

Karyology of Ten Vespertilionid Bats (Chiroptera: Vespertilionidae) from Taiwan

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Liang-Kong Lin, Masaharu Motokawa and Masashi Harada (2002) Karyology of ten vespertilionid bats (Chiroptera: Vespertilionidae) from Taiwan. *Zoological Studies* 41(4): 347-354. The karyotypes of 10 Taiwanese vespertilionid bats were examined for the 1st time. Three *Myotis* species (*M. formosus watasei*, *M. latirostris*, and *M. taiwanensis*) have the standard *Myotis* karyotype of $2n = 44$ with $FN = 50$. The karyotypes of *Plecotus taivanus* ($2n = 32$, $FN = 50$), *Eptesicus serotinus horikawai* ($2n = 50$, $FN = 48$), *Arielulus torquatus* ($2n = 50$, $FN = 48$), and *Murina puta* ($2n = 44$, $FN = 50$) show conservative patterns when compared to other species of the same respective genus. *Scotophilus kuhlii* ($2n = 36$, $FN = 48$) and *Miniopterus schreibersii* ($2n = 46$, $FN = 50$) have karyotypes essentially identical to those previously reported from other regions. *Nyctalus* sp. from Taiwan has the same karyotype of $2n = 36$ as that of *N. velutinus* from China and should be separated from *N. noctula* ($2n = 42$) based on the different karyotypes.
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During a series of intensive bat surveys conducted in Taiwan in recent years (see Lin et al. 1997), one new bat species for Taiwan has been described (Csorba and Lee 1999) and some species were newly recorded for Taiwan (Lin et al. 1997). In addition to traditional taxonomic comparisons by morphological characters, karyotypic studies not only can provide information on phylogenetic relationships but also have great value in solving systematic problems (Yoshiyuki 1989, Yoo and Yoon 1992, Sreepada et al. 1996, Volleth et al. 2001). Until now, karyotypes of only 4 species of bats belonging to the Rhinolophidae and Hipposideridae in Taiwan have been compared with their Japanese counterparts (Ando et al. 1980).

On the other hand, vespertilionid bats (Family: Vespertilionidae) have a wide geographical distribution and constitute most of the species

in Taiwan, where there are at least 18 species of vespertilionid bats (Lin unpubl. data). Specimens of 1 species each in the genera *Barbastella*, *Pipistrellus*, *Murina*, *Myotis*, and *Kerivoula* need further taxonomic studies in order to clarify their specific status, and 2 species of *Harpiocephalus* and *Vespertilio* lack specimens for karyotyping. As for the comparisons with *Pipistrellus abramus* of Japan, the karyotype of *Pipistrellus abramus* from Taiwan was presented in another paper (Lin et al. unpubl. data). Herein we analyze the karyotypes of 10 Taiwanese vespertilionid bats.

MATERIALS AND METHODS

Thirty-seven live specimens of 10 species of vespertilionid bats were captured in central and southern Taiwan (Table 1). These specimens

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were identified according to morphological characters following Jones et al. (1971) and Lin et al. (1997). Specimens were deposited in the Department of Biology, Tunghai Univ. and the Laboratory Animal Center, Osaka City Univ. Chromosome preparations were made from primary lung tissue culture cells by the methods of Harada and Yoshida (1978). Nomenclature of chromosomes follows Levan et al. (1964). The diploid number (2n) was determined by observing 30 metaphase cells in each specimen, and the fundamental number (FN) was identified as the total

number of autosomal arms.

RESULTS AND DISCUSSION

This is the 1st report on the karyotypes of the 10 vespertilionid bat species in Taiwan (Table 2). Three species of the genus *Myotis* (*M. formosus watasei*, *M. latirostris*, and *M. taiwanensis*) all have the standard of *Myotis* karyotype (2n = 44 and FN = 50) (Fig. 1A-C). The karyotypes of the 3 species are comprised of 3 large and 1 small pairs of meta-

Table 1. Vespertilionid bats specimens examined in this study

Species	No of specimens examined		Collection site
	♂	♀	
<i>Myotis formosus watasei</i>	1	1	Nanzenshan, Manchou Township, Pingtung Co.
<i>M. latirostris</i>	2	0	Tsuifeng, Jenai Township, Nantou Co.
	1	0	Dahsueshan, Heping Township, Taichung Co.
<i>M. taiwanensis</i>	0	1	Yongfu, Chungliiao Township, Nantou Co.
	3	2	Puli Township, Nantou Co.
<i>Plecotus taivanus</i>	2	1	Tsuifeng, Jenai Township, Nantou Co.
	0	1	Dahsueshan, Heping Township, Taichung Co.
<i>Scotophilus kuhlii</i>	0	1	Hsinhua City, Tainan Co.
	1	1	Taichung City
<i>Eptesicus serotinus horikawai</i>	1	0	Chunshan, Alisan Township, Chiayi Co.
	1	0	Chichi Township, Nantou Co.
<i>Arielulus torquatus</i>	1	1	Chinchiufong, Tsaotun Township, Nantou Co.
<i>Nyctalus velutinus</i>	0	2	Dantai, Hsinyi Township, Nantou Co.
<i>Miniopterus schreibersii</i>	0	1	Paoshan, Taoyuen Township, Tainan Co.
	5	0	Puli Township, Nantou Co.
	2	0	Meishan Township, Chiayi Co.
	4	0	Kenting, Hengchun Township, Pingtung Co.
<i>Murina puta</i>	1	0	Wansheng, Hsinyi Township, Nantou Co.

Table 2. Karyotype data on 10 species of Taiwanese vespertilionids

Species	2n	FN	Autosome pair			Sex chromosomes	
			M-SM	ST	A	X	Y
<i>Myotis formosus watasei</i>	44	50	4	0	17	SM	A
<i>M. latirostris</i>	44	50	4	0	17	SM	A
<i>M. taiwanensis</i>	44	50	4	0	17	SM	A
<i>Plecotus taivanus</i>	32	50	10	0	5	SM	A
<i>Scotophilus kuhlii</i>	36	48	7	0	10	SM	A
<i>Eptesicus serotinus horikawai</i>	50	48	0	0	24	SM	A
<i>Arielulus torquatus</i>	50	48	0	0	24	SM	A
<i>Nyctalus velutinus</i>	36	52	7	2	8	M	?
<i>Miniopterus schreibersii</i>	46	50	3	0	19	SM	A
<i>Murina puta</i>	44	50	4	0	17	M	A

M, metacentrics; SM, submetacentrics; ST, subtelocentrics; A, acrocentrics.

centric or submetacentric autosomes, seventeen pairs of acrocentric autosomes ranging from middle-sized to small, a medium-sized submetacentric X chromosome, and a relatively small acrocentric Y chromosome. No differences were found in chromosome morphology among the 3 species. As pointed out by Baker and Patton (1967), *Myotis* species possess the conservative form of karyotypes and represent the primitive karyotype of ves-

perilionid bats. It has been assumed that karyotypes of many vespertilionid bats are mainly derived from that of a *Myotis*-like bat by Robertsonian translocation (centric fusion or fission) (see McBee et al. 1986, Harada 1988). Although the 3 *Myotis* species in Taiwan have a distinctive external morphology and belong to different subgenera, i.e., *M. formosus watasei* belongs to the subgenus *Chrysopteron*, *M. latirostris* to *Selysius*, and *M. tai-*

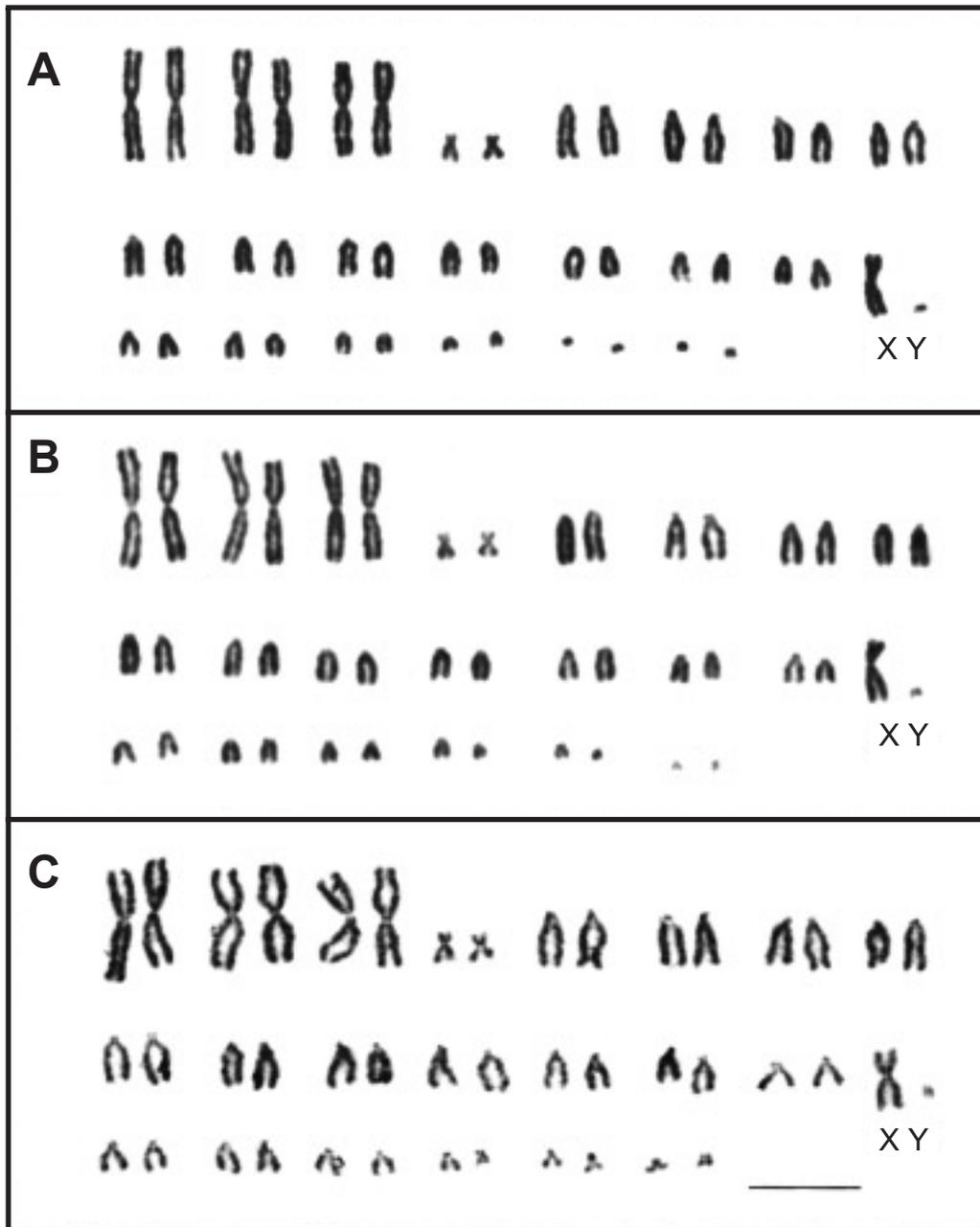


Fig. 1. Conventional karyotypes of *Myotis formosus watasei* (A), *M. latirostris* (B), and *M. taiwanensis* (C). Bar = 10 μ m.

wanensis to *Leuconoe* (see Corbet and Hill 1992), their karyotypes appear to be identical to one another in gross morphology. This fact suggests that the karyotype is an extremely stable feature in these 3 bats, whereas the external characters are much more plastic. In order to elucidate their phylogenetic relationships, molecular analyses from these 3 species should be performed in the future.

The endemic species, *Plecotus taivanus*, was discovered in 1991 (Yoshiyuki 1991). The karyotype of this species is $2n = 32$ and $FN = 50$ (Fig. 2A), and is comprised of 9 large and 1 small pairs of metacentrics or submetacentrics and 5 pairs of acrocentrics. The X chromosome is a medium-sized submetacentric, and the Y chromosome is a small acrocentric. Secondary constriction was found in the centromere region of the smallest metacentric pair. Comparing *P. taivanus* with other Asian *Plecotus* species, the karyotype of this species is similar to those of *P. auritus sacrimontis* of Japan ($2n = 32$ and $FN = 50$; Ono and Obara 1994) and *P. austriacus* of Europe ($2n = 32$ and $FN = 50$; Fedyk and Fedyk 1970). The karyotype of *Plecotus* has been regarded as having been derived from a *Myotis*-like karyotype by centric fusion (Harada 1988). By contrast, the X chromosome of *P. (Corynorhinus) mexicanus* ($2n = 32$, $FN = 50$; Bogdanowicz et al. 1998) from North America is acrocentric, and only 9 pairs of autosomes are metacentric or submetacentric (Lopez-W et al. 1995). These differences in karyotypes indicate that the species of *Plecotus* in Eurasia and North America probably have distinct lineages. Recently, a morphological and chromosomal study by Bogdanowicz et al. (1998) also confirmed that *Plecotus* species should be limited to Palaearctic areas, while the former subgenus taxon, *Corynorhinus*, should be given valid generic designation.

The karyotype of *Scotophilus kuhlii* from Taiwan is $2n = 36$ and $FN = 48$ (Fig. 2B), and has 7 large to small pairs of metacentrics or submetacentrics, and 10 medium to small pairs of acrocentrics. Secondary constriction was found in the smallest acrocentric pair. The X chromosome is a medium-sized submetacentric, and the Y chromosome is a small acrocentric. Two species of *Scotophilus* (*S. kuhlii* and *S. heathii*) are found in Asia (Corbet and Hill 1992). The karyotype of *S. temmincki* (= *kuhlii*) from Sabah, Malaysia reported by Harada and Kobayashi (1980) and *S. kuhlii* from Thailand (Harada et al. 1982a) and India (Sreepads and Gururaj 1994) share the same $2n = 36$ and $FN = 48$ karyotype with the Taiwanese

species.

Eptesicus serotinus horikawai from Taiwan has a karyotype with $2n = 50$ and $FN = 48$ (Fig. 2C). All species in the genus of *Eptesicus*, except for the small African *E. capensis*, have 24 medium to small acrocentric pairs of gradually decreasing size (see McBee et al. 1985, Volleth et al. 2001). A secondary constriction was found in the arm of 1 pair. The X chromosome is a medium-sized submetacentric, and the Y chromosome is a small acrocentric. Only the X chromosome is banded. A notable karyotypic variation in the genus *Eptesicus* is the morphology of the Y chromosome of *E. serotinus* from Poland, which is submetacentric rather than the typical acrocentric (Fedyk and Fedyk 1970), but this difference was not reported in a more recent study (see Zima and Horacek 1985).

Arielulus torquatus was first described as *Pipistrellus* sp. based on its external characters and number of premolars (Lin et al. 1997), and later described as a new species from Taiwan by Csorba and Lee (1999). The chromosomal complement of this species is comprised of 24 pairs of acrocentric chromosomes varying in size from medium to small, a medium submetacentric X, and a small acrocentric Y chromosome (Fig. 2D). According to the karyological features of *A. torquatus* and *E. serotinus horikawai* in this study, these species share the same formula of $2n = 50$ and $FN = 48$, whereas, a difference in size of the Y chromosome was found. Our results support the views of Csorba and Lee (1999) and Volleth et al. (2001), who consider *Arielulus* to be closely related to *Eptesicus*.

A species of *Nyctalus* in Taiwan was first reported as *N. noctula* (Jones 1971, Jones and Mumford 1977). No additional specimens had been reported since that capture (Lin et al. 1997), until we caught females of *Nyctalus* in mountainous areas of central Taiwan in 1999. Because males were not available, the Y chromosome could not be directly determined in this study. The chromosomal number of *Nyctalus* sp. from Taiwan is $2n = 36$. Judging from a karyotypic study of *N. velutinus* with the same $2n$ number from Anhui, China (Zhang 1990), the X chromosome was identified as the smallest metacentric pair. Autosomes of *Nyctalus* sp. from Taiwan are characterized by having 7 large pairs of metacentrics or submetacentrics, three pairs of subtelocentrics of medium to small size, and 7 pairs of medium to small acrocentrics of gradually decreasing size; the $FN = 54$ (Fig. 3A). A secondary constriction was found

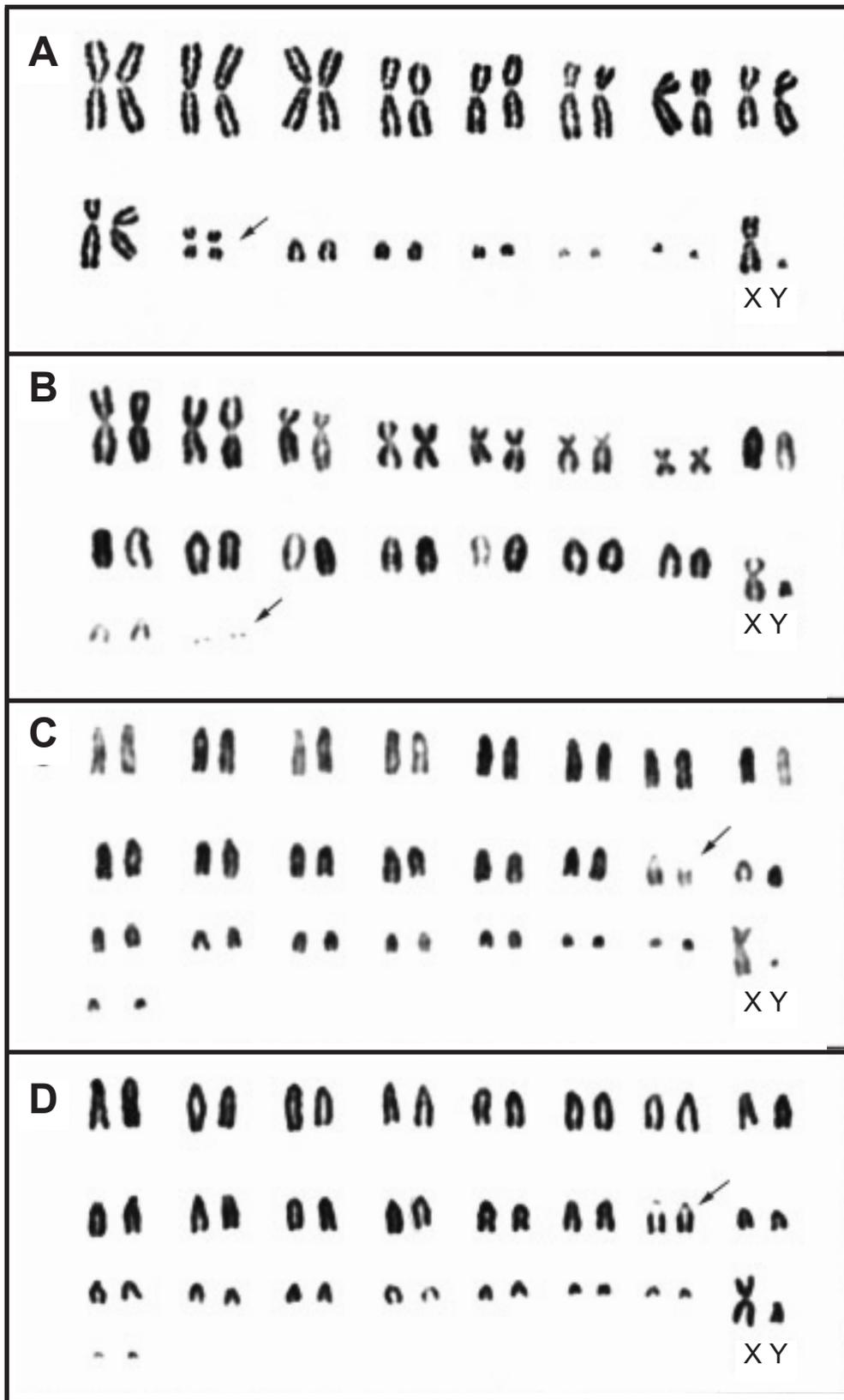


Fig. 2. Conventional karyotypes of *Plecotus taivanus* (A), *Scotophilus kuhlii* (B), *Eptesicus serotinus horikawai* (C), and *Arielulus velutinus* (D). Secondary constrictions are indicated by arrows.

within the arm of the largest acrocentric pair. The X chromosome is a medium-sized metacentric. The Y chromosome is unknown. The karyotype of *Nyctalus* sp. from Taiwan differs from known karyotypes of *Nyctalus* species such as *N. fuvvus* ($2n = 44$, FN = 52, Ando et al. 1977; $2n = 44$, FN = 50, Harada et al. 1982b), *N. lasiopterus aviator* ($2n = 42$, FN = 50, Harada 1973; $2n = 42$, FN = 50, Ando et al. 1977) from Japan, and *N. noctula* ($2n = 42$, FN = 50, Zima 1978) from Europe. The major differences among species are the number of chromosomes and the number of large autosomal pairs

of metacentrics: three pairs in *N. fuvvus*, four in *N. lasiopterus aviator* and *N. noctula*, and 7 in the Taiwanese species. Therefore, additional large metacentric pairs in the species of *Nyctalus* from Taiwan may have been caused by the centromeric fusion of 2 acrocentric elements, as previously revealed in *N. lasiopterus aviator* (Harada et al. 1982b). The karyotype reported by Zhang (1990) for *N. velutinus* from China is similar to our results for the *Nyctalus* sp. from Taiwan. Therefore, we suggest that *Nyctalus* bats ($2n = 36$) from Taiwan should be separated from *N. noctula* ($2n = 42$)

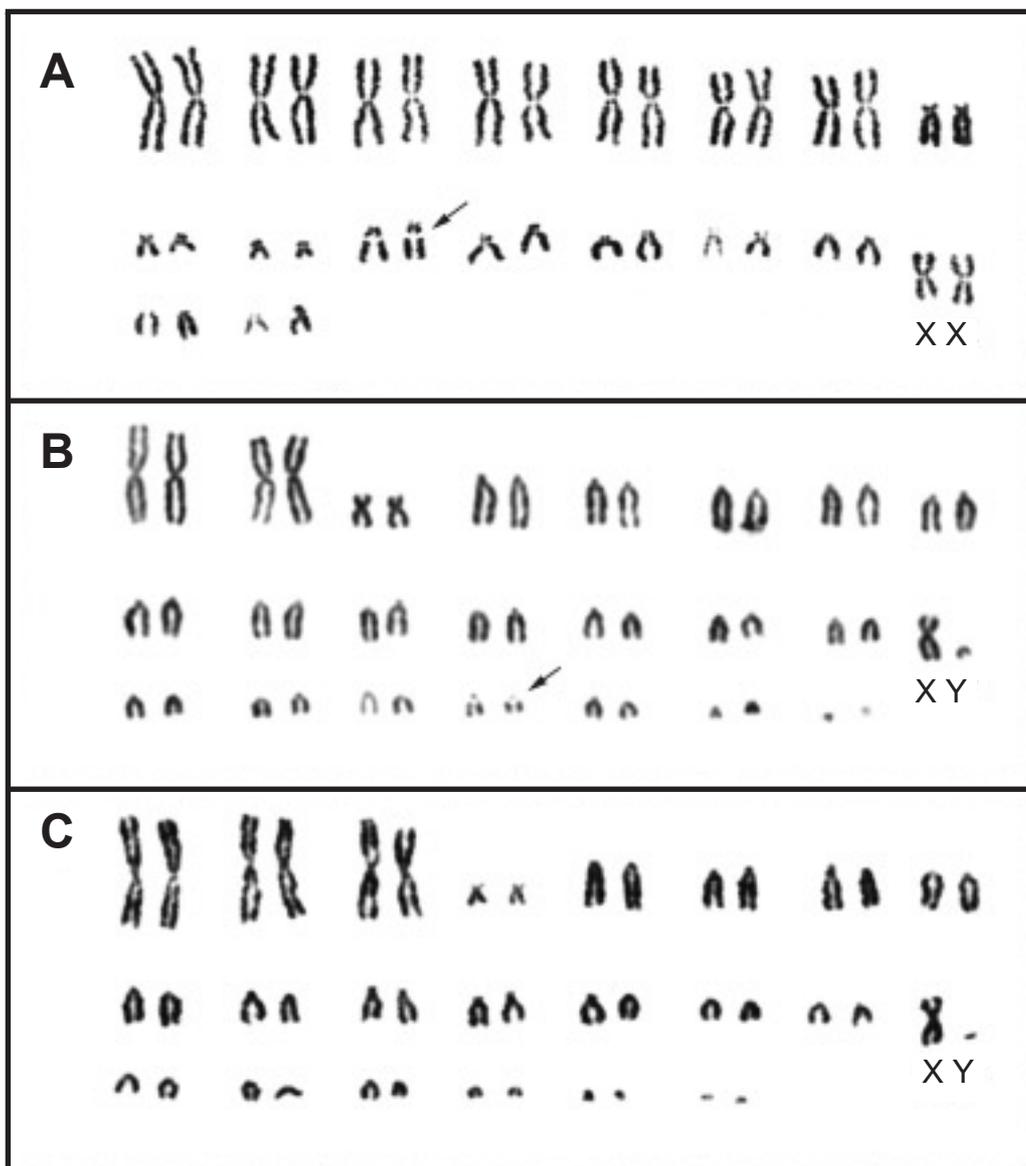


Fig. 3. Conventional karyotypes of *Nyctalus velutinus* (A), *Miniopterus schreibersii* (B), and *Murina puta* (C). Secondary constrictions are indicated by arrows.

owing to different karyotypes and be tentatively treated as *N. velutinus*.

The karyotype of *Miniopterus schreibersii* is $2n = 46$, $FN = 50$ (Fig. 3B); it has 2 large and 1 small pairs of metacentrics or submetacentrics and 19 medium-sized to small acrocentric pairs of gradually decreasing size. A secondary constriction was found in a small acrocentric pair. The X chromosome is a medium submetacentric, and the Y chromosome is a small acrocentric. Our findings on the $2n$ and FN of *M. schreibersii* from Taiwan resemble the karyotypes of conspecifics from Japan (Harada 1973, Ono and Obara 1994), Borneo (Harada and Kobayashi 1980), Korea (Oh 1975), and Europe (Zima and Horacek 1985). However, in Thailand, *M. schreibersii* has a medium-sized subtelocentric pair of chromosomes (Harada et al. 1985, McBee et al. 1986).

Murina puta ($2n = 44$ and $FN = 50$, Fig. 3C), endemic to Taiwan, has 3 large and 1 small pairs of metacentrics or submetacentrics, seventeen medium-sized to small pairs of acrocentrics of gradually decreasing size, a medium-sized metacentric X, and a small acrocentric Y chromosome. Similarities in karyotypes among *Murina* species such as *M. aurata ussuriensis* (Harada et al. 1987) and *M. leucogaster hilgendorfi* (Harada 1973, Harada et al. 1987) from Japan, and *M. leucogaster* from Thailand (McBee et al. 1986) were found.

McBee et al. (1986) suggested that karyotypic data of the family Vespertilionidae show 3 patterns of chromosomal variability among genera. The 1st pattern includes conservative genera in which all species have the same or nearly the same standard karyotypes, such as *Myotis*, *Eptesicus*, *Plecotus*, *Scotophilus*, *Miniopterus*, and *Murina*. The 2nd pattern includes the genera that exhibit interspecific variability such as *Pipistrellus* and *Nyctalus*. The 3rd pattern includes species that possess certain distinct karyotypes such as the genus *Rhogeessa*. Karyotypic attributes of the chromosome complements observed in our study may be consistent with this view. In this study, although most species show the 1st pattern and banding results are not presented, the taxonomic status of some species (e.g., *Nyctalus* from Taiwan) can be elucidated by the standard karyotypic data reported here.

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臺灣十種蝙蝠科蝙蝠之核型

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我們首次報告臺灣地區十種屬於蝙蝠科的蝙蝠之核型。三種鼠耳蝠 (*Myotis*)，即渡賴氏鼠耳蝠 (*M. formosus watasei*)、寬吻鼠耳蝠 (*M. latirostris*) 與臺灣鼠耳蝠 (*M. taiwanensis*)，都具有典型的鼠耳蝠屬的核型，為 $2n$ (雙套) 等於 44，FN (染色體臂數) 為 50。臺灣長耳蝠 (*Plecotus taivanus*) 的核型為 $2n = 32$ ，FN = 50、堀川氏棕蝠 (*Eptesicus serotinus horikawai*) 為 $2n = 50$ ，FN = 48、臺灣黃喉蝠 (*Arielulus torquatus*) 為 $2n = 50$ ，FN = 48 及臺灣管鼻蝠 (*Murina puta*) 為 $2n = 44$ ，FN = 50 皆呈現出同屬異種間所擁有保守的核型。高頭蝠 (*Scotophilus kuhlii*) ($2n = 35$ ，FN = 48) 和摺翅蝠 (*Miniopterus schreibersii*) ($2n = 46$ ，FN = 50) 的核型則與分布其他地區的相同種類所被分析過的核型一樣。臺灣地區屬於山蝠屬 (*Nyctalus*) 的物種其核型為 $2n = 36$ ，與絨山蝠 (*N. velutinus*) 相同而不同於歐洲山蝠 (*N. noctalus*) 的核型 ($2n = 42$)，故臺灣本種應屬於絨山蝠 (*N. velutinus*) 而非歐洲山蝠 (*N. noctalus*)。

關鍵詞：核型，蝙蝠科，細胞分類學。

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