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Karyotypes of Some Species of Vespertilionid Bats from Poland

[With 5 Figs. & 2 Tables]

Chromosomes of 5 species of *Vespertilionidae* are described: *Plecotus auritus* — $2N = 32$, $NF = 54$, *Plecotus austriacus*, — $2N = 32$, $NF = 54$, *Myotis daubentoni* — $2N = 44$, $NF = 54$, *Nyctalus noctula* — $2N = 46$, $NF = 54$, and *Eptesicus serotinus* — $2N = 50$, $NF = 52$. The obtained results were compared with earlier data to show geographic variability of the chromosome formula in *M. daubentoni*, and differences in the morphology of Y chromosome in *Eptesicus serotinus*. Possible mechanisms of differentiation of karyotypes in *Vespertilionidae* were also discussed and some phylogenetic conclusions presented.

I. INTRODUCTION

Until now the chromosomes of only few species of Palearctic *Vespertilionidae* have been described. Matthey & Bovey (1948) and Bovey (1949) described karyotypes of nine species: *Myotis myotis*, *M. daubentoni*, *M. mystacinus*, *M. emarginatus*, *Pipistrellus pipistrellus*, *P. nathusi*, *Miniopterus schreibersi*, *Plecotus auritus*, *Barbastella barbastellus*. The results of these studies were not accurate since in this period colchicine was not yet in use. For this reason some of the above mentioned species were reinvestigated (Capanna & Civitelli, 1964; 1966; 1967; Capanna, Conti & Renzis, 1968; Radjabl *et al.*, 1969). Also several other Palearctic species of *Vespertilionidae* were described: *Myotis capaccini* (Capanna, Civitelli & Spagnuolo, 1968), *M. nattereri* (Strelkov & Volobuev, 1969), *Nyctalus noctula* (Dulić *et al.*, 1967), *Pipistrellus tralatus* (Takayama, 1959), *Vespertilio murinus*, *V. superans* and *Eptesicus serotinus* (Vorontsov *et al.* 1969).

Baker & Patton (1967) described karyotypes of 32 species of North American *Vespertilionidae*. They found that some species (*e.g.* belonging to *Myotis* genus) have identical karyotypes as Eurasian species, but other (genera *Pipistrellus* and *Plecotus*) differ in the respect of karyotypes from Palearctic species.

In the present study karyotypes of 5 species of Palearctic *Vespertilionidae* are described: *Plecotus auritus* (Linnaeus, 1758), *Plecotus austriacus* Fischer, 1829, *Myotis daubentoni* (Kuhl, 1819), *Nyctalus leisleri* (Kuhl, 1818), and *Eptesicus serotinus* (Schreber, 1774).

II. MATERIAL AND METHOD

Altogether 13 individuals of vespertilionid bats from Poland were examined. Place of capture, sex and number of individuals of each species are given in Table 1. Skulls and skins of these specimens used for investigations of chromosomes are preserved in the collection of the Mammals Research Institute, Polish Academy of Sciences, at Białowieża.

Chromosome preparations were obtained from the spleen by a method described by Dulić (1966). One hour before sacrifice of the animals intraperitoneal injections of 0.5 ml Colcemid (Ciba) were given. A hypotonic shock (0.75% water solution of sodium citrate) was applied for 30 min. Slides were stained with lacto-aceto-orceine.

III. RESULTS

A summary of the results of this study (sex, chromosome number, fundamental number and morphological types of chromosomes) is shown in Table 2. A detailed karyotype description for each species is given below.

Table 1.
Specimens examined.

Species	No. of specimens (♂♂ + ♀♀)	Trapping locality
<i>Plecotus auritus</i>	2 —	Białowieża (N 52°42' E 23°51')
	2 —	Raciążek (N 52°52' E 18°49')
<i>Plecotus austriacus</i>	— 1	Kowal (N 52°32' E 19°09')
	1 —	Raciążek
<i>Myotis daubentoni</i>	— 1	Białowieża
<i>Nyctalus leisleri</i>	— 1	Białowieża
<i>Eptesicus serotinus</i>	2 1	Kowal
	1 1	Białowieża

Plecotus auritus (Fig. 1). ($2N = 32$, $NF = 54$). Autosomes consist of 9 pairs of large metacentrics and submetacentrics, one pair of small metacentrics, and 5 pairs of small acrocentrics. The X chromosome is an average size submetacentric, Y is a dot-like.

Plecotus austriacus (Fig. 2). ($2N = 32$, $NF = 54$). The karyotype of this species is identical with that described above of *P. auritus*.

Myotis daubentoni (Fig. 3). ($2N = 44$, $NF = 54$). This species has 3 pairs of large and one pair of small metacentrics, and 17 pairs of acrocentrics of gradually decreasing dimensions — from medium to small. The X chromosome is a medium size submetacentric.

Nyctalus leisleri (Fig. 4). ($2N = 46$, $NF = 54$). This species has 3 pairs of large metacentric autosomes, similar to those in *Myotis daubentoni*. All the remaining autosomes (19 pairs) are acrocentric, of gradually

diminishing size, from medium to small. The X chromosome is a medium size submetacentric.

Eptesicus serotinus (Fig. 5). ($2N = 50$, $NF = 52$). All autosomes (24 pairs) are acrocentric grading in size. The X chromosome is a large submetacentric, Y — small submetacentric.

IV. DISCUSSION

The great progress of cytologic techniques in recent years allows now for a detailed analysis of mammalian karyotypes. For this reason reinvestigations of chromosomes of some species described earlier are very useful. On the other hand it is also essential to carry on cytogenetic studies in various locations of a given species to detect geographic variability of the karyotype.

Table 2.

Somatic chromosome numbers and morphology of chromosomes.

Species	Sex	2N	NF	Autosomes ¹⁾		Heterochromosomes ²⁾	
				M+SM, A		X	Y
<i>Plecotus auritus</i> , 4 ♂		32	54	10	5	SM	dot-like
<i>Plecotus austriacus</i> , 1 ♂, 1 ♀		32	54	10	5	SM	dot-like
<i>Myotis daubentoni</i> , 1 ♀		44	54	4	17	SM	?
<i>Nyctalus leisleri</i> , 1 ♀		46	54	3	19	SM	?
<i>Eptesicus serotinus</i> , 3 ♂, 2 ♀		50	52	—	24	SM	SM

¹⁾ No. of pairs of autosomes, ²⁾ Morphology of sex chromosomes, M — metacentric, SM — submetacentric, A — acrocentric.

Bovey (1949) gave the first description of the *Plecotus auritus* karyotype. The present study confirms these results and points out the identity of chromosome formula of *P. auritus* with the karyotype of *P. austriacus*. The same karyotype was found also in *Barbastella barbastellus* (Capanna, Conti & Renzis, 1968). On the other hand North American species from the *Plecotus* genus show karyotypes differing from those found in species of the Old World. These differences indicate the effect of geographic isolation on the karyotype evolution.

Bovey (1949) found in the karyotype of *Myotis daubentoni* from the terrain of Switzerland 4 pairs of large metacentrics ($2N = 42$, $NF = 54$). On the other hand in the Soviet Union other karyotype of this species was reported: $2N = 44$, $NF = 56$ (Strelkov & Volobuev, 1969). These authors classify as metacentrics, apart from three pairs of large and one pair of small chromosomes, also one of the two smallest pairs of autosomes. The karyotype of *M. daubentoni* described here differs

from the descriptions given above for the area of Switzerland and Soviet Union but is identical with the description of karyotypes in all species in the *Myotis* genus, i. e. $2N = 44$, $NF = 54$ (Capanna, Civitelli & Spagnuolo, 1968; Baker & Patton, 1967, Hsu & Benirschke, 1967; 1968).

In the *Nyctalus* genus until now one karyotype of *N. noctula* (Dulić *et al.*, 1967) has been described in Yugoslavia: $2N = 42$, $NF = 54$. *Nyctalus leisleri* is lacking two pairs of metacentric autosomes in comparison with *N. noctula*. The karyotype of *N. leisleri* is more primitive, shows a higher number of acrocentric chromosomes. The karyotype of *N. noctula* evolved from this karyotype by means of two centric fusions ($2N = 46 \rightarrow 2$ fusions $\rightarrow 2N = 42$).

In the description of chromosomes of *Eptesicus serotinus* from the Soviet Union (Vorontsov *et al.*, 1969) the identity of karyotypes in this species and North American forms (*Eptesicus fuscus*, *E. andinus*, *E. furinialis*) was demonstrated, both in the number of chromosomes and their morphology ($2N = 50$, $NF = 52$). All chromosomes are acrocentric, X — chromosome — submetacentric, Y — small acrocentric. In *Eptesicus serotinus* from Poland Y chromosome is submetacentric (Fig. 5a). Differences in the morphology of this chromosome in the two populations may suggest the presence of pericentric inversion (biarmed Y \rightarrow inversion \rightarrow uniarmed Y, or inversely), or deletion of a shorter arm of the Y chromosome in the American species and *E. serotinus* from the Soviet Union. It seems, however, that differences in the morphology of Y chromosome appeared in some other way. First of all it should be pointed out that NF in species from the *Eptesicus* genus is lower by two units in comparison with remaining *Vespertilionidae*. This difference might appear in the following manner:

1. elimination of one pair of acrocentrics from the karyotype,
2. centric fusion of one acrocentric chromosome with a primarily acrocentric Y chromosome resulting in the formation of bi-armed Y chromosome, while the homologue of transformed acrocentric autosome (univalent) becomes eliminated from the karyotype.

It seems that the second alternative is more probable. This is supported by the fact that in all known vespertilionid bats Y chromosome is acrocentric. On the other hand morphological differentiation of the Y chromosome of *E. serotinus* should be explained by deletion of a shorter arm of this chromosome in populations from the Soviet Union and in North American species, as indicated by much smaller dimensions of Y chromosome in these latter. The existence of sub-species variability in the morphology of heterochromosomes has been described earlier (Baker & Patton, 1967) in *Lasiurus ega*.

V. PHYLOGENETIC CONCLUSIONS

It is known from comparative studies that in karyological respect *Vespertilionidae* from the Old World are very uniform. All Palearctic species of this family, with the exception of the *Eptesicus* genus, have the same number of chromosome arms ($NF = 54$). Bovey (1949) suggested the importance of Robertson processes in the evolution of *Vespertilionidae* karyotypes. These suggestions were confirmed by Capanna (1968b), and Capanna, Conti & Renzis (1968). Capanna (1968a), referring to the existence of three pairs of large metacentric chromosomes in the karyotypes of species that are phylogenetically old and possess many primitive morphological features (*Myotis*), assumes the possibility of two directions of radiation in the evolution of karyotype: a) centric dissociations (fissioning) — leading to a decreased number or total elimination of large metacentrics (*Miniopterus* and *Eptesicus*), b) centric fusions leading to an increased number of large metacentrics (*Barbastella* and *Plecotus*).

It should be emphasized, however, that the existence of processes of fissioning is questionable from the point of view of cellular mechanisms since until now the origin of the additional centromere essential for fissioning has not been explained (cf. White, 1954; 1961). Moreover, no proof is available that some contemporary species showing many primitive morphological features, have also primitive karyotypes (cf. Matthey, 1958).

For this reason it should be assumed that:

- 1) evolution of karyotypes in *Vespertilionidae* occurred mainly by centric fusions and may be explained by cellular mechanics,
- 2) karyotype of initial forms of *Vespertilionidae* was certainly more primitive than the karyotype of contemporary *Myotis*,
- 3) the number of radiational groups was much higher than suggested by Capanna (1968a). *Myotis*, *Barbastella* and *Plecotus* constitute a common group as confirmed by paleontological data (Handley, 1959). On the other hand, genera *Miniopterus*, *Pipistrellus*, *Vespertilio*, *Eptesicus* and *Nyctalus* represent separate radiational groups.

The most primitive karyotype is that of genus *Eptesicus*. Apparently in this genus the most favourable and well balanced gene system evolved relatively promptly and any aberrations disturbing this system were disadvantageous to the evolution of the species. Genus *Eptesicus* represents an example of lack of relationship between the evolution of karyotype and of morphological features.

The presence of various number of chromosomes in species belonging

to genera *Pipistrellus* and *Nyctalus* indicate that processes of divergence of karyotypes occur at present in these genera.

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KARIOTYPY KILKU GATUNKÓW *VESPERTILIONIDAE* Z TERENU POLSKI

Streszczenie

Zbadano 13 okazów nietoperzy należących do pięciu gatunków z rodziny *Vespertilionidae*, pochodzących z trzech miejscowości (Tabela 1) z terenu Polski. Opisano kariotypy: *Plecotus auritus* — $2N = 32$, $NF = 54$, *Plecotus austriacus* — $2N = 32$, $NF = 54$, *Myotis daubentoni* — $2N = 44$, $NF = 54$, *Nyctalus leisleri* — $2N = 46$, $NF = 54$, *Eptesicus serotinus* — $2N = 50$, $NF = 52$. (Fig. 1—5).

Na podstawie porównań z wcześniejszymi danymi wykazano zmienność geograficzną autosomów, podlegającą procesom Robertsona, u *Myotis daubentoni*, oraz zróżnicowanie morfologiczne chromosomu Y u *Eptesicus serotinus* między różnymi populacjami tego gatunku. W dyskusji odrzucono możliwość ewolucji kariotypu *Vespertilionidae* na drodze dysocjacji centrycznych, podkreślając możliwość ewolucji opartej jedynie na fuzjach centrycznych.

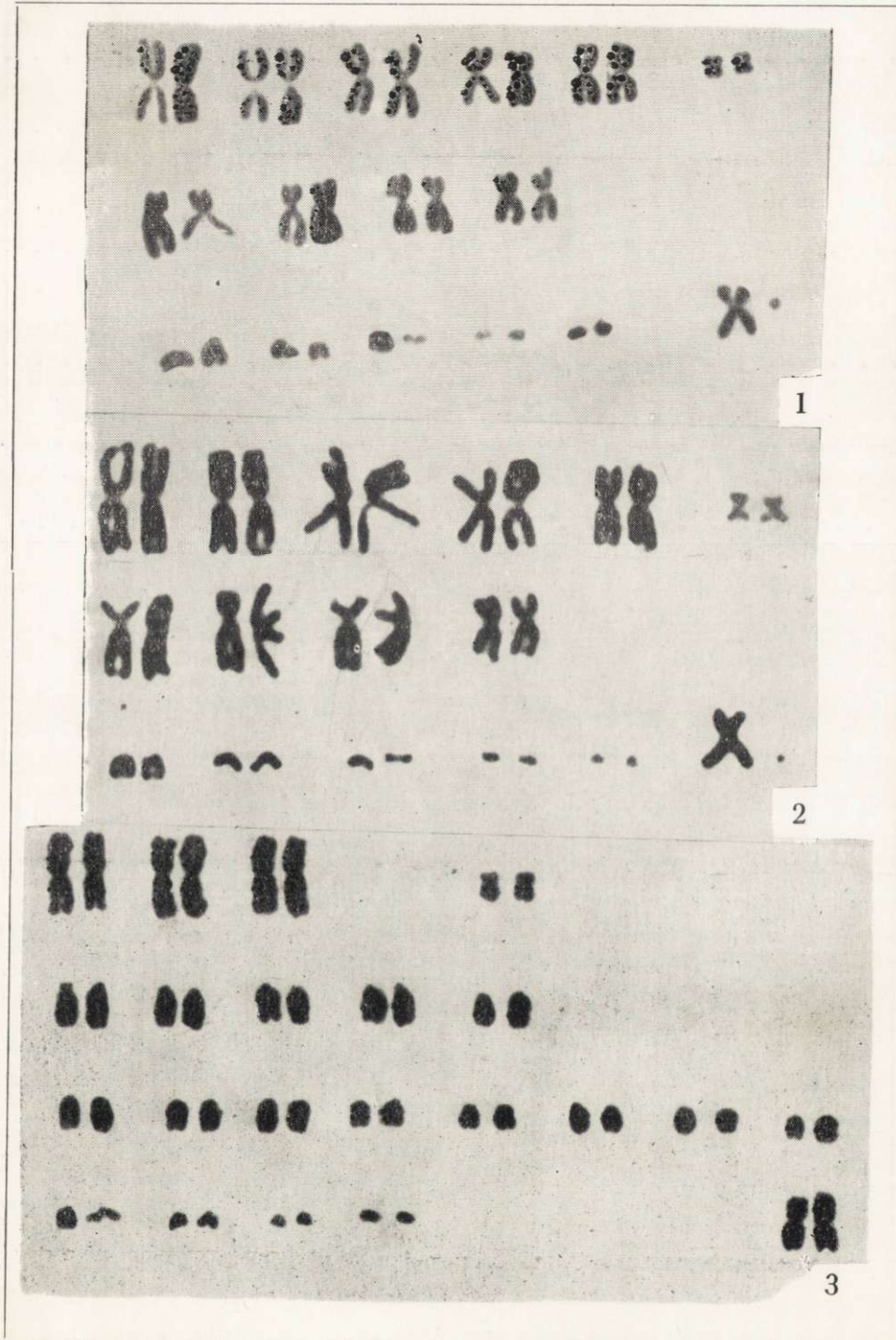
EXPLANATION OF PLATES

Plate IV.

- Fig. 1. *Plecotus auritus*, male karyotype.
Fig. 2. *Plecotus austriacus*, male karyotype.
Fig. 3. *Myotis daubentoni*, female karyotype.

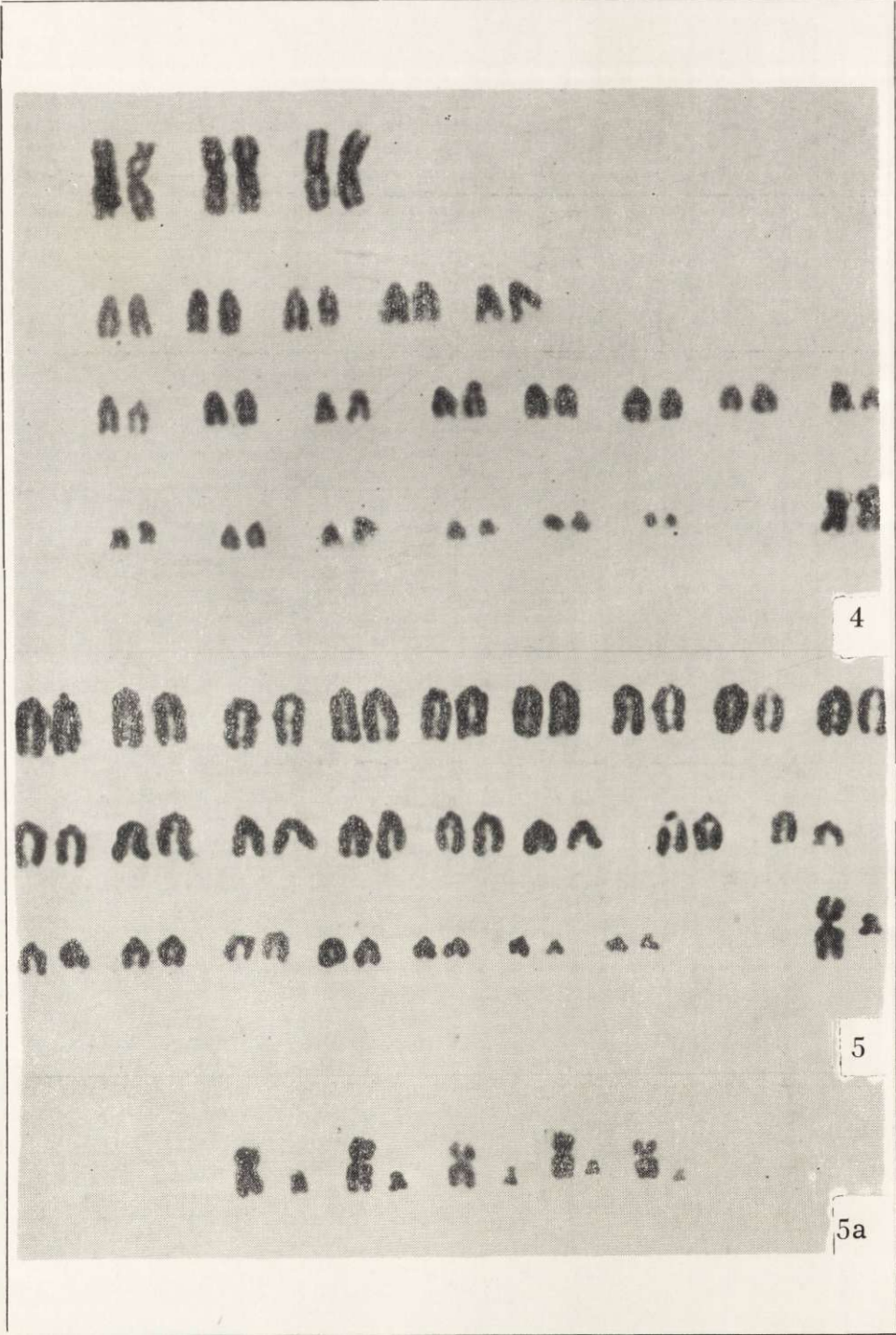
Plate V.

- Fig. 4. *Nyctalus leisleri*, female karyotype.
Fig. 5. *Eptesicus serotinus*, male karyotype.
Fig. 5a. XY pairs from five metaphases plates.



A. Fedyk & S. Fedyk.

auctor phot.



A. Fedyk & S. Fedyk.

auctor phot.