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Age, Growth and Longevity of Kotschy's Gecko, *Mediodactylus kotschyi* (Steindachner, 1870) (Reptilia, Gekkonidae) from Central Anatolia, Turkey

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Abstract: We studied the age structure of Kotschy's gecko, *Mediodactylus kotschyi*, from the Sultan Mountains (Central Anatolia, Turkey) using the skeletochronological method. We examined the humeral diaphyseal cross sections of a total of 19 (six males and 13 females) museum specimens. Our results showed that the age structure ranged from three to seven years (mean = 4.2 years, SD = 1.47) in males and from two to eight years (mean = 4.5 years, SD = 1.81) in females. Both sexes reached sexual maturity after their second hibernation, and no statistically significant difference in age composition was observed between the sexes. There was a strong positive correlation between SVL and age (Spearman's correlation coefficient, r = 0.93) and Von Bertalanffy growth curves fitted to the relationships between age (k = 0.80) and SVL (SVL_{max} = 42.4). In conclusion, *M. kotschyi* is a short-lived species with a high growth rate.

Keywords: Skeletochronology, growth, longevity, *Mediodactylus kotschyi*, Central Anatolia

Introduction

The genus *Mediodactylus* Szczerbak and Golubev, 1977 is widely distributed from the Mediterranean region to Central Asia (Ananjeva *et al.* 2006). It contains about 13 species (Uetzi, Hojbek 2014). The East Mediterranean species *Mediodactylus kotschyi* (Steindachner, 1870) is distributed on the Aegean Islands, in Greece, Albania, Macedonia, Bulgaria, Georgia, Iran, Syria, Lebanon, Israel, Cyprus, Southeastern Italy, South Crimea, and in Turkey (Beutler 1981, Kasapidis *et al.* 2005, Ananjeva *et al.* 2006). The species is divided into 31 subspecies (Uetz, Hojbek 2014); is nine nominal subspecies are represented in Turkey (Baran, Atatür 1998). The subspecies *Mediodactylus kotschyi danilewskii* (Strauch, 1887) inhabits the Sultan Mountains (Afsar 2006).

Generally, Kotschy's gecko prefers the ground in dry, rocky or stony places. It can be found also from scrubland, under the bark of old juniper trees, on cliffs or stone walls, and outside and inside buildings (Başoğlu, Baran 1977, Böhme *et al.* 2009). The species reaches a body size of about 80 mm (Ananjeva *et al.* 2006). Kotschy's gecko is listed in the least concern category of the IUCN Red List;with the major threats to this species, especially and namely to for its southern populationsits, are being the deforestation triggered by urbanisation and agricultural development (Böhme *et al.* 2009).

The global extinction and decline of amphibian and reptile populations have been followed with an increased anxiety since the 1900s (e.g. Blaustein,

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Wake 1990, Alford, Richards 1999, Gibbons *et al.* 2000). Habitat loss and degradation, introduced invasive species, environmental pollution, disease, over-exploitation, UV-B radiation and global climate change are among the main factors which trigger extinction (Alford, Richards 1999, Gibbons *et al.* 2000, AmphibiaWeb 2014). For the sustainability of endangered populations, it is quite important to know life history traits such as longevity, age at sexual maturity, growth rate, reproductive characteristics, life cycle and survival rate.

Bone is a model tissue for understanding life history and activity periods of species. The studies carried out so far have defined two types of bone structure in ectotherms and endotherms. Bone development is rather slow and regular in ectotherm animals and it is called a lamellar zonal bone, whereas bone development is quite fast in endotherm animals due to the very high metabolic activity and it is called a fibrolamellar bone (Margerie 2002). The bone tissue, where metabolic activity can be observed easily, is used in age determination studies. Skeletochronology is a widely used method for age estimates of amphibians and reptiles depending on the annual growth rings (LAGs) developed in long bones (Castanet, Smirina 1990, Castanet *et al.* 1993, Castanet 1994). This technique is quite successful in determining the age compositions of lizards (e.g. Castanet *et al.* 1988, Castanet, Baez 1991, Çiçek *et al.* 2012).

Despite the availability of detailed studies on the ecology and life history characteristics of Kotschy's gecko (e.g. Werner 1989, Szczerbak, Golubev 1996, Werner 1993, Werner *et al.* 1993), information on the age composition of the Anatolian population is still lacking. The objective of the present study was to determine the age structure, age at maturity, growth, and longevity through the skeletochronological study of humerus of *M. kotschyi*.

Materials and Methods

We examined 19 (six males and 13 females) specimens registered at the Zoology Department of Ege University (ZDEU) and deposited at the Zoology Department, Faculty of Arts and Sciences, Celal Bayar University. The specimens were previously used to determine the herpetofauna of the Sultan Mountains (Afsar 2006). They were collected from the Dereçine Village (Afyonkarahisar, latitude 38.487605°, longitude 31.258342°, altitude 1,038 m), Akşehir (Konya, lat.: 38.355911°, long.: 31.415409°, alt. 1,036 m) and the Çarıksaray Village (Isparta, lat.: 38.119419°, long.: 31.415617°, alt. 1,265 m) from the Sultan Mountains in early April 2003. For all individuals, the snout-vent length (hereinafter 'the SVL') was measured with a dial calliper to the nearest 0.01 mm. Sexes were determined using secondary sexual characters.

Humerus bones were dissected from the specimens, fixed in neutral buffered formalin and then washed with distilled water. After fixation, decalcification was performed in 5% nitric acid for 1-2 hours depending on the bone size. The tissues were dehydrated with ethanol series, cleared with xylene and embedded in paraffin. The humeral diaphyseal cross sections were stained with Ehrlich's haematoxylin for 15 min and then examined under a light microscope. For each humerus, we selected a minimum of three cross sections at the mid-diaphyseal level, with the smallest marrow cavity. The lines of arrested growth (LAGs) were counted by two of the authors (K.Ç. and M.K.). In all cases, the observers were not informed about the identification of the individuals. All mount preparations were photographed with an Olympus LC-20 Soft-Imaging System.

Normality of the SVL distributions for sexes was tested with Kolmogorov-Smirnov D test. Since they were normally distributed (Kolmogorov–Smirnov D test, $P \geq 0.05$), parametric t-test was used for comparison. The non-parametric Mann-Whitney U test was used to compare the medians, and Spearman's rank correlation was used to estimate the relationship between SVL and age, as data which were not normally distributed ($P \leq 0.05$). The alpha level was set at 0.05, and the mean values were provided with their standard deviations.

Growth was estimated using von Bertalanffy's equation (1983) that was previously used in several studies on saurians (e.g. Wabstra *et al.* 2011, Roitberg, Smirina 2006, Guarino *et al.* 2010). The modified growth formula is $SVL_t = SVL_{max}-(SVL_{max} - SVL_0)e^{-k(t)}$, where k is the Brody growth rate coefficient (units are yr^{-1}); t is the number of growing seasons experienced (age); SVL is the average length in the age groups calculated; SVL_{max} is the average length at the maximum age; and SVL_0 is the average SVL at hatching. The SVL at hatching, approximately 18 mm, was estimated for *M. kotschyi danilewskii* by Szczerbak, Golubev (1996). SVL_{max}, *K*, and their 95% confidence intervals were estimated through the nonlinear least-square regression method using the FSA package in R version 3.0.2 (R Development Core Team 2014).

Results

In the 19 individuals examined (six males and 13 females), the mean SVL was 38.5 mm (SD = 2.40, range = 35.5-41.8) in males and 38.1 mm (5.52, 26.3-45.0) in females. Although males were larger than females, no statistical difference in the SVL values ($t = 0.217$, $P \leq 0.831$) was observed between sexes.

When bones were examined histologically, we found that the bone structure was of lamellar zonal type. As bone development was rather slow and regular, the bone cortex at the diaphysis was avascular and formed of a compact parallel fibered bone tissue (Fig. 1). Primary or periosteal bone was preserved throughout the life of the species. The marrow cavity was largely or entirely covered by the periosteal bone. Secondary bone development, defined as endosteal resorption, is rather slow. The endosteal bone is located in a very narrow space around the marrow cavity, and endosteal and periosteal surfaces of bone are separated from each other by the resorption line.

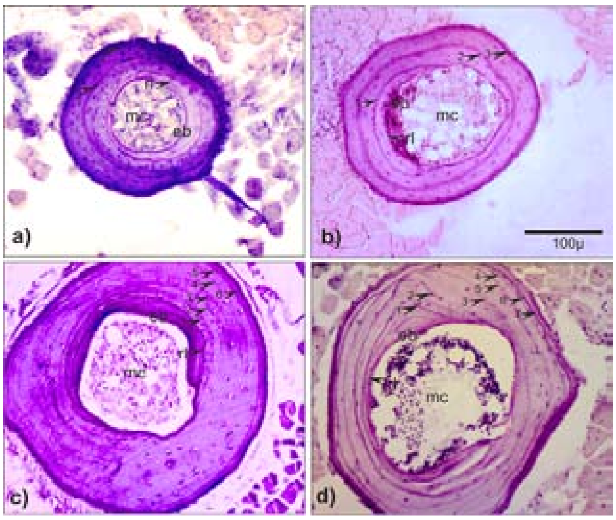

Fig. 1. The cross-sections of *Mediodactylus kotschyi* from Sultan Mountains (Central Anatolia). **(a)** three-year old male (SVL = 37.1 mm), **(b)** three-year old female (37.5 mm), **(c)** six-year old female (41.7 mm) and **(d)** seven-year old male (48.8 mm). Abbreviations: mc – marrow cavity, eb – endosteal bone, rl – resorption line. Arrows point to lines of arrested growth and resorption lines. Horizontal bar = 100 µm

All humeral cross sections showed well-defined lines of arrested growth (LAGs) in *M. kotschyi.* A LAG takes place in *M. kotschyi* after each hibernation, as in the other reptile species from the temperate zones. The LAGs are generally in the form of thin and darkly stained concentric chromophilic rings and located outside the endosteal bone (Fig. 1). On the basis of the osteometric analysis (LAG1 ≤ mean LAG1+2SD > LAG2), it was observed that the first LAG was partially (Fig. 1) or completely resorbed (for seven or 37% of the specimens: three males and four females) by the endosteal bone in all individuals.

The mean age was calculated as 4.2 years (SD = 1.47, range = 3-7) in males and as 4.5 years (SD = 1.81, range = 3-8) in females (Table 1, Fig. 2a). When compared in terms of the age structure, no statistically significant difference was observed between the sexes (Mann-Whitney U test, M-W = 32.50, $P \leq 0.579$). As suggested by Kleineberg, Smirina (1969), the first convergence in the distance between the LAGs was considered the sign of reaching sexual maturity. Both sexes reached sexual maturity after their second hibernation. The longevity of the Central Anatolian population was calculated

Table 1. Summary statistics of snout-vent length (SVL) related to age of *Mediodactylus kotschyi* from Sultan Mountains (Central Anatolia)

Age	N	Mean	SD	Range	SE
2	2	26.7	0.48	26.3 – 27.0	0.34
3	4	36.9	0.94	35.5 – 37.5	0.47
4	5	38.7	1.27	37.0 – 40.4	0.57
5	4	40.0	0.51	39.4 – 40.6	0.25
6	1	41.7	-	-	-
7	2	42.6	1.07	41.8 – 43.4	0.75
8	1	45.0	-	-	-

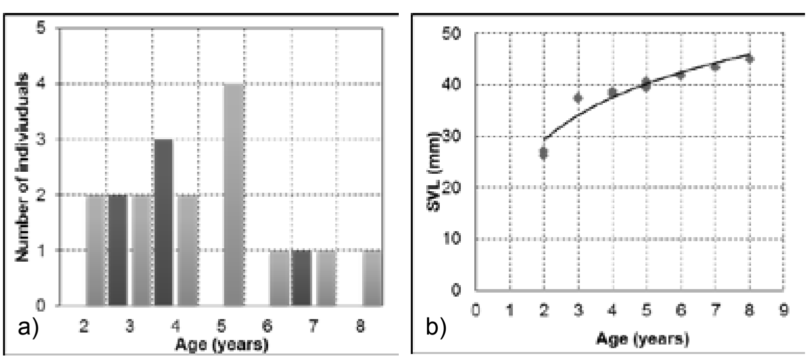

Fig. 2. **(a)** The age distribution and **(b)** growth curve of *Mediodactylus kotschyi* from Sultan Mountains (Central Anatolia). $SVL_t = 41.6 - (41.6\text{-}18.0)\, e^{-0.92(t)}$

as seven years for males and eight years for females (Table 1, Fig. 2a).

We observed that SVL and age (Spearman's correlation coefficient, r = 0.93, P ≤ 0.001) were highly correlated. According to the von Bertalanffy's formula, the estimated asymptotic SVL was calculated as 42.4 mm (SE = 0.77, CI = 40.7-44.0) and growth coefficient k as 0.80 (SE = 0.14, CI = 0.51-0-1.09), and this value was slightly lower than the maximum SVL measured (Fig. 2b).

Discussion

It was determined that the males of *Mediodactylus kotschyi* from the Sultan Mountains examined in this study were in the age range three – seven while the age of the females ranged between two and eight years. We found that they reached sexual maturity at the age of two. In terms of SVL and age, the females were longer and older than the males. It was also previously reported that females of this species were larger than males (Werner 1993). It was stated that in the Israel population, hatchlings had reached a SVL of 23-33 mm by their first autumn, a SVL of 37-43 mm by their second autumn, and a minimum SVL of 44 mm in their third summer (Werner 1993). In the Crimean population of *Mediodactylus kotschyi*, hatchlings were 17.8-18.5 mm long and two-year-old lizards reached 39 mm and sexual maturity (Szczerbak, Golubev 1996). In Israel, the young lizards reached sexual maturity when they were a year (10-11 months) old and continued to grow slowly and after the age of tree, they already ceased reproduction, probably not surviving beyond this age (Werner, 1993).

Szczerbak, Golubev (1996) stated that individuals reached the maximum length at five years (43.5 mm for males and 50.5 mm for females). Werner (1993) observed the maximum SVL of 48 mm in the Israeli population of the species. The maximum SVL measured in the population of the Sultan Mountains was 41.8 mm in males and 45 mm in females. Werner (1993) reported that a female adult taken under captivity in Jerusalem lived for about six years but the adults taken under captivity generally lived for up to four years. Werner (1993) found that when the species reached sexual maturity in its first year in Jerusalem, the maximum longevity was seven years. In the population of the Sultan Mountains, longevity was recorded as seven years for males and as eight years for females. In general, the northern populations of the species tend to have a higher body size and longevity than their southern populations (e.g. Wabstra *et al*. 2001, Roitberg, Smirina 2006, Guarino *et al.* 2010). Reaching reproductive maturity earlier for the populations from Israel might be ascribed to the fact that the inhabitants of hot places in the south are shorter and reach reproductive maturity earlier.

Among other Geckonidae, the longevity was reported was of 14 years and ecological longevity of eight-nine years for *Tarentola mauritanica* (Linnaeus, 1758) (see Slavens, Slavens 1991). Werner *et al.* (1993) reviewed the maximum longevity of 55 geckonid species and subspecies. They found that under captivity, the maximum longevity was between two and 37 [for *Woodworthia maculatus* (Gray, 1845)] years in the group. Martinez Rica (1974) stated that the longevity of *Hemidactylus turcicus* (Linnaeus, 1758) from the Iberian Peninsula was four years in the wild and eight years in captivity. The South-east Anatolian population of *H. turcicus* reached sexual maturity in two to three years, and age distribution was four to eight years for males and four to nine years for females (Kanat, Tok 2014). Piantoni *et al.* (2006) estimated the age and growth of *Homonata darwinii* Boulenger, 1885 from Argentina and found that the species reached sexual maturity at up to five-nine years and that males were in the range nine-fourteenyears and females in the range five-fifteen years.

As seen from the examined humeral diaphyseal cross sections of *Mediodactylus kotschyi*, its bone structure was not different from that of the other reptile species (e.g., Castanet *et al.* 1993, Roitberg, Smirina 2006, Guarino *et al.* 2010, Çiçek *et al.* 2012). The endosteal resorption in lizards is low, and these LAGs are often lost even in the oldest individuals (Castanet *et al.* 1988, Castanet 1994). The LAGs remain throughout life because the majority of reptiles have a non-vascularised bone tissue and the second remodelling of the bone tissue does not occur (Smirina, Ananjeva 2007). The first LAG was partially resorbed by endosteal resorption in only 37% of the lizards.

Some lizard species can live for about 60 years, while life expectancy is generally between five and 20 years (Castanet 1994). Therefore, they are known as short-lived species and many species cannot live for more than one or two years in the wild (Castanet 1994). As most ectotherms do, reptiles have a temperature-depended energy metabolism, and the longevity of species depends largely on environmental factors (Castanet 1994). Growth rates could reflect environmental conditions such as temperature and food availability (e.g. Jenssen, Andrews 1984, Smith, Ballinger 1994). In Geckonidae, longevity is associated with the body size and the ecological niche breadth (Werner *et al*. 1993). In conclusion, *Mediodactylus kotschyi* is short-lived as many other lizards, and its growth rate is quite high. Comparative studies among populations are needed to provide more information on the life history traits of the Anatolian population of the species.

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