

# Karyotypes of Bats (Chiroptera: Rhinolophidae, Vespertilionidae) from Jordan

الأنماط الصبغية لنوعين من الخفاشيات (فصيلة الخفاشيات

النضوية وفصيلة الخفاشيات الليلية) من الأردن

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**Abstract:** The karyotypes of four bat species of two families (Rhinolophidae and Vespertilionidae) were examined from Jordan. Bat specimens were captured by use of mist-nets and hand-nets from three localities in Jordan in June 2006. Chromosomes were obtained from femoral bone marrow cells and about 20–30 metaphase cells of each animal were examined. The diploid number of chromosomes (2n) and the fundamental numbers of chromosomal arms (NFa) were determined to be 2n=58 and NFa=60 for *Rhinolophus ferrumequinum*, *Rh. euryale* and *Rh. blasii*, and 2n=44, NFa=50 for *Pipistrellus kuhlii*. Karyology of *Rh. euryale* and *P. kuhlii* is investigated from Jordan for the first time. The karyotypes of Jordanian bats show similarity to other populations in other countries with respect to the diploid chromosome number, however, the fundamental chromosome number and the number of autosomal arms are quite different. **Keywords:** karyology, *Rhinolophus ferrumequinum*, *Rhinolophus euryale*, *Rhinolophus blasii*, *Pipistrellus kuhlii*, Chiroptera, Jordan.

**المستخلص:** دُرست الأنماط الصبغية لأربعة أنواع تنتمي لفصيلتين من رتبة الخفاشيات في الأردن (فصيلة الخفاشيات النضوية Rhinolophidae وفصيلة الخفاشيات الليلية Vespertilionidae). جمعت عينات الخفافيش بواسطة شبكة أو باليد من 3 مناطق في الأردن في شهر حزيران لعام 2006. تم الحصول على الصبغيات من خلايا نخاع عظم الفخذ وتم فحص 20-30 خلية في الطور الاستوائي. تبين أن العدد الصبغي المضاعف (2n=58) والعدد الأساسي للأذرع الصبغية الجسمية (NFa=60) لكل من الخفاش النضوي الكبير *Rhinolophus ferrumequinum* و خفاش البحر المتوسط النضوي *Rh. euryale* وخفاش بيتر النضوي *Rh. Blasii*. بينما كان العدد الصبغي المضاعف (2n=44) والعدد الأساسي للأذرع الصبغية الجسمية (NFa=50) لخفاش كهلي *Pipistrellus kuhlii*. للمرة الأولى يدرس النمط الصبغي للنوعين *Rh. euryale* و *P. kuhlii* من الأردن. لوحظ تشابه العدد الصبغي المضاعف (2n) للخفاشيات المدروسة مع مثيلاتها المدروسة في مناطق أخرى من العالم، بينما كان العدد الأساسي للأذرع الصبغية الجسمية (NFa) وعدد أذرع الصبغيات الجسمية مختلف تماماً. **كلمات مدخلية:** الأنماط الصبغية، الخفاشيات، *Rhinolophus ferrumequinum*, *Rhinolophus euryale*, *Rhinolophus blasii*, Chiroptera, *Pipistrellus, kuhlii*. الأردن.

## INTRODUCTION

Although Jordan occupies a small part of the Middle East, it has a relatively rich bat fauna with 24 species belonging to eight families (Qumsiyeh, *et al.* 1998; Amr, *et al.* 2006). Jordan enjoys a very important zoogeographic location, being a meeting point of four zoogeographic regions; Mediterranean, Irano-Turanian, Afro-tropical and Saharo-Arabian (Amr, *et al.* 2006). In the past, Qumsiyeh, *et al.* (1986) and Qumsiyeh and Baker (1985) studied the karyotype for 8 bat species in Jordan.

Within the Middle East, few studies were conducted on the karyotype of bats. In Turkey, Karataş, *et al.* (2004; 2006b) examined the karyotypes for several bat species including *Rhinolophus ferrumequinum*, *Rhinolophus hipposideros* and other vespertilionid bats. The karyotype of *Rh. Ferrumequinum* was studied in Iran (Karataş, *et al.* 2006a). On the other hand, many studies addressed the karyotypes for several species in Europe; Switzerland (Bovey, 1949), Italy (Capanna and Civitelli, 1964; 1966), former Yugoslavia (Đulić, 1966; 1967), former Czechoslovakia (Zima, 1982a), Greece (Zima, *et al.* 1991), where as many of these species occur in the Middle East.

In addition to traditional taxonomical studies of morphological characteristics, karyological comparisons can also provide very useful data on phylogenetic and taxonomic problems (Volleth, *et al.* 2001). Therefore, the aim of the present study is to provide karyotypic data for *Rhinolophus ferrumequinum*, *Rh. euryale*, *Rh. blasii*, and *Pipistrellus kuhlii* from Jordan and to compare data obtained with those recorded from other areas in the Palaearctic Region.

## MATERIALS AND METHODS

Bat specimens were captured by use of mist-nets and hand-nets at three localities in Jordan in June 2006. They were examined with respect to karyological characteristics. Localities and sample size of animals were examined as follows: *-Rh. ferrumequinum*:

Irbid Prov., a big cave in Dibbin National Park (32°14'33.9" N; 35°50'00.1" E) (628 m a.s.l.), 15.VI.2006: 1 ad. ♀; *-Rh. euryale*: Irbid Prov., artificial caves in Zubiya forest (32°26'11.9" N; 35°44'36.3" E) (811 m a.s.l.), 15.VI.2006: 2 ad. ♂♂; *-Rh. blasii*: Irbid Prov., artificial caves in Zubiya forest (32°26'11.9" N; 35°44'36.3" E) (811 m a.s.l.), 15.VI.2006: 2 ad. ♂♂; *-P. kuhlii*: Ma'an Prov., Wadi Musa (Ca. 15 km N of Petra), Al Bayda Wastewater Treatment Station (30°27' N; 35°26' E) (Ca. 900 m a.s.l.), 13.VI.2006: 2 ♀♀.

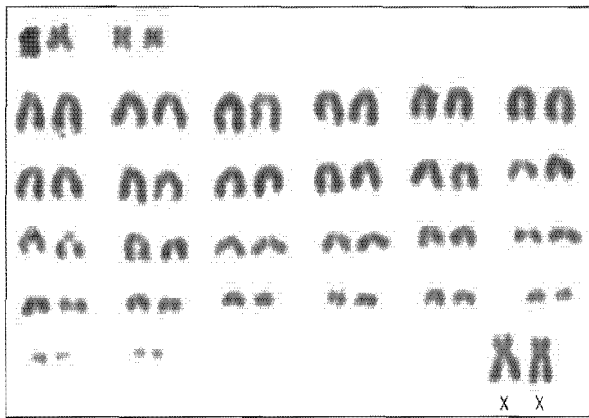
Chromosomes were obtained from femoral bone marrow cells and processed as described by Tjio and Whang (1992). By examining the photographs of about 20–30 metaphase cells of each animal, the diploid number of chromosomes (2n), the fundamental number of chromosomal arms (NF) and the number of autosomal arms (NFa) were determined along with metacentrics, submetacentrics, and acrocentrics with respect to centromere positions. The fundamental number (FN) was counted after constructing the standard karyotypes, only metacentric and submetacentric chromosomes were considered as bi-armed chromosomes (Volobouev, *et al.* 1995). All specimens were skinned in the standard museum type, and their skins, skulls and karyotype preparations have been deposited at the Department of Biology, Jordan University of Science & Technology, Irbid (Jordan).

## RESULTS AND DISCUSSION

### *Rhinolophus ferrumequinum* (Schreber, 1774)

The karyotype of a female from Dibbin (Irbid prov.) was comprised of 58 chromosomes (2n). The number of autosomal arms (NFa) was 60, and the number fundamental (NF) was 64. The X chromosome is one of the largest chromosomes in the set and submetacentric. The Y chromosome was not determined since the animal examined was female. The autosomal set can be divided into two groups based on the centromere position: two pairs of metacentrics, 26 pairs of acrocentrics (Fig.1).

The karyotype of *Rh. ferrumequinum* has been recorded from many countries in Europe,



**Fig. 1.** The Karyotype of a Female *Rh. ferrumequinum* from Dibbin (Irbid prov.) ( $2n = 58$ ,  $NFa = 60$ ).

Asia, and Africa (Table 1). In all populations analysed the  $2n$  value is 58, however, the  $NFa$  value varies between 56 and 62, based on the differences in the number of bi-armed autosomal chromosomes. The karyotype of Jordanian population,  $2n = 58$  and  $NFa = 60$ , is the most common form and also it is consistent with that previously reported from Jordan (Qumsiyeh, *et al.* 1986), former Czechoslovakia (Zima, 1982b), and Kyrgyzstan (Zima, *et al.* 1992). This confirms that the Jordanian population of this bat is resident and the karyotype of the present work falls within the range for this species.

**Table 1.** Karyotypic Records of *Rhinolophus ferrumequinum*, *Rh. euryale*, *Rh. blasii*, and *Pipistrellus kuhlii* (M/Sm: Meta-/Submetacentric, M: Metacentric, Sm: Submetacentric, St: Subtelocentric, A: Acrocentric (Incl. Dot-Like),  $D_M$ : Dot-Like Metacentric,  $D_A$ : Dot-Like Acrocentric) in Different Parts of its Distribution Range.

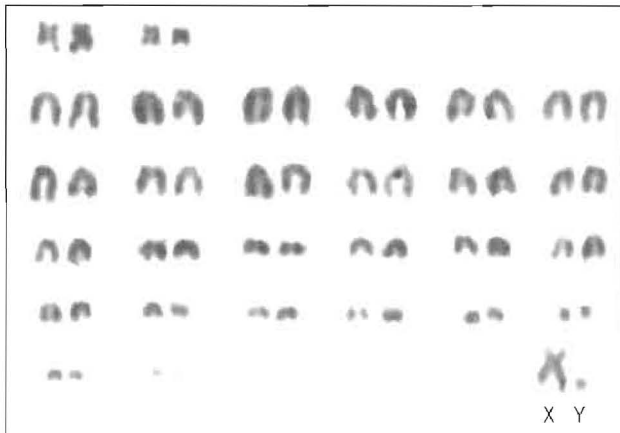
Species/Subspecies	$2n$	M/Sm	St	A	$NFa$	X	Y	Location and Reference
<i>Rh. ferrumequinum</i>	58	0	0	56	56	M	$D_A$	Switzerland (1)
	58	4	0	52	60	M	$D_A$	Italy (2), Former Yugoslavia (3)
	58	4	0	52	60	Sm	A	Tunisia (4), Azerbaijan (5)
	58	6	0	50	62	Sm	A	Azerbaijan (6)
	58	4	0	52	60	M	A	Former Czechoslovakia (7), Jordan (10)
	58	4	0	52	60	Sm	$D_M$	Kyrgyzstan (11a)
	58	4	0	52	60	Sm	-	This study
<i>Rh. f. ferrumequinum</i>	58	4	0	52	60	Sm	$D_A$	Turkey (13c)
<i>Rh. f. creticum</i>	58	6	2	48	64	Sm	A	Crete (8b)
<i>Rh. f. irani</i>	58	2	0	54	58	Sm	$D_A$	Iran (13b)
<i>Rh. euryale</i>	58	0	0	56	56	M	$D_A$	Switzerland (1)
	58	4	0	52	60	Sm	$D_A$	Italy (2a), Former Yugoslavia (3b)
	58	4	0	52	60	Sm	A	Azerbaijan (6)
	58	6	0	48	60	M	$D_M$	Former Czechoslovakia (7a)
	58	4	0	52	60	Sm	$D_M$	Greece (11a)
	58	4	0	52	60	Sm	$D_A$	This study
<i>Rh. blasii</i>	58	4	0	52	60	Sm	A	Italy (2b), Former Yugoslavia (3), Jordan (10)
	58	4	0	52	60	Sm	$D_A$	This study
<i>P. kuhlii</i>	44	8	0	34	50	M	A	Italy (2b), Tunisia (4), Azerbaijan (6), Former Czechoslovakia (7b), Greece (9, 12); Turkey (13a)
	44	10	0	32	52	M	A	Azerbaijan (5)
	44	8	0	34	-	M	-	Ukraine (11a)
	44	8	0	34	50	M	-	This study

(1) Bovey (1949); (2) Capanna and Civitelli (1964, 1966); (3) Đulić (1966, 1967); (4) Baker, *et al.* (1975); (5) Kuliev and Fattaev (1975); (6) Fattaev (1978); (7) Zima (1982a, 1982b); (8) Iliopoulou-Georgudaki (1986); (9) Iliopoulou-Georgudaki and Giagia (1986); (10) Qumsiyeh, *et al.* (1986); (11) Zima, *et al.* (1991, 1992); (12) Volleth, *et al.* (2001); (13) Karatas, *et al.* (2004, 2006a, 2006b).



***Rhinolophus euryale* (Blasius, 1853)**

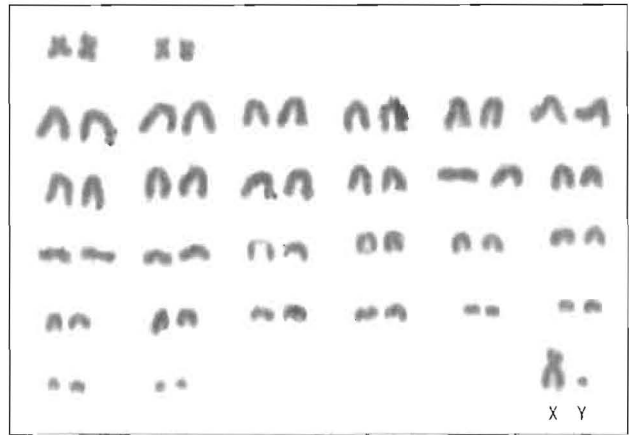
The karyotypes of specimens from Zubiya (Irbid prov.) consisted of 58 chromosomes ( $2n$ ). The number of autosomal arms (NFa) was 60, and the number of fundamental arms (NF) was 64. The autosomal set contained four pairs of medium sized metacentrics, 26 pairs of acrocentrics. The X chromosome was the largest chromosome in the set and submetacentric, and the Y chromosome is dot-like acrocentric (Fig. 2). The morphology of chromosomes is identical with those of *Rh. ferrumequinum* and *Rh. blasii* of Jordan. It is indistinguishable from those reported from Italy, Former Yugoslavia, and Azerbaijan (Capanna and Civitelli, 1964; Đulić, 1967; Fattaev, 1978), but it is different from those from Switzerland (Bovey 1949), former Czechoslovakia (Zima, 1982a) and Greece (Zima, *et al.* 1991) with respect to numbers of chromosomal arms (Table 1).



**Fig. 2.** The Karyotype of a Male *Rh. euryale* from Zubiya (Irbid prov.) ( $2n=58$ , NFa=60).

***Rhinolophus blasii* (Peters, 1866)**

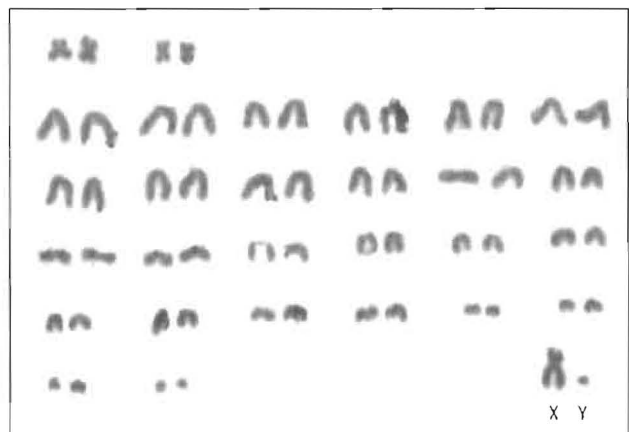
The karyotype of two males of Blasius's horseshoe bat was composed of 58 chromosomes ( $2n$ ). The number of autosomal arms (NFa) was 60, and the number fundamental arms (NF) was 64. The autosomal set contained four pairs of medium sized metacentrics, 26 pairs of acrocentrics. The X chromosome is one of the largest chromosomes in the set and submetacentric, and the Y chromosome is dot-like acrocentric (Fig.3). Karyotype of specimens from Zubiya (Irbid Prov.) is consistent with those reported from Italy, Former Yugoslavia and Jordan (Capanna and Civitelli, 1966; Đulić, 1966, 1967; Qumsiyeh, *et al.* 1986).



**Fig. 3.** The Karyotype of a Male *Rh. blasii* from Zubiya (Irbid prov.) ( $2n=58$ , NFa=60).

***Pipistrellus kuhlii* (Kuhl, 1819)**

The karyotype of two females of Kuhl's Pipistrelles from Wadi Musa near Petra was examined and the diploid number of chromosomes was determined to be  $2n=44$ . The autosomal set contained three pairs of large metacentrics, one pair of submetacentric and 17 pairs of acrocentrics (Fig. 4). The NF and NFa were 54 and 50, respectively. The X chromosome was middle sized metacentric. The Y chromosome was not determined since animal examined was female. The  $2n=44$  karyotype was similarly reported from Italy (Capanna and Civitelli, 1966), Tunisia (Baker, *et al.* 1975), Azerbaijan (Kuliev and Fattaev, 1975; Fattaev, 1978), Former Czechoslovakia (Zima, 1982b), Greece (Iliopoulou-Georgudaki and Giagia, 1986; Volleth, *et al.* 2001), Ukraine (Zima, *et al.* 1991) and Turkey (Karataş, *et al.* 2004). But variations in the number of autosomal arms have been reported in the literature, and also NFa varying from 50 to 52 (Table 1).



**Fig. 4.** The Karyotype of a Male *Rh. blasii* from Zubiya (Irbid prov.) ( $2n = 58$ , NFa = 60).

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