

# A karyomorphological study on four species of *Meconopsis* Vig. (Papaveraceae) from the Hengduan Mountains, SW China

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**Abstract** — A karyomorphological analysis on four species of *Meconopsis* (Papaveraceae) from the Hengduan Mountains, SW China was carried out for the first time. First chromosome counts are notified in two species: *M. racemosa* Maxim ( $2n=56=6m+20sm+30st$ ) and *M. pseudovenusta* Tayl. ( $2n=56=4m+36sm+16st$ ). Furthermore, the somatic chromosome number of *M. borridula* Hook.f.et Thoms. ( $2n=56=4m+16sm+36st$ ) is also confirmed. For *M. integrifolia* (Maxim.) Franch is presented an unreported chromosome number ( $2n=76=6m[2sat]+26sm+44st$ ). The karyotypes of all analyzed species belong to 3B type according to Stebbins' asymmetry classification. The most common chromosome base number of  $x=7$  is thought to be tentative with another possibility of  $x=14$ .

**Key words:** chromosome base number, karyotype, *Meconopsis*, Papaveraceae

## INTRODUCTION

*Meconopsis* Vig. (Papaveraceae), the Himalayan poppy, consists of about 49 species distributed primarily in the Himalayan area with one species, *M. cambrica* (L.) Vig., in western Europe (WU and CHUANG 1980; KADEREIT *et al.* 1997). About 38 species occur in China and most of them are distributed in the Hengduan Mountains and Sino-Himalayan region of SW China (WU 1999; WU *et al.* 2003). These are perennial monocarpic or polycarpic herbs with only basal or basal and cauline leaves which grow in forest floor and various alpine habitats between 1800 and 5500 m altitude (KADEREIT *et al.* 1997; JORK and KADEREIT 1995). Comprehensive taxonomic treatments of *Meconopsis* have been produced by TAYLOR (1934) and WU and CHUANG (1980). It was subdivided into two subgenera: Subgen. *Meconopsis* and *Discogyne*, including five sections and nine series mainly based on the characters of its inflorescence, stem, leaf, and root (WU and CHUANG 1980; CHUANG 1981).

Chromosome data are available for 18 *Meconopsis* species (TABLE 1). None of them had been karyomorphologically studied. These observations almost originated from the cultivation in

the Royal Botanical Garden, Edinburgh (RATTER 1968). The chromosome numbers of this genus vary greatly ( $n=14, 16, 28, 41$  or  $42, 37, 59$  or  $60$ ) (Table 1). Although most species of *Meconopsis* are distributed in China, only one species was reported chromosome number from China up to now. It is clear that much was unknown and further studies to obtain basic information were badly needed. In the present paper, we firstly examined the original chromosome numbers with karyotype data of four *Meconopsis* species from Hengduan Mountains, SW China.

## MATERIAL AND METHODS

The species' names, as well as their origins and habitats, are listed in TABLE 2. The plants were collected mainly from Hengduan Mountains, SW China. Voucher specimens are deposited in the Herbarium of the Kunming Institute of Botany, Chinese Academy of Sciences (KUN).

All cytological observations were made from root tips. Seeds were stored for 4 months at 4°C refrigerator. They were soaked overnight in distilled water at room temperature and were allowed to germinate on wet filter paper in petri dishes. Fresh root tips about 1cm long were cut, pretreated in 0.002M 8-hydroxyquinoline at 24°C for 3-4 hours, then fixed and stored in 3:1 ethanol-acetic acid at 4°C for 30 minutes. Prior to

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TABLE 1 — Previously published chromosome counts in *Meconopsis*.

Taxon	<i>n</i>	<i>2n</i>	Reference
Sect. Cambricae			
Ser. Chelidonifoliae			
Prain			
<i>M. chelidonifolia</i> Bur. et Franch.	14		RATTER 1968
Ser. Meconopsis			
<i>M. cambrica</i> (L.) Vig.	14		SUGIURA 1940
	14		RATTER 1968
		28	SAFONOVA 1991
<i>M. gracilipes</i> Tayl.	28		RATTER 1968
<i>M. dhowjii</i> Tayl. ex Hay	28		
<i>M. regia</i> Tayl.	28		
<i>M. longipetiolata</i> Tayl. ex Hay	28		
<i>M. paniculata</i> (D. Don) Prain	28		RATTER 1968
	14		SUGIURA 1940
<i>M. napaulensis</i> DC.	28		RATTER 1968
Sect. Racemosae			
Ser. Grandes Prain			
<i>M. integrifolia</i> (Maxim.) Franch.	37		RATTER 1968
<i>M. villosa</i> (Hook. f.) Tayl.	16		RATTER 1968
<i>M. betonicifolia</i> Franch.	41		RATTER 1968
<i>M. grandis</i> Prain	59-60		RATTER 1968
Ser. Racemosae			
<i>M. latifolia</i> (Prain) Prain	28		RATTER 1968
<i>M. speciosa</i> Prain	14	56	XIE 1999
<i>M. aculeate</i> Royle	28		SUGIURA 1937
			RATTER 1968
		28	JEE <i>et al.</i> 1989
Sect. Simplicifolia			
Ser. Simplicifoliae			
<i>M. simplicifolia</i> (D. Don) Walp.	41-42		RATTER 1968
<i>M. quintuplinervia</i> Regel	42		RATTER 1968
		76 or 84	HUANG <i>et al.</i> 1996
Ser. Delavayanae Tayl.			
<i>M. Delavayi</i> (Franch.) Franch. ex Prain		56	XIE 1999
<i>M. Horridula</i> Hook.f. et Thoms.	28		RATTER 1968

staining, the root tips were hydrolyzed in 1:1 1N HCl: 45% acetic acid at 60°C for 30 seconds, and then were squashed and stained in 1% aceto-orcein. Permanent slides were made by the standard liquid nitrogen method. Observations were made on nuclei at the somatic mitotic interphase, prophase and metaphase, and measurements of chromosome arms were taken from at least five well-spread metaphases. The karyomorphological classification of the resting and mitotic prophase

TABLE 2 — Origins of the material for chromosome investigation in *Meconopsis*.

Taxon	Locality and altitude	Voucher specimens
Sect. Racemosae		
Ser. Grandes Prain		
<i>M. integrifolia</i> (Maxim.) Franch.	Zhongdian, Yunnan, China, 4150m	Xie H.Y. 001
Ser. Racemosae		
<i>M. racemosa</i> Maxim.	Zhongdian, Yunnan, China, 4100m	Xie H.Y. 002
Sect. Simplicifoliae		
Ser. Delavayanae Tayl.		
<i>M. hurridula</i> Hook. f. et Thoms.	Weixi, Yunnan, China, 4100m	Xie H.Y. 003
<i>M. pseudovenusta</i> Tayl.	Zhongdian, Yunnan, China, 4400m	Xie H.Y. 004

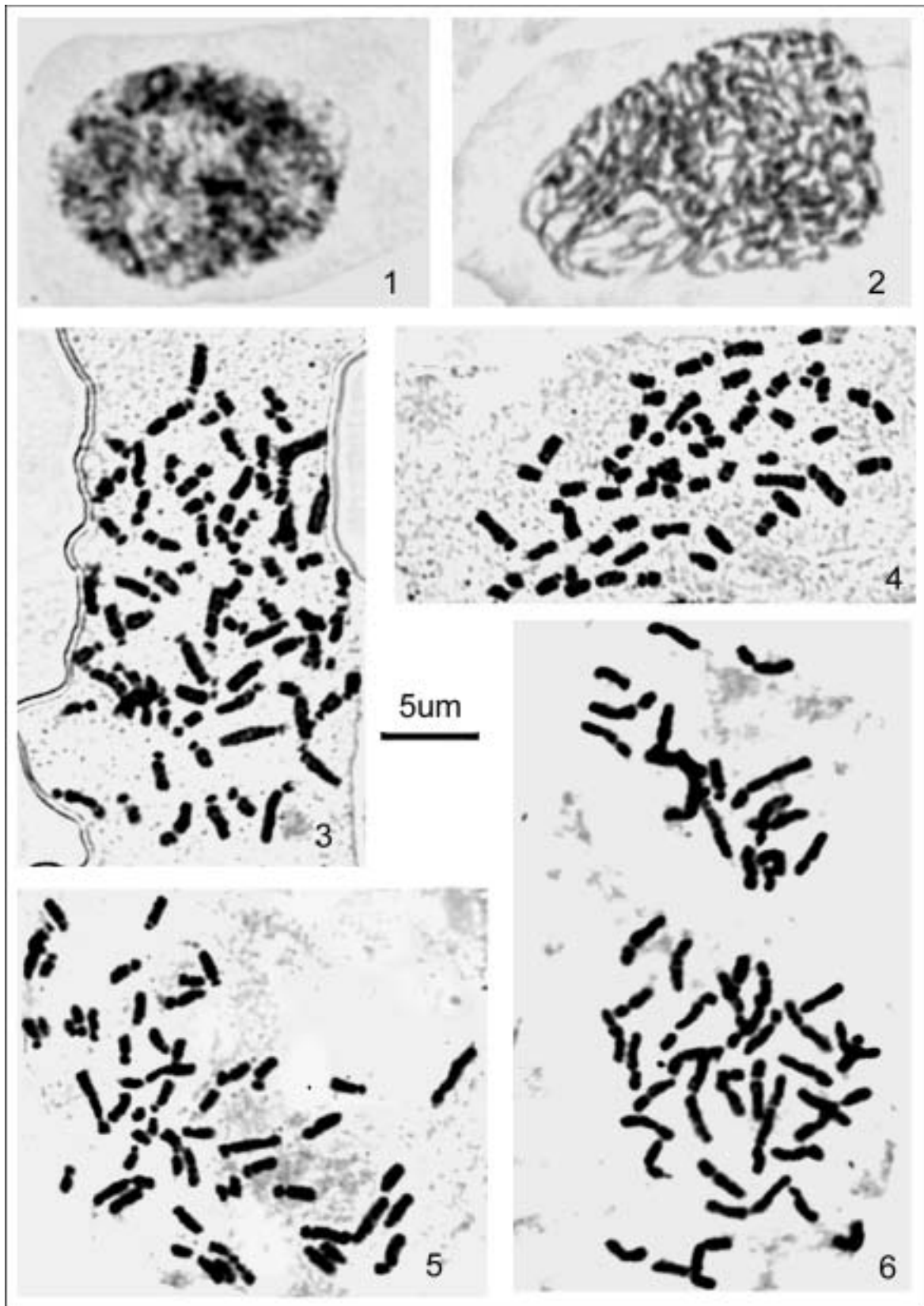
chromosomes follows TANAKA (1971, 1977). The symbols for the description of metaphase chromosomes follow LEVAN *et al.* (1964). The asymmetry of the karyotype is according to STEBBINS (1971).

## RESULTS

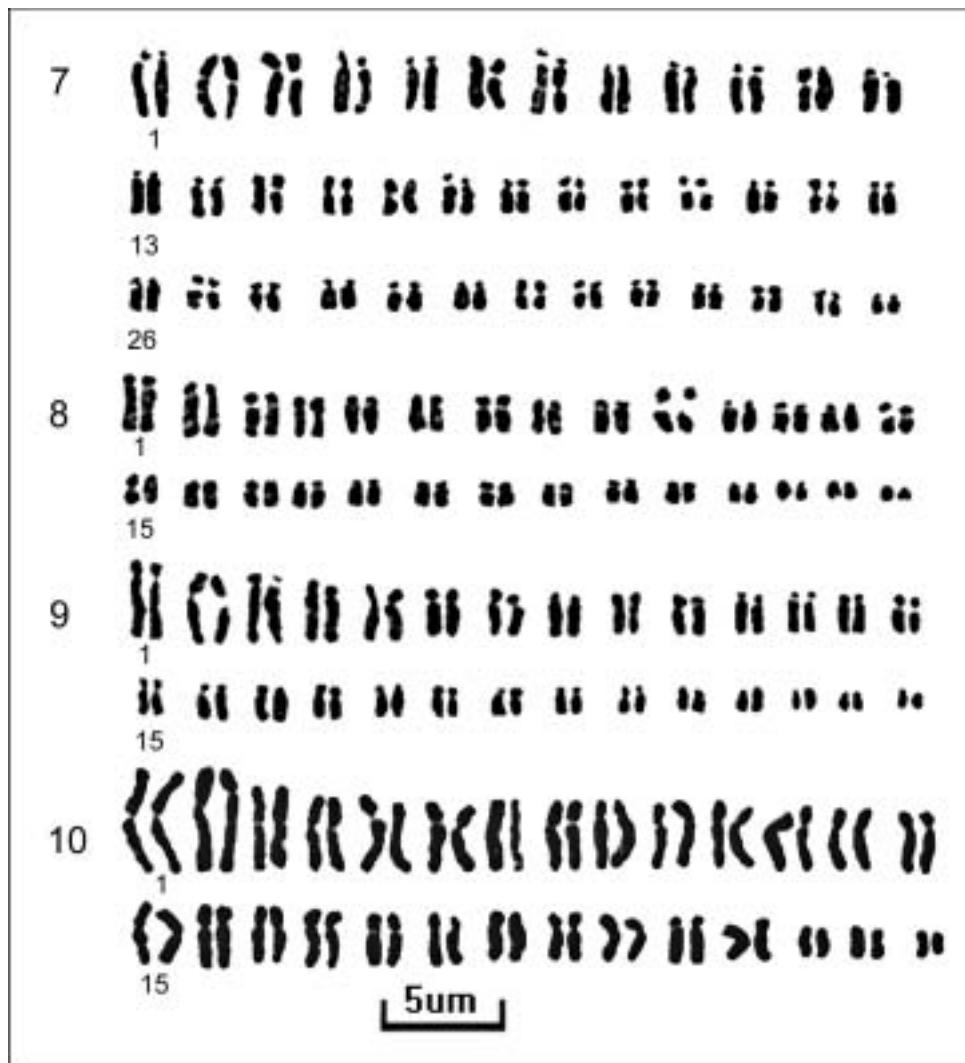
All *Meconopsis* species studied in this investigation belong to the subgenus *Meconopsis*. The chromosomes at resting stage, mitotic prophase, and metaphase of the four species examined are showed in Figs. 1-10. The resting nucleus in somatic cells of all studied species are of the complex chromocenter type and the prophase nucleus in mitotic cells are of the interstitial type. The karyomorphological characteristics are summarized as follows.

1. *Meconopsis integrifolia* (Maxim.) Franch — The chromosome numbers for this species is  $2n=76$ , with a karyotype formula  $2n=76=6m(2sat)+26sm+44st$ . The chromosome number of *M. integrifolia* disagreed with the previous report  $n=37$  (RATTER 1968). The relative lengths of the chromosomes range from 2.25 to 0.68. The 7th pair of chromosomes each has a satellite on the short arm. The ratio of the longest to the shortest chromosome is 3.31 and 82% of the chromosomes had an arm ratio of more than 2.00. The asymmetry of the karyotype is 3B-type.

2. *M. racemosa* Maxim — The chromosome numbers for this taxon is  $2n=56$ . The karyotype formula is  $2n=56=6m+20sm+30st$ . The relative lengths of the chromosomes range from 3.19 to 0.89. The ratio of the longest to the shortest chromosome is 3.58 and 82% of the chromosomes had



Figs. 1-6 — The resting nuclear, prophase and somatic metaphase chromosomes of *Meconopsis*. 1 resting nucleus; 2 prophase; 3 *M. integrifolia*; 4 *M. racesoma*; 5 *M. horridula*; 6 *M. pseudovenusta*.



Figs. 7-10 — Ideograms of somatic metaphase chromosome of *Meconopsis*. 7 *M. integrifolia*; 8 *M. racesoma*; 9 *M. horridula*; 10 *M. pseudovenusta*.

an arm ratio over 2.00. The asymmetry of the karyotype is 3B-type.

3. *M. horridula* Hook.f. et Thoms. — The chromosome number of *M. horridula* ( $2n=56$ ) is consistent with RATTER's (1968) report ( $n=28$ ). The karyotype formula is  $2n=56=4m+16sm+36st$ . The relative lengths of the chromosomes range from 3.44 to 0.88. The ratio of the longest to the shortest chromosome is 3.91 and 89% of the chromosomes had an arm ratio of more than 2.00. The asymmetry of the karyotype is 3B-type.

4. *M. pseudovenusta* Tayl. — The karyotype formula is  $2n=56=4m+36sm+16st$ . The relative lengths of the chromosomes range from 2.91 to 0.90. The ratio of the longest to the shortest chromosome is 3.23 and 75% of the chromosomes had an arm ratio of more than 2.00. The asymmetry of the karyotype is 3B-type.

## DISCUSSION

All *Meconopsis* species that have been cytologically reported belong to the large subgenus *Meconopsis*, the other being subgenus *Discogyne* with only two species (WU and CHUANG 1980). Previous cytological studies indicated some confusion concerning the basic chromosome number of the genus, and some different chromosome number were observed, such as  $n=14$ , 16, 28, 37, 41 or 42, and 59 or 60. For example, the chromosome number found in *M. integrifolia* ( $2n=76$ ) differ substantially from the  $n=37$  determined by RATTER (1968). Although a possible explanation for this disagreement could be the occurrence of aneuploidy reduction from a duodecaploid condition, this type of high haploid chromosome

TABLE 3 — Measurements of somatic chromosome at metaphase in the representative karyotypes in four species of *Meconopsis*.

Chrom.	<i>M. integrifolia</i> 2n=76=6m(2sat)+ 26sm+44st			<i>M. racemosa</i> 2n=56=6m+20sm+30st			<i>M. horridula</i> 2n=56=4m+16sm+36st			<i>M. pseudovenusta</i> 2n=56=4m+36sm+16st		
	RL	AR	PC	RL	AR	PC	RL	AR	PC	RL	AR	PC
1	2.25	6.69	st	3.19	4.95	st	3.44	3.96	st	2.91	1.30	m
2	2.18	1.75	sm	3.08	3.96	st	3.19	1.93	sm	2.87	2.32	sm
3	2.13	2.58	sm	2.44	3.09	st	3.02	6.83	st	2.34	3.09	st
4	2.03	3.33	st	2.33	1.69	m	2.63	5.83	st	2.21	3.00	st
5	1.80	6.06	st	2.25	3.90	st	2.46	2.84	sm	2.09	2.78	sm
6	1.77	2.48	sm	2.16	3.03	st	2.22	3.33	st	2.01	1.37	m
7	1.67	1.27	m*	2.11	2.40	sm	2.05	3.00	st	2.02	2.44	sm
8	1.62	5.33	st	2.03	1.50	m	1.95	4.69	st	2.01	2.63	sm
9	1.63	6.19	st	1.89	4.08	st	1.88	2.67	sm	2.01	1.98	sm
10	1.60	4.14	st	1.89	1.92	sm	1.84	3.33	st	1.90	3.03	st
11	1.50	1.72	sm	1.84	4.67	st	1.82	6.08	st	1.87	4.06	st
12	1.43	5.94	st	1.76	3.71	st	1.80	6.00	st	1.85	5.00	st
13	1.42	6.14	st	1.68	4.17	st	1.78	3.90	st	1.85	1.74	sm
14	1.36	4.33	st	1.68	2.44	sm	1.71	3.03	st	1.83	2.79	sm
15	1.30	1.79	sm	1.73	1.78	sm	1.67	3.33	st	1.8	2.71	sm
16	1.29	1.76	sm	1.62	1.61	m	1.63	5.33	st	1.76	2.15	sm
17	1.26	1.62	m	1.62	2.53	sm	1.63	3.75	st	1.75	2.16	sm
18	1.22	4.07	st	1.57	3.83	st	1.50	3.13	st	1.70	1.88	sm
19	1.18	3.61	st	1.52	6.00	st	1.45	3.25	st	1.68	2.03	sm
20	1.18	2.46	sm	1.52	3.67	st	1.43	2.72	sm	1.59	2.55	sm
21	1.18	2.77	sm	1.52	2.11	sm	1.33	2.44	sm	1.21	3.38	st
22	1.15	2.38	sm	1.46	2.38	sm	1.28	3.35	st	1.51	2.79	sm
23	1.16	4.13	st	1.41	3.33	st	1.24	2.63	sm	1.56	2.21	sm
24	1.13	2.09	sm	1.30	3.00	st	1.22	1.48	m	1.44	1.96	sm
25	1.08	3.48	st	1.24	2.83	sm	1.09	1.32	m	1.21	4.83	st
26	1.11	3.88	st	1.19	2.67	sm	0.98	2.83	sm	1.18	2.99	sm
27	1.05	3.11	st	1.08	2.33	sm	0.90	2.50	sm	0.95	3.58	st
28	1.05	3.11	st	0.89	3.75	st	0.88	3.17	st	0.90	1.89	sm
29	1.02	5.00	st									
30	1.02	3.50	st									
31	1.02	3.50	st									
32	1.02	1.57	m									
33	0.96	3.25	st									
34	0.91	3.00	st									
35	0.88	4.17	st									
36	0.88	2.44	sm									
37	0.87	2.39	sm									
38	0.68	2.48	sm									

RL: relative length; AR: arm ratio; PC: position of centromere; \* satellite-chromosomes.

number has been not reported previously within *Meconopsis* species. But, the chromosome numbers of  $2n=28$ ,  $56$  and/or  $n=14$ ,  $28$  are most prevalent in *Meconopsis* (TABLE 1 and TABLE 3).

ERNST (1965) and RATTER (1968) suggested  $x=7$  and  $8$  as the basic chromosome numbers of *Meconopsis*. But hitherto, none was found to be  $2n=14$  or  $16$  in the examination available of all the species, which covered the most morphological range in *Meconopsis*. Thus, it seems tentatively to accept that the chromosome base number is  $x=7$  and  $8$ . According to chromosome behavior in meiosis (SUGIURA 1940; RATTER 1968), the most common basic chromosome number of *Meconopsis* is also possible to be  $x=14$ . Four species stud-

ied in this paper (*M. racemosa*, *M. horridula*, and *M. pseudovenusta*) should be tetraploid with  $2n=4x=56$  except for *M. integrifolia* with various chromosome numbers ( $2n=76$ ) and various basic chromosome number. Anyway, whether the main base number is  $7$  or  $14$ , it seems that polyploidy evolution has been played an important role on the genus.

*M. integrifolia* is placed in Ser. Grandes Prain with various chromosome numbers ( $n=37$ ,  $41$ ,  $59$  or  $60$ ) (RATTER 1968). *M. racemosa* belongs to Ser. Racemosae ( $n=28$  or  $2n=56$ ). *M. horridula* and *M. pseudovenusta* belong to Ser. Delavayanae Tayl. ( $n=28$  or  $2n=56$ ). From the present cytological data, it seems that the chromosome numbers of

most species in Ser. Meconopsis, Ser. Racemosae, and Ser. Delavayanae Tayl. are  $n=28$  or  $2n=56$ . However, the most primitive species of Ser. Chelidoniifoliae were found with a lower ploidy level ( $n=14$ ). The chromosome numbers of Ser. Grandes Prain and Ser. Simplicifoliae are not sure up to now. The chromosome data support WU and CHUANG's (1980) taxonomic system.

The karyomorphological characters of four species studied first here were almost similar to each other, such as the chromosomes in the complement gradually decreasing in size, and karyotypes asymmetry of 3B-type. But, there are differences in the number of m-, sm-, and st- chromosomes as well as satellites among species.

*M. integrifolia* and *M. racemosa* belong to Ser. Grandes and Ser. Racemosae in the Sect. Racemosae, respectively, characterized by the basal or cauline leaves, racemose. Both *M. horridula* and *M. pseudovenusta* belong to Ser. Delavayanae of the Sect. Simplicifoliae, characterized by no cauline leaves, single flower. The former two species possessed 6 m- and different sm- and st- chromosomes. The latter two possessed 4 m- chromosomes and varied in sm- and st- chromosomes. It seems that the number of m- chromosomes is an important feature in the classification on the section or series level.

Because most of the previous cytological reports were from cultivated species without karyotype data, karyomorphological study on all these species from the wild as well as other unreported species are badly needed to furnish essential information in order to understand the taxonomy and evolution of the genus.

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