyological analyses on males and other sexual forms are much needed.

2. **Amana edulis** (Miq.) Honda (=*Tulipa edulis* (Miq.) Baker) (Figs. 3 and 4).

Sato (1942) reported that this species possesses 2n=48 chromosomes. Although Oikawa (1961) reported n=30 from this species, this number needs to be reconfirmed.

All the plants investigated in this study revealed to have 2n=48 somatic chromosomes. Of the 24 pairs of somatic chromosomes, six pairs (J_1) are large with a subterminal constriction, and two pairs (J_2)

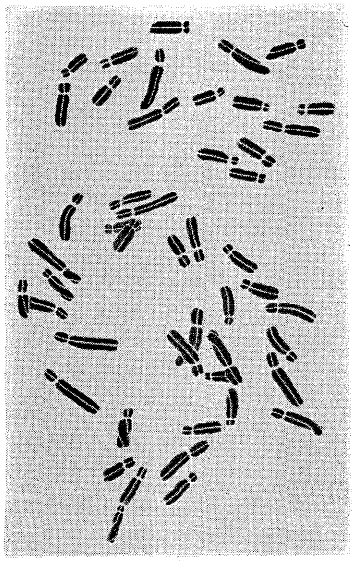


Fig. 3. Somatic chromosomes of *Amana edulis* from Akitsu, Higashi-Murayama, Tokyo.

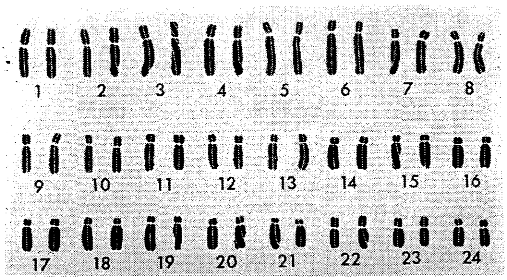


Fig. 4. The somatic chromosome complement of *Amana edulis*. Reproduced from Fig. 3.

are slightly shorter in length with a subterminal constriction. Of the remainings of the complement, five pairs (I_1) are i-shaped with a subterminal constriction, and eight pairs (I_2) are slightly shorter than the I_1-group. The three pairs (Nos. 22, 23, and 24) possess a submedian constriction.

The karyotype formula of this species may roughly be expressed as follows:

$$K(2n)=48=12J_1+4J_2+10I_1+16I_2+6j$$

3a. **Cardiocrinum cordatum** (Thunb.) Makino (Figs. 5 and 6).

The typical variety of this species, *Cardiocrinum cordatum* var. *cordatum*, is characterized by having 3 to 6 flowers per scape, oblong-ovate leaves with cordate sinus, but var. *glehni* is much more robust in general appearance, having broad-ovate leaves and more numerous flowers up to 20 per scape. The geographical distribution of var. *glehni* is extending chiefly on the Japan Sea side of northern and central Honshu, Hokkaido, Sakhalin, and S. Kuriles; on the other hand, var. *cordatum* grows in the undergrowth of warm-temperate forests in south-western Honshu, Shikoku, and Kyushu.

The present species was first examined karyologically by Takamine (1915), who reported it to be n=12 and 2n=24, but it is not certain whether the materials examined by him belong to typical var. *cordatum* or var. *glehni*.

In this investigation, the plants of *C. cordatum* var. *cordatum* collected from the undergrowth of evergreen warm-temperate forests of *Quercus myrsinaefolia* in Akitsu, Higashi-Murayama City, Tokyo, were karyologically examined. All the materials examined turned out to possess 2n=24 somatic chromosomes. Of the 12 somatic chromosome pairs, two large pairs (Nos. 1 and 2) are metacentric, No. 2 of which, however, possesses a clear secondary constriction. The four pairs (Nos. 3, 4, 5, and 6) are subterminal, but two pairs (Nos. 3 and 5) have a secondary constriction at the distal end of the long arm. The three pairs (Nos. 7, 8, and 9) are terminal, and of the remaining three pairs (Nos. 10, 11, and 12), two pairs, Nos. 10 and 11, are also terminal, but No. 11 possesses a secondary constriction at the distal end of the long arm; the pair, No. 12, is acrocentric (cf. Levan, Fregda and Sandberg, 1964).

The karyotype formula of this plant may be expressed as follows:

$$K(2n)=24=2V+2V^{cs}+4j+4j^{cs}+6I+2i+2i^{cs}+2h$$

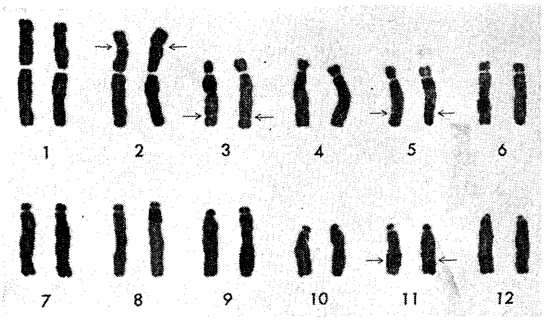


Fig. 6. The somatic chromosome complement of *Cardiocrinum cordatum* var. *cordatum*. Reproduced from Fig. 5.

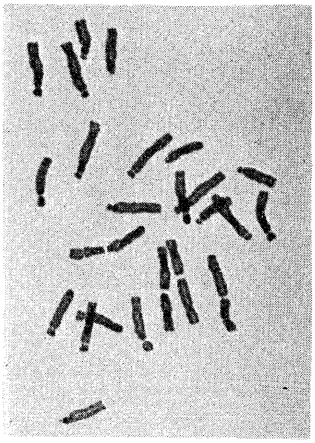


Fig. 5. Somatic chromosomes of *Cardiocrinum cordatum* var. *cordatum* from Akitsu, Higashi-Murayama, Tokyo.

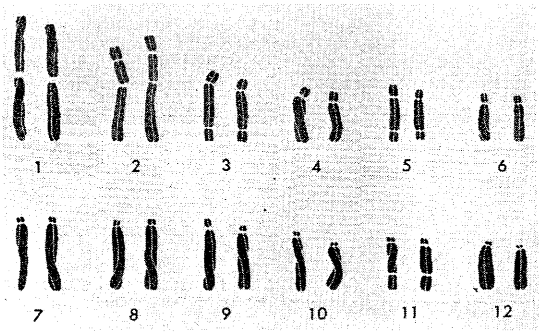


Fig. 7. The somatic chromosome complement of *Cardiocrinum cordatum* var. *glehni* from Ohoiwa, Kamiichi-machi, Toyama.

3b. **Cardiocrinum cordatum** var. **glehni** (Fr. Schm.) Hara (Fig. 7).

The typical form of var. *glehni* collected from Ohoiwa, Kamiichi-machi, Toyama was karyologically examined. The somatic chromosome number of $2n=24$ was counted in all materials examined.

The karyotype of this variety turned out to be exactly the same as that of *C. cordatum* var. *cordatum*, and may be expressed as follows:

$$K(2n)=24=2V+2V^{cs}+4j+4j^{cs}+6I+2i+2i^{cs}+2h$$

Lately, Kurosawa (1966) reported the chromosome number of *Cardiocrinum giganteum* from Darjeeling, 2200 m in Sikkim to be $2n=24$, also.

The karyogram illustrated by her shows that this Himalayan species possesses essentially the same basic karyotype as *C. cordatum*.

4. **Disporum sessile** Don (Figs. 8 and 9).

The present species has so far been studied karyologically by various authors (Hasegawa, 1933; Matsuura & Suto, 1935; Washiashi, 1935; Therman, 1956; Kayano, 1960, 1961; Arano & Nakamura, 1967; Lee, 1967; Fujishima & Kurita, 1973), and all these authors reported this species to be diploid with 2n=16 somatic chromosomes.

A recent extensive cytological study made by Fujishima & Kurita (1973) revealed that the karyotype of this species is basically identical, but considerably differs in number and position of satellites.

All the materials examined in this study had 2n=16 chromosomes. Of the eight pairs of the chromosome complement, two pairs (Nos. 1 and 2) are large, two pairs (Nos. 3 and 4) are somewhat shorter, of which one pair, No. 4, possesses a clear secondary constriction at the distal end of the long arm. The remaining four pairs (Nos. 5, 6, 7, and 8) are small, of which No. 8 is much shorter in length and has a submedian constriction.

The general karyotype composition of the plants examined in the present study is nearly identical with that reported previously (*l. c.*), but is different in the chromosome morphology of the pair, No. 4, in which a clear secondary constriction is observed at the distal end of the long arm. No satellite was observed in all materials examined in this study.

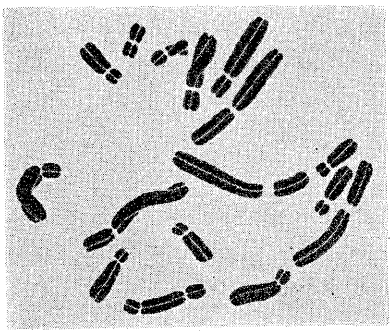


Fig. 8. Somatic chromosomes of *Disporum sessile* from Tochiori, Toyama.

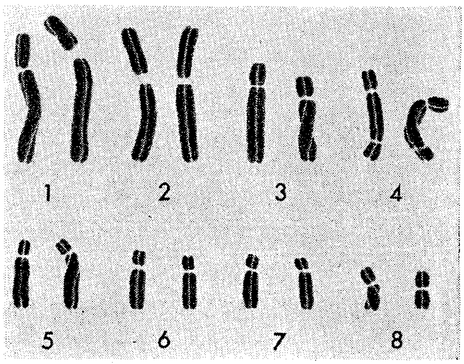


Fig. 9. The somatic chromosome complement of *Disporum sessile*. Reproduced from Fig. 8.

The karyotype formula may be expressed as follows:

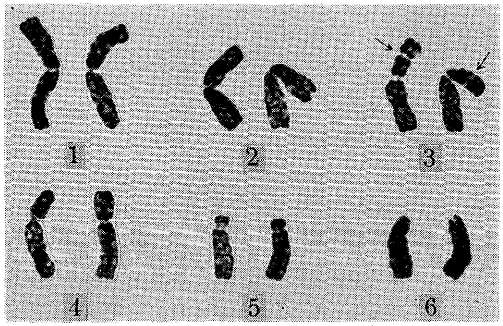
$$K(2n)=16=2L_1+2L_2+2L_3+2L_4^{cs}+6S_1+2S_2$$

(cf. Fujishima & Kurita, 1973)

5. **Diphylleia grayi** Fr. Schm. (Figs. 10 and 11).

The chromosome number of the present species is reported to be 2n=12 (Matsuura & Suto, 1935; Soeda, 1942; Kuroki, 1967; Noda & Fujimura, 1970). Lee (1967) reported the chromosome number of 2n=16 from Korean materials, but further confirmation is needed.

Fig. 10. Somatic chromosomes of *Diphylleia grayi* from Nikko, Tochigi.



Fi.g. 11. The somatic chromosome complement of *Diphylleia grayi*. Reproduced from Fig. 10.

All the materials examined in the present study also proved to possess 2n=12 somatic chromosomes. Of the six pairs of the chromosome complement, two pairs (Nos. 1 and 2) are large and metacentric, one pair, No. 3, is submedian, but has a secondary constriction at the distal end of the short arm (cf. Fig. 11), and one pair, No. 4, is subterminal. Of the remaining two pairs of small chromosomes, one pair, No. 5, has a terminal constriction, but one pair, No. 6, is telocentric.

The karyotype formula of this species may be expressed as follows:

$$K(2n)=12=4V_1+2V_2^{cs}+2J+2j+2h$$

6. **Adoxa moschatellina** L. (Figs. 12 and 13).

This is a northern circumpolar species. The chromosome number of European and North American plants is reported to be 2n=36 (Sokolovskaja, 1966; Zhukova, 1967; Johnson & Packer, 1968; Packer, 1968; Lepper, 1970).

From Japanese plants, two different chromosome numbers, 2n=45 and 54, were reported by Hara (1956). Hara (*l.c.*) regards our Japanese plants to be distinguishable from European plants by stoloniferous and bulbiferous characters, and gave a varietal epithet, i.e., var. *japonica* Hara. As was noted by Hara, the vegetative reproduction is very prominent in our Japanese plants. They usually start to elongate underground stolons in April and subsequently in May and June about 20 to 30 bulbils are formed at the tip of each stolon. However, when the aerial parts become withered in June, stolons are also decayed and each bulbil becomes separate. Bulbils stay in the dormant state until next spring. The entire matter preserved in each bulb is consumed for growth of new aerial organs (cf. Kawano, 1971).

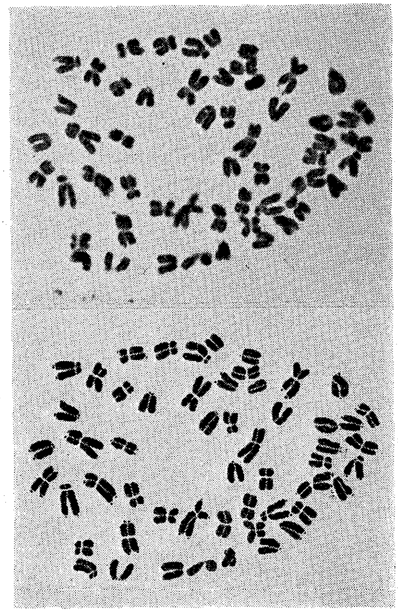


Fig. 12. Somatic chromosomes of *Adoxa moschatellina* from Akitsu, Higashi-Murayama, Tokyo.

All the materials examined in the present study proved to be triploid with 2n=54 somatic chromosomes. It is very evident from the karyotypic

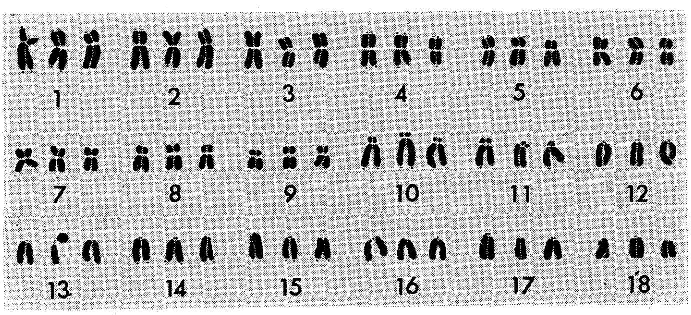


Fig. 13. The somatic chromosome complement of *Adoxa moschatellina*. Reproduced from Fig. 12.

constitution, as illustrated in Fig. 13, that our plants no doubt represent triploid. It is also very charcteristic that numerous satellites are to be observed often on both short and long arms of the chromosomes (cf. Fig. 13). Of the 2n=54 somatic chromosomes, eighteen chromosomes (Nos. 1-6) are metacentric, nine chromosomes (Nos. 7, 8, and 9) have a subterminal constriction, six chromosomes (Nos. 10 and 11) are acrocentric, and the remaining twenty-one chromosomes (Nos. 12-18) are typical telocentric. As to the number, position, and distribution of satellites, much more critical future studies are needed.

The karyotype of this particular material (Fig. 13) may roughly be expressed as follows:

$$K(2n)=54=18V(6V_1+6V_2+6V_3)+9J+6I+21h$$

References

Arano, H. & Nakamura, T. 1967. Cytological studies in family Liliaceae of Japan. I. The karyotype analysis and its karyological considerations in some species of *Polygonatum*, *Disporum*, and *Smilacina*. La Kromosomo **68**: 2205-2214. Brat, S. V. 1965. Genetic systems in *Allium* I. Chromosome variation. Chromosoma **16**: 486-489. Fujishima, H. & Kurita, M. 1973. Variation in number, size and location of satellite of *Disporum sessile* Don. Jap. J. Gen. **48**: 271-278. Hara, H. 1956. Contributions to the study of variations in the Japanese plants closely related to those of Europe or North America. Part 2. J. Fac. Sci., Univ. Tokyo, Sect. III, Bot. **6**: 343-391. Hasegawa, N. 1932. Comparison of chromosome type in *Disporum*. Cytologia **3**: 350-368. —— 1933. Chromosome studies in diploid and triploid forms of *Disporum sessile*. Jap. J. Gen. **9**: 9-14. Johnson, A. W. & Packer, J. G. 1968. Chromosome numbers in the flora of Ogotruk Creek, N. W. Alaska. Bot. Notis. **121**: 403-456. Kawano, S. 1961. Brief notes on the chromosomes of some Japanese plants (1). Jap. J. Bot. **36**: 29-32. —— 1963. —— (2). Jap. J. Bot. **38**: 47-50. —— 1965. Application of pectinase and cellulase in an orcein squash method. Bot. Mag. Tokyo **78**: 36-42. —— 1970. Species problems viewed from productive and reproductive biology. I. J. Coll. Lib. Arts, Toyama Univ. **3**: 181-213. Kayano, H. 1960. Chiasma studies in structural hybrids. III. Reductional and equational separation in *Disporum sessile*. Cytologia **25** : 461–467. —— 1961. Failure of chromosome pairing and unreduced diploid pollen grains in *Disporum sessile*. Chrom. Inf. Serv. **2** : 5–7. Kuroki, Y. 1967. Chromosome study in seven species of Berberidaceae. Mem. Ehime Univ. Sect. II, Ser. B, **5** : 175–181. Kurosawa, S. 1966. Cytological studies on some Eastern Himalayan plant. *In* : The Flora of Eastern Himalaya, Univ. of Tokyo Press, pp. 658–670. Lee, Y. N. 1967. Chromosome numbers of flowering plants in Korea (1). J. Korean Cult. Res. Inst. **11** : 455–478. Lepper, L. 1970. Beiträge zur Chromosomenzahlen-Dokumentation. Wiss. Z. Friedrich-Schiller Univ., Jena, Math.-Naturwiss. Reihe **19** : 369–376. Levan, A., Fregda, K. & Sandberg, A. A. 1964. Nomenclature for centromeric position on chromosomes. Hereditas **52** : 201–220. Matsuura, H. & Suto, T. 1935. Contributions to the idiogram study in phanerogamous plants. I. J. Fac. Sci., Hokkaido Univ. Ser. V, **5** : 33–75. Nagai, Y. 1972. Reproductive biology of *Allium monanthum* Maxim. J. Geobot. **20** : 84–91 (in Japanese). Noda, S. & Fujimura, T. 1970. Karyotypes in root-tip cell and endosperm nucleus of *Diphylleia grayi*. La Kromosomo **79–80** : 2548–2551. Oikawa, K. 1961. The embryosac development in *Amana edulis* (Miq.) Honda. Sci. Rep. Tohoku Univ. Series IV, **27** : 155–158. Packer, J. G. 1968. *In* : IOPB chromosome number report XVII. Taxon **17** : 285–288. Sato, D. 1942. Karyotype alternation and phylogeny in Liliaceae and allied families. I & II. Jap. J. Bot. **12** : 57–161. Soeda, T. 1942. On the chromosomes of *Diphylleia grayi*. Jap. J. Gen. **18** : 47–48 (in Japanese). Sokolovskaja, A. P. 1966. Geograficheskoe rasprostranenie poliploidnykk vidov rasteniy. Vestnik Leningr. Univ. Ser. Biol. **3** : 92–106. Takamine, N. 1915. Über die Prophasen der Kernteilungen von *Cardiocrinum cordatum* Makino. Bot. Mag. Tokyo **29** : 17–23. Therman, E. 1956. Cytotaxonomy of the tribe Polygonatae. Amer. J. Bot. **43** : 134–142. Tjio, J. H. & Levan, A. 1950. The use of oxyquinoline in chromosome analysis. Ann. Estac. Exp. Aula Dei **2** : 21–64. Washiashi, F. 1935. Cytological studies on the influence of low temperature upon the pollen formation in *Disporum sessile*. Jap. J. Gen. **11** : 60–70. Zhukova, P. G. 1967. Chromosome numbers in some species of plants of the north-eastern part of the USSR. II. Bot. Zhur. **52** : 983–987 (in Russian).

* * * *

本報ではユリ科のヒメニラ，アマナ，ウバユリ，オオウバユリ，ホウチャクソウ，ミヤオソウ科のサンカヨウ，レンプクソウ科のレンプクソウの染色体数および核型について報告した。ヒメニラは雌および性型不明の栄養型の個体について調べたが，いずれも3倍体で 2n=24 であった。この種より3倍体が見いだされたのは今回が初めてである。アマナからは 2n=48，ウバユリ，オオウバユリからは 2n=24 の染色体が観察された。ウバユリとオオウバユリでは基本核型は同一であることが見いだされた。ホウチャクソウはすべて 2n=16 で他の報告者の結果と一致する。しかし，核型は第4対 (No. 4) の染色体が2次狭窄を長腕の末端部をもつ点で異なっている。サンカヨウは 2n=12 で既報の結果と一致する。Lee (1967) は朝鮮の植物で 2n=16 を報告しているが，詳細な核型分析の結果が待たれる。レンプクソウはすべて 2n=54 で3倍体であった。各染色体組に数多くの satellite が見られるのが特色である。satellite の正確な数と分布に関してはさらに詳細な研究が必要であろう。

○おしば標本の新らしい貼付法（金井弘夫） Hiroo KANAI: Heat-seal, a new mounting technique of herbarium specimen

先に本誌 47: 120 (1972) において本法の概要を発表しておいたが，その後改良を加え満足すべき結果を得たので報告する。既報の如く本法の骨子はポリエチレンラミネート紙のテープをハンダ鏝を用いて台紙に熔着するというものである。

まずラミネート紙であるが，これは市販品があることを知った。きわめて薄い和紙又は化繊紙の片面にポリエチレンをラミネートしたもので，南国パルプ工業の製品はヒートロンとよばれ，東京加工紙製のものはラミコートという。この種の紙は最中などの和菓子の包紙によく用いられているので，少量ならこれを利用できる。紙質はヒートロンでは PCS 30 g/m² が適当だった。この紙は何枚も重ねると互に滑ってしまって切り難い欠点があり，後述のような商品としてテープを製作する場合には幅 7 mm より細くは切れない。テープを手製するならば 4 mm と 8 mm の2種類を用意するとよい（図 D, E)。次にハンダ鏝であるが 60～80 W のラジオ用がよい。先の太いものは先端を約 30° に斜に削り，面を長方形にヤスリで仕上げる（図 C)。先が細い鏝は先端をたゝきつぶしてから角度をつけて曲げる。先端は約 90° の角度をつけて左

図. 1 説明は本文中. Fig. 1. A. Mountnig instrument. a: Pilot lamp. b: Switch. c: Regulator. d: Mounting iron (Made by Futaba Seisakusho). B, C. Tips of mounting iron. D. Mounting tapes on sale (width 7 mm and 10 mm) (Made by Futaba Seisakusho). E. Mounting tapes made of the wrapper of sweet cake. F. How to mount specimen. G. How to cut tape. H-J. How to cover fragile portion. H: Place polyethylene sheet over the portion and heat seal it with the aid of mounting tape. I: Cut off the excess portion of sheet with the tip of mounting iron. J. Heat seal the tape again firmly with the bottom of iron.