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Author: Sharifi, Mozafar

Source: Acta Chiropterologica, 6(1) : 155-161

Published By: Museum and Institute of Zoology, Polish Academy of Sciences

URL: https://doi.org/10.3161/001.006.0112

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Acta Chiropterologica, 6(1): 155–161, 2004
PL ISSN 1508-1109 © Museum and Institute of Zoology PAS

Postnatal growth and age estimation in the Mehely's horseshoe bat (*Rhinolophus mehelyi*)

MOZAFAR SHARIFI

Department of Biology, Faculty of Science, Razi University, Kermanshah 67149, Iran
E-mail: sharifmozafar@hotmail.com

Empirical growth curves were derived by measuring length of forearms, body mass and length of the total gap of the fourth metacarpal-phalangeal joint in a free ranging population of the Mehely's horseshoe bat (*Rhinolophus mehelyi*) in a maternity roost in western Iran. Length of forearm and body mass followed a linear pattern of growth until day 14 and subsequently decreased to reach a stable level. The length of the gap of the metacarpal-phalangeal joint showed an increase up to 10 days and decreased until it had closed at over 55 days. Initiation of flight occurred approximately four weeks after birth. During the first two weeks of postnatal growth, the body mass and the length of forearm of pups increased linearly at mean rates of 0.58 g/day and 1.55 mm/day, respectively. A method of estimation of age was derived from values of forearm length and the total gap of the four metacarpal-phalangeal joint during the preflight and post flight periods.

Key words: *Rhinolophus mehelyi*, postnatal growth, age estimation, body mass, forearm length, epiphyseal gap

INTRODUCTION

The time in which a young mammal develops appropriate sensory and locomotors skills necessary to become independent from its mother is defined as the postnatal growth period (Baptista *et al*., 2000). Growth and development of bats have been studied both under natural (e.g., Buchler, 1980; Burnet and Kunz, 1982; Kunz and Anthony, 1982; Kunz and Robson, 1995) and captive conditions (e.g., Jones, 1967; Kleiman, 1969; De Paz, 1986; Taft and Handley, 1991; Hughes *et al*., 1995). These studies have shown that measurement of body size, length of forearm and length of the gap of the fourth metacarpal-phalangeal joint (Burnet and Kunz, 1982; Kunz and Anthony, 1982; de Fanis and Jones, 1995; Hayssen and Kunz, 1996; Hoying and Kunz, 1998; Stern and Kunz, 1998), as well as changes in tooth development (Anthony, 1988) can be used to estimate age of bats during the early postnatal period. Moreover, studies on postnatal development facilitate investigation on various aspects of early development such as initiation of flight and foraging (Kunz and Anthony, 1996), wing morphology and ontogeny (Powers *et al*., 1991; Stern *et al*., 1997), echolocation (Moss *et al*., 1997) and feeding and breeding behavior (Rajan and Marimuthu, 1999).

Different patterns of growth and development have been reported for various species and families of bat (Kunz and Stern, 1995). Interspecific variation observed in postnatal growth in different groups of bats may reflect environmental

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factors, availability of food, as well as biological factors such as size at birth, sex, metabolic rate and foraging success (Tuttle and Stevenson, 1982; Kunz and Stern, 1995). Generally, bats with larger litter size have slower growth rates compared with those with smaller litter size (Kunz, 1987).

Most studies on postnatal growth have focused on vespertilionids in temperate zones in Europe and North America. It was, therefore, my aim to provide data on the pattern of postnatal growth for a sub-tropical species of rhinolophid bat. This was accomplished by quantifying body mass, length of forearm and the epiphyseal gap to derive age-specific growth equations for *Rhinolophus mehelyi* Matschie, 1901 in a maternity roost in western Iran. *Rhinolophus mehelyi* is a widespread bat ranging from North Africa and southern Europe through Asia Minor to Transcaucasia and Iran (DeBlase, 1980; Rodrigues and Palmeirim, 1999). It has been reported in eight localities in Iran (DeBlase, 1980; Hemmati, 2000), which range down the Zagross Mountain from north at the border with Turkey to south in the Fars Province. This species probably occurs throughout the Zagross Range and may occur in the Elbourz Mountains as well (DeBlase, 1980). Based on an attempt made to estimate the conservation status of bats from Iran *R. mehelyi* is ranked as a very rare species (Sharifi *et al*., 2000).

Materials and Methods

The present study was conducted at Mahidasht Cave (33°23'N and 47°30'E) in the mid western Zagros Range, Kermanshah Province, Iran during mid-May until late August 2001. The Mahidadht cave is occupied by a nursery colony, and not used as a hibernaculum presumably because of high winter temperature. Observations performed during 1999–2001 indicated no visible signs of bats during January and February in either years (M. Sharifi, B. Kiabi, and K. Faizollahi, unpubl. data); nevertheless, gradually until mid-March colonies of at least three species (*Myotis blythii*, *R. mehelyi* and *Miniopterus schreibersii*) aggregate there. Counts of emerging bats performed in August 2001 provided an estimate of 550 individuals, including 350 *M. blythii*, 140 *M. schreibersii* and 60 *R. mehelyi*.

Following nightly departure of females, pups were captured by hand at their natal roosts. Observations and measurements of pups were carried out as soon as possible after they were captured. Flightless pups were returned to their roost usually before females returned from foraging. Visits to the roost were made twice a week during the first two weeks of the study and then once a week until mid-August 2001. In this study, pups with an umbilical cord still attached were judged to be 1-day old. Pups with eye slits still closed were assumed to be born at mid time between last visit and the visit thereafter.

Length of forearm was measured to the nearest 0.1 mm with Vernier calipers. Body mass was measured to the nearest 0.1 g using an electronic balance. Length of the gap of the fourth metacarpal-phalangeal joint was measured to the nearest 0.1 mm using Vernier calipers while the wing of the bat was spread over a transparent solid plastic sheet over a strong torch in order to make the gap visible.

Growth data of young bats were fitted to the following three models: the Von Bertalanffy equation, the Gompertz equation and the logistic equation (e.g., Swift, 2001). The three equations are as follows:

logistic model, $w = A/\{1+\beta\, e^{-kt}\}$;

Gompertz model, $w = A \exp\{-\beta\, e^{-kt}\}$;

Von Bertalanffy model, $w = \{A^{1-\beta} - \theta\, e^{-kt}\}^{1/(1-\beta)}$;

where A = asymptotic value, w = body mass (g) at age (t), and k = growth rate constant ($days^{-1}$).

The parameters A and k in each model were estimated for the growth of mass and length of forearm in the neonate population. Similar equations have been used for forearm.

The least-squares linear regression model was applied to derive age-predictive growth equations from pooled data on the length of forearm (1–10 days), body mass (1–14 days) and total epiphyseal gap (12–50 days). To derive an age-predictive equation for these three parameters the axes on the growth curves for the identified period of time were reversed, and age for the specific periods was considered as the dependent variable (Kunz and Anthony, 1982).

Results

On June 3rd two pups with umbilical cords appeared in the colony. On June 7th,

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nine additional pups were found, none of which had attached umbilical cords; therefore it appears that these pups had been born during the four days between the two successive visits. All females observed with young had single offspring. Average mass of two pups at birth and nine pups, which were born thereafter represents 28.5% of the mother's body mass (obtained from asymptotic values in the growth curve). Bats at birth were naked with eyes closed. Eye slits opened 4–5 days after birth. Fur was visible from the 2nd week onward, predominantly on the dorsal and ventral side of the body. However, the back of the neck remained conspicuously naked until the fourth week. Young were able to flutter and glide when dropped from hands at 21 days of age. At 30 days of age young were able to fly freely inside the cave. It was not possible to catch young *R. mehelyi* in the nursery site after four weeks of age.

During the first 10 days, both forearm length and body mass increased in a linear fashion, with a growth rate of 1.55 mm and 0.58 g per day (Fig. 1A and 1B), respectively. Subsequently, the rate of growth in these two characters decreased. In contrast, the length of gap of the metacarpal-phalangeal joint showed linear increase until 10 days. The gap disappeared on ca. day 55, indicating epiphyseal closure (Fig. 1C). Judging from the highly significant coefficient of determination for relationships between age and the length of forearm ($r^2 = 0.98$, $n = 11$, $P < 0.001$), and the length of total epiphyseal gap ($r^2 = 0.95$, $n = 11$, $P < 0.001$), these equations may be used to estimate the age of *R. mehelyi* (Fig. 2).

Both forearm length and body mass were fitted to three non-linear growth models. Among these models, the logistic curve gave the best fit for body mass, while the Gompertz equation provided the best fit for forearm length. With the use of the logistic and von Bertalanffy equations the asymptotic sizes for body mass and forearm length were calculated to be 11.44 g and 53.35 mm, respectively. The logistic equation gave a growth rate constant for body mass of 0.27 for all pups. According to the Gompertz equation, growth rate constant for forearm was 0.09 (Table 1).

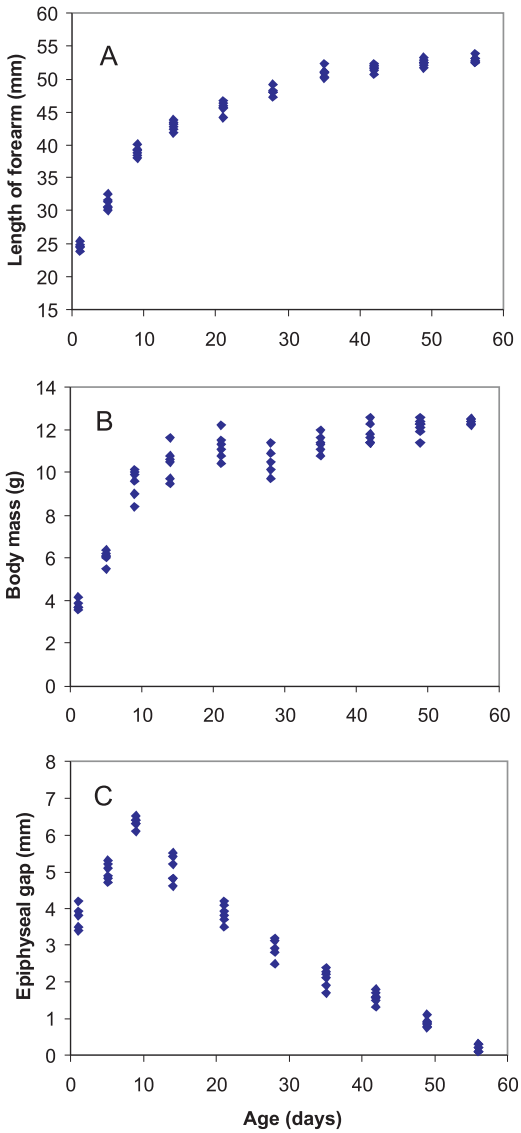


FIG. 1. Empirical growth curve for length of forearm (A), body mass (B), and the fourth metacarpal-phalangeal joint (C) of young *R. mehelyi* from day 1 to 55 of age

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Discussion

Studies on postnatal growth of free ranging bats commonly involve longitudinal (mark-recapture) sampling and/or cross-sectional (grab) sampling (reviewed by Baptista *et al*., 2000). In longitudinal sampling, day-old young are identified (based on presence of the umbilical cord) and marked. Subsequently measuring body mass and linear dimensions of individuals carries on throughout the postnatal period. In grab sampling, it is assumed that average changes in body mass or linear dimensions of individuals can be adequately represented by data collected from bats captured at different times. In this method there is potentially a risk for part of the population to be over-represented by multiple capturing. It is equally possible that parts of population are under-represented simply by not being captured adequately. There are not many studies that show apparent differences in growth rates derived from longitudinal and cross-sectional sampling methods (reviewed by Baptista *et al*., 2000). Several authors have assessed postnatal growth using cross-sectional sampling (e.g., Dwyer, 1963; Pagels and Jones, 1974; Stangel *et al*., 1996); nevertheless, growth rates derived from the longitudinal method are faster than those based on the cross-sectional one (reviewed by Baptista *et al*., 2000). Baptista *et al*. (2000) also demonstrated that growth derivatives for free-ranging *Myotis lucifugus* obtained by a cross-sectional

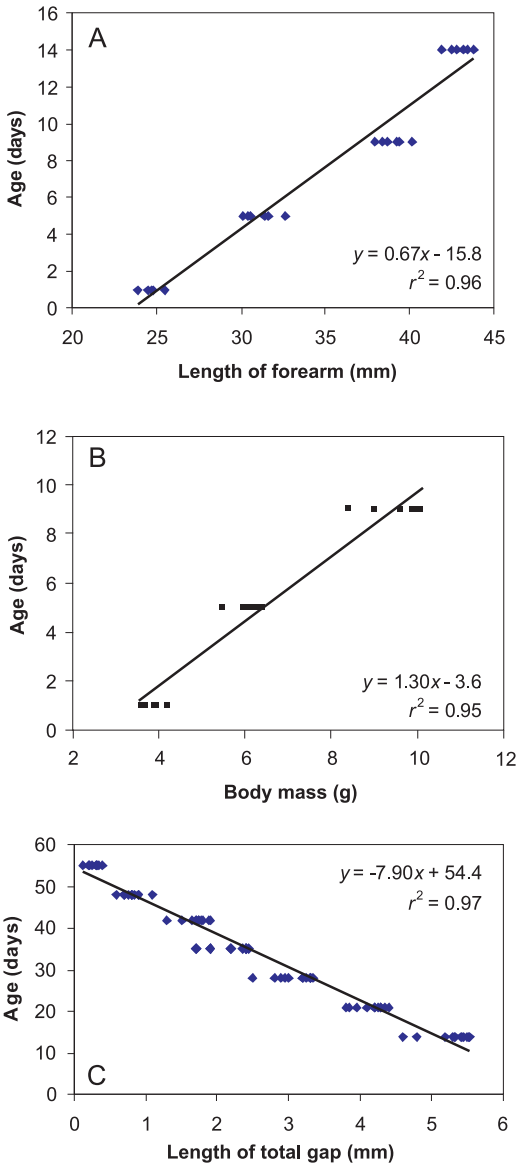


Fig. 2. Regression line estimating the age of *R. mehelyi* from the values of length of forearm (A) and body mass (B) for the first two weeks after birth; and from the length of gap of the fourth metacarpal-phalangeal joint (C) from day 10 to 55

Table 1. Parameters obtained from fitted growth curves to forearm and body mass data of 11 pups of *R. mehelyi* in the Mahidasht cave in western Iran. Abbreviations: k = growth rate constant (days^{-1}); A = asymptotic mass (g) or length (mm); r^2 = coefficient of determination, $P < 0.001$ for all data

Model	Parameter	Body mass ($\bar{x}$ ± SE)	Forearm length ($\bar{x}$ ± SE)
Logistic	A	11.44 ± 0.093	52.58 ± 0.334
	k	0.27 ± 0.020	0.10 ± 0.005
	r^2	0.92	0.96
Gompertz	A	11.51 ± 0.092	53.35 ± 0.359
	k	0.21 ± 0.010	0.09 ± 0.004
	r^2	0.91	0.97
Von Bertalanffy	A	11.51 ± 0.092	54.6 ± 0.425
	k	0.20 ± 0.010	0.07 ± 0.004
	r^2	0.91	0.95

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sampling differ significantly from those obtained by a longitudinal sampling.

As observed in other studies on bats (e.g., Kleiman, 1969; Krátký, 1970, 1981; Maeda, 1972; Kunz, 1973; Hoying and Kunz, 1998), some pups of *R. mehelyi* experienced a loss of body mass on the fourth week near the onset of flight (Fig. 2). Kunz (1987) suggested that such recession of body mass may reflect depletion of fat reserves following the onset of flight or a shift by young animals from a diet of milk to one composed of insects. However, this recession in body mass has also been reported in captive bats (e.g., Hoying and Kunz, 1998), and this phenomenon cannot be explained completely by increasing energy demands associated with early flight. *Rhinolophus mehelyi* starts flying at time when the length of forearm in young bats, while compared with the asymptotic values of the forearm curve, is approximately 92% of adult's size. At that time the average mass of young bats is about 87% of the body mass at the final stage of the growth. It appears that although both wing dimensions and body mass approach adult proportions very rapidly (Fig. 1A and 1B), body mass remains proportionally lower. This causes a lower wing loading for young adults, which in turn increases maneuverability and decreases the cost of flight at a time when young bats are learning how to fly, detect and capture flying insects (Huges *et al*., 1995). Similar disproportional development of wing dimensions and body mass have been reported for *Pipistrellus subflavus* (Hoying and Kunz, 1998). The pattern of growth in body mass during the postnatal period in *R. mehelyi* was more variable when compared to growth of the forearm. This has been reported in other studies (e.g., Thomas and Marshall, 1984), and is believed to be the result of higher responsiveness of body mass to environmental conditions than forearm length (Kunz and Robson, 1995).

Various non-linear models have been used to characterize postnatal growth rates in bats (see Hoying and Kunz, 1998). Recent investigations on postnatal growth in bats have compared growth parameters derived from the logistic and other non-linear models and have concluded that the logistic model provides the best fit to empirical data for *Pipistrellus pipistrellus*, *Plecotus auritus* (de Fanis and Jones, 1995), and *Tadarida brasiliensis* (Kunz and Robson, 1995). The pattern of postnatal growth in *R. mehelyi* with a rapid linear increment in the first two weeks and an asymptote soon after is similar to that of several species of bats (Kunz and Stern, 1995). Among three species of bat reproducing in the Mahidasht cave, the degree of synchrony at birth and the subsequent variation in size during the course of the neonatal growth in *R. mehelyi* is similar to *Miniopterus schreibersii*, which is a similarly sized. However, *M. blythi*, which is a bigger species in the same cave, has more unsynchronized birth timing. Available information on postnatal growth and age estimation of *M. blythii* also demonstrates more variation in timing of parturition and subsequent growth pattern in this species (M. Sharifi, B. Kiabi, and K. Faizollahi, unpubl. data). Observations on fledging and foraging of young of the three bat species indicate that, although the bigger bat species (*M. blythii*) gives birth earlier, higher growth rate in smaller species causes an overall synchronization in foraging and other activities of the bat community in late summer and early autumn. Further research is needed to determine whether the present variation in parturition in these species is a part of the species reproductive strategy or just a reflection to the environmental variation.

Acknowledgements

This study was carried out with a grant provided by the Department of Environment in Kermanshah.

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I would like to thank Mr K. Faizolahi for his invaluable field assistance. I also thank two anonymous reviewers and Dr. Wiesław Bogdanowicz for their useful suggestions and help.

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Received 27 June 2003, accepted 04 January 2004

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