

Notes on morphology, ecology, behavior and systematics of *Bufo luristanicus* SCHMIDT, 1952 (Anura: Bufonidae)

Bemerkungen zu Morphologie, Ökologie, Verhalten und Systematik
von *Bufo luristanicus* SCHMIDT, 1952
(Anura: Bufonidae)

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KURZFASSUNG

Bufo luristanicus SCHMIDT, 1952 ist ein Endemit des Südwest-Iran. Wir untersuchten die Morphologie, Ökologie und das Verhalten adulter und juveniler Exemplare von verschiedenen Fundorten in den Provinzen Lorestan und Khuzestan.

Die Tiere unterschieden sich hinsichtlich der Größe und des Typs der Tuberkel auf der Dorsalseite von Rumpf und Extremitäten. Wir unterschieden folgende zwei Tuberkeltypen: (a) einfache bestachelte Tuberkel und (b) warzenförmige bestachelte Tuberkel. Alle untersuchten Exemplare zeigten Asymmetrien in Größe und Form der Ohrdrüsen. Jungtiere hatten kleine Tuberkel, Trommelfell und Ohrdrüsen waren bei ihnen nicht feststellbar.

Das Taxon bevorzugt im wesentlichen zwei Lebensraumtypen, Tal- und Gebirgslandschaften, mit einer Reihe von Mikrohabitaten wie Tunneln und Gesteinsspalten.

Die grobmorphologische Untersuchung der im Rahmen dieser Untersuchung aufgesammelten Tiere legt nahe, *Bufo luristanicus* eher als Unterart von *B. surdus* denn als eigenständige Art aufzufassen. Es erscheint sinnvoll, *B. luristanicus* als Angehörigen des *B. surdus* Komplexes zu betrachten, der dann drei Formen umfassen würde: *B. surdus surdus*, *B. s. annulatus* and *B. s. luristanicus*.

ABSTRACT

Bufo luristanicus SCHMIDT, 1952 is an endemic toad of southwest Iran. We studied the morphology, ecology and behavior of adult and juvenile specimens collected from several localities in the provinces of Lorestan and Khuzestan.

The specimens varied in terms of the size and type of their tubercles on the dorsal side of trunk and limbs. We recognized two types as follows: (a) simple spiny tubercles and (b) nipple-shaped spiny tubercles. All specimens studied displayed some asymmetry in size and shape of the parotoid glands. The juveniles displayed small tubercles; tympanic membrane and parotoid glands were not observed in these specimens.

This taxon prefers two major habitat types, valleys and mountains with several microhabitats such as tunnels and clefts of stones.

Gross morphological examination of the specimens, which were collected and examined in this research, suggests that the taxon *B. luristanicus* should be considered a subspecies of *B. surdus* rather than an independent species. According to this it is more reasonable to put *B. luristanicus* in the *B. surdus* complex which, thus, would consist of three subspecies: *B. surdus surdus*, *B. s. annulatus* and *B. s. luristanicus*.

KEY WORDS

Amphibia: Anura: Bufonidae: *Bufo luristanicus*; morphology, systematics, taxonomy, ecology, behavior, Iran

INTRODUCTION

Bufo luristanicus SCHMIDT, 1952 is a small toad endemic to the western Iranian provinces of Khuzestan and Lorestan (SCHMIDT 1952; ANDERSON 1963; SCHMIDTLER & SCHMIDTLER 1969; BALOUTCH & KAMI 1995). This toad is distinct from other species of the collective genus *Bufo* inhabiting Iran by possessing square parotoid glands, a small

tympanum, and lacking conspicuous blotches on the back (SCHMIDT 1952; ANDERSON 1963; BALOUTCH & KAMI 1995). *Bufo luristanicus* is currently classified Least Concern on the IUCN Red List of threatened species (STÖCK et al. 2004).

Very little information is available on various aspects of the natural history of this

amphibian. In this paper we provide information about the morphological variation,

ecology, and conservation status of this toad.

MATERIALS AND METHODS

The studies focused on two localities: (a) Hosseiniye region, 15 km north of Andimeshk City, northern Khuzestan province (48°15'E, 32°41'N), an area with elevations between 350-500 m, and (b) a small waterfall, three km north of Poledokhtar City, southern Lorestan province (47°40'E, 33°13'N). This is a mountainous area with elevations between 850-1300 m. The localities are within the range shown by the IUCN (2008) map of *B. luristanicus*. Our fieldwork in the province of Khuzestan started on 22 April 2007 and lasted until 27 April.

Similarly, fieldwork conducted in southern Lorestan began on 3 June 2007 and continued until 8 June.

We collected 37 adult and 16 juvenile specimens from these localities; the toads were preserved in 96 % methanol and stored in Farhang Torki Herpetological Museum (FTHM) at Farhang Torki Ecology and Herpetology Center for Research (FTEHCR, Nourabad City). Direct observations were made during several days and nights in order to collect information on various aspects of behavior and natural history of this species.

RESULTS AND DISCUSSION

Morphological variation in adults (Figs. 1 and 2)

Besides the large tubercles which were found on the metacarpus and metatarsus, adult specimens had two types of spiny tubercles (ST) in which a small spine is visible at the top of each tubercle. ST may be present as (a) Simple Spiny Tubercles (SST, simple, not swollen and uniform in color) and (b) Nipple shaped Spiny Tubercles (NST, salient, swollen, and bicolor, i. e., the end of the tubercle is brown); this form has much similarity to a nipple, and the shape of the tubercle is similar to a breast (Fig. 1b). Size and volume were bigger in NST than in SST. Distribution of the types of tubercles across the body was different among specimens. The dorsum could be covered with both NST and SST forms of tubercles. In some specimens forelimb tubercles were SST only and exclusively found on the joints, while in other specimens SST were distributed over all parts of the dorsal forelimbs. Hindlimbs in some specimens were covered by both NST and SST, but in many specimens only SST were found. Distribution of ST on hindlimbs was similar as in forelimbs and unique in each specimen. The upper head was without any tubercles, but SST

and NST were present on the upper eyelids. In addition, SST surrounded the tympanum. However, tubercles were not found from the lower eyelids as far as to the end of the rostrum (Fig. 1a). In most specimens, NST were visible on the posterior lips. Parotoid glands were covered by both ST forms, especially NST. The diameter of NST on the parotoids was larger than on the other parts of the body. ST were lacking on the ventral side of the body. This was in contrast to the presence of ST on both metacarpus and metatarsus where they were readily visible.

Parotoid glands were roughly square in shape, small in size, and in one specimen did not even exist. Webbing membranes were not found on the forelimbs, but were present on the hindlimbs. The membrane reached less than 7% of the length of the 4th toe.

Large blotches were present on the limbs and on the sides of the head while they were absent on the dorsal body and head region in most specimens (Fig. 2); in some specimens small spots were surrounded by many tubercles. The end of each spiny-tubercle was needle-like, its color was blackish. Tubercle density was higher in the dorsal body than in other regions and lower on hindlimbs than on forelimbs. Dorsolateral tubercles were largest. All specimens in this

Table 1: Measurements of adult specimens of *Bufo luristanicus* SCHMIDT, 1952 ($n = 37$). BS - body size (= snout-vent length); HW - head width; HL - head length; HD - head depth; TD - tympanum diameter; ED - eye diameter; PL - parotoid length; PW - parotoid width; EE - inter-orbital diameter (distance between eyes); PP - distance between parotoids; ER - distance from anterior corner of eye to rostrum; TR - distance from anterior edge of tympanum to rostrum; IL - distance between insertions of fore and hind limbs; NN - distance between nostrils; RF - distance from rostrum to insertion of fore limb; SEM - Standard error of the mean.

Tab. 1: Maße adulter Exemplare von *Bufo luristanicus* SCHMIDT, 1952 ($n = 37$). BS - Kopf-Rumpf-Länge; HW - Kopfbreite; HL - Kopflänge; HD - Kopfhöhe; TD - Trommelfelldurchmesser; ED - Augendurchmesser; PL - Parotidenlänge; PW - Parotidenbreite; EE - Abstand zwischen den Augen; PP - Abstand zwischen den Parotiden; ER - Entfernung Vorderer Augenwinkel - Schnauzenspitze; TR - Entfernung vorderer Trommelfellrand - Schnauzenspitze; IL - Abstand zwischen den Insertionen von Vorder- und Hinterextremität; NN - Nasenlochabstand; RF - Entfernung Schnauzenspitze - Insertion der Vorderextremität; SEM - Standardfehler des Mittelwertes.

	BS	HW	HL	HD	TD	ED	PL	PW
Mean	39.58	13.77	12.62	5.80	1.16	3.95	3.30	2.87
SEM	0.89	0.35	0.41	0.28	0.22	0.24	0.47	0.36
(Min./Max.)	(33.6/42.7)	(11.8/14.9)	(10.5/14.9)	(4.6/7.2)	(0.0/1.9)	(3.03/5.3)	(0.0/5.3)	(0.0/3.9)
	EE	PP	ER	TR	IL	NN	RF	
Mean	10.16	6.93	5.08	9.46	14.69	2.88	14.81	
SEM	0.17	0.91	0.21	1.14	0.98	0.11	0.42	
(Min./Max.)	(9.2/10.9)	(0.0/10.6)	(3.7/5.9)	(0.0/12.8)	(11.7/22.3)	(2.1/3.3)	(13.3/17.5)	

study demonstrated an asymmetry in parotoid gland size, as well as distribution and density of tubercles. Morphometric details are given in Table 1.

Juvenile morphology

In all juvenile toadlets, with or without tail residues, parotid glands were not yet visible, but their place was occupied by large spiny tubercles and by this was distinguishable from other regions (e.g., pyramid region). For additional morphological data see Table 2.

Tympanic membrane and parotoid glands were not observed in juvenile specimens. Juveniles had small and large tubercles. The number of small tubercles was

higher than of large tubercles. Distribution of both types of tubercles was more or less homogenous on dorsum, but not in other parts of the body. The pattern of tubercles distributed on the body was unique in each specimen. For example, in one specimen the forelimbs were covered only by small tubercles, in contrast the hind limbs were covered by both types of tubercles. In individuals of larger body size (more than 21 mm), large tubercles were spiny, similar to adult specimens.

Based on all juveniles available ($n = 16$) four steps of growth of spiny tubercles can be described relative to the body size: (1) primary tubercles: (knobs, distinguishable from small tubercles), color white; (2) simple shaped tubercles: color yellowish; (3) nip-

Table 2: Measurements of juvenile specimens of *Bufo luristanicus* SCHMIDT, 1952 ($n = 16$). BS - body size (= snout-vent length); IL - distance between insertions of fore and hind limbs; HW - head width; HL - head length; HD - head depth; LB - diameter of large blotch on dorsum; RF - distance from rostrum to insertion of fore limb. SEM - Standard error of the mean.

Tab. 2: Maße juveniler Exemplare von *Bufo luristanicus* SCHMIDT, 1952 ($n = 16$). BS - Kopf-Rumpf-Länge; IL - Abstand zwischen den Insertionen von Vorder- und Hinterextremität; HW - Kopfbreite; HL - Kopflänge; HD - Kopfhöhe; LB - Durchmesser eines großen Rückenfleckes; RF - Entfernung Schnauzenspitze - Insertion der Vorderextremität; SEM - Standardfehler des Mittelwertes.

	BS	IL	HW	HL	HD	LB	RF
Mean / Mittel	18.52	7.20	6.56	6.47	3.56	1.11	7.91
SEM	1.39	0.59	0.45	0.46	0.21	0.19	0.64
Minimum	12.80	4.30	5.00	4.90	2.90	.40	5.70
Maximum	25.50	9.20	8.40	8.40	4.70	1.90	10.60

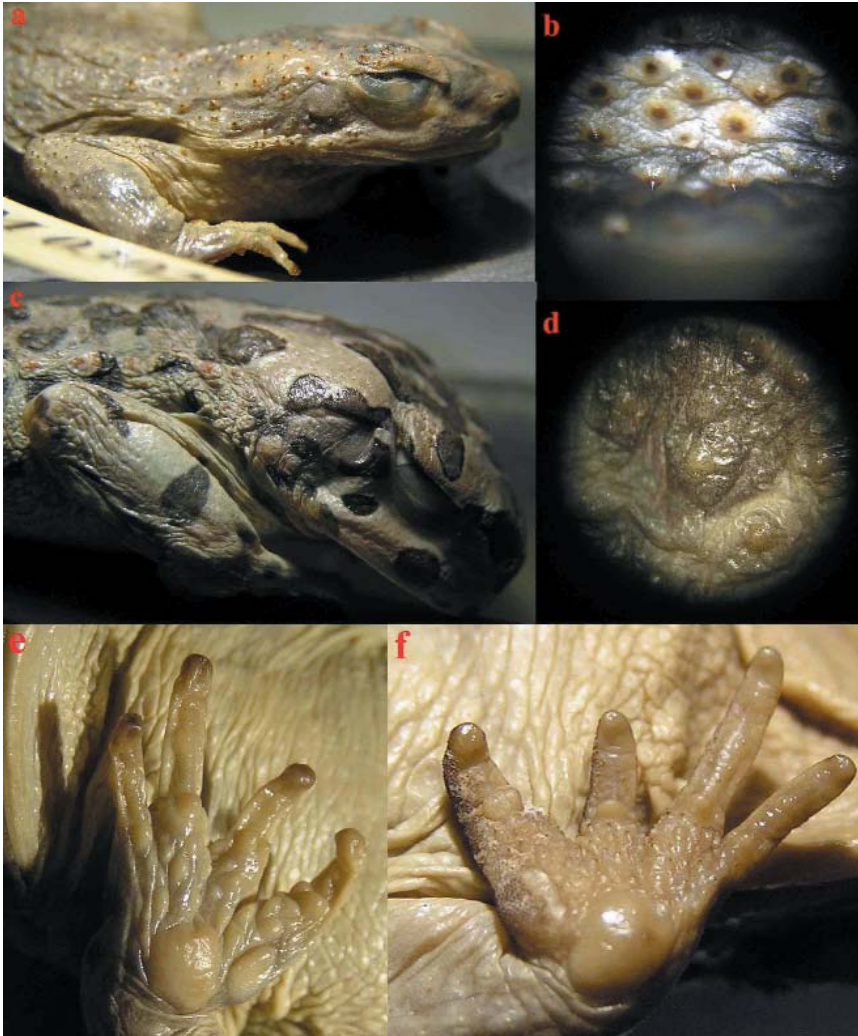


Fig. 1: Comparison of *Bufo luristanicus* SCHMIDT, 1952 with *Bufo viridis* LAURENTI, 1768 from the study area. a - lateral view of *B. luristanicus*; b - spiny tubercles in *B. luristanicus*; c - lateral view of *B. viridis*; d - tubercle structure of *B. viridis*; e - palm of *B. viridis*; f - palm of *B. luristanicus*.

Abb. 1: Vergleich von *Bufo luristanicus* SCHMIDT, 1952 mit *Bufo viridis* LAURENTI, 1768 des Untersuchungsgebietes. a - Seitenansicht von *B. luristanicus*; b - stachelige Tuberkel bei *B. luristanicus*; c - Seitenansicht von *B. viridis*; d - Tuberkelstruktur bei *B. viridis*; e - Handfläche von *B. viridis*; f - Handfläche von *B. luristanicus*.

ple-shaped tubercles: yellowish, surrounded by a white ring; in this stage tubercles become large; and (4) thorn shaped tubercles: tubercles become spiny. In early juvenile stages, the number of small tubercles was high, and strongly decreased with age.

Spiny tubercles at the sides and on/ around the parotoid glands matured earlier

than in other regions. The venter was not smooth, but covered by many knob-shaped tubercles which were different from dorsal tubercles.

Pattern

The number of blotches and body size were inversely proportional. At a smaller



Fig. 2: Variation in color-pattern and distribution of spiny tubercles on the dorsal and lateral side of some *Bufo luristanicus* SCHMIDT, 1952 in the FTHM collection (Nourabad City).

Abb. 2: Variabilität in Färbung und Verteilung der stacheligen Tuberkel auf dem Rücken und den Seiten einiger *Bufo luristanicus* SCHMIDT, 1952 in der Sammlung des FTHM (Nourabad City).

body size (less than 15 mm), blotch size on head, limbs and dorsum was similar; but at a larger body size (more than 16 mm), blotches on head and limbs were large, while they were small and well separated on the dorsum. In all juvenile specimens the color of the end of the fingers (in both hind and fore limbs) was white; this was in contrast to adult specimens where the finger tips were brownish.

Juveniles versus adults

In juvenile specimens the tubercles on the metacarpus are relatively larger than in

adults. In most juveniles the pattern is characterized by many small blotches on the body while in most adult specimens blotches can not be recognized. Parotoid glands and tympanum are not distinct in juvenile specimens. However, in some adult specimens the tympanum is very small or parotoid glands are not diagnosable.

Habitat (Fig. 3)

The authors found *B. luristanicus* in two distinct habitat types: (a) valley habitat characterized by rock outcrops (Hosseiniye region, 350-500 m a.s.l.); (b) mountainous

habitat containing a small waterfall and ponds (southern Lorestan province, 850-1300 m a.s.l.). Like other toads, *B. luristanicus* utilizes freshwater, especially ponds (Talab in Farsi) for reproduction (e.g., BALOUTCH & KAMI 1995). Maximum population densities of *B. viridis* (LAURENTI, 1768) and *Pelophylax ridibundus* (PALLAS, 1771) were observed near and inside ponds, while *B. luristanicus* was found most frequently at some distance from the water.

Microhabitat and variation in annual biological activity

Bufo luristanicus took shelter in cavities provided by accumulations of big or small stones, cracks of large stones and natural tunnels. Depth of these tunnels was approximately 30 to 150 cm; many of them were interconnected and represented a cavernous network. One to four specimens were seen in each tunnel and one or two specimens could be found together under small stones. Under one stone (about 50 x 30 cm) more than 20 juvenile specimens of *B. luristanicus* were burrowed.

We observed a timing gradient in the annual biological activity favoring the Khuzestan population of *B. luristanicus*. In Lorestan, biological activity was clearly shorter (early spring to early autumn) than in Khuzestan (mid-winter to late autumn), obviously due to different climate and ecological conditions: (1) higher average annual temperature, lower elevation, hilly habitat type in the Khuzestan locality, and (2) lower average annual temperature, higher elevation, mountainous habitat type in the Lorestan locality.

Sympatric amphibian and reptile species observed

In northern Khuzestan the authors observed *B. luristanicus* sympatric with *Bufo surdus* BOULENGER, 1891, *B. viridis* LAURENTI, 1768, *Pelophylax ridibundus* (PALLAS, 1771), *Asaccus elisae* (F. WERNER, 1895), *Cyrtopodion scabrum* (HEYDEN, 1827), *Tropiocolotes helenae* (NIKOLSKY, 1907), *Ophisops elegans* (MENETRIES, 1832) and *Trachylepis aurata* (LINNAEUS, 1758). In southern Lorestan *B. luristanicus* co-occurred with the following herpetological species: *Asaccus nasrullahi* Y. WERNER,

2006, *Laudakia nupta* (DE FILIPPI, 1843), *T. aurata*, *O. elegans*, *Malpolon monspessulanus* (HERMANN, 1804), *Natrix tessellata* (LAURENTI, 1768) and *Platyceps karelini* (BRANDT, 1838).

Behavior

Activities of *B. luristanicus* started at sunset and lasted until morning; a few specimens were observed by day (before mid-day). Locomotion was slower in *B. luristanicus* than in *B. viridis*. *Bufo luristanicus* presents a kind of discreet, almost timid behavior and we think this trait is special for this species.

Calls were exclusively produced by night and were an important factor for observing this species in nature. The calls produced by *B. luristanicus* were shrill, strong, communicative, and significantly different from those of *B. viridis* and *P. ridibundus*. Based on our observations each male *B. luristanicus* produced sequences of 3-8 single calls, which lasted 3-10 seconds.

After seizing of the animal, a defense call and behavior such as abdomen puffing was performed. The call was similar as in *B. viridis* but abdomen puffing occurred before catching, in contrast to *B. viridis* (our observation). Abdomen puffing (before catching) seemed to be a special and vital defense behavior in *B. luristanicus*. When seized the animal discharged cloacal fluid.

Systematics of the *Bufo surdus* BOULENGER, 1891 complex

FROST & AMNH (1998-2008) reviewed the synonymy of *B. luristanicus* and commented on the history of its systematic allocation: *B. luristanicus* was placed into the *B. viridis* group by INGER (1972), and the *B. surdus* subgroup by STÖCK, GÜNTHER & BÖHME (2001). SCHMIDTLER & SCHMIDTLER (1969) and EISELT & SCHMIDTLER (1973) considered *luristanicus* a subspecies of *surdus*. Later *luristanicus* was removed from the synonymy of *B. surdus* by ANDERSON (1985) and STÖCK, GÜNTHER & BÖHME (2001).

According to SCHMIDT (1952), ANDERSON (1963), BALOUTCH & KAMI (1995) and STÖCK et al. (2004) *B. luristanicus* is a full species and *B. surdus* consists of two sub-

species: *B. s. surdus* and *B. s. annulatus* SCHMIDTLER & SCHMIDTLER, 1969; both being distinct from *B. luristanicus* by tympanum and dorsal pattern characteristics. Some authors considered *B. luristanicus* to belong to the *B. surdus* group (SCHMIDTLER & SCHMIDTLER 1969; STÖCK et al. 2001).

However, the tympanic membrane is present in *B. luristanicus* and *B. s. annulatus*, but in both species it is very small or covered by skin; in *B. s. surdus* there is no tympanum at all (SCHMIDT 1952; BALOUTCH & KAMI 1995). Concerning the color-pattern, *B. s. surdus* lacks any green-olive circular spots on the dorsum, while such spots are present in *B. s. annulatus*.

The present study revealed that the variation in tympanum structure and distributional pattern of blotches on the body overlap considerably among the three species of the collective genus *Bufo* (i. e. *B. s. surdus*; *B. s. annulatus*; *B. luristanicus*). On one of the *B. luristanicus* specimens there is not a single blotch on the dorsum and the tympanum is absent (as in *s. surdus*); some specimens have small blotches (ring blotches) on the dorsum (as in *s. annulatus*); one of these specimens is without tympanum (as in *s. surdus*) but two have a tympanic membrane (typical to *s. annulatus* and *luristanicus*); moreover, parotoid glands are not visible in one specimen (Table 1).

Gross morphological examination of the specimens collected in this study suggested that *B. luristanicus* should be considered a subspecies of *B. surdus* rather than a species of its own leading to the inclusion of *B. luristanicus* into the *B. surdus* complex which thus would consist of three subspecies: *B. surdus surdus*, *B. s. annulatus* and *B. s. luristanicus*. This hypothesis on the taxonomic position of *B. luristanicus* is similar to the opinion of other authors (SCHMIDTLER & SCHMIDTLER 1969; STÖCK et al. 2001).

STÖCK et al. (2006) suggested that *B. luristanicus* and *B. surdus* were close to *B. viridis*, but studies based on ND (NADH dehydrogenase) and tRNA (transfer RNA) have shown that *B. luristanicus* is more closely related to *B. v. viridis* and *B. variabilis* (PALLAS, 1769) in that all three taxa cluster in a single clade. A study based on ND and tRNA analysis (STÖCK et al. 2006) revealed that *B. surdus* was much different from *B. luristanicus*. This is in contrast to the results provided by the present morphological study, where *B. luristanicus* showed much morphological overlap with *B. surdus*.

On the other hand we found that *B. luristanicus* has well developed spiny tubercles. This trait plays an important role in behavioral and adaptation aspects stabilized during evolution and is not found in *B. viridis*.

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Fig. 3: Habitats of *Bufo luristanicus* SCHMIDT, 1952 in the region of Hosseiniye (a-b) and in southern Lorestan province (c-d), Iran.

Abb. 3: Habitate von *Bufo luristanicus* SCHMIDT, 1952 im Gebiet von Hosseiniye (a-b) und im Süden der Provinz Lorestan (c-d), Iran.

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DATE OF SUBMISSION: February 4, 2008

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Digitale Literatur/Digital Literature

Zeitschrift/Journal: [Herpetozoa](#)

Jahr/Year: 2009

Band/Volume: [21_3_4](#)

Autor(en)/Author(s): Javari Majid, Torki Farhang

Artikel/Article: [Notes on morphology, ecology, behavior and systematics of Bufo luristanicus SCHMIDT, 1952 \(Anura: Bufonidae\) 171-178](#)