

## Karyomorphological Studies in *Phragmipedium*, Orchidaceae

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### Introduction

The genus *Phragmipedium* is considered one of the most primitive genus of the Orchidaceae as same as *Cypripedium*, *Paphiopedilum* and *Selenipedium*, which are classified into Cypripedioideae (Garay 1960).

We are studying karyomorphologically on Cypripedioideae for the elucidation of the origin of Orchidaceae and of the phylogenetic relationships of this sub-family. This paper is a report dealing with *Phragmipedium* following *Paphiopedilum* (Karasawa 1979).

The reports on the chromosome of *Phragmipedium* are in three species: *P. caudatum*  $2n=32$  (Hoffman 1929, cited in Duncan 1959), *P. longifolium* var. *hartwegii*  $2n=20$  (Brown 1959, cited in Duncan 1959) and *P. boisseranum*  $2n=18$  (Karasawa & Tanaka 1976). However, there, is no report on the detail of the karyomorphological analysis.

In the present investigation the morphology of chromosomes was studied in 14 species and one variety in *Phragmipedium*.

### Acknowledgements

I would like to acknowledge the continuing guidance and encouragement of Professor Dr. Ryuso Tanaka of Hiroshima University. I also wish to thank Dr. Yoshihiko Yonezawa for his helpful advice and Dr. Leslie A. Garay for identification of specimen. My special thanks are also due to Mr. Sigeaki Ichijo, Mr. Hiromichi Miyahara, Mr. Yoshio Udagawa, Mr. Kazuhito Uchida and Mr. Nobuhiro Hata for supplying the materials.

### Materials and Methods

Materials, sources and the number of clones investigated are listed in Table 1. Taxonomical treatment was followed Garay (1979). Observation of somatic chromosomes in the root tip cells was made according to the previous report (Karasawa & Tanaka 1976).

Karyotype analysis was made on the chromosomes at interphase, prophase and metaphase. In interphase, the morphology of the condensed and diffused chromatin was

analysed, and the location of early condensing chromatin in prophase. In the metaphase chromosomes, the length of both long and short arms was measured, and relative length and arm ratio were estimated. The position of centromere was expressed by the terms of median, submedian, subterminal and terminal according to Levan *et al.* (1964).

Table 1. Sources, number of clones and chromosome number of the species of *Phragmipedium* studied

Species	Source	No. of clone	Chromosome number (2n)
Section MICROPETALUM			
<i>schlimii</i>	Colombia	4	30*
× <i>stenophyllum</i> ***		1	40*
Section PLATYPETALUM			
<i>lindleyanum</i>	Venezuela	2	22*
<i>sargentianum</i>	Brazil	4	22*
Section PHRAGMIPEDIUM			
<i>caudatum</i>	Peru, Ecuador	4	28**
<i>wallisii</i>	Ecuador	1	28*
<i>lindenii</i>	Venezuela, Colombia	4	28*
Section HIMANTOPETALUM			
<i>ecuadorensis</i>	Peru	2	22*
<i>pearcei</i>	Peru	3	20*, 21*, 22*
Section LORIFOLIA			
<i>boisseranum</i>	Peru	3	18
<i>vittatum</i>	Brazil	2	18*
<i>hartwegii</i>	Ecuador	2	20
<i>longifolium</i>	Colombia, Panama	2	23*(20+3f)
<i>longifolium</i> var. <i>gracile</i>	(Rands Orchids)***	1	21*(20+1f)
<i>roezlii</i>	Panama	2	22*(20+2f)

\* first time record

\*\* differ from previous record

\*\*\* cultured

### Observations

In the preliminary study, the morphological changes of chromosomes in mitotic cell cycle were observed in root tip cells of *Phragmipedium hartwegii*, 2n=20 (Fig. 1). In the

interphase nuclei (Fig. 1A, B), many, darkly stained chromocenters were observed. These chromocenters occasionally showed aggregations forming several, large heteropycnotic blocks. These nuclei were categorized the complex chromocenter type of Tanaka's classification of resting nuclei (Tanaka, 1971). In early prophase, chromosomes changed to fibrous thread stained almost homogeneously (Fig. 1C), while in middle or late prophase, early condensing segments were observed at the proximal regions of both arms (Fig. 1D, E). These early condensing segments transformed abruptly into late condensing segments

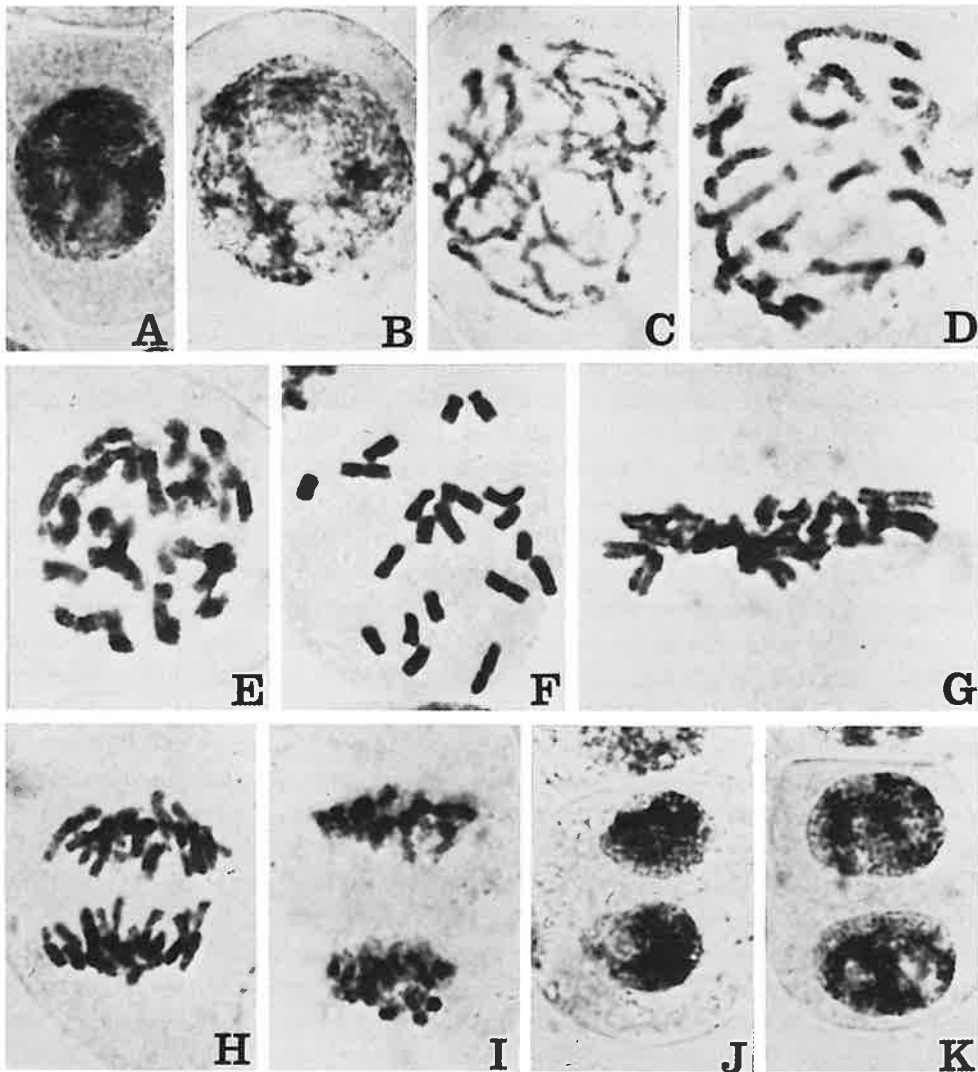


Fig. 1. Chromosomes in mitotic cell cycle in root-tip cells of *Phragmipedium hartwegii*,  $2n=20$ . A and B, interphase. C and D, prophase. E, prometaphase. F and G, metaphase. H, anaphase. I, telophase. J and K, early interphase of daughter cells.  $\times 1600$ .

located at the distal regions. These prophase nuclei were categorized the proximal type on the distribution of the early condensing chromatin (Tanaka 1977).

At metaphase pretreated with 8-hydroxyquinoline,  $2n=20$  chromosomes were counted, and each chromosome was distinguished by their length and centromere position (Fig. 1F).

At anaphase to next interphase, no karyomorphological characteristic was observed (Fig. 1G-K).

Therefore, karyotype analysis was made in the chromosomes at interphase, prophase and metaphase as mentioned in Materials and Methods.

## I. MICROPETALUM

### 1) *Phragmipedium schlimii* (Lindl. & Rchb. f.) Rolfe, $2n=30$

Varidated specimen No. BG-31, BG-32, M-1, I-1.

This species is native to Colombia and has the following distinct morphological characteristics: The dorsal sepal is oval. The petals are broad and not pointed. The upper sepal and petals are pinkish white in color. The natural spread of flower is about 4 cm in width. The pouch is oval and white with deep pinkish stripes. The middle part of rhombate staminode is brownish saffron-yellow in color (Fig. 2A).

The interphase nuclei were similar to those of *P. hartwegii* mentioned above showing the complex chromocenter type (Fig. 2B). Prophase chromosomes were the proximal type (Fig. 2C). At metaphase,  $2n=30$  chromosomes were counted in four clones (Fig. 2D). The measurements of chromosome length and arm ratio are shown in Table 2.

Among the 30 chromosomes the two longest chromosomes (Nos. 1, 2) were distinguished. They were  $5.8\mu\text{m}$  and  $5.2\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 3.5 and 3.0, and the position of centromeres were subterminal and submedian, respectively. Four chromosomes (Nos. 3-6) ranged from  $4.0$  to  $3.6\mu\text{m}$  in length and the position of centromeres of these chromosomes were terminal. The 7th and 8th chromosomes were both  $3.5\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.9, and the position of centromeres were submedian. Two chromosomes (Nos. 9, 10) were both  $3.4\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 11, 12) were both  $3.3\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.5, and the position of centromeres were median. Eight chromosomes (Nos. 13-20) ranged from  $3.3$  to  $2.9\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 21, 22) had the satellites at the distal region of long arms. They were both  $2.9\mu\text{m}$  ( $2.0+0.9\mu\text{m}$ ), and the position of centromeres were terminal. The remaining eight chromosomes (Nos. 23-30) ranged from  $2.9$  to  $2.1\mu\text{m}$  in length, and the position of centromeres were terminal.

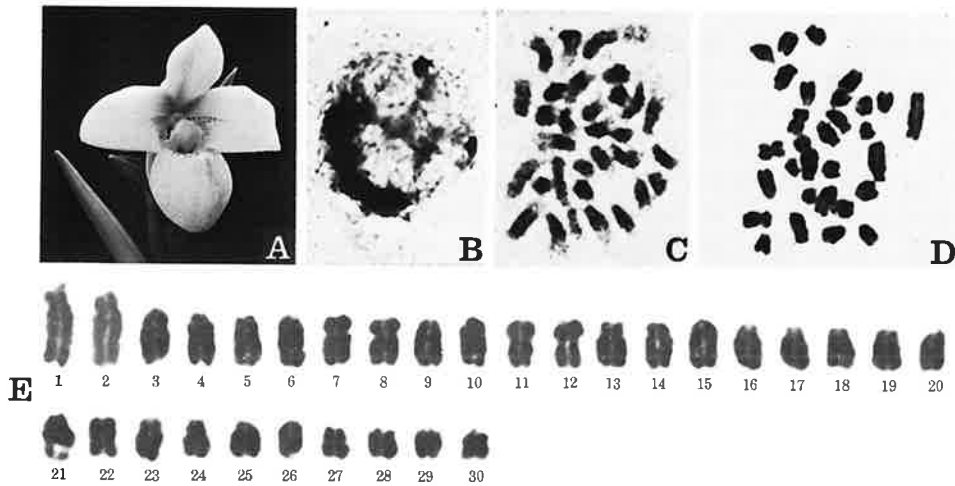


Fig. 2. *Phragmipedium schlimii*,  $2n=30$ . A, flower. B, interphase nucleus. C, prometaphase. D and E, metaphase. A,  $\times 0.7$ . B-D,  $\times 1200$ . E,  $\times 1800$ .

As mentioned above, the  $2n=30$  chromosomes were divided into two quantitative groups showing bimodal karyotype: The one consisted of the two longest metacentric chromosomes and the other consisted of 28 small chromosomes which decreased gradually in size. Two telocentric chromosomes which had satellites were the marker chromosomes for the karyotype of this species.

## 2) *Phragmipedium x stenophyllum*, $2n=40$

Varidated specimen No. 3014.

This species was nearly recorded as the new species in the magazine of the Orchid Digest (42: 232, 1978). In the horticultural world it was introduced as *P. schlimii*, but Garay (1979) considered it *P. x stenophyllum* based on the examination of the flower which showed to be identical with the original specimen of *P. x stenophyllum* in Reichenbach Herbarium (Fig. 3A).

In the present investigation the chromosome number of this species was examined to be  $2n=40$ . The chromosomes at prometaphase and metaphase are shown in Fig. 3B-D. The results of the measurement of chromosome length and arm ratio are shown in Table 3.

Among the 40 chromosomes the longest chromosome (No. 1) was  $6.0\mu\text{m}$  in length. Arm ratio of this chromosome was 3.3, and the position of centromere was subterminal. The 2nd chromosome was  $5.1\mu\text{m}$  in length and had the satellite on its long arm. The short arm was  $2.1\mu\text{m}$  and the long arm was  $3.0\mu\text{m}(2.3+0.7\mu\text{m})$  in length. Arm ratio of this chromosome was 1.4, and the position of centromere was median. The 3rd chromosome was  $4.9\mu\text{m}$  in length. Arm ratio of the chromosome was 2.8, and the position of centro-

mere was submedian. The 4th chromosome was  $4.3\mu\text{m}$  in length. Arm ratio of this chromosome was 1.3, and the position of centromere was median. The 5th chromosome was  $4.1\mu\text{m}$  in length. Arm ratio of this chromosome was 1.3, and the position of centromere was median. The 6th chromosome was  $4.1\mu\text{m}$  in length. Arm ratio of this chromosome was 2.4, and the position of centromere was submedian. The 7th chromosome was  $4.0\mu\text{m}$  in length and had the small constriction near the middle region of the long arm. The position of centromere of this chromosome was terminal. The 8th chromosome was  $4.0\mu\text{m}$  in length. Arm ratio of the chromosome was 2.1, and the position of centromere was submedian. Two chromosomes (Nos. 9, 10) were both  $4.0\mu\text{m}$  in length. The position of centromeres of these chromosomes were terminal. The 11th chromosome was  $4.0\mu\text{m}$  in length. Arm ratio of this chromosome was 1.2, and the position of centromere was median. Two chromosomes (Nos. 12, 13) were both  $3.9\mu\text{m}$  in length. The position of centromeres of these chromosomes were terminal. The 14th chromosome was  $3.6\mu\text{m}$  in length. Arm ratio of this chromosome was 1.4, and the position of centromere was median. The 15th chromosome was  $3.6\mu\text{m}$  in length. Arm ratio of this chromosome was 2.0, and the position of centromere was submedian. The 16th chromosome was  $3.6\mu\text{m}$  in length. The position of centromere of this chromosome was terminal. Two chromosomes (Nos. 17, 18) were both  $3.5\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.9, and the position of centromeres were submedian. Two chromosomes (Nos. 19, 20) were both  $3.4\mu\text{m}$  and  $3.3\mu\text{m}$  in length, respectively. The position of centromeres of these chromosomes were terminal. Two chromosomes (Nos. 21, 22) were both  $3.3\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.5, and the position of centromeres were median. The eight chromosomes (Nos. 23-30) ranged from  $3.0$  to  $2.8\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 31, 32) were both  $2.8\mu\text{m}$  in length,

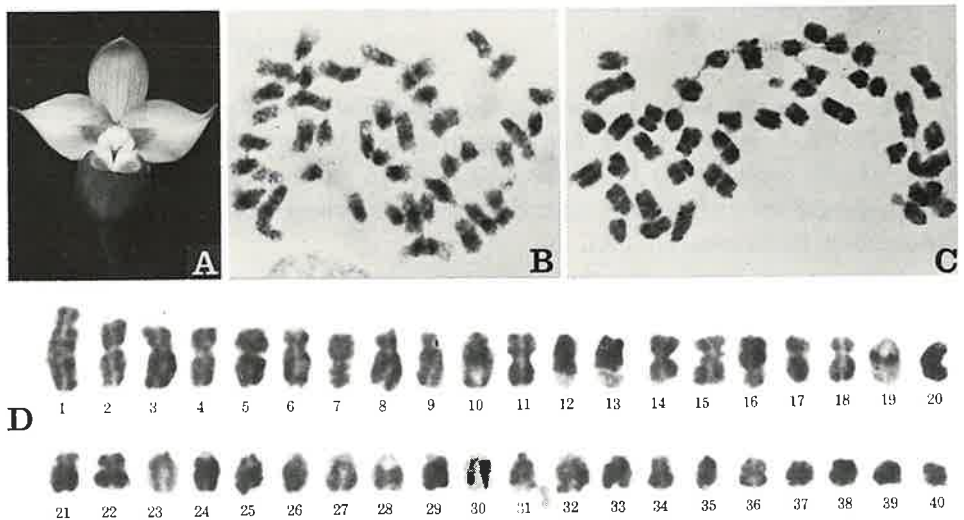


Fig. 3. *Phragmipedium*  $\times$  *stenophyllum*,  $2n=40$ . A, flower. B, prophase. C and D, metaphase. A,  $\times 0.4$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

and had satellites at the distal region of long arms. The position of centromeres of these chromosomes were terminal. The eight chromosomes (Nos. 33-40) ranged from 2.8 to  $2.0\mu\text{m}$  in length and gradually decreased in size. The position of centromeres of these chromosomes were terminal.

According to the morphology of chromosome complement, the  $2n=40$  chromosomes of this species consisted of 14 metacentric chromosomes and 26 telocentric chromosomes. The 16 chromosomes (Nos. 1-16) of them were heterogeneous pairs which did not make the matched chromosomes, while the rest 24 chromosomes (Nos. 17-40) of them were homogeneous pairs which made 12 matched chromosomes.

## II. PLATYPETALUM

### 1) *Phragmipedium lindleyanum* (Schomb. ex Lindl.) Rolfe, $2n=22$

Varidated specimen No. BG-30, I-2

This large-sized species grow in Guyana and in Venezuela. External characteristics of this species are as follows: Leaves are lustrous and leathery, the tip of leaf is pointed and 50-60cm in length. The dorsal sepal is long oval. The petals are beltshape and their ends are round. The staminode is nearly lozenge (Fig. 4A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 4B). Prophase chromosomes were the proximal type (Fig. 4C). At metaphase,  $2n=22$  chromosomes were counted in two clones (Fig. 4D, E). The measurements of chromosome length and arm ratio are shown in Table 4.

Two chromosomes (Nos. 1, 2) which had the small constrictions near the middle of their short arms were both  $5.7\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.3, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) which had the satellites at the distal region of long arms were  $5.3\mu\text{m}$  in length. The long arms of these chromosomes were  $2.9\mu\text{m}$  ( $1.8+1.1\mu\text{m}$ ) and the short arms were  $2.4\mu\text{m}$  in length. Arm ratio of these chromosomes were 1.2, and the position of centromeres were median. Two chromosomes (Nos. 5, 6) were both  $5.2\mu\text{m}$  in length. Arm ratio of these chromosomes were 3.0, and the position of centromeres were submedian. These chromosomes had the small constrictions near the middle region of their long arms. Two chromosomes (Nos. 7, 8) were both  $5.2\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.2, and the position of centromeres were median. Two chromosomes (Nos. 9, 10) were both  $4.2\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.2, and the position of centromeres were submedian. Two chromosomes (Nos. 11, 12) were  $3.4\mu\text{m}$  and  $3.1\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 13, 14) were both  $3.1\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.6, and the position of centromeres were median. The remaining eight chromosomes (Nos. 15-22) ranged from 2.9 to  $2.6\mu\text{m}$  in length. These chromosomes were decreased gradually in size and the position of centromeres were all terminal.

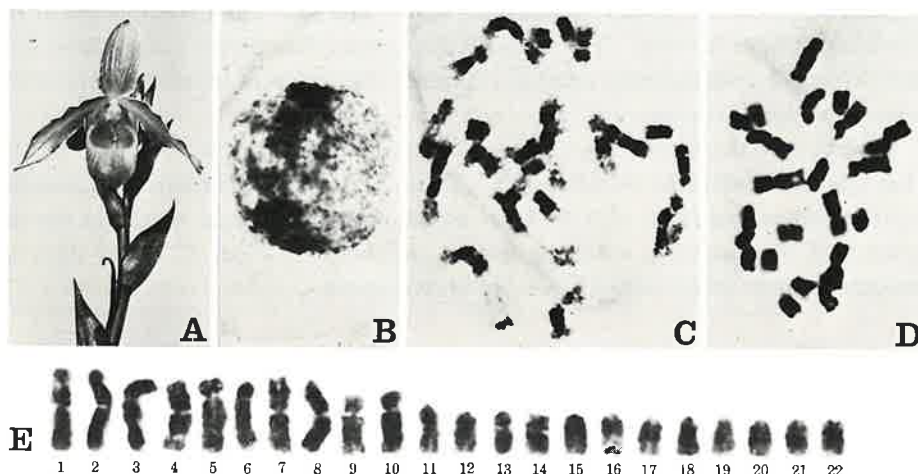


Fig. 4. *Phragmipedium lindleyanum*,  $2n=22$ . A, flower. B, interphase nucleus. C, prometaphase. D and E, metaphase. A,  $\times 0.3$ . B-D, 1200. E,  $\times 1800$ .

As mentioned above, the chromosome complement of this species consisted of 22 chromosomes which could be divided into two groups showing a bimodal karyotype: The one consisted of 10 large chromosomes and the other consisted of 12 small chromosomes which decreased gradually in size. Among the 12 small chromosomes, 10 chromosomes were terminal. Two large chromosomes (Nos. 3, 4) had the satellites at the distal regions of long arms and the 11th chromosome was longer than the 12th chromosome. The small constrictions were observed in the middle region of the short arms of the first and the second chromosomes and near the middle region of the long arms of the fifth and the sixth chromosomes.

## 2) *Phragmipedium sargentianum* (Rolfe) Rolfe, $2n=22$

Varidated specimen No. 3013, BG-35, M-2, M-3.

This species, native to Brazil, is similar to above *P. lindleyanum*. Color of the flower is more purplish red than the *P. lindleyanum*. The dorsal sepal has red stripes. The petals are 1.5 to 2.0 times as long as the dorsal sepal. The staminode is broad and nearly trapezoid (Fig. 5A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type. Prometaphase chromosomes were the proximal type (Fig. 5B). At metaphase,  $2n=22$  chromosomes were counted in four clones (Fig. 5C, 5D). The measurements of chromosome length and arm ratio are shown in Table 5.

Two chromosomes (Nos. 1, 2) which had small constrictions on the middle region of the short arms were both  $5.9\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.3, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) which had satellites in their long arms were both  $5.4\mu\text{m}$  in length. The short arms were  $2.2\mu\text{m}$  and the



long arms were  $3.2\mu\text{m}$  ( $2.2+1.0\mu\text{m}$ ) in length. Arm ratio of these chromosomes were both 1.5, and the position of centromeres were median. Two chromosomes (Nos. 5, 6) were both  $5.4\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.9, and the position of centromeres were submedian. There were small constrictions near the middle region of their long arms of these two chromosomes. Two chromosomes (Nos. 7, 8) were both  $4.8\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. Two chromosomes (Nos. 9, 10) were both  $4.6\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.1, and the position of centromeres were submedian. The 11th chromosome which was  $3.5\mu\text{m}$  in length was longer than the 12th chromosome which was  $3.2\mu\text{m}$  in length. The position of centromeres of these two chromosomes were both terminal. Two chromosomes (Nos. 13, 14) were both  $2.9\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.4, and the position of centromeres were median. The remaining eight chromosomes (Nos. 15-22) which were decreased gradually in size ranged from  $2.9$  to  $2.4\mu\text{m}$  in length, and the position of centromeres were terminal.

According to the morphology of chromosome complement, this species was extremely similar to above *P. lindleyanum*.

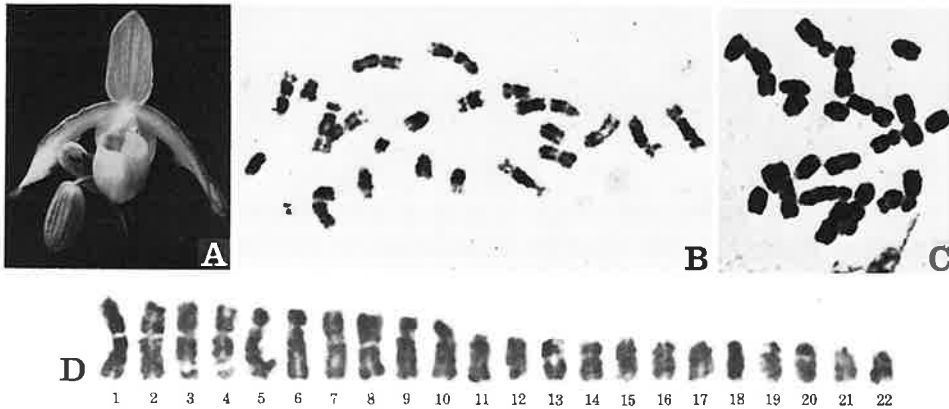


Fig. 5. *Phragmipedium sargentianum*,  $2n=22$ . A, flower. B, prometaphase. C and D, metaphase. A,  $\times 0.3$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

### III. PHRAGMIPEDIUM

#### 1) *Phragmipedium caudatum* (Lindl.) Rolfe, $2n=28$

Varidated specimen No. 1156, 3006, 3020, BG-33.

This species grows in Central America, Ecuador, Peru and Colombia. The height of this species is different in accordance with the habitat. Several flowers which are on the erect peduncle bloom at the same time. The dorsal sepal is acute triangle and its edge is wave. Petals are narrow twisted ribbon and hang down about 50-80 cm in length (Fig. 6A).

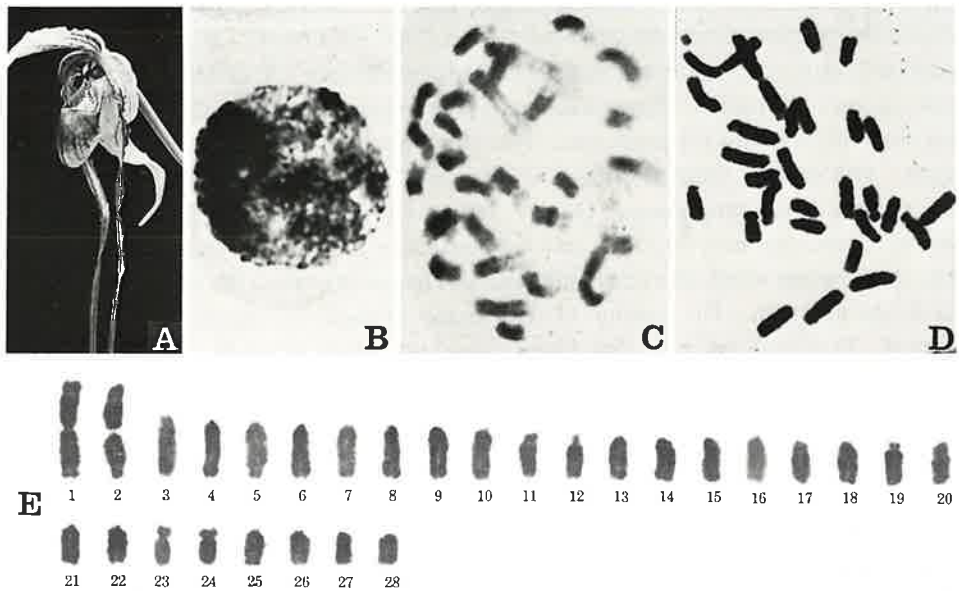


Fig. 6. *Phragmipedium caudatum*,  $2n=28$ . A, flower. B, interphase nucleus. C, prophase. D and E, metaphase. A,  $\times 0.3$ . B and C,  $\times 1200$ . D and E,  $\times 1800$ .

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 6B). Prophase chromosomes were the proximal type (Fig. 6C). Chromosome number of  $2n=32$  in this species was previously reported by Hoffman (1929, cited in Duncan 1959). In the present investigation the number of chromosomes were examined in four clones to be  $2n=28$  (Fig. 6D, E). The measurements of chromosome length and arm ratio are shown in Table 6.

Among the 28 chromosomes the two longest chromosomes (Nos. 1, 2) were distinguished. The other 26 chromosomes (No. 3-28) ranged from  $4.5$  to  $2.5\mu\text{m}$  in length, and decreased gradually in size. The 1st and 2nd chromosomes were both  $7.0\mu\text{m}$  in length, and had satellites at the distal region of long arms. The short arms of them were  $3.4\mu\text{m}$  and the long arms were  $3.6\mu\text{m}(2.9+0.7\mu\text{m})$  in length. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. There were small constrictions near the middle region of their arms of these two chromosomes. Eight chromosomes (Nos. 3-10) ranged from  $4.5$  to  $3.7\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 11, 12) were  $3.5\mu\text{m}$  in length. Arm ratio of these chromosomes were 4.0, and the position of centromeres were subterminal. Six chromosomes (Nos. 13-18) ranged from  $3.3$  to  $3.1\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 19, 20) were both  $3.1\mu\text{m}$  in length. Arm ratio of these chromosomes were 3.4, and the position of centromeres were subterminal. Two chromosomes (Nos. 21, 22) were both  $2.9\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 23, 24) were both  $2.9\mu\text{m}$  in length. Arm ratio of these chromosomes were

2.2, and the position of centromeres were submedian. The rest four chromosomes (Nos. 25-28) ranged from 2.7 to 2.5 $\mu$ m in length, and the position of centromeres were terminal.

As mentioned above, the chromosome complement of this species consisted of 28 chromosomes which could be divided into two groups showing a bimodal karyotype: The one consisted of two large metacentric chromosomes and the other consisted of 26 small chromosomes which were decreased gradually in size. Among the 26 chromosomes six chromosomes (Nos. 11, 12, 19, 20, 23 and 24) were metacentric chromosomes and the rest 20 chromosomes were all telocentric chromosomes. Two large chromosomes (Nos. 1, 2) had the satellites at the distal region of long arms and small constrictions were in the middle region of their short arms.

2) *Phragmipedium wallisii* (Rchb. f.) Garay, 2n=28

Varidated specimen No. M-4.

This species is native to Ecuador. The morphology of the flower is similar to that of *P. caudatum*, but lateral lobes of the staminode of this species is more acute (Fig. 7A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 7B). Prophase chromosomes were the proximal type. At metaphase, 2n=28 chromosomes were counted in one clone (Fig. 7C, D). The measurements of chromosome length and arm ratio are shown in Table 7.

Among the 28 chromosomes the two longest chromosomes (Nos. 1, 2) which were both 6.5 $\mu$ m length were clearly distinguished. The other 26 chromosomes (Nos. 3-28) ranged from 4.1 to 2.3 $\mu$ m in length, and decreased gradually in size. The longest two

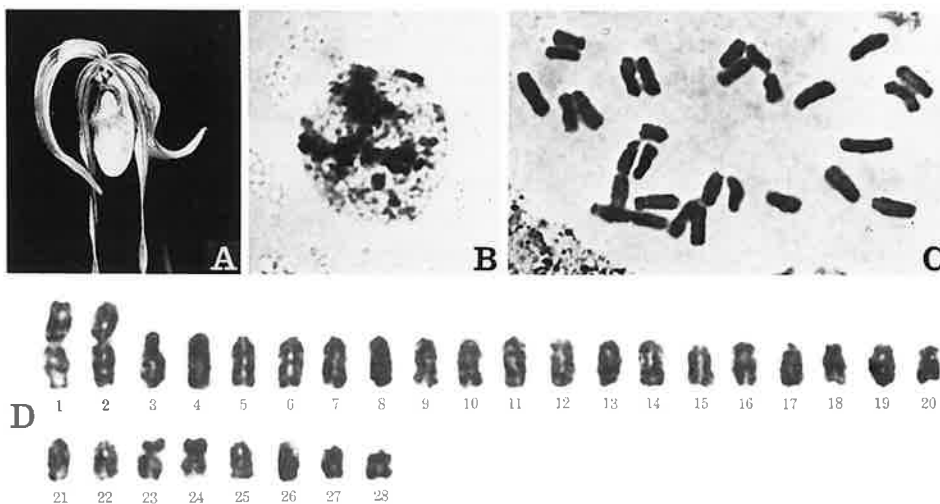


Fig. 7. *Phragmipedium wallisii*, 2n=28. A, flower. B, interphase nucleus. C and D, metaphase. A,  $\times 0.3$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

chromosomes (Nos. 1, 2) had satellites at the distal region of long arms. The short arms of them were  $3.0\mu\text{m}$  and the long arms were  $3.5\mu\text{m}(2.5+1.0\mu\text{m})$  in length. Arm ratio of these chromosomes were both 1.2, and the position of centromeres were median. There were small constrictions near the middle region of their short arms of these two chromosomes. Eight chromosomes (Nos. 3-10) ranged from 4.1 to  $3.7\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 11, 12) were both  $3.7\mu\text{m}$  in length. Arm ratio of these chromosomes were both 3.1, and the position of centromeres were subterminal. Six chromosomes (Nos. 13-18) ranged from 3.5 to  $3.0\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 19, 20) were both  $3.0\mu\text{m}$  in length. Arm ratio of these chromosomes were both 3.3, and the position of centromeres were subterminal. Two chromosomes (Nos. 21, 22) were  $3.0\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 23, 24) were both  $3.0\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.3, and the position of centromeres were submedian. The remaining four chromosomes (Nos. 25-28) ranged from 2.9 to  $2.3\mu\text{m}$  in length, and the position of centromeres were terminal.

According to the morphology of chromosome complement, this species was similar to *P. caudatum* mentioned above.

3) *Phragmipedium lindenbergii* (Lindl.) Dressler & N. Wms.,  $2n=28$

Varidated specimen No. BG-34, BG-36, BG-37, I-3.

This species distributes in Venezuela, Colombia and Ecuador. The petals are all same form and color, and does not have a pouch (Fig. 8A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type. Prometaphase chromosomes were the proximal type (Fig. 8B). At metaphase,  $2n=28$  chromosomes were counted in four clones (Fig. 8C, D). The measurements of chromosome length and arm ratio are shown in Table 8.

Among the 28 chromosomes the two longest chromosomes (Nos. 1, 2) which were both  $6.7\mu\text{m}$  in length were clearly distinguished. The other 26 chromosomes (Nos. 3-28) ranged from 4.7 to  $2.4\mu\text{m}$  in length, and decreased gradually in size. The 1st and 2nd chromosomes had satellites at the distal region of long arms. The short arms of them were  $3.0\mu\text{m}$  and the long arms were  $3.7\mu\text{m}(2.9+0.8\mu\text{m})$  in length. Arm ratio of these chromosomes were both 1.2, and the position of centromeres were median. There were small constrictions near the middle region of their arms of these two chromosomes. Eight chromosomes (Nos. 3-10) ranged from 4.7 to  $3.7\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 11, 12) were both  $3.6\mu\text{m}$  in length. Arm ratio of these chromosomes were both 4.1, and the position of centromeres were subterminal. Six chromosomes (Nos. 13-18) ranged from 3.6 to  $3.3\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 19, 20) were both  $3.3\mu\text{m}$  in length. Arm ratio of these chromosomes were both 3.1, and the position of centromeres were subterminal. Two chromosomes (Nos. 21, 22) were both  $3.3\mu\text{m}$  in length, and the position of cen-

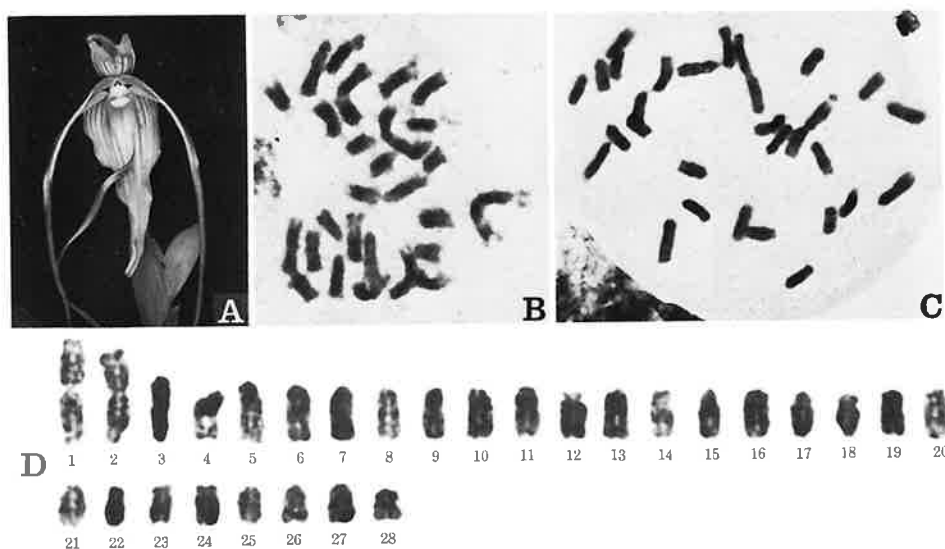


Fig. 8. *Phragmipedium lindenii*,  $2n=28$ . A, flower. B, prometaphase. C and D, metaphase. A,  $\times 0.3$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

tromeres were terminal. Two chromosomes (Nos. 23, 24) were both  $3.0\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.8, and the position of centromeres were submedian. The remaining four chromosomes (Nos. 25-28) ranged from  $2.7$  to  $2.4\mu\text{m}$  in length, and the position of centromeres were terminal.

According to the morphology of chromosome complement, this species was similar to *P. caudatum* mentioned above.

#### IV. HIMANTOPETALUM

##### 1) *Phragmipedium ecuadorensis* Garay, $2n=22$

Varidated specimen No. 3010, BG-38.

This species which distributes in Ecuador and in Peru is smaller than *P. longifolium* belonging to Sect. Lorifolia. The length of the leaves and peduncle are about half of *P. longifolium*. The color of flower is greenish pink. Petals are ribbon-type and twistingly hang down. The staminode has thick black hairs (Fig. 9A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 9B). Prometaphase chromosomes were the proximal type (Fig. 9C). At metaphase,  $2n=22$  chromosomes were counted in two clones (Fig. 9D, E). The measurements of chromosome length and arm ratio are shown in Table 9.

The 22 chromosomes of this species ranged from  $5.6$  to  $2.4\mu\text{m}$  in length, and de-

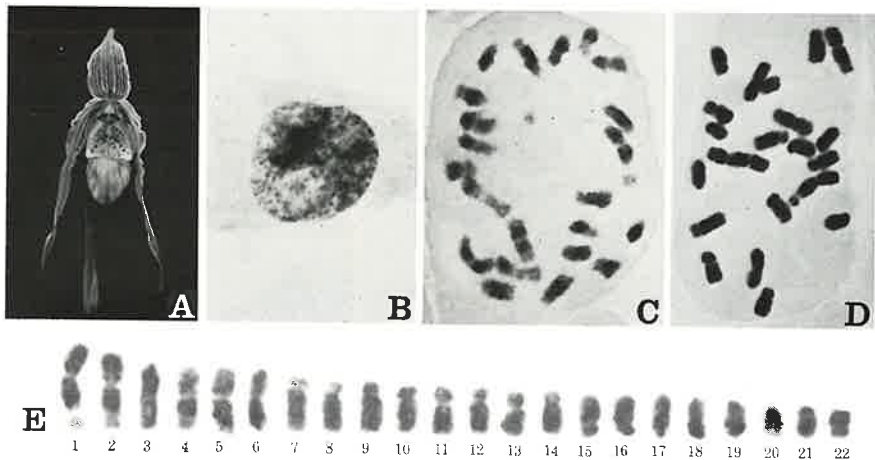


Fig. 9. *Phragmipedium ecuadorensis*,  $2n=22$ . A, flower. B, interphase nucleus. C, prometaphase. D and E, metaphase. A,  $\times 0.5$ . B-D,  $\times 1200$ . E,  $\times 1800$ .

creased gradually in size. The 1st and 2nd chromosomes had satellites at the distal region of long arms and were  $5.6\mu\text{m}$  and  $5.5\mu\text{m}$  in length, respectively. The short arms of them were each  $2.3\mu\text{m}$  and  $2.2\mu\text{m}$  and the long arms were both  $3.3\mu\text{m}(2.5+0.8\mu\text{m})$  in length. Arm ratio of these chromosomes were 1.4 and 1.5 and the position of centromeres were median. Four chromosomes (Nos. 3-6) ranged from  $5.1$  to  $4.9\mu\text{m}$  in length. Arm ratio of these chromosomes were 1.2 and 1.1, and the position of centromeres were median. Two chromosomes (Nos. 7, 8) were each  $4.3$  and  $4.2\mu\text{m}$  in length. Arm ratio of these chromosomes were each 3.3 and 3.2, and the position of centromeres were subterminal. Four chromosomes (Nos. 9-12) ranged from  $3.8$  to  $3.5\mu\text{m}$  in length. Arm ratio of these chromosomes varied from 2.8 to 2.9, and the position of centromeres were submedian. Two chromosomes (Nos. 13, 14) were both  $3.4\mu\text{m}$  in length. Arm ratio of these chromosomes were 1.6, and the position of centromeres were median. Eight chromosomes (Nos. 15-22) which had the small constriction in the middle region of long arms varied from  $3.3$  to  $2.4\mu\text{m}$  in length, and the position of centromeres were terminal.

As mentioned above, the chromosome complement of this species consisted of 22 chromosomes which decreased gradually in size. The two longest chromosomes (Nos. 1, 2) had the satellites at the distal region of long arms. Among the 22 chromosomes eight small chromosomes were the telocentric chromosomes.

2) *Phragmipedium pearcei* (Rchb. f.) Rauh & Senghas, Type 1,  $2n=20$   
 Varidated specimen No. 3016.

This species is native to Ecuador and Peru and has been classified into two types based on the differences of the shape and the color of flower by Garay (1979). The specimen in-

vestigated is suit to Type 1 and its peduncle reaches about 60 cm in height. Color of the dorsal sepal and petals are brownish green with brownish veins. The staminode of this species is a half-moon shape and its edge bears black hairs (Fig. 10A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 10B). Prophase chromosomes were the proximal type (Fig. 10C). In the present investigation the chromosome number of this Type 1 was examined to be  $2n=20$  (Fig. 10D, E). The measurements of chromosome length and arm ratio are shown in Table 10.

Among the 20 chromosomes the 1st chromosome (No. 1) was  $7.2\mu\text{m}$  in length and was distinguished. The other 19 chromosomes (Nos. 2-20) ranged from  $4.4$  to  $2.1\mu\text{m}$  in length, and decreased gradually in size.

Arm ratio of the 1st chromosome was 1.2, and the position of centromere was median. This chromosome had the small constrictions in the middle region of long and short arms. The 2nd chromosome had a satellite at the distal region of long arm. The short arm was  $1.9\mu\text{m}$  in length and the long arm was  $2.5\mu\text{m}(1.7+0.8\mu\text{m})$  in length. Arm ratio of this chromosome was 1.3, and the position of centromere was median. The 3rd chromosome was  $4.3\mu\text{m}$  in length and had the small constriction in the middle region of the long arm. Arm ratio of the chromosome was 1.0, and the position of centromere was median. The 4th chromosome was  $4.3\mu\text{m}$  in length and had the small constriction in the middle region of the long arm. Arm ratio of this chromosome was 2.3, and the position of centromere was submedian. The 5th chromosome was  $4.3\mu\text{m}$  in length and had the satellite at the distal region of the long arm. The short arm was  $1.2\mu\text{m}$  in length and the long arm was  $3.1\mu\text{m}$

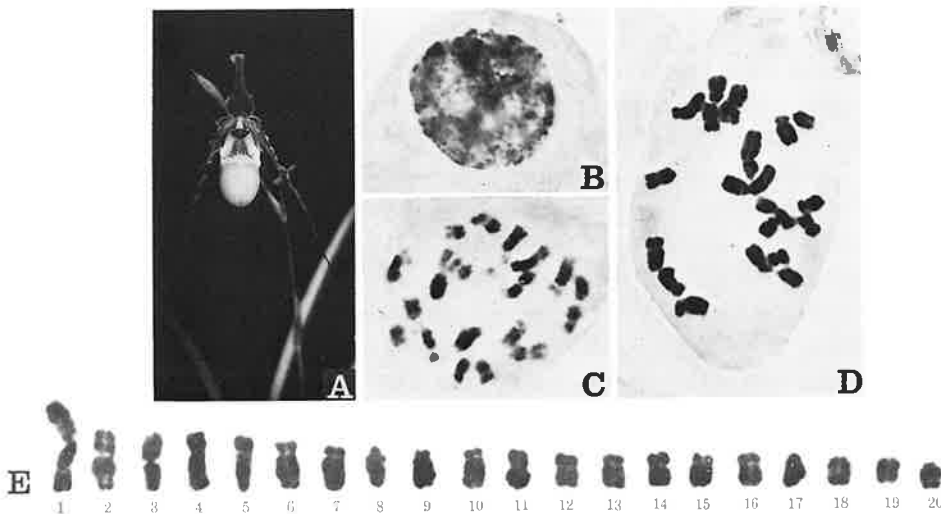


Fig. 10. *Phragmipedium pearcei* type 1,  $2n=20$ . A, flower. B, interphase nucleus. C, prometaphase. D and E, metaphase. A,  $\times 0.4$ . B and C,  $\times 900$ . D,  $\times 1200$ . E,  $\times 1800$ .

( $2.3+0.8\mu\text{m}$ ). Arm ratio of this chromosome was 2.6, and the position of centromere was submedian. Two chromosomes (Nos. 6, 7) were  $3.7\mu\text{m}$  and  $3.3\mu\text{m}$  in length and had small constrictions near the middle region of their long arms. Their arm ratio were 2.4 and 2.9, and the position of centromeres were submedian. The 8th chromosome was  $3.1\mu\text{m}$  in length. Arm ratio was 1.4, and the position of centromere was median. Two chromosomes (Nos. 9, 10) were both  $3.0\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.8, and the position of centromeres were submedian. The 11th chromosome was  $2.9\mu\text{m}$  in length. Arm ratio of this chromosome was 1.1, and the position of centromere was median. The 12th chromosome was  $2.7\mu\text{m}$  in length. Arm ratio of this chromosome was 1.5, and the position of centromere was median. The 13th chromosome was  $2.7\mu\text{m}$  in length. Arm ratio of this chromosome was 2.0, and the position of centromere was submedian. Three chromosomes (Nos. 14-16) ranged from  $2.7$  to  $2.5\mu\text{m}$  in length. Arm ratio of these chromosomes varied from 1.3 to 1.5, and the position of centromeres were median. Four chromosomes (Nos. 17-20) ranged from  $2.4$  to  $2.1\mu\text{m}$  in length, and the position of centromeres were terminal. Two chromosomes (Nos. 18, 20) among these four chromosomes had small constrictions in the middle region of the long arms.

As mentioned above the  $2n=20$  chromosomes of this species consisted of one large chromosome and 19 small chromosomes which decreased gradually in size. The position of centromeres of the four shortest chromosomes were terminal. These 20 chromosomes did not form the pair of matched chromosomes morphologically, and were heterogeneous.

### 3) *Phragmipedium pearcei*, Type 2, $2n=21$

Varidated specimen No. 3033.

Comparing with Type 1, the leaves and the peduncle are shorter. The color of the flower is more purplish-red and the petals are shorter, too. Furthermore the petals of this species do not hang and do not firmly twist (Fig. 11A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 11B). Prophase chromosomes were the proximal type. At metaphase,  $2n=21$  chromosomes were counted (Fig. 11C, D). The measurements of chromosome length and arm ratio are shown in Table 11.

The length of the 21 chromosomes of Type 2 varied from the longest to the shortest chromosomes. Two chromosomes (Nos. 1, 2) had the satellites at the distal region of their long arms and were both  $5.3\mu\text{m}$  in length. The short arms were  $2.3\mu\text{m}$  and the long arms were  $3.0\mu\text{m}$  ( $2.3+0.7\mu\text{m}$ ) in length. Arm ratio of these chromosomes were both 1.3, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) were  $4.9\mu\text{m}$  and  $4.8\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 1.1 and 1.2, and the position of centromeres were median. These chromosomes had the small constrictions in the middle region of their long arms. Two chromosomes (Nos. 5, 6) were  $4.4\mu\text{m}$  and  $4.2\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 1.3 and 1.2, and the position of centromeres were median. These chromosomes had the small constrictions in



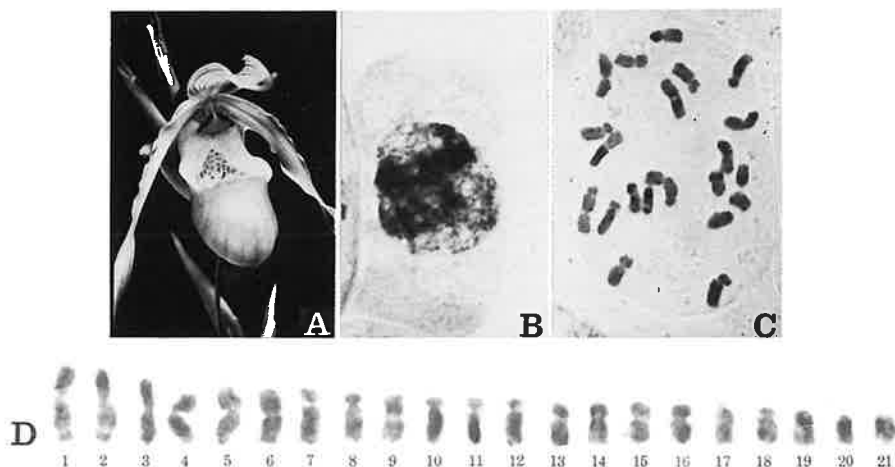


Fig. 11. *Phragmipedium pearcei* type 2,  $2n=21$ . A, flower. B, interphase nucleus. C and D, metaphase. A,  $\times 0.6$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

the middle region of their long arms. Two chromosomes (Nos. 7, 8) were  $3.8\mu\text{m}$  and  $3.7\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 2.5 and 2.7, and the position of centromeres were submedian. The 9th chromosome was  $3.7\mu\text{m}$  in length. Arm ratio of the chromosome was 1.2, and the position of centromere was median. The 10th chromosome was  $3.6\mu\text{m}$  in length. Arm ratio of this chromosome was 3.0, and the position of centromere was submedian. Two chromosomes (Nos. 11, 12) were both  $3.6\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.6, and the position of centromeres were submedian. Two chromosomes (Nos. 13, 14) were both  $3.3\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.8, and the position of centromeres were submedian. Two chromosomes (Nos. 15, 16) were both  $3.2\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.5, and the position of centromeres were median. The 17th chromosome was  $3.2\mu\text{m}$  in length, and the position of centromere was terminal. The 18th chromosome was  $3.0\mu\text{m}$  in length. Arm ratio of this chromosome was 1.5, and the position of centromere was median. The rest three chromosomes (Nos. 19-21) ranged from  $2.8$  to  $2.3\mu\text{m}$  in length, and the position of centromeres were terminal.

According to the morphology of chromosome complement, the  $2n=21$  chromosomes did not formed the pair of matched chromosomes morphologically. Especially two pairs, Nos. 9-10 and Nos. 17-18 were heterogenous. The karyotype of Type 2 was different from that of above *P. pearcei* Type 1.

4) *Phragmipedium pearcei*, Type 3,  $2n=22$ 

Varidated specimen No. 3020.

The external characteristics of this type are similar to those of above Type 2, and could not be distinguished each other. Petals of this type are longer than those of above Type 2 and twist (Fig. 12A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type. Prophase chromosomes were the proximal type (Fig. 12B). At metaphase,  $2n=22$  chromosomes were counted in one clone (Fig. 12C, D). The measurements of chromosome length and arm ratio are shown in Table 12.

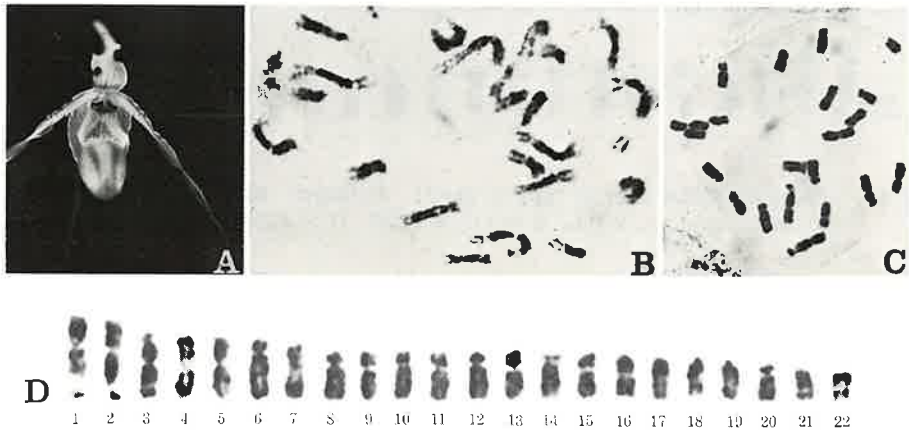


Fig. 12. *Phragmipedium pearcei* type 3,  $2n=22$ . A, flower. B, prometaphase. C and D, metaphase. A,  $\times 0.4$ . B and C,  $\times 900$ . D,  $\times 1800$ .

The length of the 22 chromosomes of Type 3 varied from the longest to the shortest chromosomes. Two chromosomes (Nos. 1, 2) had the satellites at the distal region of the long arms and were  $5.3\mu\text{m}$  in length. The short arms were  $2.2\mu\text{m}$  and the long arms were  $3.1\mu\text{m}(2.4+0.7\mu\text{m})$  in length. Arm ratio of these chromosomes were both 1.4, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) were  $4.7\mu\text{m}$  and  $4.3\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 1.1 and 1.2, and the position of centromeres were median. These two chromosomes had the small constrictions in the middle region of the short arm of the 3rd chromosomes and near the middle region of the short arm of the 4th chromosome. The 5th chromosome was  $4.3\mu\text{m}$  in length. Arm ratio of this chromosome was 1.2, and the position of centromere was median. The 6th chromosome was  $4.3\mu\text{m}$  in length. Arm ratio of this chromosome was 3.3, and the position of centromere was subterminal. The 7th chromosome was  $4.0\mu\text{m}$  in length. Arm ratio of this chromosome was 1.4, and the position of centromere was median. Two chromo-

somes (Nos. 8, 9) were  $3.6\mu\text{m}$  and  $3.5\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 2.3 and 1.9, and the position of centromeres were submedian. The 10th chromosome was  $3.4\mu\text{m}$  in length. Arm ratio of this chromosome was 2.4, and the position of centromere was submedian. The 11th chromosome was  $3.4\mu\text{m}$  in length. Arm ratio of this chromosome was 2.1, and the position of centromere was submedian. The 12th chromosome was  $3.4\mu\text{m}$  in length. Arm ratio of this chromosome was 2.4, and the position of centromere was submedian. The 13th chromosome was  $3.4\mu\text{m}$  in length. Arm ratio of this chromosome was 1.6, and the position of centromere was median. The 14th chromosome was  $3.2\mu\text{m}$  in length. Arm ratio of this chromosome was 3.6, and the position of centromere was subterminal. The 15th chromosome was  $2.9\mu\text{m}$  in length. Arm ratio of this chromosome was 1.9, and the position of centromere was submedian. The 16th chromosome was  $2.9\mu\text{m}$  in length. Arm ratio of this chromosome was 1.4, and the position of centromere was median. Three chromosomes (Nos. 17-19) ranged from  $2.9$  to  $2.7\mu\text{m}$  in length, and the position of centromeres were terminal. These three chromosomes had the small constrictions on the middle region of their long arms. The 20th chromosome was  $2.6\mu\text{m}$  in length. Arm ratio of this chromosome was 2.7, and the position of centromere was submedian. The remaining two chromosomes (Nos. 21, 22) were both  $2.3\mu\text{m}$  in length, and the position of centromeres were terminal. These two chromosomes had the small constrictions near the middle region of the long arm of the 21th chromosome and in the middle region of the long arm of the 22th chromosome.

According to the morphology of chromosome complement, the  $2n=22$  chromosomes did not form the pair of matched chromosomes morphologically. Especially two pairs, Nos. 5-6, and 7-8 were heterogenous. The karyotype of this Type was different from that of *P. pearcei* Type 1, while it was similar to that of *P. pearcei* Type 2.

## V. LORIFOLIA

### 1) *Phragmipedium boisseranum* (Rchb. f.) Rolfe, $2n=18$

Varidated specimen No. 3034, I-5, I-6.

This species distributes in Peru. External characteristics of this species are as follows: Leaves are broad and are about 10cm in length. The peduncle is about 50-60cm in length and the width of flower is about 15cm. The color of flower is green. The dorsal sepal is lanceolate and the edge is wave. Petals are narrow and the edges are wave. The petals twist and obliquely spread towards the down ward. The staminode is rhombic-triangular (Fig. 13A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 13B). Prophase chromosomes were the proximal type (Fig. 13C). The chromosome number of  $2n=18$  in this species was previously reported by Karasawa & Tanaka (1976) except the details of the chromosome complement. The metaphase chromo-

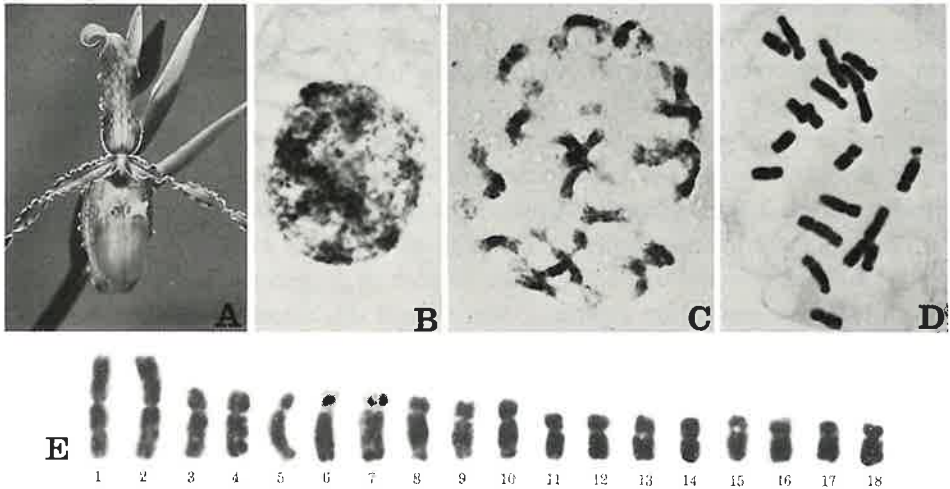


Fig. 13. *Phragmipedium boisseranum*,  $2n=18$ . A, flower. B, interphase nucleus. C, pro-metaphase. D and E, metaphase. A,  $\times 0.3$ . B D,  $\times 1200$ . E,  $\times 1800$ .

somes are shown in Fig. 13D and E. The measurements of chromosome length and arm ratio are shown in Table 13.

Among the 18 chromosomes the two longest chromosomes (Nos. 1, 2) which were both  $7.3\mu\text{m}$  in length were clearly distinguished. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. These two chromosomes had small constrictions on the middle region of the long and short arms. Two chromosomes (Nos. 3, 4) were both  $5.0\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.9, and the position of centromeres were submedian. These two chromosomes had small constrictions near the middle region of their long arms. Two chromosomes (Nos. 5, 6) which had the small constrictions near the middle region of their long arms were both  $4.7\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.6, and the position of centromeres were submedian. Two chromosomes (Nos. 7, 8) which had the satellites at the distal region of long arms were both  $4.7\mu\text{m}$  in length. The short arms of these chromosomes were  $1.0\mu\text{m}$  and the long arms were  $3.7\mu\text{m}(2.9+0.8\mu\text{m})$  in length. Arm ratio of these chromosomes were both 3.6, and the position of centromeres were subterminal. Two chromosomes (Nos. 9, 10) were both  $4.5\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.8, and the position of centromeres were submedian. These two chromosomes had small constrictions near the distal region of their long arms. Six chromosomes (Nos. 11-16) ranged from  $3.5$  to  $3.3\mu\text{m}$  in length. Arm ratio of these chromosomes varied from 1.2 to 1.7, and the position of centromeres were median. Two chromosomes (Nos. 17, 18) were  $3.1\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.8, and the position of centromeres were submedian.

Thus, the chromosome complement of this species consisted of 18 chromosomes which could be divided mainly into three groups showing a trimodal karyotype: The one consisted of two largest chromosomes and the second consisted of 8 medium-size chromosomes which were decreased gradually in size and the rest consisted of 8 small chromosomes which were decreased gradually in size, too. One pair of matched chromosomes (Nos. 7, 8) had the satellites on their long arms. Two large chromosomes (Nos. 1, 2) had the small constrictions on the long and short arms and another six chromosomes (Nos. 3-6, 9, 10) had them on their long arms.

2) *Phragmipedium vittatum* (Vell.) Rolfe,  $2n=18$

Varidated specimen No. 3018, BG-39.

This species, distributing in Brazil, is similar to above *P. boisseranum*. Color of the edge of leaves is yellow. The edge of ventral sepals is not wave. The staminode is cordate-triangular (Fig. 14A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 14B). Prometaphase chromosomes were the proximal type (Fig. 14C). At prometaphase and metaphase,  $2n=18$  chromomes were counted in two clones (Fig. 14D, E). The measurements of chromosome length and arm ratio are shown in Table 14.

Among the 18 chromosomes the two longest chromosomes (Nos. 1, 2) were distinguished. They were both  $7.5\mu\text{m}$  in length. Their arm ratio were both 1.2, and the position of centromeres were median. These two chromosomes had the small constrictions in

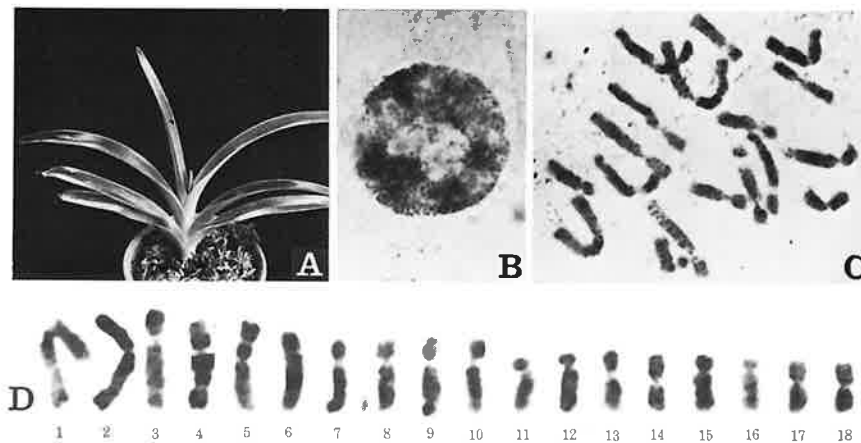


Fig. 14. *Phragmipedium vittatum*,  $2n=18$ . A, seedling. B, interphase nucleus. C, prometaphase. D, metaphase. A,  $\times 0.1$ . B and C,  $\times 1200$ . D,  $\times 1800$ .

middle region of each long and short arms. Two chromosomes\* (Nos. 3, 4) were both  $5.5\mu\text{m}$  in length. Arm ratio of these chromosomes were 2.1, and the position of centromeres were submedian. These two chromosomes had the small constrictions near the end of the long arms. Two chromosomes (Nos. 5, 6) were both  $5.5\mu\text{m}$  in length. Arm ratio of these chromosomes were 2.3, and the position of centromeres were submedian. These two chromosomes had the small constrictions near the centromere of the long arms. Two chromosomes (Nos. 7, 8) which had the satellites at the distal region of the long arms were both  $5.3\mu\text{m}$  in length. The short arms of these chromosomes were  $1.3\mu\text{m}$  and long arms of them were  $4.0\mu\text{m}(3.2+0.8\mu\text{m})$  in length. Arm ratio of these chromosomes were both 3.1, and the position of centromeres were subterminal. Two chromosomes (Nos. 9, 10) were both  $4.9\mu\text{m}$  in length. Their arm ratio were both 2.5, and the position of centromeres were submedian. Two chromosomes (Nos. 11, 12) were both  $4.2\mu\text{m}$  in length. Their arm ratio were both 3.2, and the position of centromeres were subterminal. Two chromosomes (Nos. 13, 14) were both  $3.9\mu\text{m}$  in length. Their arm ratio were 1.3 and 1.2, respectively, and the position of centromeres were median. Two chromosomes (Nos. 15, 16) were both  $3.9\mu\text{m}$  in length. Their arm ratio were both 2.5, and the position of centromeres were submedian. The remaining two chromosomes (Nos. 17, 18) were  $3.8\mu\text{m}$  and  $3.7\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were both 1.2, and the position of centromeres were median.

The similarity between the chromosome complement of this species and that of above *P. boisseranum* was observed. Thus, the chromosome complement of this species could be divided into three groups showing a trimodal karyotype: The first group consisted of two largest chromosomes and the second consisted of 8 medium-sized chromosomes and the rest consisted of 8 small chromosomes. The two longest chromosomes had the small constrictions on the middle region of the long and short arms. Six medium-sized chromosomes (Nos. 3-6, 9, 10) had the small constrictions and two chromosomes (Nos. 7, 8) had the satellites on their long arms.

Compared with the karyotype of *P. boisseranum*, two subtelocentric chromosomes of this species (Nos. 11, 12) were the characteristics.

3) *Phragmipedium hartwegii* (Rchb. f.) L. O. Wms.,  $2n=20$

Varidated specimen No. 3035, BG-40.

*Phragmipedium hartwegii* is native to Ecuador. The leaves are linear, archedly curved and 30cm in length. The erect peduncle bears several flowers which bloom by turns. The flower is horizontally about 7cm in diameter. The dorsal sepal is lanceolate and pale green, suffused with pale redish-purple and has redish-purple veins (Fig. 15A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type. Prophase chromosomes were the proximal type. The chromosome number of  $2n=20$  in this species was previously reported by Brown (1959, cited in Duncan

1959). In the present investigation the number of chromosomes was reexamined to be  $2n=20$  (Fig. 15 B-F). The measurements of chromosome length and arm ratio are shown in Table 15.

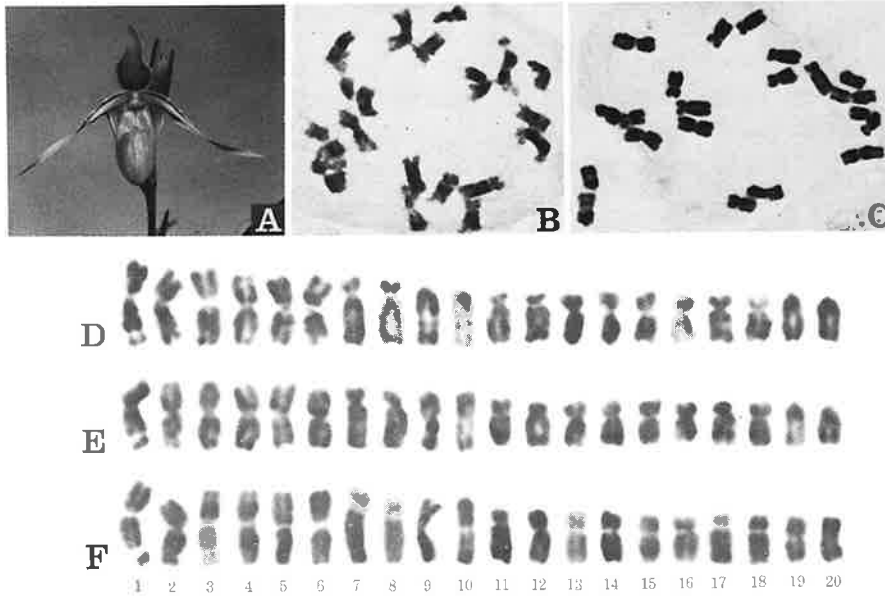


Fig. 15. *Phragmipedium hartwegii*,  $2n=20$ . A, flower. B, prometaphase. C-F, metaphase. A,  $\times 0.5$ . B,  $\times 1200$ . C,  $\times 1000$ . D-F,  $\times 1800$ .

The 20 chromosomes (Nos. 1-20) ranged from  $5.4$  to  $3.4\mu\text{m}$  in length, and decreased gradually in size. The two longest chromosomes (Nos. 1, 2) had the satellites at the distal region of the long arms. The short arms of these chromosomes were  $2.4\mu\text{m}$  and the long arms were  $3.0\mu\text{m}(2.3+0.7\mu\text{m})$  in length. Arm ratio of these chromosomes were 1.2, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) were both  $4.9\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.2, and the position of centromeres were median. These two chromosomes had small constrictions near the middle region of the short and long arms. Two chromosomes (Nos. 5, 6) were both  $4.9\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. Two chromosomes (Nos. 7, 8) were  $4.8\mu\text{m}$  and  $4.7\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 3.0 and 2.9, and the position of centromeres were submedian. These chromosomes had the small constrictions near the end of their long arms. The 9th and 10th chromosomes were both  $4.4\mu\text{m}$  in length, and the position of centromeres were terminal. These chromosomes had the small constrictions near the middle region of the long arms. Two chromosomes (Nos. 11, 12) were both  $3.8\mu\text{m}$  in length. Arm ratio of these chromosomes were 2.8, and the position of centromeres were submedian. Four chromosomes (Nos. 13-16) ranged from  $3.7$  to  $3.5\mu\text{m}$  in length. Arm ratio of these chro-

mosomes varied from 1.6 to 1.7, and the position of centromeres were median. Two chromosomes (Nos. 17, 18) were both  $3.5\mu\text{m}$  in length. Arm ratio of these chromosomes were 2.2, and the position of centromeres were submedian. The rest two chromosomes (Nos. 19, 20) were  $3.5\mu\text{m}$  and  $3.4\mu\text{m}$  in length, respectively, and the position of centromeres were terminal. These chromosomes had the small constrictions near the middle region of the long arms.

The chromosome complement of this species consisted of 10 pairs of morphologically matched chromosomes which decreased gradually in size. The longest pair of matched chromosomes had the satellites at the distal region of the long arms. Four chromosomes (Nos. 9, 10, 19 and 20) were telocentric chromosomes.

4) *Phragmipedium longifolium* (Rchb. f. & Warsc.) Rolfe,  $2n=23$  (=20+3f)  
Varidated specimen No. 1241, I-6.

The distribution of this species is recorded in Costa Rica, Panama, Colombia and Ecuador. External characteristics of this species are as follows: Leaves are linear lanceolate and are 40-60cm in length. The peduncle is 50-60cm in length. Natural spread of the flower is 7-10cm in width and its color is green or brownish-pinked green. The dorsal sepal is lanceolate and its edge curls toward outside. The petals are linear lanceolate and twistingly hang down slant. The upper edge of staminode has thick black hairs (Fig. 16A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 16B). Prophase chromosomes were the proximal type (Fig. 16C). At metaphase,  $2n=23$  chromosomes were counted in two clones (Fig. 16D, E). The measurements of chromosome length and arm ratio are shown in Table 16.

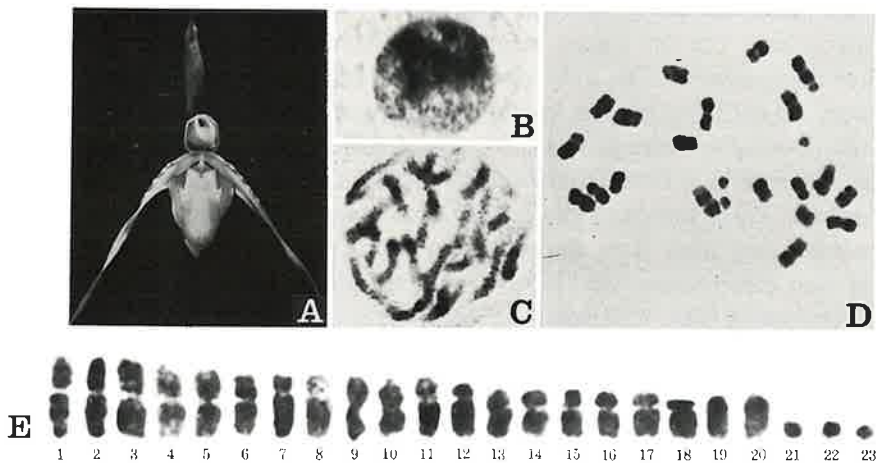


Fig. 16. *Phragmipedium longifolium*,  $2n=23$ . A, flower. B, interphase nucleus. C, prophase. D and E, metaphase. A,  $\times 0.3$ . B-D,  $\times 900$ . E,  $\times 1800$ .



Among the 23 chromosomes, 20 chromosomes ranged from 5.4 to 3.0 $\mu\text{m}$  in length, and decreased gradually in size. The two longest chromosomes (Nos. 1, 2) had the satellites at the distal region of the long arms. The short arms of them were 2.3 $\mu\text{m}$  and the long arms were 3.1 $\mu\text{m}$ (2.1+1.0 $\mu\text{m}$ ) in length. Arm ratio of these chromosomes were 1.3, and the position of centromeres were median. Four chromosomes (Nos. 3-6) ranged from 5.0 to 4.8 $\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 1.2, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) had the small constrictions near the middle region of the short and long arms. Two chromosomes (Nos. 7, 8) were both 4.4 $\mu\text{m}$  in length. Arm ratio of these chromosomes were 3.0, and the position of centromeres were submedian. Two chromosomes (Nos. 9, 10) were both 4.1 $\mu\text{m}$  in length, and the position of centromeres were terminal. These chromosomes had the small constrictions near the middle region of long arms. Two chromosomes (Nos. 11, 12) were 4.0 and 3.9 $\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 2.1 and 2.3, and the position of centromeres were submedian. Two chromosomes (Nos. 13, 14) were both 3.5 $\mu\text{m}$  in length. Arm ratio of these chromosomes were 1.7, and the position of centromeres were median. Four chromosomes (Nos. 15-18) ranged from 3.2 to 3.0 $\mu\text{m}$  in length. Arm ratio of them varied from 1.9 to 2.3, and the position of centromeres were submedian. Two chromosomes (Nos. 19, 20) were both 3.0 $\mu\text{m}$  in length, and the position of centromeres were terminal. The remaining three chromosomes (Nos. 21-23) ranged from 1.5 to 1.4 $\mu\text{m}$  in length, and the position of centromeres were terminal.

As mentioned above, the 23 chromosomes of this species consisted of 20 chromosomes which decreased in descending order of size and three minute chromosomes. Among the 23 chromosomes, 20 chromosomes (Nos. 1-20) of this species were similar to those of *P. hartwegii*. The difference between the chromosome complement of this species and that of *P. hartwegii* was the presence of three minute chromosomes in this species.

5) *Phragmipedium longifolium* (Rchb. f. & Warsc.) Rolfe var. *gracile* Rolfe,  
2n=21 (=20+1f)

Varidated specimen No. 3015.

The specimen was obtained from Rands Orchids. The leaves of this species are more narrow and longer than those of above *P. longifolium*.

The interphase nuclei were similar to those of above species showing the complex chromocenter type. Prophase chromosomes were the proximal type. At prometaphase and metaphase, 2n=21 chromosomes were counted in one clone (Fig. 17A-C). The measurements of chromosome length and arm ratio are shown in Table 17.

Among the 21 chromosomes, 20 chromosomes (Nos. 1-20) ranged from 5.5 to 3.7 $\mu\text{m}$  in length, and decreased gradually in size. The 1st and 2nd chromosomes were both 5.5 $\mu\text{m}$  in length, and had the satellites at the distal region of the long arms. The short arms of their chromosomes were both 2.3 $\mu\text{m}$  and the long arms were both 3.2 $\mu\text{m}$ (2.3+0.9 $\mu\text{m}$ ) in

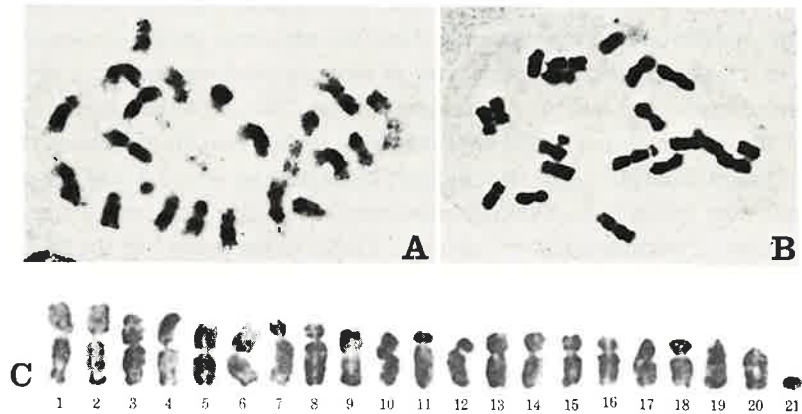


Fig. 17. *Phragmipedium longifolium* var. *gracile*,  $2n=21$ . A, prometaphase. B and C, metaphase. A and B,  $\times 1200$ . C,  $\times 1800$ .

length. Arm ratio of these chromosomes were 1.4, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) were both  $5.3\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.3, and the position of centromeres were median. These two chromosomes had the small constrictions near the middle region of each short and long arms. Two chromosomes (Nos. 5, 6) were both  $4.7\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. Two chromosomes (Nos. 7, 8) were both  $4.5\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.8, and the position of centromeres were submedian. Two chromosomes (Nos. 9, 10) were both  $4.3\mu\text{m}$  in length, and the position of centromeres were terminal. These two chromosomes had the small constrictions near the middle region of the long arms. Two chromosomes (Nos. 11, 12) were both  $4.2\mu\text{m}$  in length. Arm ratio of these chromosomes were both 2.8, and the position of centromeres were submedian. Two chromosomes (Nos. 13, 14) were both  $4.1\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.6, and the position of centromeres were median. Four chromosomes (Nos. 15-18) ranged from  $3.8\mu\text{m}$  to  $3.7\mu\text{m}$  in length. Arm ratio of these chromosomes ranged from 1.9 to 1.8, and the position of centromeres were submedian. Two chromosomes (Nos. 19, 20) were both  $3.7\mu\text{m}$  in length, and the position of centromeres were terminal. The 21th chromosome was  $1.0\mu\text{m}$  in length, and the position of centromere was terminal.

As mentioned above, among the 21 chromosomes 20 chromosomes (Nos. 1-20) of this species were similar to those of *P. hartwegii*. The difference between the chromosome complement of this species and that of *P. hartwegii* was the presence of minute chromosome in this species.

6) *Phragmipedium roezlii* (Rchb. f.) Garay,  $2n=22$  (=20+2f)

Varidated specimen No. 3017, BG-41.

This species distributes in Costa Rica, Panama and Colombia. Compared with *P. longifolium*, the shape of this species is closely similar but a little small. Petals are short and spread horizontally. Color of dorsal sepal and petals are light yellowish green with red veins. The staminode is cordate and its upper edge has thick black hairs. (Fig. 18A).

The interphase nuclei were similar to those of above species showing the complex chromocenter type (Fig. 18B). Prophase chromosomes were the proximal type (Fig. 18C). At metaphase,  $2n=22$  chromosomes were ocunted in two clones (Fig. 18D, E). The measurements of chromosome length and arm ratio are shown in Table 18.

Among the 22 chromosomes, 20 chromosomes (Nos. 1-20) ranged from  $5.2$  to  $2.9\mu\text{m}$  in length, and decreased gradually in size. Two chromosomes (Nos. 1, 2) had the satellites at the distal region of long arms. These chromosomes were both  $5.2\mu\text{m}$  in length. The short arms of their chromosomes were  $2.2\mu\text{m}$  and the long arms were  $3.0\mu\text{m}(2.1+0.9\mu\text{m})$  in length. Arm ratio of their chromosomes were 1.4, and the position of centromeres were median. Two chromosomes (Nos. 3, 4) were both  $5.1\mu\text{m}$  in length. Arm ratio of these chromosomes were both 1.1, and the position of centromeres were median. Two chromosomes (Nos. 5, 6) were  $5.0\mu\text{m}$  and  $4.5\mu\text{m}$  in length, respectively. Arm ratio of these chromosomes were 1.1 and 1.0, and the position of centromeres were median. Two chro-

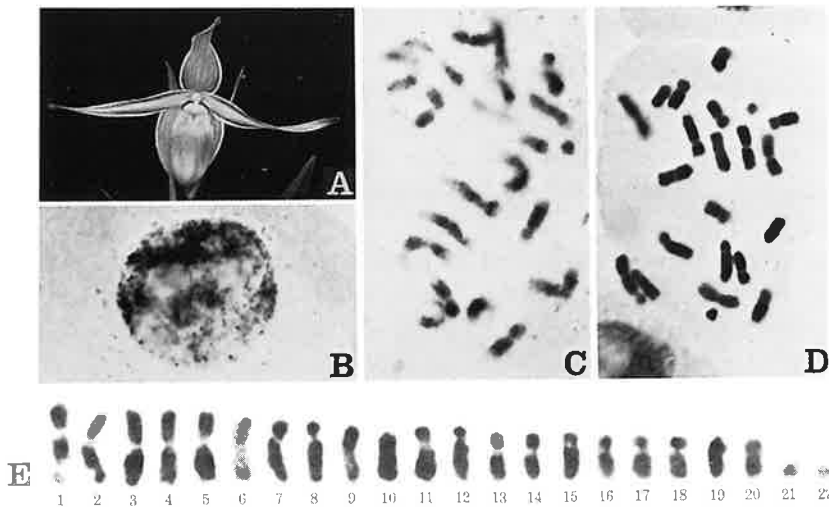


Fig. 18. *Phragmipedium roezlii*,  $2n=22$ . A, flower. B, interphase nucleus. C, prophase. D and E, metaphase. A,  $\times 0.5$ . B-D,  $\times 1200$ . E,  $\times 1800$ .

mosomes (Nos. 7, 8) were 4.5 $\mu$ m and 4.3 $\mu$ m in length, respectively. Arm ratio of these chromosomes were 2.5 and 2.6, respectively, and the position of centromeres were submedian. Two chromosomes (Nos. 9, 10) were 4.3 $\mu$ m and 4.1 $\mu$ m in length, respectively, and the position of centromeres were terminal. Two chromosomes (Nos. 11, 12) were both 3.9 $\mu$ m in length. Arm ratio of these chromosomes 2.5 and 2.9, respectively, and the position of centromeres were submedian. Two chromosomes (Nos. 13, 14) were both 3.4 $\mu$ m in length. Arm ratio of these chromosomes were both 1.4, and the position of centromeres were median. Four chromosomes (Nos. 15-18) ranged from 3.2 to 3.0 $\mu$ m in length. Arm ratio of these chromosomes were 2.2, and 2.0, respectively, and the position of centromeres were submedian. Two chromosomes (Nos. 19, 20) were 3.0 $\mu$ m and 2.9 $\mu$ m in length, respectively, and the position of centromeres were terminal. The rest two chromosomes (Nos. 21, 22) were 1.1 $\mu$ m and 1.0 $\mu$ m in length, respectively, and the position of centromeres were terminal.

As mentioned above, among the 22 chromosomes, 20 chromosomes (Nos. 1-20) of this species were similar to those of *P. hartwegii*, and the presence of two minute chromosomes was the characteristic of this species.

## Discussion

### I. Karyomorphological characteristics of *Phragmipedium*

All interphase nuclei of 15 taxa of *Phragmipedium* investigated were categorized into the complex chromocenter type (cf. Tanaka 1971). This is the same as the taxa of *Paphiopedilum* (Karasawa 1979), but the prophase chromosomes showing the proximal type (cf. Tanaka 1971) clearly differ from those of *Paphiopedilum* showing the interstitial type (Karasawa 1979).

Small constrictions were observed in certain chromosomes at prometaphase in all species of *Phragmipedium*. These small constrictions were more general and clear than those of *Paphiopedilum* (Karasawa 1979). In addition, two satellite chromosomes were observed in 14 taxa, and three satellite chromosomes were observed in one taxa investigated. These satellites were almost same size in 15 taxa, and existed in the long arms of metacentric chromosomes except for *P. schlimii* and *P. x stenophyllum* having in telocentric chromosomes.

The chromosome numbers of  $2n=18$  and  $2n=20$  in *Phragmipedium* were reexamined and those of  $2n=21$ ,  $2n=22$ ,  $2n=28$ ,  $2n=30$ ,  $2n=20+1f$ ,  $2n=20+2f$ ,  $2n=20+3f$  and  $2n=40$  were newly added. The  $2n=32$  in *P. caudatum* by Hoffman (1929) was corrected as  $2n=28$ .

The chromosome complements of *P. boisseranum* and *P. vittatum* with  $2n=18$  consisted of nine pairs of matched chromosomes with median, submedian and subterminal centromere, while those of the species with over  $2n=20$  had several chromosomes with terminal centromere besides on above three types (Fig. 19).

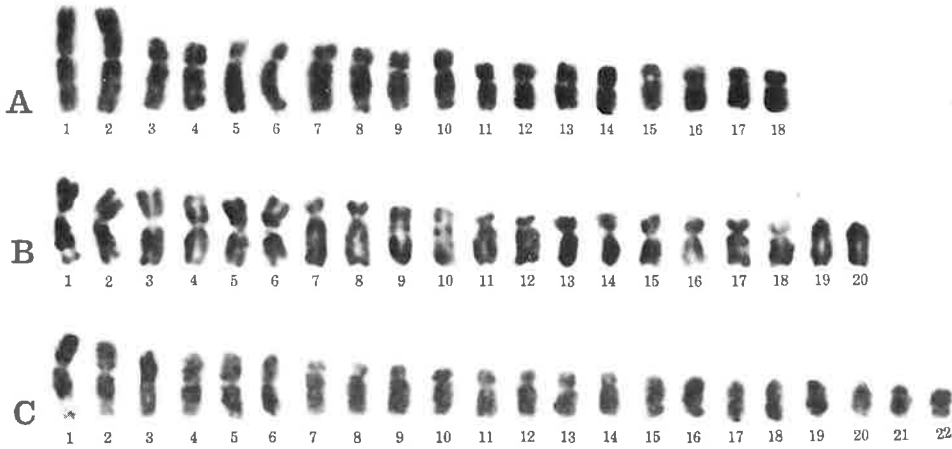


Fig. 19. Metaphase chromosomes of three species of *Phragmipedium*. A, *P. boisseranum*  $2n=18$ . B, *P. hartwegii*  $2n=20$ . C, *P. ecuadorensis*  $2n=22$ .  $\times 1800$ .

When we express the metacentric, submetacentric and subtolocentric chromosome as 'V' and the telocentric chromosome as 'I', the karyotype of each species can be shown as  $2n=18=18V$  in *P. boisseranum* and *P. vittatum*, as  $2n=20=16V+4I$  in *P. hartwegii*, as  $2n=22=14V+8I$  in *P. ecuadorensis*, as  $2n=28=8V+20I$  in *P. caudatum*, *P. wallisii* and *P. lindenii*, as  $2n=30=6V+24I$  in *P. schlimii*. By this method of karyotype expression, it is found that the increase of 'I' chromosome originates from the decrease of 'V' chromosome. When we assume that two 'I' chromosomes derive from one 'V' chromosome by centric fission,  $2n=20=16V+4I$  in *P. hartwegii* is converted into  $2n=18V$ . Similarly,  $2n=22=14V+8I$  in *P. ecuadorensis* is converted into  $2n=18V$ ,  $2n=28=8V+20I$  in *P. caudatum*, *P. wallisii* and *P. lindenii* into  $2n=18V$  and  $2n=30=6V+24I$  in *P. schlimii* into  $2n=18V$ , while  $2n=22=12V+10I$  in *P. lindleyanum* and *P. sargentianum* of section *Platypetalum* is converted into  $2n=17V$ . This may be suggests the presence of the difference basic karyotype in *Phragmipedium*. However, the species with the karyotype of  $2n=17=17V$  has not been found. Therefore, we assume that the karyotype of  $2n=17$  is derive not from the different basic karyotype but from the loss of telocentric chromosomes after centric fission, like *Paphiopedilum* (Karasawa 1979).

*P. longifolium* var. *gracile* ( $2n=20+1f$ ), *P. roezlii* ( $2n=20+2f$ ) and *P. longifolium* ( $2n=20+3f$ ) had small fragment chromosomes of different number, while the morphology of rest 20 chromosomes were almost similar to those of *P. hartwegii* ( $2n=20$ ). Consequently, the karyomorphological difference among them was due to the number of fragment chromosomes.

## II. Cytotaxonomical comparisons in the species of *Phragmipedium*

The genus *Phragmipedium* had been classified into three subgenus and 11 species by Brieger (1971), while it has been classified into five sections, 12 species and one variety by Fowlie (1970). Recently Garay (1979) rearranged into five sections and 21 species. In the present study, karyomorphological relationships in this genus will be discussed according to Garay's classification.

### (1) MICROPETALUM

*P. schlimii* is only one species in this section. The sepal and petals of this species are oval and their top are round. The chromosome number of  $2n=30$  is the largest in this genus except for *P. x stenophyllum*, and the complement consists of two large metacentric chromosomes and 28 small chromosomes which are divided into four metacentric and 24 telocentric chromosomes. Thus, the karyotype of this species can be expressed as  $2n=30=6V+24I$ , and it could be converted into  $2n=18V$ . Satellites being at the distal region of the telocentric chromosomes are characteristics of this species.

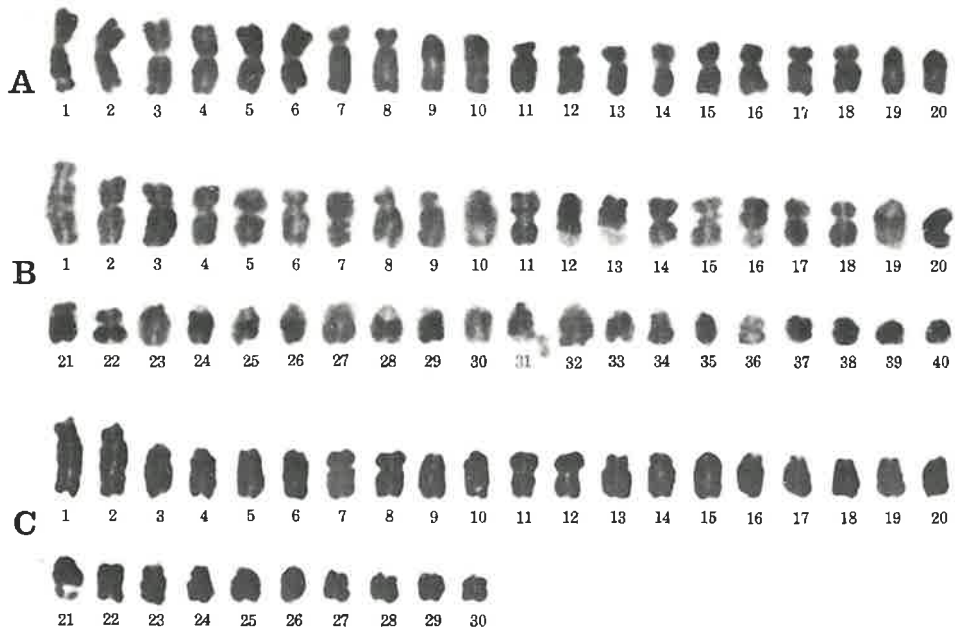


Fig. 20. Metaphase chromosomes of three species of *Phragmipedium*. A, *P. hartwegii*  $2n=20$ . B, *P. x stenophyllum*  $2n=40$ . C, *P. schlimii*  $2n=30$ .  $\times 1800$ .

The large-sized species which has been recognized as *P. x stenophyllum* by Garay (1979) is bigger than *P. schlimii*. The karyomorphological characteristics are as follows; Figure 20 shows the metaphase chromosomes of *P. hartwegii* ( $2n=20$ ), *P. x stenophyllum* ( $2n=40$ ) and *P. schlimii* ( $2n=30$ ). Among the 40 chromosomes of *P. x stenophyllum*, 24 chromosomes (Nos. 17-40) were similar to the 24 chromosomes (Nos. 7-30) of *P. schlimii*. Six chromosomes (Nos. 1, 3, 9, 10, 12 and 13) in the other 16 chromosomes were similar to the six chromosomes (Nos. 1-6) of *P. schlimii* and the rest 10 chromosomes which consisted of one metacentric chromosome with satellite (No. 2), seven metacentric chromosomes (Nos. 4, 5, 6, 8, 11, 14 and 15) and two telocentric chromosomes (Nos. 7, 16) were similar to a half set of the complement of *P. hartwegii* ( $2n=20$ ).

Namely the chromosome complement of *P. x stenophyllum* might be consist of one set of the complement of *P. schlimii* and a half set of the complement of *P. hartwegii* or another  $2n=20$  species. Thus we assume that *P. x stenophyllum* is complex triploid as same as hybrid of *Paphiopedilum* (Atwood 1980).

## (2) PLATIPETALUM

The species of section Platypetalum have an oval dorsal sepal, and petals are not sharp and longer than dorsal sepal. Two species of this section, *P. lindleyanum* and *P. sargentianum*, had the same chromosome number of  $2n=22$ , and their karyotype were similar to each other.

The complements of these species consisted of 11 pairs of matched chromosomes, and could be divided into two groups showing a bimodal karyotype. The karyotypes of these species can be expressed as  $2n=22=12V+10I$  and can be converted into  $2n=17V$ . This is the characteristic of this section and differ from other sections.

## (3) PHRAGMIPEDIUM

The ribbon-type petals are morphological characteristic of the species of this section. Fowlie (1970) recognized one species and one variety in this section; *P. caudatum* and *P. caudatum* var. *lindenii*, while Garay (1979) recognized four species by the difference of staminode; *P. caudatum*, *P. wallisii*, *P. warscewizii* and *P. lindenii*.

The chromosome numbers of three species, *P. caudatum*, *P. wallisii* and *P. lindenii*, were all  $2n=28$ , and the karyotype of these species were similar to each other. Therefore, we could not distinguish karyomorphologically among them.

The karyotype of these species could be expressed as  $2n=28=8V+20 I$  and could be converted into  $2n=18V$ .

## (4) HIMANTOPETALUM

The difference between Himantopetalum and following Lorifolia is the shape of lip. The opening of lip of this section had not a pair of hornlike protuberances on side.

The karyotype of *P. ecuadorensis*  $2n=22$  are extremely similar to that of *P. hartwegii*  $2n=20$  which is included section Lorifolia. The karyotype of *P. ecuadorensis* can be expressed as  $2n=22=14V+8I$  and can be converted into  $2n=18V$ .

Two types of *P. pearcei* which were presented by Garay (1979) were different from each other in the color and the shape of flowers. Chromosome numbers of these types were  $2n=20$ , 21 and 22, respectively, and the chromosome complements of them consisted of heterogenous pairs suggesting hybrids.

Figure 21 shows the metaphase chromosomes of *P. boisseranum* ( $2n=18$ ), *P. pearcei* Type 1 ( $2n=20$ ) and *P. ecuadorensis* ( $2n=22$ ). The largest chromosome (No. 1) and the satellited chromosome (No. 5) of *P. pearcei* Type 1 were similar to the largest chromosome (No. 1 or 2) and satellited chromosome (No. 7 or 8) of *P. boisseranum*, while the meta-centric chromosome with satellite (No. 2) and small telocentric chromosomes (Nos. 7, 8, 9 and 20) of *P. pearcei* Type 1 were similar to those of No. 1 or 2 and No. 15 or 16, 17 or 18, 19 or 20 and 21 or 22 of *P. ecuadorensis* ( $2n=22$ ).

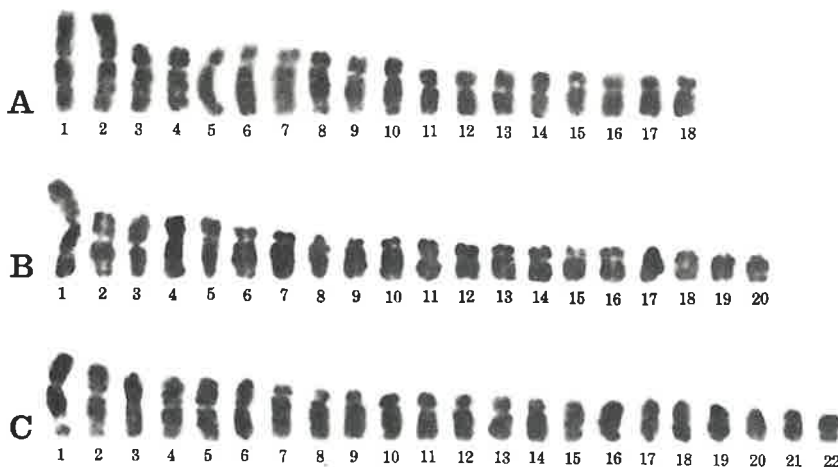


Fig. 21. Metaphase chromosomes of three species of *Phragmipedium*. A, *P. boisseranum*  $2n=18$ . B, *P. pearcei* Type 1  $2n=20$ . C, *P. ecuadorensis*  $2n=22$ .  $\times 1800$ .

Namely, the chromosome complement of *P. pearcei* Type 1 might consisted of a half set of the complement of both *P. boisseranum* and *P. ecuadorensis*. Thus we assume that *P. pearcei* Type 1 is F1 hybrid between *P. boisseranum* and *P. ecuadorensis*.

The chromosome complements of *P. pearcei* Type 2 ( $2n=21$ ) and Type 3 ( $2n=22$ ) also showed heterogenous karyotype. These two types did not have any large metacentric chromosomes but had medium-sized metacentric chromosomes with satellite and four or five small telocentric chromosomes. According to their karyomorphological characteris-



tics, we assume that these two types are hybrids originated to the  $2n=20$  species (e. g. *P. hartwegii*) and the  $2n=22$  species (e. g. *P. ecuadorensis*).

Thus the species which is classified as *P. pearcei* may be hybrid group.

### (5) LORIFOLIA

The morphological characteristic of this section is the opening of lip with a pair of horn-like protuberances on side.

*P. boisseranum*  $2n=18$  and *P. vittatum*  $2n=18$  which have the lowest chromosome number in this genus are included in this section. The complement of *P. boisseranum* consisted of nine pairs of matched chromosomes, and could be divided into three groups showing a trimodal karyotype: The first consisted of two large chromosomes and the second eight medium-sized chromosomes and the third eight small chromosomes. The position of centromeres were median, submedian and subterminal (Fig. 13E).

The karyotype of *P. vittatum*  $2n=18$  showed a trimodal karyotype similar to *P. boisseranum*, except for centromeric position of each chromosome.

The complement of *P. hartwegii*  $2n=20$  consisted of 16 metacentric and four telocentric chromosomes, and showed a gradient karyotype (Fig. 15D-F). The karyotype of this species can be expressed as  $2n=20=16V+4I$  and can be converted into  $2n=18V$ , showing a trimodal karyotype.

Three specimen, *P. longifolium*  $2n=20+3f$ , *P. roezlii*  $2n=20+2f$  and *P. longifolium* var. *gracile*  $2n=20+1f$ , are very similar to *P. hartwegii*  $2n=20$  in external morphology, and their karyotype of 20 chromosomes (Nos. 1-20) except for fragment chromosomes were also similar.

As a conclusion of karyomorphological analysis in the *Phragmipedium*, each distinct karyotype is found to be useful for not only the validation species but also sectioning of genus. Especially section Himantopetalum and Lorifolia are closely related in the morphological characteristics. Thus, the phylogenetic differentiation of *Phragmipedium* can be considered as follow: The species with the chromosome number of  $2n=18$  might be the most primitive. Karyomorphological differentiation in this genus might be led mainly by centric fission of chromosomes and partly by certain structural changes, e. g. chromosome loss.

### Summary

1. Karyomorphological investigations were carried out in 15 taxa of *Phragmipedium*.
2. The chromosome numbers of 15 taxa investigated are as follows: *P. schlimii*  $2n=30$ , *P. x stenophyllum*  $2n=40$ , *P. lindleyanum*  $2n=22$ , *P. sargentianum*  $2n=22$ , *P. wallisii*  $2n=28$ , *P. lindenii*  $2n=28$ , *P. ecuadorensis*  $2n=22$ , *P. pearcei*  $2n=20, 21, 22$ , *P. vittatum*  $2n=18$ , *P. longifolium*  $2n=23 (=20+3f)$ , *P. longifolium* var. *gracile*  $2n=21 (=20+1f)$  and

- P. roezlii*  $2n=22$  ( $=20+2f$ ); (new count), *P. caudatum*  $2n=28$ ; (revised count), *P. boisseranum*  $2n=18$  and *P. hartwegii*  $2n=20$ ; (confirmed count).
3. *P. boisseranum* and *P. vittatum* with the lowest chromosome numbers of  $2n=18$  showed trimodal karyotype: The first group consisted of two large metacentric chromosomes and the second eight medium-sized metacentric chromosomes and the third eight small metacentric chromosomes.
  4. The species with the chromosome number  $2n=20, 22, 28$  and  $30$  including sect. Micropetalum, Phragmipedium, Himantopetalum and Lorifolia, showed the decrease of metacentric chromosomes and the increase of telocentric chromosomes. It might be caused directly by centric fission on a pair of metacentric chromosomes.
  5. Converting every two telocentric chromosomes into one metacentric chromosome, the chromosome numbers of the species of section Micropetalum, Phragmipedium, Himantopetalum and Lorifolia, can be connected to  $2n=18$ , while the chromosome number of  $2n=22$  of the species of section Platypetalum can be connected to  $2n=17$ . It is assumed that the species with the chromosome number of  $2n=22$  in section Platypetalum is derived from the loss of telocentric chromosomes after centric fission.
  6. The chromosome numbers of three types of *P. pearcei*, section Himantopetalum, of which color and shape of flowers were slightly different were examined as  $2n=20, 21$  and  $22$ , respectively. These types were considered to be hybrids because the karyotypes of their chromosomes were heterogenous. By the comparison of karyotype among three types, it was found that the  $2n=20$  type and the  $2n=21, 22$  types were derived from different parental species.
  7. *P. x stenophyllum* ( $2n=40$ ) was considered to be the complex triploid originated from the 30 chromosomes of *P. schilimii* ( $2n=30$ ) and the 10 chromosomes of *P. hartwegii* ( $2n=20$ ).
  8. Fragment chromosomes were observed in *P. longifolium*, *P. longifolium* var. *gracile* and *P. roezlii* including section Lorifolia. The 20 chromosomes except for fragment chromosomes of these three taxa were very similar to those of *P. hartwegii*.
  9. All species of *Phragmipedium* showed distinct karyotypes, and section Himantopetalum was resemble to section Lorifolia, karyomorphologically.
  10. The phylogenetic differentiation of *Phragmipedium* was considered as follows: The species with  $2n=18$  might be the most primitive. Karyomorphological differentiation in this genus might be led mainly by centric fission of chromosomes and partly by certain structural changes, e. g. chromosome loss.

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Table 2. Measurements of somatic chromosomes of *Phragmipedium schlimii* at metaphase,  $2n=30$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	1.3+4.5= 5.8	6.0	3.5	s t
2	1.3+3.9= 5.2	5.4	3.0	sm
3	d+4.0= 4.0	4.1	$<\infty$	t
4	d+3.7= 3.7	3.8	$<\infty$	t
5	d+3.6= 3.6	3.7	$<\infty$	t
6	d+3.6= 3.6	3.7	$<\infty$	t
7	1.2+2.3= 3.5	3.6	1.9	sm
8	1.2+2.3= 3.5	3.6	1.9	sm
9	d+3.4= 3.4	3.5	$<\infty$	t
10	d+3.4= 3.4	3.5	$<\infty$	t
11	1.3+2.0= 3.3	3.4	1.5	m
12	1.3+2.0= 3.3	3.4	1.5	m
13	d+3.3= 3.3	3.4	$<\infty$	t
14	d+3.3= 3.3	3.4	$<\infty$	t
15	d+3.3= 3.3	3.4	$<\infty$	t
16	d+3.3= 3.3	3.4	$<\infty$	t
17	d+3.3= 3.3	3.4	$<\infty$	t
18	d+3.0= 3.0	3.1	$<\infty$	t
19	d+2.9= 2.9	3.0	$<\infty$	t
20	d+2.9= 2.9	3.0	$<\infty$	t
21	d+2.0+0.9=2.9*	3.0	$<\infty$	t
22	d+2.0+0.9=2.9*	3.0	$<\infty$	t
23	d+2.9= 2.9	3.0	$<\infty$	t
24	d+2.9= 2.9	3.0	$<\infty$	t
25	d+2.7= 2.7	2.8	$<\infty$	t
26	d+2.5= 2.5	2.6	$<\infty$	t
27	d+2.3= 2.3	2.4	$<\infty$	t
28	d+2.1= 2.1	2.2	$<\infty$	t
29	d+2.1= 2.1	2.2	$<\infty$	t
30	d+2.1= 2.1	2.2	$<\infty$	t

\* Chromosome with secondary constriction  
d : dot

Table 3. Measurements of somatic chromosomes of *Phragmipedium*  
*× stenophyllum* at metaphase,  $2n=40$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	1.4+4.6= 6.0	4.4	3.3	s t
2	2.1+2.3+0.7=5.1 *	3.8	1.4	m
3	1.3+3.6= 4.9	3.7	2.9	sm
4	1.9+2.4= 4.3	3.2	1.3	m
5	1.8+2.3= 4.1	3.0	1.3	m
6	1.2+2.9= 4.1	3.0	2.4	sm
7	d+4.0= 4.0	3.0	< $\infty$	t
8	1.3+2.7= 4.0	3.0	2.1	sm
9	d+4.0= 4.0	3.0	< $\infty$	t
10	d+4.0= 4.0	3.0	< $\infty$	t
11	1.8+2.2= 4.0	3.0	1.2	m
12	d+3.9= 3.9	2.9	< $\infty$	t
13	d+3.9= 3.9	2.9	< $\infty$	t
14	1.5+2.1= 3.6	2.7	1.4	m
15	1.2+2.4= 3.6	2.7	2.0	sm
16	d+3.6= 3.6	2.7	< $\infty$	t
17	1.2+2.3= 3.5	2.6	1.9	sm
18	1.2+2.3= 3.5	2.6	1.9	sm
19	d+3.4= 3.4	2.5	< $\infty$	t
20	d+3.3= 3.3	2.4	< $\infty$	t
21	1.3+2.0= 3.3	2.4	1.5	m
22	1.3+2.0= 3.3	2.4	1.5	m
23	d+3.0= 3.0	2.2	< $\infty$	t
24	d+3.0= 3.0	2.2	< $\infty$	t
25	d+2.9= 2.9	2.1	< $\infty$	t
26	d+2.9= 2.9	2.1	< $\infty$	t
27	d+2.9= 2.9	2.1	< $\infty$	t
28	d+2.9= 2.9	2.1	< $\infty$	t
29	d+2.8= 2.8	2.1	< $\infty$	t
30	d+2.8= 2.8	2.1	< $\infty$	t
31	d+2.0+0.8=2.8 *	2.1	< $\infty$	t
32	d+2.0+0.8=2.8 *	2.1	< $\infty$	t
33	d+2.8= 2.8	2.1	< $\infty$	t
34	d+2.8= 2.8	2.1	< $\infty$	t
35	d+2.5= 2.5	1.8	< $\infty$	t
36	d+2.5= 2.5	1.8	< $\infty$	t
37	d+2.2= 2.2	1.6	< $\infty$	t
38	d+2.2= 2.2	1.6	< $\infty$	t
39	d+2.0= 2.0	1.5	< $\infty$	t
40	d+2.0= 2.0	1.5	< $\infty$	t

\* Chromosome with secondary constriction  
d : dot

Table 4. Measurements of somatic chromosomes of *Phragmipedium lindleyanum* at metaphase,  $2n=22$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	2.5+3.2= 5.7	6.6	1.3	m
2	2.5+3.2= 5.7	6.6	1.3	m
3	2.4+1.8+1.1=5.3 *	6.2	1.2	m
4	2.4+1.8+1.1=5.3 *	6.2	1.2	m
5	1.3+3.9= 5.2	6.1	3.0	sm
6	1.3+3.9= 5.2	6.1	3.0	sm
7	2.4+2.8= 5.2	6.1	1.2	m
8	2.4+2.8= 5.2	6.1	1.2	m
9	1.3+2.9= 4.2	4.9	2.2	sm
10	1.3+2.9= 4.2	4.9	2.2	sm
11	d+3.4= 3.4	4.0	$<\infty$	t
12	d+3.1= 3.1	3.6	$<\infty$	t
13	1.2+1.9= 3.1	3.6	1.6	m
14	1.2+1.9= 3.1	3.6	1.6	m
15	d+2.9= 2.9	3.4	$<\infty$	t
16	d+2.9= 2.9	3.4	$<\infty$	t
17	d+2.8= 2.8	3.3	$<\infty$	t
18	d+2.8= 2.8	3.3	$<\infty$	t
19	d+2.7= 2.7	3.1	$<\infty$	t
20	d+2.7= 2.7	3.1	$<\infty$	t
21	d+2.6= 2.6	3.0	$<\infty$	t
22	d+2.6= 2.6	3.0	$<\infty$	t

\* Chromosome with secondary constriction  
d : dot

Table 5. Measurements of somatic chromosomes of *Phragmipedium sargentianum* at metaphase,  $2n=22$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	2.6+3.3= 5.9	6.8	1.3	m
2	2.6+3.3= 5.9	6.8	1.3	m
3	2.2+2.2+1.0=5.4 *	6.2	1.5	m
4	2.2+2.2+1.0=5.4 *	6.2	1.5	m
5	1.4+4.0= 5.4	6.2	2.9	sm
6	1.4+4.0= 5.4	6.2	2.9	sm
7	2.3+2.5= 4.8	5.5	1.1	m
8	2.3+2.5= 4.8	5.5	1.1	m

Table 5. (continued)

9	1.5+3.1= 4.6	5.3	2.1	sm
10	1.5+3.1= 4.6	5.3	2.1	sm
11	d+3.5= 3.5	4.0	<∞	t
12	d+3.2= 3.2	3.7	<∞	t
13	1.2+1.7= 2.9	3.3	1.4	m
14	1.2+1.7= 2.9	3.3	1.4	m
15	d+2.9= 2.9	3.3	<∞	t
16	d+2.9= 2.9	3.3	<∞	t
17	d+2.8= 2.8	3.2	<∞	t
18	d+2.8= 2.8	3.2	<∞	t
19	d+2.8= 2.8	3.2	<∞	t
20	d+2.8= 2.8	3.2	<∞	t
21	d+2.5= 2.5	2.9	<∞	t
22	d+2.4= 2.4	2.8	<∞	t

\* Chromosome with secondary constriction  
d: dot

Table 6. Measurements of somatic chromosomes of *Phragmipedium caudatum* at metaphase, 2n=28

Chromosome	Length (μm)	Relative length	Arm ratio	Form
1	3.4+2.9+0.7=7.0 *	6.9	1.1	m
2	3.4+2.9+0.7=7.0 *	6.9	1.1	m
3	d+4.5= 4.5	4.5	<∞	t
4	d+4.3= 4.3	4.3	<∞	t
5	d+4.2= 4.2	4.2	<∞	t
6	d+4.0= 4.0	4.0	<∞	t
7	d+3.9= 3.9	3.9	<∞	t
8	d+3.9= 3.9	3.9	<∞	t
9	d+3.8= 3.8	3.8	<∞	t
10	d+3.7= 3.7	3.7	<∞	t
11	0.7+2.8= 3.5	3.5	4.0	st
12	0.7+2.8= 3.5	3.5	4.0	st
13	d+3.3= 3.3	3.3	<∞	t
14	d+3.3= 3.3	3.3	<∞	t
15	d+3.3= 3.3	3.3	<∞	t
16	d+3.3= 3.3	3.3	<∞	t
17	d+3.1= 3.1	3.1	<∞	t
18	d+3.1= 3.1	3.1	<∞	t

Table 6. (continued)

19	0.7+2.4=	3.1	3.1	3.4	st
20	0.7+2.4=	3.1	3.1	3.4	st
21	d+2.9=	2.9	2.9	<∞	t
22	d+2.9=	2.9	2.9	<∞	t
23	0.9+2.0=	2.9	2.9	2.2	sm
24	0.9+2.0=	2.9	2.9	2.2	sm
25	d+2.7=	2.7	2.7	<∞	t
26	d+2.7=	2.7	2.7	<∞	t
27	d+2.6=	2.6	2.6	<∞	t
28	d+2.5=	2.5	2.5	<∞	t

\* Chromosome with secondary constriction

d : dot

Table 7. Measurements of somatic chromosomes of *Phragmipedium wallisii* at metaphase, 2n=28

Chromosome	Length (μm)	Relative length	Arm ratio	Form
1	3.0+2.5+1.0=6.5 *	6.5	1.2	m
2	3.0+2.5+1.0=6.5 *	6.5	1.2	m
3	d+4.1=	4.1	<∞	t
4	d+4.1=	4.1	<∞	t
5	d+3.8=	3.8	<∞	t
6	d+3.8=	3.8	<∞	t
7	d+3.8=	3.8	<∞	t
8	d+3.8=	3.8	<∞	t
9	d+3.7=	3.7	<∞	t
10	d+3.7=	3.7	<∞	t
11	0.9+2.8=	3.7	3.1	st
12	0.9+2.8=	3.7	3.1	st
13	d+3.5=	3.5	<∞	t
14	d+3.5=	3.5	<∞	t
15	d+3.3=	3.3	<∞	t
16	d+3.3=	3.3	<∞	t
17	d+3.0=	3.0	<∞	t
18	d+3.0=	3.0	<∞	t
19	0.7+2.3=	3.0	3.3	st
20	0.7+2.3=	3.0	3.3	st
21	d+3.0=	3.0	<∞	t
22	d+3.0=	3.0	<∞	t



Table 7. (continued)

23	0.9+2.1= 3.0	3.0	2.3	sm
24	0.9+2.1= 3.0	3.0	2.3	sm
25	d+2.9= 2.9	2.9	<∞	t
26	d+2.9= 2.9	2.9	<∞	t
27	d+2.4= 2.4	2.4	<∞	t
28	d+2.3= 2.3	2.3	<∞	t

\* Chromosome with secondary constriction

d : dot

Table 8. Measurements of somatic chromosomes of *Phragmipedium lindenii* at metaphase, 2n=28

Chromosome	Length (μm)	Relative length	Arm ratio	Form
1	3.0+2.9+0.8=6.7 *	6.4	1.2	m
2	3.0+2.9+0.8=6.7 *	6.4	1.2	m
3	d+4.7= 4.7	4.5	<∞	t
4	d+4.5= 4.5	4.3	<∞	t
5	d+4.4= 4.4	4.2	<∞	t
6	d+4.3= 4.3	4.1	<∞	t
7	d+4.0= 4.0	3.8	<∞	t
8	d+3.9= 3.9	3.7	<∞	t
9	d+3.7= 3.7	3.5	<∞	t
10	d+3.7= 3.7	3.5	<∞	t
11	0.7+2.9= 3.6	3.4	4.1	st
12	0.7+2.9= 3.6	3.4	4.1	st
13	d+3.6= 3.6	3.4	<∞	t
14	d+3.6= 3.6	3.4	<∞	t
15	d+3.6= 3.6	3.4	<∞	t
16	d+3.6= 3.6	3.4	<∞	t
17	d+3.3= 3.3	3.2	<∞	t
18	d+3.3= 3.3	3.2	<∞	t
19	0.8+2.5= 3.3	3.2	3.1	st
20	0.8+2.5= 3.3	3.2	3.1	st
21	d+3.3= 3.3	3.2	<∞	t
22	d+3.3= 3.3	3.2	<∞	t
23	0.8+2.2= 3.0	2.9	2.8	sm
24	0.8+2.2= 3.0	2.9	2.8	sm
25	d+2.7= 2.7	2.6	<∞	t
26	d+2.7= 2.7	2.6	<∞	t

Table 8. (continued)

27	d+2.7= 2.7	2.6	<∞	t
28	d+2.4= 2.4	2.3	<∞	t

\* Chromosome with secondary constriction

d : dot

Table 9. Measurements of somatic chromosomes of *Phragmipedium ecuadorensis* at metaphase, 2n=22

Chromosome	Length (μm)	Relative length	Arm ratio	Form
1	2.3+2.5+0.8=5.6 *	6.6	1.4	m
2	2.2+2.5+0.8=5.5 *	6.5	1.5	m
3	2.3+2.8= 5.1	6.0	1.2	m
4	2.3+2.8= 5.1	6.0	1.2	m
5	2.3+2.6= 4.9	5.8	1.1	m
6	2.3+2.6= 4.9	5.8	1.1	m
7	1.0+3.3= 4.3	5.1	3.3	st
8	1.0+3.2= 4.2	5.0	3.2	st
9	1.0+2.8= 3.8	4.5	2.8	sm
10	1.0+2.8= 3.8	4.5	2.8	sm
11	0.9+2.6= 3.5	4.2	2.9	sm
12	0.9+2.6= 3.5	4.2	2.9	sm
13	1.3+2.1= 3.4	4.0	1.6	m
14	1.3+2.1= 3.4	4.0	1.6	m
15	d+3.3= 3.3	3.9	<∞	t
16	d+3.3= 3.3	3.9	<∞	t
17	d+3.2= 3.2	3.8	<∞	t
18	d+3.2= 3.2	3.8	<∞	t
19	d+2.7= 2.7	3.2	<∞	t
20	d+2.7= 2.7	3.2	<∞	t
21	d+2.5= 2.5	3.0	<∞	t
22	d+2.4= 2.4	2.8	<∞	t

\* Chromosome with secondary constriction

d : dot

Table 10. Measurements of somatic chromosomes of *Phragmipedium pearcei* Type 1. at metaphase,  $2n=20$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$3.3+3.9= 7.2$	10.9	1.2	m
2	$1.9+1.7+0.8=4.4 *$	6.7	1.3	m
3	$2.1+2.2= 4.3$	6.5	1.0	m
4	$1.3+3.0= 4.3$	6.5	2.3	sm
5	$1.2+2.3+0.8=4.3 *$	6.5	2.6	sm
6	$1.1+2.6= 3.7$	5.6	2.4	sm
7	$0.9+2.6= 3.5$	5.3	2.9	sm
8	$1.3+1.8= 3.1$	4.7	1.4	m
9	$0.8+2.2= 3.0$	4.6	2.8	sm
10	$0.8+2.2= 3.0$	4.6	2.8	sm
11	$1.4+1.5= 2.9$	4.4	1.1	m
12	$1.1+1.6= 2.7$	4.1	1.5	m
13	$0.9+1.8= 2.7$	4.1	2.0	sm
14	$1.2+1.5= 2.7$	4.1	1.3	m
15	$1.1+1.6= 2.7$	4.1	1.5	m
16	$1.0+1.5= 2.5$	3.8	1.5	m
17	$d+2.4= 2.4$	3.6	$<\infty$	t
18	$d+2.3= 2.3$	3.5	$<\infty$	t
19	$d+2.1= 2.1$	3.2	$<\infty$	t
20	$d+2.1= 2.1$	3.2	$<\infty$	t

\* Chromosome with secondary constriction  
d : dot

Table 11. Measurements of somatic chromosomes of *Phragmipedium pearcei* Type 2. at metaphase,  $2n=21$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$2.3+2.3+0.7=5.3 *$	6.8	1.3	m
2	$2.3+2.3+0.7=5.3 *$	6.8	1.3	m
3	$2.3+2.6= 4.9$	6.3	1.1	m
4	$2.2+2.6= 4.8$	6.2	1.2	m
5	$1.9+2.5= 4.4$	5.7	1.3	m
6	$1.9+2.3= 4.2$	5.4	1.2	m
7	$1.1+2.7= 3.8$	4.9	2.5	sm
8	$1.0+2.7= 3.7$	4.8	2.7	sm
9	$1.7+2.0= 3.7$	4.8	1.2	m
10	$0.9+2.7= 3.6$	4.6	3.0	sm
11	$1.0+2.6= 3.6$	4.6	2.6	sm

Table 11. (continued)

12	1.0+2.6=	3.6	4.6	2.6	sm
13	1.2+2.1=	3.3	4.3	1.8	sm
14	1.2+2.1=	3.3	4.3	1.8	sm
15	1.3+1.9=	3.2	4.1	1.5	m
16	1.3+1.9=	3.2	4.1	1.5	m
17	d+3.2=	3.2	4.1	< $\infty$	t
18	1.2+1.8=	3.0	3.9	1.5	m
19	d+2.8=	2.8	3.6	< $\infty$	t
20	d+2.3=	2.3	3.0	< $\infty$	t
21	d+2.3=	2.3	3.0	< $\infty$	t

\* Chromosome with secondary constriction

d : dot

Table 12. Measurements of somatic chromosomes of *Paphiopedilum pearcei* Type 3. at metaphase, 2n=22

Chromosome	Length ( $\mu$ m)	Relative length	Arm ratio	Form	
1	2.2+2.4+0.7=5.3*	6.8	1.4	m	
2	2.2+2.4+0.7=5.3*	6.8	1.4	m	
3	2.2+2.5=	4.7	6.1	1.1	m
4	2.0+2.3=	4.3	5.6	1.2	m
5	2.0+2.3=	4.3	5.6	1.2	m
6	1.0+3.3=	4.3	5.6	3.3	s t
7	1.7+2.3=	4.0	5.2	1.4	m
8	1.1+2.5=	3.6	4.7	2.3	sm
9	1.2+2.3=	3.5	4.5	1.9	sm
10	1.0+2.4=	3.4	4.4	2.4	sm
11	1.1+2.3=	3.4	4.4	2.1	sm
12	1.0+2.4=	3.4	4.4	2.4	sm
13	1.3+2.1=	3.4	4.4	1.6	m
14	0.7+2.5=	3.2	4.1	3.6	s t
15	1.0+1.9=	2.9	3.7	1.9	sm
16	1.2+1.7=	2.9	3.7	1.4	m
17	d+2.9=	2.9	3.7	< $\infty$	t
18	d+2.7=	2.7	3.5	< $\infty$	t
19	d+2.7=	2.7	3.5	< $\infty$	t
20	0.7+1.9=	2.6	3.4	2.7	sm
21	d+2.3=	2.3	3.0	< $\infty$	t
22	d+2.3=	2.3	3.0	< $\infty$	t

\* Chromosome with secondary constriction

d : dot

Table 13. Measurements of somatic chromosomes of *Phragmipedium boisseranum* at metaphase,  $2n=18$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$3.4+3.9= 7.3$	9.2	1.1	m
2	$3.4+3.9= 7.3$	9.2	1.1	m
3	$1.7+3.3= 5.0$	6.3	1.9	sm
4	$1.7+3.3= 5.0$	6.3	1.9	sm
5	$1.3+3.4= 4.7$	5.9	2.6	sm
6	$1.3+3.4= 4.7$	5.9	2.6	sm
7	$1.0+2.9+0.8=4.7 *$	5.9	3.7	s t
8	$1.0+2.9+0.8=4.7 *$	5.9	3.7	s t
9	$1.6+2.9= 4.5$	5.7	1.8	sm
10	$1.6+2.9= 4.5$	5.7	1.8	sm
11	$1.3+2.2= 3.5$	4.4	1.7	m
12	$1.3+2.2= 3.5$	4.4	1.7	m
13	$1.6+1.9= 3.5$	4.4	1.2	m
14	$1.6+1.9= 3.5$	4.4	1.2	m
15	$1.3+2.0= 3.3$	4.2	1.5	m
16	$1.3+2.0= 3.3$	4.2	1.5	m
17	$1.1+2.0= 3.1$	3.9	1.8	sm
18	$1.1+2.0= 3.1$	3.9	1.8	sm

\* Chromosome with secondary constriction

Table 14. Measurements of somatic chromosomes of *Phragmipedium vittatum* at metaphase,  $2n=18$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$3.4+4.1= 7.5$	8.4	1.2	m
2	$3.4+4.1= 7.5$	8.4	1.2	m
3	$1.8+3.7= 5.5$	6.2	2.1	sm
4	$1.8+3.7= 5.5$	6.2	2.1	sm
5	$1.7+3.8= 5.5$	6.2	2.3	sm
6	$1.7+3.8= 5.5$	6.2	2.3	sm
7	$1.3+3.2+0.8=5.3 *$	6.0	3.1	s t
8	$1.3+3.2+0.8=5.3 *$	6.0	3.1	s t
9	$1.4+3.5= 4.9$	5.5	2.5	sm
10	$1.4+3.5= 4.9$	5.5	2.5	sm
11	$1.0+3.2= 4.2$	4.7	3.2	s t
12	$1.0+3.2= 4.2$	4.7	3.2	s t
13	$1.7+2.2= 3.9$	4.4	1.3	m

Table 14. (continued)

14	1.8+2.1= 3.9	4.4	1.2	m
15	1.1+2.8= 3.9	4.4	2.5	sm
16	1.1+2.8= 3.9	4.4	2.5	sm
17	1.7+2.1= 3.8	4.3	1.2	m
18	1.7+2.0= 3.7	4.2	1.2	m

\* Chromosome with secondary constriction

Table 15. Measurements of somatic chromosomes of *Phragmipedium hartwegii* at metaphase, 2n=20

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	2.4+2.3+0.7=5.4 *	6.4	1.3	m
2	2.4+2.3+0.7=5.4 *	6.4	1.3	m
3	2.2+2.7= 4.9	5.8	1.2	m
4	2.2+2.7= 4.9	5.8	1.2	m
5	2.3+2.6= 4.9	5.8	1.1	m
6	2.3+2.6= 4.9	5.8	1.1	m
7	1.2+3.6= 4.8	5.7	3.0	sm
8	1.2+3.5= 4.7	5.6	2.9	sm
9	d+4.4= 4.4	5.2	$<\infty$	t
10	d+4.4= 4.4	5.2	$<\infty$	t
11	1.0+2.8= 3.8	4.5	2.8	sm
12	1.0+2.8= 3.8	4.5	2.8	sm
13	1.4+2.3= 3.7	4.4	1.6	m
14	1.4+2.3= 3.7	4.4	1.6	m
15	1.3+2.2= 3.5	4.1	1.7	m
16	1.3+2.2= 3.5	4.1	1.7	m
17	1.1+2.4= 3.5	4.1	2.2	sm
18	1.1+2.4= 3.5	4.1	2.2	sm
19	d+3.5= 3.5	4.1	$<\infty$	t
20	d+3.4= 3.4	4.0	$<\infty$	t

\* Chromosome with secondary constriction

d : dot

Table 16. Measurements of somatic chromosomes of *Phragmipedium longifolium* at metaphase,  $2n=23$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$2.3+2.1+1.0=5.4$ *	6.4	1.3	m
2	$2.3+2.1+1.0=5.4$ *	6.4	1.3	m
3	$2.3+2.7= 5.0$	5.9	1.2	m
4	$2.3+2.7= 5.0$	5.9	1.2	m
5	$2.2+2.6= 4.8$	5.7	1.2	m
6	$2.2+2.6= 4.8$	5.7	1.2	m
7	$1.1+3.3= 4.4$	5.2	3.0	sm
8	$1.1+3.3= 4.4$	5.2	3.0	sm
9	$d+4.1= 4.1$	4.8	$<\infty$	t
10	$d+4.1= 4.1$	4.8	$<\infty$	t
11	$1.3+2.7= 4.0$	4.7	2.1	sm
12	$1.2+2.7= 3.9$	4.6	2.3	sm
13	$1.3+2.2= 3.5$	4.1	1.7	m
14	$1.3+2.2= 3.5$	4.1	1.7	m
15	$1.1+2.1= 3.2$	3.8	1.9	sm
16	$1.1+2.1= 3.2$	3.8	1.9	sm
17	$1.0+2.1= 3.1$	3.7	2.1	sm
18	$0.9+2.1= 3.0$	3.5	2.3	sm
19	$d+3.0= 3.0$	3.5	$<\infty$	t
20	$d+3.0= 3.0$	3.5	$<\infty$	t
21	$d+1.3= 1.3$	1.5	$<\infty$	t
22	$d+1.3= 1.3$	1.5	$<\infty$	t
23	$d+1.2= 1.2$	1.4	$<\infty$	t

\* Chromosome with secondary constriction

d : dot

Table 17. Measurements of somatic chromosomes of *Phragmipedium longifolium* var. *gracile* at metaphase,  $2n=21$ 

Chromosome	Length ( $\mu\text{m}$ )	Relative length	Arm ratio	Form
1	$2.3+2.3+0.9=5.5$ *	6.2	1.4	m
2	$2.3+2.3+0.9=5.5$ *	6.2	1.4	m
3	$2.3+3.0= 5.3$	6.0	1.3	m
4	$2.3+3.0= 5.3$	6.0	1.3	m
5	$2.2+2.5= 4.7$	5.3	1.1	m
6	$2.2+2.5= 4.7$	5.3	1.1	m
7	$1.2+3.3= 4.5$	5.1	2.8	sm

Table 17. (continued)

8	1.2+3.3=	4.5	5.1	2.8	sm
9	d+4.3=	4.3	4.9	<∞	t
10	d+4.3=	4.3	4.9	<∞	t
11	1.1+3.1=	4.2	4.7	2.8	sm
12	1.1+3.1=	4.2	4.7	2.8	sm
13	1.6+2.5=	4.1	4.6	1.6	m
14	1.6+2.5=	4.1	4.6	1.6	m
15	1.3+2.5=	3.8	4.3	1.9	sm
16	1.3+2.5=	3.8	4.3	1.9	sm
17	1.3+2.4=	3.7	4.2	1.8	sm
18	1.3+2.4=	3.7	4.2	1.8	sm
19	d+3.7=	3.7	4.2	<∞	t
20	d+3.7=	3.7	4.2	<∞	t
21	d+1.0=	1.0	1.1	<∞	t

\* Chromosome with secondary constriction

d : dot

Table 18. Measurements of somatic chromosomes of *Phragmipedium roezlii* at metaphase, 2n=22

Chromosome	Length (μm)	Relative length	Arm ratio	Form
1	2.2+2.1+0.9=5.2 *	6.3	1.4	m
2	2.2+2.1+0.9=5.2 *	6.3	1.4	m
3	2.4+2.7= 5.1	6.2	1.1	m
4	2.4+2.7= 5.1	6.2	1.1	m
5	2.4+2.6= 5.0	6.1	1.1	m
6	2.2+2.3= 4.5	5.5	1.0	m
7	1.3+3.2= 4.5	5.5	2.5	sm
8	1.2+3.1= 4.3	5.2	2.6	sm
9	d+4.3= 4.3	5.2	<∞	t
10	d+4.1= 4.1	5.0	<∞	t
11	1.1+2.8= 3.9	4.7	2.5	sm
12	1.0+2.9= 3.9	4.7	2.9	sm
13	1.4+2.0= 3.4	4.1	1.4	m
14	1.4+2.0= 3.4	4.1	1.4	m
15	1.0+2.2= 3.2	3.9	2.2	sm
16	1.0+2.2= 3.2	3.9	2.2	sm
17	1.0+2.0= 3.0	3.6	2.0	sm
18	1.0+2.0= 3.0	3.6	2.0	sm



Table 18. (continued)

19	d+3.0= 3.0	3.6	< $\infty$	t
20	d+2.9= 2.9	3.5	< $\infty$	t
21	d+1.1= 1.1	1.3	< $\infty$	t
22	d+1.0= 1.0	1.2	< $\infty$	t

\* Chromosome with secondary constriction

d : dot