

# 日本産オドリコソウ属(広義) の細胞分類学的研究

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## Norihit Miura<sup>1, 2</sup> and Yoshikane Iwatsubo<sup>3\*</sup> : Cytotaxonomic study of *Lamium sensu lato* (Labiatae) in Japan

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### Abstract

We report the karyotypes of the following six species of Japanese *Lamium* : *L. album* var. *barbatum* ( $2n = 18 = 16m + 2sm$ ), *L. amplexicaule* ( $2n = 18 = 6m + 1^t sm + 7sm + 2^t st + 2st$ ), *L. purpureum* ( $2n = 18 = 2^t m + 3m + 6^t sm + 6sm + 1st$ ), *L. humile* ( $2n = 34 = 26m + 8sm$ ), *L. tuberiferum* ( $2n = 32 = 18m + 6sm + 8st$ ) and *L. ambiguum* ( $2n = 26 = 14m + 4^t sm + 6sm + 2st$ ). The present study determined, for the first time, chromosome numbers of *L. tuberiferum* and *L. ambiguum*. Our findings showed that *L. album* var. *barbatum*, *L. amplexicaule* and *L. purpureum* had a basic chromosome number of  $x = 9$ , *L. ambiguum* had  $x = 13$ , *L. tuberiferum* had a basic chromosome number of  $x = 16$ , and *L. humile* had a basic chromosome number of  $x = 17$ . These differences in basic chromosome numbers support the taxonomic treatments proposed by Makino (1905, 1915) where these *Lamium* species are treated as *Ajugoides*, *Lamium* s.s., *Loxocalyx* and *Matsumurella*, respectively.

**Key words :** chromosome number, Japanese *Lamium*, karyotype, Labiatae.

Six species of *Lamium* L. sensu lato occur in Japan (Murata 1981; Ohwi and Kitagawa 1992). Of the six species, *L. humile* (Miq.) Maxim., *L. tuberiferum* (Makino) Ohwi and *L. ambiguum* (Makino) Ohwi are often divided into the following three genera: *L. humile* as *Ajugoides humilis* (Miq.) Makino (Makino 1915); *L. tuberiferum* as *Matsumurella tuberifera* (Makino) Makino (Makino 1915); and *L. ambiguum* as *Loxocalyx ambiguum* (Makino) Makino (Makino 1905). The taxonomic treatments were followed by Kudo (1929), Ryding (2004) and Mabberley (2008). Murata and Yamazaki (1993) treat only *L. ambiguum* as *Loxocalyx ambiguum* and the other species as *Lamium*.

Worldwide, various chromosome numbers of  $n = 9$ , 17, 18, and  $2n = 16$ , 18, 32, 34, 36 have been identified for *Lamium* (Appendix). Basic chromosome numbers are considered to be  $x = 8$ , 9, 17 and 18 (Darlington and Wylie 1955; Singh 1995; Miura et al. 2005). Chromosome numbers for Japanese *Lamium* taxa includ-

ing naturalized species are also variable. For example,  $2n = 18$  for *L. album* L. var. *barbatum* (Siebold et Zucc.) Franch. et Sav. (Tanaka 1974; Terasaka and Tanaka 1974 (as  $n = 9$ ); Nishikawa 1981; Miura et al. 2005), *L. amplexicaule* L. (Miura et al. 2005) and *L. purpureum* L. (Miura et al. 2005), 34 for *L. humile* (Miura et al. 2005), and 36 chromosomes for *L. hybridum* Vill. (Miura et al. 2005). Among the Japanese *Lamium*, chromosome numbers for *L. ambiguum* and *L. tuberiferum* remain unexamined. Among plants, Makino (1905) treated *L. humile* as *Ajugoides humilis*, *L. ambiguum* as *Loxocalyx ambiguum* and *L. tuberiferum* as *Matsumurella tuberifera*, respectively.

To clarify the taxonomic relationships of six species of Japanese *Lamium* s.l., we examined their karyotypes and report them here.

### Materials and methods

A total of 40 plants representing six species of Japanese *Lamium* s.l. used in this study were collected from 10 localities (Table 1).

Table 1. Chromosome number and collected locality of Japanese *Lamium* sensu lato taxa used in the study

Taxon	Chromosome number (2n)	Observed number	Collected locality
<i>L. album</i> var. <i>barbatum</i>	18	1	Nishikamisawa, Tome City, Miyagi Pref.
		3	Shimonagano, Kanuma City, Tochigi Pref.
		1	Kyushiojiri, Shiojiri City, Nagano Pref.
<i>L. ambiguum</i>	26	1	Kongosanchi, Chihaya-akasaka-mura, Osaka Pref.
<i>L. amplexicaule</i>	18	1	Sutamacho-egusa, Hokuto City, Yamanashi Pref.
		12	Gofuku, Toyama City, Toyama Pref.
<i>L. humile</i>	34	4	Ryusozan, Aoi-ku, Shizuoka City, Shizuoka Pref.
		2	Suyama, Susono City, Shizuoka Pref.
		4	Tomiyama, Toyone-mura, Kitashitara-gun, Aichi Pref.
<i>L. purpureum</i>	18	7	Gofuku, Toyama City Toyama Pref.
<i>L. tuberiferum</i>	32	4	Mikegawa, Shirahama-cho, Nishimuro-gun, Wakayama Pref.

Plants were cultivated in the experimental garden at the Faculty of Science, University of Toyama. Karyotype was determined using meristematic cells obtained from a root tip from each individual that was subjected to the ordinary squash technique. Plants came from the following localities: *L. album* var. *barbatum*, collected at Shimonagano, Kanuma City, Tochigi Pref.; *L. ambiguum*, collected at Kongosanchi, Chihaya-akasaka-mura, Minamikawachi-gun, Osaka Pref.; *L. amplexicaule*, collected at Sutamachoegusa, Hokuto City, Yamanashi Pref.; *L. humile*, collected at Tomiyama, Toyone-mura, Kitashitara-gun, Aichi Pref.; *L. purpureum*, collected at Gofuku, Toyama City, Toyama Pref.; *L. tuberiferum*, collected at Mikegawa, Shirahama-cho, Nishimuro-gun, Wakayama Pref.

Newly formed root tips were collected, pre-treated in 1.8–2.2 mM 8-hydroxyquinoline at room temperature (ca. 25°C) for 1 h, and incubated at 5°C for 15 h. Root tips were fixed with a mixture of glacial acetic acid and ethyl alcohol (1 : 3) for 1 h, soaked in 1N hydrochloric acid at room temperature for 1 h, macerated in 1N hydrochloric acid at 60°C for 10 minutes, washed in tap water, and then stained in a drop of 2% lacto-propionic orcein on a slide glass. Chromosome forms were described according to nomenclature proposed by Levan et al. (1964). Voucher specimens of the plants examined were deposited at the Toyama Science Museum (TOYA).

## Results and discussion

### *Lamium album* var. *barbatum*:

We observed  $2n = 18$  chromosomes, and those at metaphase ranged from 1.9 to 2.4 μm in length and from 1.1 to 1.8 in arm ratio (Figs. 1A, 2A; Table 2). In the chromosome complement, 16 chromosomes were metacentric and two were submetacentric (Table 2). The karyotype is thus formulated as  $2n = 18 = 16m + 2sm$  and shows that the basic chromosome number is  $x = 9$ .

### *Lamium ambiguum*:

We observed  $2n = 26$  chromosomes, and those at metaphase ranged from 1.5 to 2.7 μm in length and from 1.1 to 3.5 in arm ratio (Figs. 1B, 2B; Table 3). In this chromosome complement, 14 chromosomes were metacentric, eight were submetacentric and the other four chromosomes were subtelocentric (Table 3). In this chromosome complement, two submetacentric chromosomes and two subtelocentric chromosomes had satellites on their short arms. The karyotype is thus formulated as  $2n = 26 = 14m + 4^{st}sm + 6sm + 2st$  and shows the basic chromosome number as  $x = 13$ .

### *Lamium amplexicaule*:

We observed  $2n = 18$  chromosomes, and those at metaphase ranged from 1.4 to 1.7 μm in length and from 1.0 to 3.0 in arm ratio (Figs. 1C, 2C; Table 4). In the chromosome complement, six chromosomes were metacentric, eight chromosomes were submetacentric, and the other four were subtelocentric (Table 4). In the chromosome complement, one submetacentric

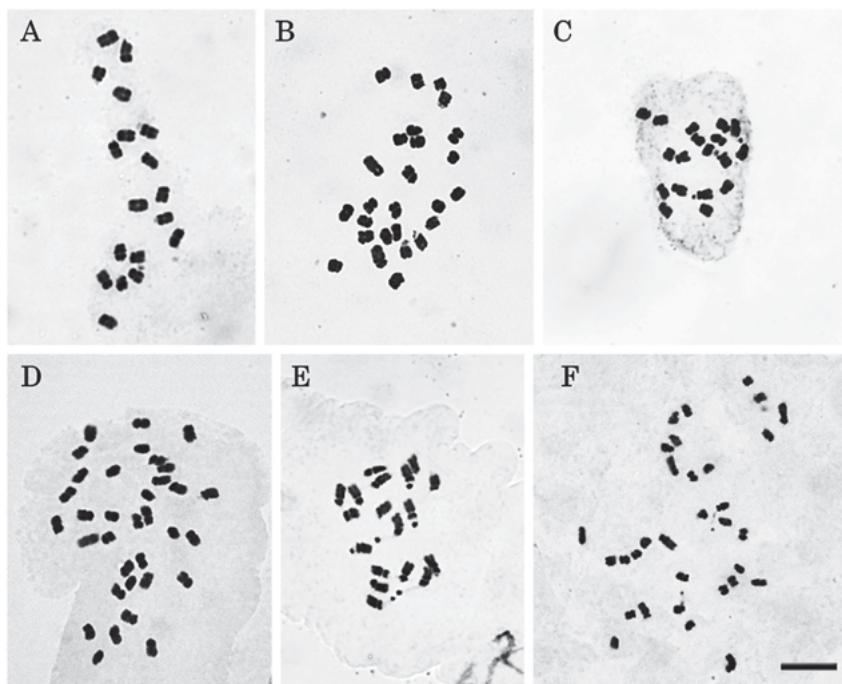


Fig. 1. Photographs of somatic metaphase chromosomes of Japanese *Lamium*. A: *L. album* var. *barbatum* ( $2n = 18$ ), B: *L. ambiguum* ( $2n = 26$ ), C: *L. amplexicaule* ( $2n = 18$ ), D: *L. humile* ( $2n = 34$ ), E: *L. purpureum* ( $2n = 18$ ), F: *L. tuberiferum* ( $2n = 32$ ). Bar indicates  $5 \mu\text{m}$ .

chromosome and two subtelocentric chromosomes had satellites on their short arms. The karyotype was thus formulated as  $2n = 18 = 6m + 1^t\text{sm} + 7\text{sm} + 2^t\text{st} + 2\text{st}$  and shows the basic chromosome number as  $x = 9$ .

#### *Lamium humile:*

We observed  $2n = 34$  chromosomes. Chromosomes at metaphase ranged from 1.5 to  $2.4 \mu\text{m}$  in length and from 1.1 to 2.4 in arm ratio (Figs. 1D, 2D; Table 5). In the chromosome complement, 26 chromosomes were metacentric and the other eight chromosomes were submetacentric (Table 5). The karyotype is thus formulated as  $2n = 34 = 26m + 8\text{sm}$  and shows the basic chromosome number as  $x = 17$ .

#### *Lamium purpureum:*

We observed  $2n = 18$  chromosomes, and those at metaphase ranged from 2.0 to  $2.4 \mu\text{m}$  in length and from 1.1 to 3.0 in arm ratio (Figs. 1E, 2E; Table 6). In the chromosome complement, five chromosomes were metacentric, 12 were submetacentric and the other one was subtelocentric (Table 6). In the somatic chro-

Table 2. Measurements of somatic metaphase chromosomes of *Lamium album* var. *barbatum*

No	Length ( $\mu\text{m}$ )	Arm ratio	Form
1	$1.1 + 1.3 = 2.4$	1.2	m
2	$1.1 + 1.3 = 2.4$	1.2	m
3	$0.9 + 1.5 = 2.4$	1.7	m
4	$0.9 + 1.5 = 2.4$	1.7	m
5	$0.9 + 1.4 = 2.3$	1.6	m
6	$0.9 + 1.4 = 2.3$	1.6	m
7	$0.9 + 1.4 = 2.3$	1.6	m
8	$0.9 + 1.4 = 2.3$	1.6	m
9	$0.9 + 1.4 = 2.3$	1.6	m
10	$0.9 + 1.4 = 2.3$	1.6	m
11	$0.8 + 1.4 = 2.2$	1.8	sm
12	$0.8 + 1.4 = 2.2$	1.8	sm
13	$0.8 + 1.3 = 2.1$	1.6	m
14	$0.8 + 1.3 = 2.1$	1.6	m
15	$1.0 + 1.1 = 2.1$	1.1	m
16	$1.0 + 1.1 = 2.1$	1.1	m
17	$0.8 + 1.1 = 1.9$	1.4	m
18	$0.8 + 1.1 = 1.9$	1.4	m



Fig. 2. Karyotypes of Japanese *Lamium*. A: *L. album* var. *barbatum* ( $2n = 18$ ), B: *L. ambiguum* ( $2n = 26$ ), C: *L. amplexicaule* ( $2n = 18$ ), D: *L. humile* ( $2n = 34$ ), E: *L. purpureum* ( $2n = 18$ ), F: *L. tuberiferum* ( $2n = 32$ ). Bar indicates 5  $\mu\text{m}$ . Arrows indicate chromosomes with satellite.

mosome complement, two metacentric chromosomes and six submetacentric chromosomes had satellites on their short arms. The karyotype is thus formulated as  $2n = 18 = 2^t\text{m} + 3\text{m} + 6^t\text{sm} + 6\text{sm} + 1\text{st}$  and shows the basic chromosome number as  $x = 9$ .

#### *Lamium tuberiferum*:

We observed  $2n = 32$  chromosomes. Chromosomes at metaphase ranged from 1.2 to 2.4  $\mu\text{m}$  in length and from 1.1 to 6.0 in arm ratio (Figs. 1F, 2F; Table 7). In the chromosome complement, 18 chromosomes were metacentric, six

Table 3. Measurements of somatic metaphase chromosomes of *Lamium ambiguum*

No	Length (μm)	Arm ratio	Form
1	0.6 + 2.1 = 2.7	3.5	st
2	0.6 + 2.1 = 2.7	3.5	st
3	0.9 + 1.3 = 2.2	1.4	m
4	0.9 + 1.3 = 2.2	1.4	m
5	0.8 + 1.4 = 2.2	1.8	sm
6	0.8 + 1.4 = 2.2	1.8	sm
7	1.0 + 1.1 = 2.1	1.1	m
8	1.0 + 1.1 = 2.1	1.1	m
9	1.0 + 1.1 = 2.1	1.1	m
10	1.0 + 1.1 = 2.1	1.1	m
11	0.8 + 1.2 = 2.0	1.5	m
12	0.8 + 1.2 = 2.0	1.5	m
13	0.9 + 1.0 = 1.9	1.1	m
14	0.9 + 1.0 = 1.9	1.1	m
15	0.8 + 1.0 = 1.8	1.3	m
16	0.8 + 1.0 = 1.8	1.3	m
17	0.6 + 1.2 = 1.8	2.0	sm
18	0.6 + 1.2 = 1.8	2.0	sm
19	t-0.5 + 1.2 = 1.7	2.4	sm
20	t-0.5 + 1.2 = 1.7	2.4	sm
21	0.5 + 1.2 = 1.7	2.4	sm
22	0.5 + 1.2 = 1.7	2.4	sm
23	t-0.4 + 1.1 = 1.5	2.8	sm
24	t-0.4 + 1.1 = 1.5	2.8	sm
25	0.7 + 0.8 = 1.5	1.1	m
26	0.7 + 0.8 = 1.5	1.1	m

t: satellite.

Table 4. Measurements of somatic metaphase chromosomes of *Lamium amplexicaule*

No	Length (μm)	Arm ratio	Form
1	0.5 + 1.2 = 1.7	2.4	sm
2	t-0.5 + 1.2 = 1.7	2.4	sm
3	t-0.4 + 1.2 = 1.6	3.0	st
4	t-0.4 + 1.2 = 1.6	3.0	st
5	0.4 + 1.2 = 1.6	3.0	st
6	0.4 + 1.2 = 1.6	3.0	st
7	0.4 + 1.1 = 1.5	2.8	sm
8	0.4 + 1.1 = 1.5	2.8	sm
9	0.6 + 0.9 = 1.5	1.5	m
10	0.6 + 0.9 = 1.5	1.5	m
11	0.4 + 1.1 = 1.5	2.8	sm
12	0.4 + 1.1 = 1.5	2.8	sm
13	0.7 + 0.8 = 1.5	1.1	m
14	0.7 + 0.8 = 1.5	1.1	m
15	0.7 + 0.7 = 1.4	1.0	M
16	0.7 + 0.7 = 1.4	1.0	M
17	0.4 + 1.0 = 1.4	2.5	sm
18	0.4 + 1.0 = 1.4	2.5	sm

t: satellite.

Table 5. Measurements of somatic metaphase chromosomes of *Lamium humile*

No	Length (μm)	Arm ratio	Form
1	0.8 + 1.6 = 2.4	2.0	sm
2	0.8 + 1.6 = 2.4	2.0	sm
3	1.1 + 1.3 = 2.4	1.2	m
4	1.1 + 1.3 = 2.4	1.2	m
5	1.0 + 1.3 = 2.3	1.3	m
6	1.0 + 1.3 = 2.3	1.3	m
7	1.1 + 1.2 = 2.3	1.1	m
8	1.1 + 1.2 = 2.3	1.1	m
9	0.8 + 1.4 = 2.2	1.8	sm
10	0.8 + 1.4 = 2.2	1.8	sm
11	1.0 + 1.2 = 2.2	1.2	m
12	1.0 + 1.2 = 2.2	1.2	m
13	0.8 + 1.3 = 2.1	1.6	m
14	0.8 + 1.3 = 2.1	1.6	m
15	0.8 + 1.3 = 2.1	1.6	m
16	0.8 + 1.3 = 2.1	1.6	m
17	1.0 + 1.1 = 2.1	1.1	m
18	1.0 + 1.1 = 2.1	1.1	m
19	0.9 + 1.1 = 2.0	1.2	m
20	0.9 + 1.1 = 2.0	1.2	m
21	0.9 + 1.0 = 1.9	1.1	m
22	0.9 + 1.0 = 1.9	1.1	m
23	0.9 + 1.0 = 1.9	1.1	m
24	0.9 + 1.0 = 1.9	1.1	m
25	0.8 + 1.0 = 1.8	1.3	m
26	0.8 + 1.0 = 1.8	1.3	m
27	0.5 + 1.2 = 1.7	2.4	sm
28	0.5 + 1.2 = 1.7	2.4	sm
29	0.5 + 1.2 = 1.7	2.4	sm
30	0.5 + 1.2 = 1.7	2.4	sm
31	0.7 + 0.9 = 1.6	1.3	m
32	0.7 + 0.9 = 1.6	1.3	m
33	0.7 + 0.8 = 1.5	1.1	m
34	0.7 + 0.8 = 1.5	1.1	m

were submetacentric, and the other eight were subtelocentric (Table 5). The karyotype is thus formulated as  $2n = 32 = 18m + 6sm + 8st$  and shows the basic chromosome number as  $x = 16$ .

This study shows that Japanese *Lamium* s.l. has four basic chromosome numbers of  $x = 9, 13, 16$  and  $17$ . Of these counts,  $x = 13$  and  $16$  found in *L. ambiguum* and *L. tuberiferum*, respectively, represent new basic chromosome numbers in *Lamium* s.l. The five species except for *L. ambiguum* have gradual and symmetric karyotypes in common. Of the five species, *L. tuberiferum* has a high-pitched and symmetric karyotype while *L. ambiguum* has a bimodal karyotype.

Differences among basic chromosome numbers of *L. ambiguum*, *L. humile* and *L. tuberiferum*

Table 6. Measurements of somatic metaphase chromosomes of *Lamium purpureum*

No	Length (μm)	Arm ratio	Form
1	t-0.8 + 1.7 = 2.5	2.1	sm
2	t-0.8 + 1.7 = 2.5	2.1	sm
3	0.9 + 1.6 = 2.5	1.8	sm
4	0.9 + 1.6 = 2.5	1.8	sm
5	t-0.9 + 1.5 = 2.4	1.7	m
6	t-0.9 + 1.5 = 2.4	1.7	m
7	1.1 + 1.2 = 2.3	1.1	m
8	1.1 + 1.2 = 2.3	1.1	m
9	t-0.7 + 1.6 = 2.3	2.3	sm
10	t-0.7 + 1.6 = 2.3	2.3	sm
11	0.7 + 1.5 = 2.2	2.1	sm
12	0.7 + 1.5 = 2.2	2.1	sm
13	t-0.6 + 1.5 = 2.1	2.5	sm
14	t-0.6 + 1.5 = 2.1	2.5	sm
15	0.6 + 1.5 = 2.1	2.5	sm
16	0.6 + 1.5 = 2.1	2.5	sm
17	0.5 + 1.5 = 2.0	3.0	st
18	0.9 + 1.1 = 2.0	1.2	m

t: satellite.

Table 7. Measurements of somatic metaphase chromosomes of *Lamium tuberiferum*

No	Length (μm)	Arm ratio	Form
1	0.7 + 1.7 = 2.4	2.4	sm
2	0.7 + 1.7 = 2.4	2.4	sm
3	0.4 + 1.9 = 2.3	4.8	st
4	0.4 + 1.9 = 2.3	4.8	st
5	0.3 + 1.8 = 2.1	6.0	st
6	0.3 + 1.8 = 2.1	6.0	st
7	0.4 + 1.6 = 2.0	4.0	st
8	0.4 + 1.6 = 2.0	4.0	st
9	0.4 + 1.5 = 1.9	3.8	st
10	0.4 + 1.5 = 1.9	3.8	st
11	0.5 + 1.3 = 1.8	2.6	sm
12	0.5 + 1.3 = 1.8	2.6	sm
13	0.7 + 1.1 = 1.8	1.6	m
14	0.7 + 1.1 = 1.8	1.6	m
15	0.8 + 0.9 = 1.7	1.1	m
16	0.8 + 0.9 = 1.7	1.1	m
17	0.7 + 1.0 = 1.7	1.4	m
18	0.7 + 1.0 = 1.7	1.4	m
19	0.6 + 1.0 = 1.6	1.7	m
20	0.6 + 1.0 = 1.6	1.7	m
21	0.7 + 0.8 = 1.5	1.1	m
22	0.7 + 0.8 = 1.5	1.1	m
23	0.5 + 0.9 = 1.4	1.8	sm
24	0.5 + 0.9 = 1.4	1.8	sm
25	0.6 + 0.8 = 1.4	1.3	m
26	0.6 + 0.8 = 1.4	1.3	m
27	0.6 + 0.7 = 1.3	1.2	m
28	0.6 + 0.7 = 1.3	1.2	m
29	0.6 + 0.7 = 1.3	1.2	m
30	0.6 + 0.7 = 1.3	1.2	m
31	0.5 + 0.7 = 1.2	1.4	m
32	0.5 + 0.7 = 1.2	1.4	m

support the taxonomic treatment of the three genera as *Loxocalyx ambiguus*, *Ajugoïdes humilis* and *Matsumurella tuberifera* as proposed by Makino (1905, 1915).

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### 三浦憲人<sup>1,2</sup>・岩坪美兼<sup>3</sup>：日本産オドリコソウ属（広義）の細胞分類学的研究

日本産オドリコソウ属（広義）*Lamium* s.l. にはオドリコソウ*L. album* var. *barbatum*, マネキグサ*L. ambiguum*, ホトケノザ*L. amplexicaule*, ヤマジオウ*L. humile*, ヒメオドリコソウ*L. purpureum*, ヒメキセワタ*L. tuberiferum*の6種が知られている (Murata 1981, Ohwi and Kitagawa 1992)。Makino (1905, 1915)はマネキグサ, ヤマジオウおよびヒメキセワタをそれぞれ *Loxocalyx ambiguum*, *Ajugoidea humilis*, *Matsumurella tuberifera* としてオドリコソウ属（狭義）*Lamium* s.s. とは別属として扱い, Kudo (1929), Ryding (2004), Mabberley

(2008) もこの見解を採用している。Murata and Yamazaki (1993) はマネキグサのみを別属の *Loxocalyx ambiguus* とし、他の5種をオドリコソウ属として扱っている。このように日本産オドリコソウ属（広義）は分類的扱いにいくつかの見解が存在している。

オドリコソウ属（広義）は過去の染色体数の報告から  $x = 9$  であることが知られていたが (Darlington and Wylie 1955; Singh 1995), 日本産ヤマジオウでは新たに  $x = 17$  (Miura et al. 2005) も知られている。本研究は、日本産オドリコソウ属（広義）内の類縁関係を明らかにするために、本属（広義）6種の染色体観察を行った。

観察の結果、染色体数および核型式はオドリコソウ :  $2n = 18 = 16m + 2sm$ , マネキグサ :  $2n = 26 = 14m + 4^t sm + 6sm + 2st$ , ホトケノザ :  $2n = 18 = 6m + 1^t sm + 7sm + 2^t st + 2st$ , ヤマジオウ :  $2n = 34 = 26m + 8sm$ , ヒメオドリコソウ :  $2n = 18 = 2^t m + 3m + 6^t sm + 6sm + 1st$ , ヒメキセワタ :  $2n = 32 = 18m + 6sm + 8st$  であった。核型はマネキグサではもっとも長い1対の染色体が極端に長いという特徴をもっていた。一方、他の

5種（オドリコソウ、ホトケノザ、ヒメオドリコソウ、ヒメキセワタ、ヤマジオウ）は、すべて勾配的な核型であった。そしてこれら5種のうち、ヒメキセワタでは最も短い染色体は最も長い染色体の半分の長さしかない急勾配的核型を持ち、他の4種とは異なっていた。ヒメキセワタの染色体基本数は、核型分析の結果から  $x = 16$ 、マネキグサは  $x = 13$  であると判断され、日本産オドリコソウ属（広義）の基本数には  $x = 9, 13, 16, 17$  の4種類が存在することがわかった。オドリコソウ属は  $x = 9$  が基本数とされているが、日本では基本数が複数存在していることが明らかとなり、この染色体基本数の違いは Makino (1905, 1915) の分類学的見解と一致している。

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**Appendix.** Chromosome numbers reported of *Lamium*

Taxon	Chromosome number		Reference (Locality)
	n	2n	
<i>L. album</i>	8		Marchal 1920 (cult.)
	9		Jörgensen 1927 (unknown); Tischler 1934 (Germany); Rohweder 1937 (Germany); Sorsa 1962 (Finland); Gill 1981 (Canada), 1983 (Canada); Bir and Saggoo 1982 (India); Saggoo and Bir 1983a (India); 1983b (India)
	9, 9+1B	18	Gill 1970 (India), 1984 (India) Heitz 1926 (unknown); Turesson 1938 (Russia, Sweden); Delay 1948 (unknown); Pólya 1949 (Hungary); Löve and Löve 1956 (Iceland); Gadella and Kliphuis 1963 (Netherland); Skalińska et al. 1968 (Poland); Pogosyan et al. 1971 (Armenia); Morton 1973 (U.K.); Belaeva and Siplivinsky 1975 (Russia); Májovský et al. 1978 (Slovakia); Zukowski and Slowinska 1979 (Poland); Dmitriyeva 1985 (Byelorussia); Al-Bermani 1993 (U.K.)
<i>L. album</i> var. <i>barbatum</i>	9		Terasaka and Tanaka 1974 (Japan)
	18		Tanaka 1974 (Japan); Nishikawa 1981 (Japan); Miura et al. 2005 (Japan)
<i>L. amplexicaule</i>	9		Jörgensen 1927 (unknown); Tischler 1934 (Germany), 1937 (North Sea); Gill 1981 (Canada), 1983 (Canada); Saggoo and Bir 1983a (India), 1983b (India); Ortega-Olivencia and Ruiz-Téllez 1990 (Spain)
	18		Pólya 1950 (Hungary); Bernström 1952 (Sweden); Heiser and Whitaker 1948 (U.S.A.); Löve and Löve 1956 (Iceland); Strid 1965 (Greece); Gadella and Kliphuis 1966 (Netherland); Skalińska et al. 1968 (Poland); Májovský et al. 1970 (Slovakia); Dahlgren et al. 1971 (Bolearic Islands); Fernandes and Queirós 1971 (Portugal); Löve and Kjellqvist 1974 (Spain); Feráková 1972 (Slovakia); Morton 1973 (U.K.); Markova and Thu 1974 (Bulgaria); Aryavand 1977 (Iran); Van den Brand et al. 1979 (France); Strid and Franzén 1981 (Greece); Fernandes and Leitão 1984 (as <i>L. amplexicaule</i> subsp. <i>amplexicaule</i> , Portugal); Elena-Rosselló et al. 1988 (Spain); Miura et al. 2005 (Japan)
<i>L. barbatum</i> Siebold et Zucc.	18		Sokolovskaya 1966 (Russia); Starodubtsev 1985 (Russia);

## Appendix. Continued

Taxon	Chromosome number		Reference (Locality)
	n	2n	
<i>L. bifidum</i> Cirillo	9	18	Sokolovskaya et al. 1986 (Russia); Rudyka 1995 (Russia)
		18	Elena Rosselló et al. 1988 (Spain)
		18	Ortega Olivencia and Ruiz Tellez 1990 (as <i>L. bifidum</i> subsp. <i>bifidum</i> , Spain)
<i>L. corsicum</i> Gren. et Godr.		18	Diana-Corrias 1980 (Italy)
<i>L. dissectum</i> With.	18		Jörgensen 1927 (unknown); Tischler 1934 (Germany)
<i>L. flexuosum</i> Ten.		18	Van den Brand et al. 1979 (Belgium)
<i>L. galeobdolon</i> (L.) Crantz	16		Marchal 1920 (cult.)
		18	Jörgensen 1927 (unknown); Pólya 1949 (Hungary);
<i>L. galeobdolon</i> (L.) L.		36	Gadella and Kliphuis 1966 (Netherlands)
<i>L. galeobdolon</i>		36	Májovský et al. 1974 (Slovakia);
subsp. <i>montanum</i>			Strid and Franzén 1981 (Greece);
(Pers.) Hayek			Markova and Goranova 1995 (Bulgaria)
<i>L. galeobdolon</i> (L.) Nath.	9, 18		Polatschek 1966 (East Alps)
<i>L. garganicum</i> L.		18	Strid 1965 (Greece); Gadella and Kliphuis 1972 (Yugoslavia);
			Markova and Ivanova 1974
		36	(as <i>L. garganicum</i> subsp. <i>garganicum</i> , Bulgaria)
<i>L. garganicum</i>			Baltisberger 1991 (Greece)
subsp. <i>laevigatum</i> Arcang.		18	Van Loon 1980 (Italy);
<i>L. garganicum</i>			Markova and Goranova 1995 (Bulgaria)
subsp. <i>pictum</i>		18	Markova and Goranova 1995 (Bulgaria)
(Boiss. et Heldr.) P. W. Ball			
<i>L. garganicum</i>	9		De Montmollin 1982 (Switzerland)
subsp. <i>striatum</i>		18	Sz.-Borsos 1971 ( <i>L. garganicum</i> subsp. <i>striatum</i> )
(Sibth. et Sm.) Hayek			f. <i>sofianum</i> Stoj et Stef., Hungary)
<i>L. garganicum</i>		18	Favarger 1973 (Italy)
var. <i>grandiflorum</i>			
(Pourr.) Fiori			
<i>L. garganicum</i>	9		Favarger 1973 (Italy)
var. <i>veronicaefolium</i>			
(Benth.) Fiori			
<i>L. humile</i>		34	Miura et al. (2005)
<i>L. hybridum</i>	18		Ortega-Olivencia and Ruiz-Tellez 1990 (Spain)
		36	Löve and Löve 1956 (Iceland); Morton 1973 (U.K.);
			Fernandes and Leitão 1984 (Portugal);
			Elena-Rosselló et al. 1988 (Spain);
			Miura et al. 2005 (Japan)
<i>L. intermedium</i> Franch.	18		Jörgensen 1927 (unknown); Tischler 1934 (Germany)
<i>L. iranicum</i> Parsa	17		Aryavand 1977 (Iran)
<i>L. longiflorum</i> Ten.	9		Jörgensen 1927 (unknown)
<i>L. maculatum</i> L.	9	18	Baltisberger and Baltisberger 1995 (Albania)
			Jörgensen 1927 (unknown); Tischler 1934 (Germany);
			Gill 1981 (Canada), 1983 (Canada);
			Ortega-Olivencia and Ruiz-Tellez 1990 (Spain);
			Kliphuis and Wieffering 1972 (France);
			Van Faassen and Nadeau 1976 (U.S.A.);
			Mulligan 1984 (Canada)

**Appendix.** Continued

Taxon	Chromosome number		Reference (Locality)
	n	2n	
	9	18	Elena Rosselló et al. 1988 (Spain)
		18	Gadella and Kliphuis 1963 (Netherlands), 1973 (Netherlands); Májovský et al. 1974 (Slovakia), 1978 (as <i>L. maculatum</i> subsp. <i>maculatum</i> , Slovakia); Van den Brand et al. 1979 (Switzerland); Fernandes and Leitão 1984 (Portugal); Dmitriyeva 1985 (Byelorussia)
<i>L. maculatum</i>		18	Májovský et al. 1978 (Slovakia)
subsp. <i>cupreum</i> (Schott) Hadac			
<i>L. maculatum</i>		18	Markova and Goranova 1995 (Bulgaria)
var. <i>cupreum</i>			
(Schim. et al.) Briq.			
<i>L. maculatum</i>		18	Markova and Goranova 1995 (Bulgaria)
var. <i>nemorale</i> Rehb.			
<i>L. molucellifolium</i> Franch.		36	Morton 1973 (U.K.); Löve and Löve 1956 (Iceland)
<i>L. montanum</i> Pers.		36	Baltisberger 1991 (Greece)
<i>L. moschatum</i> Mill.		18	Strid 1965 (Greece); Baltisberger and Huber 1987 (Greece)
<i>L. orvala</i> L.	9		Jörgensen 1927 (unknown)
<b><i>L. purpureum</i></b>	9		Jörgensen 1927 (unknown); Tischler 1934 (Germany), 1937 (North Sea); Griesinger 1937 (Germany); Gill 1981 (Canada), 1983 (Canada)
		18	Heitz 1926 (unknown); Pólya 1949 (Hungary); Löve and Löve 1956 (Iceland); Sorsa 1963 (as 2n=ca. 18, Finland); Skalińska et al. 1968 (Poland); Gadella and Kliphuis 1971 (Netherlands); Morton 1973 (U.K.); Löve and Kjellqvist 1974 (Spain); Markova and Thu 1974 (Bulgaria); Májovský et al. 1976 (Slovakia); Van den Brand et al. 1979 (Netherlands); Fernandes and Leitão 1984 (Portugal); Elena-Rosselló et al. 1988 (Spain); Hill 1989 (U.S.A.); Ortega-Olivencia and Ruiz-Tellez 1990 (Spain); Miura et al. 2005 (Japan)
<i>L. rugosum</i> Ait.	9		Jörgensen 1927 (unknown)
<i>L. tomentosum</i> Willd.		18	Tumajanov and Beridze 1968 (Russia); Gagnidze and Gviniashvili 1984 (Gruzia)

Bold faced character shows taxon observed in this study.