# Morphometric Variability in Horseshoe Bat Species, *Rhinolophus ferrumequinum* and *Rhinolophus lepidus* Captured from District Malakand, Khyber Pakhtunkhwa, Pakistan

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# ABSTRACT

Specimens of *Rhinilophus ferrumequinum* (n = 2) and *R. lepidus* (n = 3) were captured from Malakand district, Khyber Pakhtunkhwa during a three years survey extending from June 2010 through May 2013. Mean body mass of the captured *R. ferrumequinum* specimens was  $18.45 \pm 0.35$  g, head and body length was  $60.11 \pm 1.42$  mm and forearm length was  $60.01 \pm 1.41$  mm. Similarly, average greatest length of skull was  $23.35 \pm 0.20$  mm while the total length of baculum of a single specimen was recorded 4.13 mm. The mean body mass, head and body length and forearm length of the *R. lepidus* specimens captured from the study area was  $3.93 \pm 0.21$  g,  $38.49 \pm 0.54$  mm and  $38.02 \pm 0.63$  mm, respectively. Average greatest length of skull of two specimens was  $15.94 \pm 0.15$  mm while their baculum was  $2.33 \pm 0.14$  mm long.



The genus *Rhinolophus* Lacepede, 1799 has 76 species worldwide of which sixteen species exist in the Indian subcontinent while five have been recorded from Pakistan which include greater horseshoe bat *R. ferrumequinum* Schreber, 1774, the lesser horseshoe bat *R. hipposideros* Bechstein, 1800, the Blasius horseshoe bat *R. blasii* Peters, 1866, the Blyth's horseshoe bat *R. lepidus* Blyth, 1844 and the big-eared horseshoe bat *R. macrotis* Blyth, 1844 (Roberts, 1997; Bates and Harrison, 1997; Simmons, 2005).

*R. ferrumequinum* is widely distributed in northern Himalayan region and extends southwards through the mountains of Waziristan and northern Balochistan. This bat seems to be rare in southern Balochistan because of the lesser supply of suitable insect prey and very dry climate which is unfavorable to the family Rhinolophidae (Roberts, 1997). Specimens have been collected from around Dir town (USNM), Abbotabad (HZM), Karakar pass (FMNH), Gilgit (Bates and Harrison, 1997) and Kululai in Swat (Roberts, 1997). These larger greyer specimens have been assigned to the subspecies *R. f. proximus* (Roberts, 1997). Small colonies found in Balochistan near Kalat, Nushki and Quetta have been assigned to *R. f. irani* (Mirza, 1965; Bates and Harrison, 1997). The greater horseshoe bat is considered to be Least Concerned worldwide (IUCN, 2008; Mahmood-ul-Hassan *et al.*, 2009) and Near Threatened in South Asia (South Asian Chiroptera C.A.M.P. Report, 2002; Walker and Molur, 2003; IUCN, 2008).

*R. lepidus* in Pakistan was captured together with *R. macrotis* during October 1985 near Abbotabad from an elevation of 1280m in Khyber Pakhtunkhwa, Pakistan (Roberts 1997; Bates and Harrison, 1997). The species has also been collected from Afghanistan (Corbet and Hill, 1992) and in the dryer parts of Rajasthan, India (Sinha, 1980). Population status of *R. lepidus* in Pakistan is unknown (Roberts, 1997).



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#### Authors' Contribution

This manuscript is part of PhD Thesis of MS. He collected and analyzed the data and prepared the document. AJ designed and supervised the entire work. FR assisted in surveys to different areas of Malakand. KJI helped in lab work regarding preparation of skull and bacula. FU Assisted in collection of bat samples.

Key words Horseshoe bat, Baculum, *R. lepidus*, Malakand, Abbottabad.

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In Pakistan, the bats are amongst the least studied group of animals. Present survey was therefore planned to ascertain the presence or absence of *Rhinolophus* species from Malakand district, Khyber Pakhtunkhwa, Pakistan.

# **MATERIALS AND METHODS**

### Study area

District Malakand owns variety of habitat types ranging from plain to mountainous areas. River Swat is the natural water body that irrigates cultivable areas in the district. Other sources include water tanks, tube wells and lift pumps. The climate is moderately cool in winter and pleasant in summer, hottest months are June, July and August. The maximum temperature during summer reaches 41 °C and minimum during winter reaches -2 °C. Rose and tulips are wildly grown. Main plant species in Malakand include Bakain (*Melia azerdarach*), Shesham (*Dalbergia sissoo*), Persian walnut (*Juglans regia*), figs (*Ficus carica*), phulai or Paloosa (*Acacia modesta*), white mulberry (*Morus alba*) black mulberry (*Morus nigra*) chilgoza pine (*Pinus gerardiana*) blue gum (*Eucalptus*) globulus) Persian poplar (*Populus caspica*) and chinar (*Platanus orientalis*) are grown in hilly as well as in plain areas. The common wild animals found in the area are jackal, deer, leopard, monkey and wolf. The wild fauna includes the markhor (*Capra falconeri*), the collared pika (*Ochotona rufescens*), the migratory hamster (*Cricetulus migratorius*), the stone marten (*Martes foina*), the forest dormouse (*Dryomys nitedula*), Persian jird (*Meriones persicus*) and the mouse-like hamster (*Calomyscus bailwardi*). The livestock animals in the district are buffalo, cow, sheep, goat, camel, horse, ass and poultry.

#### Sampling strategy

Bats were trapped over duration of three years (2010-2013) on their roosting and foraging grounds using hand net and mist nets at randomly preselected points within Malakand district. Sampling stations were decided based on a pilot study designed to assess the species composition in a variety of habitats. Mist nets of 12 m, 9 m and 6 m size were erected at 10 to 50 m intervals along trails and water bodies, and were monitored for three hours depending on weather and bats' activities.



Fig. 1. Facial (A-C) and cranial (D-G) features of Rhinolophus ferrum equinum.

Table I.- Comparison of mean body mass (g), external body, cranial (n = 2) and bacular (n = 1) measurements (mm) of *Rhinolophus ferrumequinum* (n = 2) captured from Loya Agra in Malakand district with previous studies from South Asia.

			Mea	un±SD		
	Albayrak	<b>Bates and Harrison</b>	Roberts	Benda <i>et al</i> .	Albayrak <i>et al</i> .	Present
	(1985)	(1997)	(1997)	(2010)	(2013)	Study (n = 2)
<b>Body parameters</b>						
Body mass	21.9±4.19	-	25	-	19.3±3.76	18.45±0.35
Head and body	73.2±1.98	65.9±6.6	71	73.5±2.268	72.2±3.39	60.11±1.42
Ear	24.6±2.10	24.3±2.1	24	-	25.2±1.87	$20.62 \pm 0.70$
Forearm	54.2±1.29	58.9±2.2	-	57.55±1.477	53.3±1.72	60.01±1.41
Thumb with claw	-	-	-	-	-	$4.01 \pm 0.01$
2nd metacarpal	-	-	-	-	-	39.56±3.47
1st phalanx on 2nd metacarpal	-	-	-	-	-	$4.01 \pm 0.01$
2nd phalanx on 2nd metacarpal	-	-	-	-	-	18.56±0.64
3 <sup>rd</sup> metacarpal	-	39.3±1.3	-	-	-	40.07±1.34
1 <sup>st</sup> Phalanx on 3 <sup>rd</sup> metacarpal	-	20.5±1.0	-	-	-	21.56±0.64
2 <sup>nd</sup> Phalanx on 3 <sup>rd</sup> metacarpal	-	32.6±2.1	-	-	-	31.57±0.62
3rd phalanx on 3rd metacarpal	-	-	-	-	-	4.01±0.01
4 <sup>th</sup> metacarpal	-	43.9±1.3	-	-	-	4.01±0.01
1 <sup>st</sup> Phalanx on 4 <sup>th</sup> metacarpal	-	12.5±0.9	-	-	-	13.62±0.57
2 <sup>nd</sup> Phalanx on 4 <sup>th</sup> metacarpal	-	20.1±1.3	-	-	-	13.58±0.61
5 <sup>th</sup> metacarpal	-	45.3±0.5	-	-	-	45.66±0.64
1 <sup>st</sup> Phalanx on 5 <sup>th</sup> metacarpal	-	-	-	-	-	16.07±1.33
2 <sup>nd</sup> Phalanx on 5 <sup>th</sup> metacarpal	-	-	-	-	-	18.59±0.63
Wing span	-	356.0	-	-	-	359.00±1.41
Hind foot	13.2±1.19	11.6±1.3	12	-	13.8±0.87	12.11±8.56
Tail	35.0±3.34	34.9±3.0	34	40.0±4.071	37.9±3.46	34.60±0.71
Calcar	-	-	-	-	-	6.51±0.69
Condylo-basal length	20.3±0.32	-	-	-	20.4±0.46	-
Condylo-canine length	-	21.5±0.6	-	21.31±0.443	-	20.78±0.05
Cranial parameters	-	-	-	-	-	_
Greatest length of skull	-	24.2±1.1	-	-	23.8±0.45	$23.35 \pm 0.20$
Condylo-canine length	-	$21.5\pm0.6$	-	-	-	20.78±0.05
Zvgomatic breadth	-	12.4±0.5	-	-	12.0±0.31	12.15±0.12
Interorbital constriction	-	2.6±0.2	-	-	$2.5\pm0.17$	$2.98 \pm 0.02$
Breadth of braincase	-	$9.5\pm0.4$	-	_	$10.2\pm0.26$	$9.69\pm0.23$
Anterior palatal width	-	-	-	-	-	$6.31 \pm 0.01$
Posterior palatal width	-	-	-	-	-	$8.73 \pm 0.14$
Maxillary toothrow	-	9 1±0 3	-	_	8 6±0 18	$9.18\pm0.02$
Mandible length	-	$9.9\pm0.3$	-	_	$15.9\pm0.35$	$1633\pm013$
Mandibular toothrow	-	17.0+0.6	_	_	9 2+0 25	9 86+0 01
Bacular narameters		17.0-0.0			9.2-0.23	9.00-0.01
Total baculum length	_	_	_	_	-	4 13
Width of proximal tin	-	-	-	_	-	0.33
Width of proximal extreme	-	-	-	_	-	0.8
Width of distal extreme	_	_	_	_	_	1 13
Baculum height	-	-	-	-	-	0.83

The nets used throughout the survey were the same and were opened simultaneously at sunset each evening. On either side of the mist net torches were positioned to illuminate bats approaching from either side of the net. Bats were removed and placed in cloth bags (made with a breathable material), and each bat was weighted (g) with a digital scale. The external body, cranial and bacular measurements were taken with a digital vernier caliper (Bates and Harrison, 1997; Javid, 2011).

# **RESULTS AND DISCUSSION**

During present survey, a colony of 46 Rhinolophus ferrumequinum was observed in a cave at Loya Agra tehsil Batkhela in Malakand district (Fig. 1) and two of them were captured through hand net. The length of cave was 40 ft, width of cave opening 4 ft, width of cave middle 8 ft while height of the cave was 7 ft. The captured specimens had a woolly fur, doom shaped head and curved ears that had no tragus. The outer margins of the ears however were curved round to form a prominent antitragus. The muzzle of the bat was well covered with hairs. The wings were delicate, short and rounded in outline. A well-developed interfemoral membrane is supported by calcars which have no lobe of skin beneath them. The front and side view of the nose leaf show that horseshoe apparatus is broad, sella is small and the superior connecting process is broadly triangular in these bats (Fig. 1). The inferior extremity is bluntly rounded and lancet is equilateral triangular in shape. The mean body mass of two R. ferrumequinum was  $18.45 \pm 0.35$  g, their head and body length was  $60.11 \pm$ 1.42 mm while their ear was  $20.62 \pm 0.70$  mm long. Mean thumb with claw and forearm length was  $4.01 \pm 0.01$  mm and  $60.01 \pm 1.41$  mm, respectively. Average length of  $2^{nd}$ metacarpal was  $39.56 \pm 3.47$  mm, length of 3<sup>rd</sup> metacarpal was  $40.07 \pm 1.34$  mm, length of 4<sup>th</sup> metacarpal was 43.62  $\pm$  0.57 mm while average length of 5<sup>th</sup> metacarpal was  $45.66 \pm 0.64$  mm. Average wingspan and calcar length was  $359.00 \pm 1.41$  mm and  $6.51 \pm 0.69$  mm, respectively. Their tibia, tail and hind foot lengths were  $26.06 \pm 1.34$ mm,  $34.60 \pm 0.71$  mm and  $12.11 \pm 8.56$  mm, respectively (Table I).

Average greatest length of skull (n = 2) of *R*. ferrumequinum specimens (Fig. 1) was  $23.35 \pm 0.20$  mm, breadth of braincase  $9.69 \pm 0.23$  mm and zygomatic bone of the two specimens was  $12.15 \pm 0.12$  mm long. The postorbital constriction was  $2.98 \pm 0.02$  mm, condylocanine length was  $20.78 \pm 0.05$  mm while anterior and posterior palatal widths were  $6.31 \pm 0.01$  mm and  $8.73 \pm 0.14$  mm, respectively. The maxillary toothrow length, mandibular toothrow length and mandible length was  $9.18 \pm 0.02$  mm,  $9.86 \pm 0.01$  mm and  $16.33 \pm 0.13$  mm, respectively.

Total baculum length of a single *R. ferrumequinum* specimen was 4.13 mm, width of proximal tip 0.33 mm, width of middle extreme 0.80 mm, width of distal extreme 1.13 mm while baculam height was recorded 0.83 mm (Table I, Fig. 2).



Fig. 2. Bacular features of Rhinolophus ferrumequinum.

Specimens (n = 3) of *Rhinolophus lepidus* were captured from Malakand University in Malakand district through mist nets during present survey. The horseshoe in this species is 6.0-8.0 mm broad and does not cover the whole muzzle (Fig. 3). The sella is generally narrow. The lancet is well developed. The pelage color is typically grey brown dorsally and slightly pale ventrally. Average body mass, head and body length and ear of three Blyth's horseshoe bat was  $3.93 \pm 0.21$  g,  $38.49 \pm 0.54$  mm and 14.80 $\pm 0.27$  mm, respectively. The mean thumb with claw length and forearm length was  $3.87 \pm 0.13$  mm and  $38.02 \pm 0.63$ mm, respectively (Table II). The length of 2<sup>nd</sup> metacarpal, 3<sup>rd</sup> metacarpal, 4<sup>th</sup> metacarpal and 5<sup>th</sup> metacarpal was 27.98  $\pm 0.76$  mm, 28.21  $\pm 0.01$  mm, 29.61  $\pm 0.01$  mm and 29.43  $\pm$  0.01 mm, respectively. Average wingspan, tibia, calcar, hind foot and tail length was  $232.67 \pm 1.15$  mm,  $15.83 \pm$ 0.67 mm, 8.67  $\pm$  0.58 mm, 7.86  $\pm$  0.28 mm and 18.96  $\pm$ 3.24 mm, respectively (Table II).

The mean greatest length of skull (Fig. 3) of R. *lepidus* specimens (n = 2) captured from University of Malakand was  $15.94 \pm 0.15$  mm, breadth of braincase 6.76  $\pm$  0.06 mm, zygomatic bone 7.61  $\pm$  0.01 mm, post-orbital constriction  $2.34 \pm 0.01$  mm and condylo-canine length was  $13.90 \pm 0.08$  mm. Anterior and posterior palatal width was  $3.73 \pm 0.01$  mm and  $5.72 \pm 0.01$  mm, respectively. The maxillary toothrow length, mandibular toothrow length and mandible length was  $5.86 \pm 0.02$  mm,  $6.57 \pm 0.64$  mm and  $10.34 \pm 0.04$  mm, respectively (Table II). The mean total baculum length, width of proximal extreme, shaft length, width of middle extreme, width of distal extreme and baculum height of R. lepidus (n = 2) was  $2.33 \pm 0.14$ mm,  $0.01 \pm 0.00$  mm,  $2.16 \pm 0.12$  mm,  $0.01 \pm 0.00$  mm,  $0.49 \pm 0.05$  mm and  $0.55 \pm 0.00$  mm, respectively (Table II, Fig. 4).

	Bates and Harrison, (1997)	Shahbaz <i>et al.</i> (2014) (n=10)	Present Study (n=3)
Body parameters			
Body mass	-	6.42 (5.0-8.20)	3.93±0.21(3.70-4.10)
Head and body	42.9±4.8 (35.0-54.0)	42.36 (41-44.6)	38.49±0.54(38.17-39.11)
Ear	16.9±1.3(14.5-20.6)	13.49 (11-16)	14.80±0.27 (14.61-15.11)
Horseshoe	-(6.0-8.0*)	-	6.00±0.00(6.00-6.00)
Forearm	39.8±1.0 (37.0-41.8)	40.34 (39-42)	38.02±0.63(37.39-38.64)
Thumb with claw	-	-	3.87±0.13 (3.76-4.02)
2 <sup>nd</sup> metacarpal	-	-	27.98±0.76 (27.11-28.52)
1st phalanx on 2nd metacarpal	-	-	9.00±1.00 (8.00-10.00)
3 <sup>rd</sup> metacarpal	30.4±1.3(28.2-33.3)	31.28 (30-32.4)	28.21±0.01(28.20-28.22)
1st Phalanx on 3rd metacarpal	11.8±0.6(10.0-13.3)	12.35 (11.7-13)	10.66±0.32(10.45-11.03)
2 <sup>nd</sup> Phalanx on 3 <sup>rd</sup> metacarpal	17.3±0.6(16.0-18.9)	19 (17-23)	16.02±0.01(16.01-16.02)
4 <sup>th</sup> metacarpal	31.4±1.0(29.6-33.8)	32.0 (30.8-33)	29.61±0.01(29.61-29.62)
1st Phalanx on 4th metacarpal	8.7±0.5(7.6-10.5)	9.68 (9-11)	8.82±0.23(8.67-9.08)
2 <sup>nd</sup> Phalanx on 4 <sup>th</sup> metacarpal	10.8±0.7(9.6-12.3)	11.55 (9-13)	9.78±0.29(9.61-10.11)
5 <sup>th</sup> metacarpal	31.1±0.9(29.4-33.4)	31.48 (30.4-32.7)	29.43±0.01(29.43-29.44)
1st phalanx on 5th metacarpal	-	10.61 (9.8-11.5)	9.66±0.20 (9.43-9.81)
2 <sup>nd</sup> phalanx on 5 <sup>th</sup> metacarpal	-	-	9.70±0.53 (9.09-10.02)
Wing span	244.0±12.0(232.0-256.0)	240.10 (220.9-250.2)	232.67±1.15(232.00-234.00)
Tibia	16.7±0.7(14.9-18.4)	17.19 (16.5-18.1)	15.83±0.67(15.20-16.53)
Hind foot	7.6±1.0(5.5-10.0)	8.78 (8-10.5)	7.86±0.28(7.62-8.17)
Tail	20.4±4.0(14.0-28.0)	22.79 (21-26)	18.96±3.24 (16.19-22.52)
Cranial parameters			
Condylo-canine length	14.6±0.5 (13.8-15.5)	14.2 (14-14.4)	13.90±0.08(13.84-13.96)
Maxillary toothrow	6.1±0.3(5.6-6.8)	6.75 (6.5-7)	5.86±0.02(5.84-5.87)
Mandibular toothrow	6.6±0.3(6.0-7.4)	5.6 (5.5-5.8)	6.57±0.64(6.12-7.02)
Greatest length of skull	17.2±0.7(16.2-18.4)	17.5 (16.8-17.5)	15.94±0.15(15.83-16.04)
Mandible length	11.0±0.5(10.0-12.1)	11.2 (11-11.4)	10.34±0.04(10.31-10.37)
Posterior palatal width	5.9±0.3(5.7-6.3)	6.3 (6.2-6.4)	5.72±0.01(5.71-5.72)
Zygomatic breadth	8.2±0.3(7.6-8.8)	8.5 (8.3-8.7)	7.61±0.01(7.60-7.61)
Breadth of braincase	7.1±0.3(6.5-7.8)	7.5 (7.3-7.7)	6.76±0.06(6.72-6.80)
Postorbital constriction	2.2±0.1(1.8-2.6)	2.3 (2.2-2.4)	2.34±0.01(2.33-2.34)
Anterior palatal width	4.0±0.1(3.7-4.2)	4.3 (4.2-4.4)	3.73±0.01(3.72-3.74)
Bacular parameters			
Total baculum length	-	4.35 (4.06-4.61)	2.33±0.14 (2.23-2.43)
Width of proximal extreme	-	-	0.01±0.00 (0.01-0.01)
Width of middle extreme	-	-	0.01±0.00 (0.01-0.01)
Width of distal extreme	-	-	0.49±0.05 (0.45-0.53)
Shaft length	-	3.85 (3.69-4.02)	2.16±0.12 (2.08-2.25)
Baculum height	-	1.30 (1.23-1.38)	0.55±0.00 (0.55-0.55)

# Table II.- Comparison of average body mass (g), external body (n = 3), cranial and bacular (n = 2) measurements (mm) of *Rhinolophus lepidus* (n = 3) captured from Malakand district with previous study from in South Asia.

M. Salim et al.



Fig. 3. Facial (A and B) and cranial (C-F) features of Rhinolophus lepidus.



Fig. 4. Bacular features of Rhinolophus lepidus.

Prior to the present study, *R. ferrumequinum* was reported from Gilgit, Kululai in Swat, Kohistan, Dir, Abbotabad and Karakar Pass (Roberts 1997; Bates and Harrison, 1997) in Khyber Pakhtunkhwa (KPK). In Baluchistan, the species has so far only been collected from Kalat and Nushki (Mirza, 1965). The first specimen of *R. lepidus* was captured in mid-Octuber 1985 from Abbotabad (Corbet and Hill, 1992; Roberts 1997; Bates and Harrison, 1997) in KPK. The species was reported from Gujranwala in Punjab province (Shahbaz *et al.*, 2014).

# CONCLUSION

*Rhinolophus lepidus* was reported only from Abbottabad in Khyber Pakhtunkhwa and Gujranwala in Punjab province prior to the present study. During present survey, the species were recorded from Malakand for the first time. *R. ferrumequinum* was already reported from the study area however, both the congeners can be differentiated from one another on the basis of forearm length, which is longer in *R. ferrumequinum* and bacular features. Statement of conflict of interest

Authors have declared no conflict of interest.

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