Iranian Journal of Animal Biosystematics (IJAB) Vol.11, No.1, 7-16, 2015 ISSN: 1735-434X (print); 2423-4222 (online)

# The Male Reproductive Cycle of the Bedriaga's Plate-tailed Gecko, *Teratoscincus bedriagai* in Iran

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(Received: 30 March 2015; Accepted: 23 June 2015)

The Bedriaga's Plate-tailed Gecko, *Teratoscincus bedriagai*, Nikolsky, 1900 is distributed in the northern and eastern deserts of Iran and the desert regions of southern Afghanistan. In this study, the male reproductive cycle of the lizard has been studied from 5 April to 5 August, 2013. Totally, 40 adult males were collected by hand at midnight from four adjacent stations in Damghan County, Semnan Province of Iran. Most of the lizards were observed on highly saline, loose soil covered with a thin salt crust, and were common near *Tamarix* bushes. Animals were transferred to the laboratory and their morphometric characters were measured. After anesthetizing the animals, their testes were removed and processed for morphometric and histological studies. Results showed that the spermatogenesis started after hibernation from early April and ended in August. The peak of spermatogenesis was in May. The numbers of seminiferous tubules were 24-75 and their diameter varied between 72.50 and 232.50  $\mu$ . The diameter of tunica albuginea varied between 3.05 and 8.20 microns and diameter of germinal layer varied between 20.40 and 124.30 microns. Spermatogenesis of *T. bedriagai* in Iran is seasonal and alternates with associate type.

Key words: Spermatogenesis, testis, lizard, gecko, Teratoscincus bedriagai, Iran.

#### INTRODUCTION

Three general types of reproductive cycles are observed in Lizards: constant, associated and dissociated (Pough et al., 2001). In constant reproductive cycle, gonads are active almost year-round (Jenssen and Nunez, 1994). In associated and dissociated reproductive cycles, mating season is discontinuous. In associated type, gonadal activity increases immediately prior to the mating period in both males and females simultaneously and females have no need to store sperm due to it's availability during the reproductive season (Huang, 1997; Censky, 1995). In a dissociated type, gonadal activity is low during the mating period and peaks during the non-mating period while male gonadal activity is shorter than that of females and sperm is stored by the female genital system for later fertilization (Torki, 2006).

The Gekkota is a suborder of Squamata, and consists of seven families (Gamble et al., 2008). The Sphaerodactylidae are a family of geckos distributed in North America, South America, and the Caribbean, as well as in Southern Europe, North Africa, the Middle East, and into Central Asia

(Gamble et al., 2008). The Bedriaga's Plate-tailed Gecko or Skink Gecko, *Teratoscincus bedriagai* Nikolsky, 1900, which belongs to the Sphaerodactylidae family, is a nocturnal and insectivorous lizard distributed in the northern and eastern desert basins of the Central Plateau of Iran, Sistan, and the desert regions of southern Afghanistan as far east as Kandahar. This species in Iran is distributed in Sistan and Balutchestan, Khorasan, Semnan, Tehran and Yazd Provinces (Smid et al., 2014). A description of this species was presented by Anderson (1999) which was based on five specimens. Further morphological data was given by Hojati in accordance with specimens collected in northern deserts of Central Plateau of Iran (Hojati et al., 2009, Hojati et al., 2014). The female reproductive cycle of the species in the study area has been studied as well (Mojibi and Hojati, 2014). Since no data is available on the spermatogenic cycle of the species, this research has been conducted to characterize the male reproductive cycle.

#### MATERIALS AND METHODS

The specimens were collected from deserts of four Adjacent villages including: Hassan Abad, Saleh Abad, Alian and Yazdan Abad located in south of Damghan County, Semnan Province of Iran (54°19'E, 35°55'N). The study area is located at 1170 m above sea level and north of the Central Kavir Desert. The annual average temperature is 17.2°C. According to the reports of Department of Environment of Damghan County, this area consists of alkaline saline soils comprising clay and sand and the dominant plant species are *Tamarix* sp., *Salsola* sp., *Alhaji* sp., *Peganum* sp., *Atriplex* sp. and *Astragalus* sp.

All specimens were collected periodically every month during the main activity period of this species from 5 April to 5 August 2013. Sampling occurred at night from 21:00 to 24:00 hours. In total, 40 sexually mature males were captured by hand (eight specimens per sampling period), usually from the holes in the desert sand. Most of the specimens were observed on highly saline, loose soil covered with a thin salt crust, and were common near *Tamarix* bushes (Fig. 1). Males have larger and more embossed base of tail because of the presence of the hemipenes.



FIGURE 1. Teratoscincus bedriagai in Saleh Abad, Iran.

The specimens were transferred alive to the zoology laboratory of Islamic Azad University, Damghan Branch, Iran. Table1 shows the measured characters. Length, width and diameter values were measured by dial caliper with an accuracy of 0.02 mm. Weight was measured by scale with an accuracy of 0.001 g. Volume of the testis was calculated by using the formula for the volume of an ellipsoid:  $4/3 \pi$  ab<sup>2</sup>, where  $a = \frac{1}{2}$  the longest axis, and  $b = \frac{1}{2}$  the shortest axis. Gonads, once removed, were measured for metric and meristic studies. Weight, diameter (length and width) and volume values were measured for the right and left testis separately. After fixing the testes in 10% formalin, tissues were dehydrated, cleared in xylene, infiltrated with paraffin, embedded and sectioned and stained with haematoxylin & eosin following standard histological protocols. The sections were studied by light microscopy at 400× magnification. Photographs were prepared by digital camera while the data was analyzed by SPSS 18 software and one-way ANOVA and Tukey test.

Abbreviation	Characters			
W	Body Weight			
SVL	Snout-Vent Length			
LCD	Tail Length			
HL	Head Length			
HW	Head Width			
RTL	Right Testis Length			
LTL	Left Testis Length			
RTWi	Right Testis Width			
LTWi	Left Testis Width			
RTW	Right Testis Weight			
LTW	Left Testis Weight			
RTV	Right Testis Volume			
LTV	Left Testis Volume			
RTAD	Right Tunica Albuginea Diameter			
LTAD	Left Tunica Albuginea Diameter			
RSTN	Right Seminiferous Tubules Number			
LSTN	Left Seminiferous Tubules Number			
RSTD	Right Seminiferous Tubules Diameter			
LSTD	Left Seminiferous Tubules Diameter			
RGLD	Right Germinal Layer Diameter			
LGLD	Left Germinal Layer Diameter			
RLD	Right Lumen Diameter			
LLD	Left Lumen Diameter			
RSgN	Right Spermatogonia Number			
LSgN	Left Spermatogonia Number			
RSpN	Right Spermatocytes Number			
LSpN	Left Spermatocytes Number			
RStN	Right Spermatids Number			
LStN	Left Spermatids Number			
RSN	Right Sperms Number			
LSN	Left Sperms Number			
GI	Gonadal index = gonadal weight/body weight $\times$ 100			
RHpL	Right Hemipenis Length			
LHpL	Left Hemipenis Length			
RHpW	Right Hemipenis Width			
LHpW	Left Hemipenis Width			

TABLE 1. The measured characters in Teratoscincus bedriagai.

## RESULTS

The mean body weight (W), snout-vent length (SVL), tail length (LCD), head length (HL) and head width (HW) of males were 7.56 g, 56.86 mm, 29.70 mm, 15.56 mm and 13.34 mm, respectively. The smallest SVL was 50.04 mm while the largest one was 65.73 mm. There was no significant difference in body size characters including W, SVL, LCD, HL and HW through different months (Table 1). The mean weight, length, width and volume of right testes were 0.038 g, 7.50 mm, 3.10 mm and 47.70 mm<sup>3</sup>, respectively. The mean weight, length, width and volume of left testes were 0.037 g, 7.04 mm, 3.70 mm and 65.82 mm<sup>3</sup>, respectively. The descriptive statistics of characters in T. bedriagai are shown in Tables 1 and 2. The mean number of seminiferous tubules was 52 (N=40). The mean diameters of seminiferous tubules in right and left testes were 155.34 and 155 microns, respectively. The mean diameter of the tunica albuginea, germinal layer and lumen were 6.39, 72.28 and 72.03 microns in right testes; and 6.39, 72.28 and 72.03 microns in left testes, respectively. The mean number of spermatogonia, spermatocytes, spermatids and sperms were 156.55, 294.97, 247.88, 203.95 in right testes; and 156.55, 294.97, 247.88 and 203.95 in left testes, respectively. The minimum diameter, weight and volume of testes were observed on 5 August (Figs 2 and 3). There were no significant differences in the testis and hemipenis characters between the left and right side of body (paired t-test, P > 0.05 in all cases). There were significant differences in macroscopic testicular characters between months (Table 2). They hibernate from October to March. Early spermiogenesis starts in March and continues from April to July. Observation of the large numbers of mature sperms in the tubules in April shows that the spermatogenesis starts from March. The Sertoli cells, spermatogonia and spermatocytes were observed in the seminiferous tubules during the reproductive season from April to August. The spermatids were observed in the germinal layer from April to early August, but their number was reduced after June. The spermatozoids were abundant in the lumen of tubules during May and June and there were no spermatozoids after July (Fig. 4). Tubule diameter and germinal epithelium height reached maximum sizes during May. The minimum height of germinal epithelium was observed in early August. Tunica albuginea diameter reached its maximum and minimum size in April and August, respectively. Lumen diameter increased in July and August. The maximum level of sperm production was on May (Fig. 5). The gonadal index (GI) increased from April to May and decreased from July to August (Fig. 6). There were significant differences in microscopic testicular characters between months (Table 3).

One pair of light pink hemipenes are located at the base of the cloaca. Hemipenes are club-shaped and apex is subdivided into two lobes (Fig. 7). There is an internal, ossified structure named hemibacula in each hemipenis. Their mean length and width of right hemipenis were 3.97 and 2.40 mm and those of the left side were 3.91 and 2.55 mm, respectively. There were significant differences in hemipenal characters between months (Table 2). The maximum size of hemipenes was observed in April and May while the minimum size observed in July and August (Fig. 8).

## DISCUSSION

Testicular histology showed that spermatogenesis of *Teratoscincus bedriagai* began in early April, reached its peak in early May, and ended between late July and mid August. The gonadal index (GI) increased during spring because the testis weight increased in regenerative phase after hibernation and body weight decreased during hibernation. GI decreased in summer because the testis weight was reduced as a result of the high reproductive activity in spring. The authors predict that GI will increase after August because the testis weight increases again due to entering to the silent and regenerative phase. The spermatogenic cycle of sympatric lizard, Caspian Bent-toed Gecko, *Tenuidactylus caspius* has been studied in northern parts of Iran and has showed the similar results (Hojati et al., 2013). Gonadal index of *Tenuidactylus caspius* increased from September

Characters	Minimum	Maximum	Mean ± Std. Error	Std. Deviation	Sig.
W (g)	5.45	10.23	$7.56 \pm 0.19$	1.21	0.143
SVL (mm)	50.04	65.73	$56.86 \pm 0.77$	4.88	0.084
LCD (mm)	22.76	36.54	$29.70 \pm 0.56$	3.59	0.084
HL (mm)	12.81	17.57	$15.56 \pm 0.20$	1.30	0.347
HW (mm)	10.22	16.74	$13.34 \pm 0.25$	1.62	0.075
RTW (g)	0.004	0.084	$0.038 \pm 0.003$	0.024	0.000
LTW (g)	0.003	0.078	$0.037 \pm 0.003$	0.022	0.000
RTL (mm)	4.53	10.28	$7.54 \pm 0.33$	2.09	0.000
LTL (mm)	4.51	9.53	$7.04 \pm 0.28$	1.80	0.000
RTWi (mm)	1.75	4.35	$3.14 \pm 0.14$	0.92	0.000
LTWi (mm)	1.92	5.37	$3.70 \pm 0.21$	1.35	0.000
RTV (mm3)	7.28	95.85	$47.76 \pm 5.02$	31.77	0.000
LTV (mm3)	9.09	143.45	$65.82 \pm 7.97$	50.45	0.000
RHpL (mm)	2.43	5.36	$3.97 \pm 0.11$	0.71	0.000
LHpL (mm)	3.07	4.93	$3.91 \pm 0.09$	0.59	0.000
RHpW (mm)	1.43	3.32	$2.40 \pm 0.09$	0.57	0.000
LHpW (mm)	1.51	3.62	$2.55\pm0.09$	0.59	0.000
GI	0.12	2.10	$0.95 \pm 0.09$	0.58	0.000

TABLE 2. Descriptive statistics of macroscopic characters in Teratoscincus bedriagai (N=40).

**TABLE 3.** Descriptive statistics of microscopic characters in Teratoscincus bedriagai (N=40).

Characters	Minimum	Maximum	Mean ± Std. Error	Std. Deviation	Sig.
RTAD (µ)	3.87	8.20	$6.69 \pm 0.21$	1.34	0.000
LTAD (µ)	3.05	7.55	$6.09 \pm 0.23$	1.46	0.000
RSTN	24.00	66.00	$49.25 \pm 1.92$	12.19	0.000
LSTN	38.00	75.00	$55.80 \pm 1.70$	10.80	0.000
RSTD (µ)	72.50	232.50	$154.94 \pm 8.54$	54.05	0.000
LSTD (µ)	85.00	210.00	$155.74 \pm 6.87$	43.50	0.000
RGLD (µ)	20.40	118.40	$70.48 \pm 5.67$	35.86	0.000
LGLD (µ)	24.80	124.30	$74.08 \pm 5.64$	35.67	0.000
RLD $(\mu)$	53.1	89.10	$71.29 \pm 1.29$	8.21	0.000
LLD $(\mu)$	51.70	89.60	$72.77 \pm 1.85$	11.72	0.000
RSgN	60.00	216.00	$145.12 \pm 8.09$	51.18	0.000
LSgN	78.00	263.00	$167.97 \pm 10.27$	65.01	0.000
RSpN	74.00	431.00	$255.40 \pm 21.79$	137.82	0.000
LSpN	108.00	541.00	$334.55 \pm 24.01$	151.91	0.000
RStN	54.00	479.00	$268.57 \pm 23.77$	150.34	0.000
LStN	62.00	389.00	$227.20 \pm 17.06$	107.93	0.000
RSN	0.00	425.00	$207.82 \pm 24.44$	154.57	0.000
LSN	0.00	375.00	$200.07 \pm 20.17$	127.56	0.000



FIGURE 2. Mean of Testis Length (TL) of Teratoscincus bedriagai from April to August 2013.



FIGURE 3. Mean of testis weight (TW) of Teratoscincus bedriagai from April to August 2013.



FIGURE 4. Seminiferous tubules of *Teratoscincus bedriagai*. Spermatogonia, spermatocytes, spermatids and sperms are abundant on April (A) and May (B). Seminiferous epithelial heights and tubule diameters increase on May; and testes regression with germinal epithelium reduces on July (C) and August (D). All slides are shown at 400× magnificence.



FIGURE 5. Mean of sperm number (SN) of Teratoscincus bedriagai from April to August 2013.



FIGURE 6. Mean of gonadal index (GI) of Teratoscincus bedriagai from April to August 2013.



FIGURE 7. Hemipenes of Teratoscincus bedriagai.



FIGURE 8. Mean of hemipenis length (HpL) of Teratoscincus bedriagai from April to August 2013.

to November and the peak of spermatogenesis was in May and June. All lizards of the study area hibernated from October or November to March. In *T. bedriagai*, most of testicular parameters were observed in minimum size in August. In comparison with other lizards of Iran, these two sympatric geckos, *Teratoscincus bedriagai* and *Tenuidactylus caspius*, start their reproductive activities earlier than other studied species.

Hemipenal morphology in many geckos has long been described (Russell, 1977). Hemipenes in geckos are generally characterized by a club-shaped basal part consisting of pedicel and truncus that is followed by a voluminous apex with paired lobes. The hemipenes of some gekkonids such as *Aristelliger* and *Uroplatus* have internal supportive, calcified structures named hemibacula (Rösler & Böhme, 2006). This structure has been observed in *T. bedriagai*. Also, this ossified structure was observed in *Tenuidactylus caspius* (Hojati et al., 2013).

Moreover, the female reproductive cycle of *T. bedriagai* was studied in this area and showed the synchronization between the ovarian and testicular activities. Mating occurs in spring, especially at the beginning of May, with oviposition occurring from late May to late July (Mojibi and Hojati, 2014). Approximately, 1 to 2 eggs are laid by females per clutch. The maximum ovarian activity takes place in May and continues with a little decreasing in June and more reduction in July and finally ends in August (Mojibi and Hojati, 2014). Since spermatogenesis of *T. bedriagai* occur from April to July, this species follows a seasonal and alternate reproductive cycle with associate type.

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