

Chromosome numbers of *Eusteralis stellata* and *E. yatabeana* (Lamiaceae) , vulnerable plants of Japan

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Xiao-Dong Dong¹, Masashi Nakata², Kiyotaka Kawazumi³ and Takaaki Oohara²: **Chromosome numbers of *Eusteralis stellata* and *E. yatabeana* (Lamiaceae), vulnerable plants of Japan**

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The genus *Eusteralis* (Lamiaceae) comprises about 20 species native to southeastern Asia, India and Australia (Murata 1981). Two species, *Eusteralis stellata* (Lour.) Panigrahi and *E. yatabeana* (Makino) Panigrahi occur in Japan in low wetland or rice paddy (Murata 1981; Murata and Yamazaki 1993). However, recently, they are threatened caused by land development or by using herbicides in paddy fields, thus, the two species are listed in "Red data book 2nd ed." (Environment Agency of Japan 2000) as vulnerable plants (VU).

Chromosome numbers of the genus *Eusteralis* have been reported as ; $n=14$ for *E. stellata* var. *roxburghiana* (Keng) Bennet et Raizada (Cherian and Kuriachan 1993), $n=16$ and $2n=32$ for *E. tomentosa* (Dalzell) Panigrahi var. *gracilis* (Dalz.) Bennet et Raizada (Cherian and Kuriachan 1993), $2n=24$ for *E. cruciata* (Benth.) Panigrahi (Vembu and Sampathkumar 1998 in "Index to plant chromosome numbers [=IPCN] 1998-2000", under *Dysophylla cruciata* Benth.), $2n=30$ for *E. quadrifolia* (Benth.) Panigrahi (Thoppil and Jose 1995), $2n=36$ for *E. quadrifolia* (Vembu and Sampathkumar 1998 in "IPCN 1998-2000", under *D. quadrifolia* Benth.), $2n=40$ for *E. stellata* (Vembu and Sampathkumar 1999 in "IPCN 1998-2000", under *Pogostemon stellatus* (Lour.) Kuntze) and $2n=72$ for *E. stellata* (Borgmann 1964, under *D. verticillata* Benth.), however, no information on the Japanese materials had been available up to now.

In Toyama Prefecture, *E. stellata* (Fig. 1 A) rarely grows in paddy fields, and thus, it is listed in the "Red data book Toyama" (Toyama Pref. 2002). On the other hand, *E. yatabeana*

(Fig. 1 E) had not been recorded in Toyama until the third author found it on a small abandoned paddy-field in the northern part of Toyama City. This short report deals with the cytological observations on *E. stellata* and *E. yatabeana*, endangered plants of Japan.

Materials and method

Two individuals of *E. stellata* collected in a rice-field located in the northern part of Toyama City, Toyama Pref., central Japan, were used as the material. Of *E. yatabeana* as research material were used two ramets from a presumed large clone, which grows densely on an abandoned small rice paddy also located in the northern part of Toyama City. The materials were cultivated in small plastic containers with water for several days until new roots grow. The new roots about 5 mm long were cut and pretreated by 2 mM 8-hydroxyquinoline aqueous solution at 18°C for 4 hr, then fixed with 1 : 3 acetic alcohol at 5°C for 20 hr. The fixed root tips were macerated by a mixture of 45 % acetic acid and 1 N hydrochloric acid (1 : 1, w/w) at 60°C for 15 sec, rinsed with water for 15 sec, and meristematic tissues of the root tips were cut and picked up on glass slides. They were stained with 1 % acetic orcein for 10 min, and then squashed.

The voucher specimens (*E. stellata* : 1 Oct. 2004, Nakata 24084-1, 24084-2 ; *E. yatabeana* : 11 Sep. 2004, Nakata 24071) were kept in the Botanic Gardens of Toyama (TYM).

Results and discussion

Eusteralis stellata

Somatic chromosome number was found to be

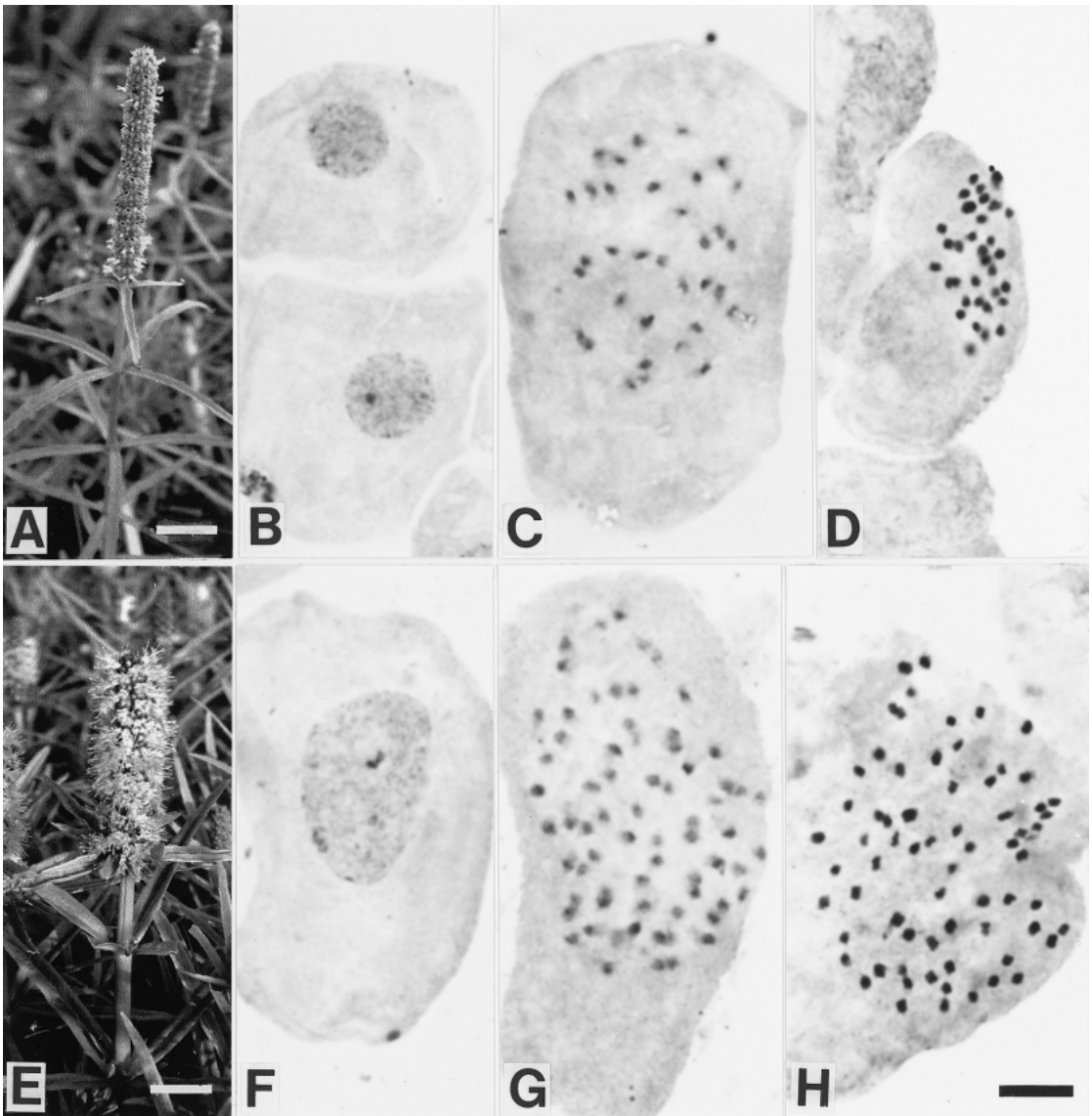


Fig. 1. *Eusteralis stellata*, $2n=34$ (A-D) and *E. yatabeana*, $2n=68$ (E-H). A and E: Plant habit. Scale bars indicate 1 cm. B and F: Interphase nuclei. C and G: Mitotic prophase chromosomes. D and H: Mitotic metaphase chromosomes. Scale bar in H indicates $5\ \mu\text{m}$ for B-D and F-H.

$2n=34$ at prophase (Fig. 1 C) and metaphase (Fig. 1 D). Chromosome numbers for *E. stellata* have been reported to be $2n=40$ (Vembu and Sampathkumar 1999 in IPCN 1998-2000, under *P. stellatus*) and $2n=72$ (Borgmann 1964, under *D. verticillata*). For var. *roxburghiana*, $n=14$ chromosomes has been reported (Cherian and Kurian 1993). The present count for *E. stellata* is new to the species. The interphase nucleus had 1-11 (mean \pm SD : 6.0 ± 2.5) darkly-stained small

heterochromatic blocks along with numerous chromomeric granules (Fig. 1 B). As the number of the chromatin blocks is less than the chromosome number, the nucleus is categorized as "the simple chromocenter type" proposed by Tanaka (1971). At prophase the chromosomes had early condensed segments at the centromeric regions. At metaphase the 34 chromosomes were small, varied in length from 0.4 to 0.9 μm . Small satellites were observed in two chromosomes. Cen-

Table 1. Chromosome numbers in *Eusteralis* and related two genera *Dysophylla* and *Pogostemon* found in Fedorov (1974) and a series of IPCN through 1967-2000 (Moore 1973 ; Goldblatt 1981, 1984, 1985, 1988 ; Goldblatt and Johnson 1990, 1994, 1996, 1998, 2003).

Taxon*	Chromosome number		Original literature**
	n=	2n=	
<i>E. quadrifolia</i>		30	Thoppil and Jose 1995
<i>E. stellata</i> var. <i>roxburghiana</i>	14		Cherian and Kuriachan 1993
<i>E. tomentosa</i> var. <i>gracilis</i>	16	32	Cherian and Kuriachan 1993
<i>D. auricularia</i>	6		Hsu 1967
		17	Saggo 1983 ; Saggo and Bir 1983 ; Bir and Saggo 1985
		34	Chuang et al. 1963 ; Cherian and Kuriachan 1984
<i>D. cruciata</i>		24	Vembu and Sampathkumar 1998
<i>D. quadrifolia</i>		36	Vembu and Sampathkumar 1998
<i>D. verticillata</i>		72	Borgmann 1964
<i>P. auricularius</i>	17	34	Cherian and Kuriachan 1993
<i>P. benghalense</i>	32		Bir and Saggo 1981 ; Saggo 1983 ; Saggo and Bir 1983
	32	64	Cherian and Kuriachan 1993
<i>P. benghalensis</i>		42	Vembu 1984 ; Vembu and Sampathkumar 1999
<i>P. cablin</i>		32	Lavana 1984
		64	Tyagi and Bahl 1990
<i>P. gardneri</i>	16		Cherian and Kuriachan 1981, 1993
<i>P. heynea</i>	16		Basavaraj and Krishnappa 1982
<i>P. heyneanus</i>	16		Krishnappa and Basavaraj 1982
	32	64	Cherian and Kuriachan 1993
		64	Cherian and Kuriachan 1984
		60	Vembu and Sampathkumar 1999
<i>P. mollis</i>	16		Cherian and Kuriachan 1981
	16	32	Cherian and Kuriachan 1993
<i>P. pachouli</i>		60	Vembu 1984
<i>P. paludosus</i>	17		Bir and Saggo 1981 ; Saggo and Bir 1982 ; Saggo 1983
<i>P. paniculatus</i>	16		Cherian and Kuriachan 1981
		32	Basavaraj and Krishnappa 1982 ; Krishnappa and Basavaraj 1982 ; Vembu 1982, 1983 ; Vembu and Sampathkumar 1999
	16	32	Cherian and Kuriachan 1993
<i>P. parviflorus</i>	32		Mehra and Gill 1968 ; Gill 1971, 1984
<i>P. plectranthoides</i>	17		Mehra and Gill 1968
	32		Gill 1971, 1984 ; Vij and Kashyap 1975, 1976 ; Thakur 1978
<i>P. pubescens</i>	16		Saggo and Bir 1982, 1983 ; Saggo 1983 ; Bir and Saggo 1985
<i>P. purpurascens</i>	16		Bir and Saggo 1981, 1982 ; Saggo and Bir 1981 ; Saggo 1983
		32	Krishnappa and Basavaraj 1982
	17	34	Cherian and Kuriachan 1993
		34	Cherian and Kuriachan 1984
<i>P. speciosis</i>		34	Krishnappa and Basavaraj 1982 ; Basavaraj and Krishnappa 1983
		40	Vembu 1984
<i>P. stellatus</i>		40	Vembu and Sampathkumar 1999
<i>P. wightii</i>	16		Cherian and Kuriachan 1981
	17		Krishnappa and Basavaraj 1982 ; Basavaraj and Krishnappa 1983
	16, 32	64	Cherian and Kuriachan 1993
		64	Cherian and Kuriachan 1984

* As original description. ** Those not appeared in the text are excluded from the references.

centromeric constrictions were obscure for almost all chromosomes.

Eusteralis yatabeana

Somatic chromosome number was found to be $2n=68$ at prophase (Fig. 1 G) and metaphase (Fig. 1 H). This is the first report of chromosome number for the species, and also new number for the genus. The interphase nucleus was similar to that of *E. stellata* in showing “the simple chromocenter type” proposed by Tanaka (1971), while it had 5-18 (mean \pm SD : 10.6 ± 4.0) heterochromatin blocks (Fig. 1 F). At prophase, as those of *E. stellata*, the chromosomes had early condensed segments at the centromeric regions. At metaphase the 68 chromosomes were small, varied in length from 0.4 to 1.0 μm . Small satellites were observed in two chromosomes. At least 20 chromosomes were observed to be symmetric in centromeric position, while the rest chromosomes were obscure.

According to Fedorov (1974) and a series of “Index to plant chromosome numbers” involving chromosome information through 1967 to 2000, variation in chromosome numbers are present in the genus *Eusteralis* and its related genera *Dysophylla* and *Pogostemon* (Table 1). Basic chromosome number for the genus *Pogostemon* had been suggested to be $x=8$ (Gill 1971; Vij and Kashyap 1976) or $x=16$ and 17 (Saggoo and Bir 1985). Based on the observations on 12 taxa of the subtribe Pogostemoninae sensu Bentham from south India, Cherian and Kuriachan (1993) considered that $x=16$ might be the primary basic number of the genus and that $x=17$ is derived by dysploidy from $x=16$. According to Cherian and Kuriachan (1993), Japanese *Eusteralis* contains diploid species (*E. stellata* with $2n=34$) and tetraploid species (*E. yatabeana* with $2n=68$). As *E. stellata* is an annual plant while *E. yatabeana* is a perennial, polyploidization might have played a role in species evolution especially in life form.

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- (Received November 14, 2004; accepted December 14, 2004)
- 董 曉東¹・中田政司²・川住清貴³・大原隆明³: 絶滅危惧種ミズネコノオおよびミズトラノオの染色体数
- ミズネコノオ *Eusteralis stellata* (Lour.) Panigrahi と ミズトラノオ *E. yatabeana* (Makino) Panigrahi は水田や低湿地に生えるシソ科の植物で、開発や土地造成で生育地が全国的に減少し、環境省の植物レッドリストでは絶滅危惧Ⅱ類 (VU) にランクされている。富山県富山市北部で最近生育が確認 (ミズトラノオは富山県初記録) されたこの2種について染色体を観察した結果、染色体数はミズネコノオが $2n=34$ 、ミズトラノオが $2n=68$ で、 $x=17$ を基本数とする、それぞれ二倍体、四倍体であることがわかった。中期染色体は $1\mu\text{m}$ 以下と小型であった。両種とも代謝期核は単純染色中央粒型、前期染色体は動原体付近に早期凝縮部を持つ基部型で、核形態は似ていた。一年草であるミズネコノオが二倍体であるのに対し多年草のミズトラノオが四倍体であることから、生活形の進化に倍数化が関与したものかもしれない。
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