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Cover: *Cuora galbinifrons*. Diaoluoshan, 18 km N. of Nanxi, Hainan Province, China. Photo by James F. Parham.

The Activity and Thermal Biology of the Fossorial Reptile, *Diplometopon zarudnyi* (Amphisbaenia: Trogonophiidae) in Central Saudi Arabia

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Abstract .- The nocturnal activity of the fossorial amphisbaenid *Diplometopon zarudnyi* was studied in the field and its thermal selection and tolerance were determined in the laboratory. During the study period (summer) the animals commenced activity at 20.00 hrs (ground temperature 30-32°C) and finish their foraging between 3.00 to 4.00 hrs (ground temperature 28-26°C) before the break of dawn. The mean activity temperature determined in the field was 31.5°C, which was usually 0.5 to 1°C higher than the ground temperature. The mean selected body temperatures in a gradient during day and night were 26.4°C (range 15-38°C) and 26.3°C (range 14-38°C) respectively. The Critical Thermal Maximum was 47.6°C and the Critical Thermal Minimum was 7°C. *D. zarudnyi* tolerates a wide range of temperatures while maintaining its mean body temperature within a narrow range. The role of Selected Body Temperature is discussed in relation to metabolism.

Key words.- Amphisbaenia, Trogonophiidae, fossorial reptile, *Diplometopon zarudnyi*, Saudi Arabia, Central Arabia, activity, thermal preference, thermal tolerance.

Introduction

The amphisbaenian family Trogonophiidae is represented in central Arabia by the single species *Diplometopon zarudnyi* (Al-Sadoon, 1988). The range of this species extends to northern Arabia and the coastal Arabian Gulf (Arnold, 1986). *D. zarudnyi* is an oscillating digger, commonly found burrowing in low sand dunes in open terrain and in sub-surface soils of date palm farms. This amphisbaenian is a nocturnal forager, sometimes occupying ant and termite tunnels.

In this study the nocturnal activity of *D. zarudnyi* was studied in the field, and its thermal selection and temperature tolerance were determined in the laboratory. A comparison of results of this study has been made with the results of other species from different geographical habitats.

Material and Methods

Several field trips were made during the summer season (July-August.) to habitats of *D. zarudnyi* located around Riyadh city (Thummama and Dilam). Nocturnal activity of the animals was observed during the activity period. Rechargeable fluorescent lanterns were employed to illuminate the area being studied. When tracks appeared on the sand surface, they were followed up to the location of the animal, which was either dug out of its subsurface position by hand or using a small hand shovel. Soil temperature (T_s), air temperature (T_a) and cloacal body temperature (T_b) were taken within 15 seconds of capture by a quick reading cloacal thermometer (Millar and Weber Co.,

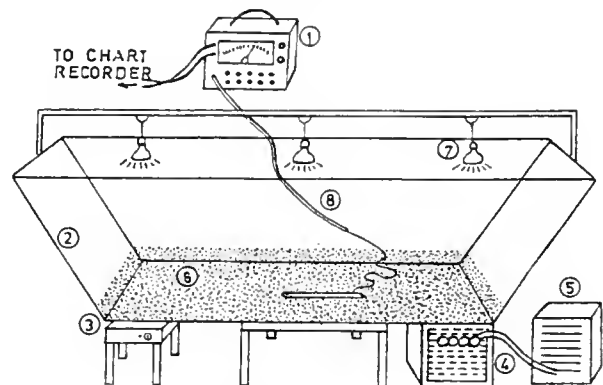


Figure 2. Setup used to measure the selected body temperature: (1) Telethermometer, (2) Thermal gradient, (3) Thermostat heater, (4) Cooling coil in water bath, (5) Refrigerant machine, (6) 10 cm thick sand layer, (7) 100 W heating bulb, (8) Thermistor probe.

U.S.A). Air and ground temperature were also monitored every hour from dusk to dawn.

Thirty *D. zarudnyi* (adult animals of both sexes) were used in the laboratory study (mean mass = 7.63 g, SD \pm 2.54; mean SVL=182 mm, SD \pm 13.4; mean VTL=14 mm, SD \pm 2.0). They were collected from various locations around Riyadh city. They were maintained for short periods in Plexiglass boxes filled with 10 cm of clean sand. The sand was sprinkled periodically with water. Mealworms and water were available *ad lib*. The laboratory temperature was 23 \pm 1.5°C.

The Selected Body Temperature (SBT) of ten amphisbaenians was determined in a metal thermal

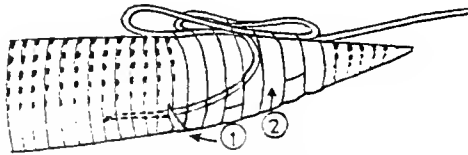


Figure 2. Method employed to tie the thermistor; the probe (1) inserted inside the animal's cloaca and looped to be held on the tail (2) with bands of adhesive tape.

gradient which measured 200 x 25 x 40 cm (Fig.1). Sand was spread evenly to a depth of 10 cm on the base of the gradient. A temperature gradient from 10-50°C was achieved by fitting a thermostatic hot plate below one end and by placing an insulated ice container constantly frozen by an immersion refrigerant coil on the other end. The surface sand at the cool end was sprinkled with water intermittently to prevent desiccation. The gradient was illuminated by two 100 watt bulbs suspended 80 cm from the surface of the sand controlled by an electronic timer to maintain the photoperiod. The animals were left in the gradient for one day prior to the start of the experiment for acclimation.

The cloacal body temperature (T_b) was monitored with high sensitivity probes (Model 511; Yellow Springs Inc.). The probes were connected to a multi-channel YSI Telethermometer, and an Omniscribe Houston Instruments continuous chart recorder. The flexible tip of the pre-calibrated probe was inserted for 1 cm into the cloaca and held in place with 3-4 mm bands of adhesive tape (Fig.2). The probes remained in place even when the animal burrowed below the sand. Twenty four hour continuous recording of T_b was obtained for three days on each animal and one measurement for each hour was recorded.

These instruments were also used to measure the Critical Thermal Maximum (CTMax) and the Critical Thermal Minimum (CTMin). CTMax is the arithmetic mean of the collective thermal points at which locomotor activity becomes disorganized and the animal loses its ability to escape from conditions that will promptly lead to its death. CTMin is the low temperature that produces cold narcosis and prevents locomotion (Pough and Gans, 1982). A sheet metal box (25 X 20 cm) filled with a layer of sand and kept above a thermostat heater was employed to measure CTMax. For CTMin, a Plexiglass box (25 X 15 cm) with a layer of sand held inside a larger insulated box surrounded by ice cubes was employed. Twenty different animals (10 for each category) were used. Each animal was gradually heated or cooled inside the experimental chambers (1°C increase or decrease per

minute). The probe remained inserted in the cloaca and the body temperature was monitored continuously while the test chamber sand was gradually heated or cooled.

To determine the CTMax and CTMin, the amphibaenian was observed until the animal lost its righting response when it was turned on its back. During the CTMin experiments, after initial cooling (9.5°C), the animal lost its ability to right itself but, after its belly was touched by a fine paint brush, the animal exhibited a wave of convulsions down the body. Further cooling led to total loss of all responses, and the temperature at which this occurred was designated as the CTMin. All but one experimental animal survived after returning to room temperature. After 10 days, the animals which were used for CTMax experiments were used to measure CTMin. The CTMin of these animals was compared to that of the first group.

Statistical analysis was performed using GLM procedure of Minitab package (version 8.2). Two-way analysis of variance (ANOVA) with interaction (unbalanced) was used for data analysis ($P < 0.05$).

Results

The nocturnal activity of the animals is clearly defined during summer nights. Emergence for foraging activity begins at 22.00 hrs when the air temperature is 30-32 °C and the ground temperature is 32-34°C. The foraging activity subsides between 03.00 to 04.00 hrs when the air temperature is 28-26°C and the ground temperature is 27-25°C. After this the animals move deeper into the sand surface (presumably to their burrows or refuges) and no kind of activity is observed during the daylight hours.

The animals were active between adjacently located shrubby small sand dunes. Most of these sand dunes hosted colonies of ant and termite mounds. Movement is typically a combination of sub-surface and surface locomotion. Undisturbed movement of the animal over the surface of the sand was observed only two instances. On the rest of the occasions the animals were tracked by their impressions in the sand. The animals move just below the surface of the sand for two or three meters by piercing and wiggling by strong head and body movement (probably in search of termites). After that they come out on the surface and effortlessly glide on the sand surface for several meters by fast subsequent spring action. This was done by making an 'S' shape of the body and flicking forward. Consequently this kind of activity leaves a distinct pattern of tracks on the sand surface which

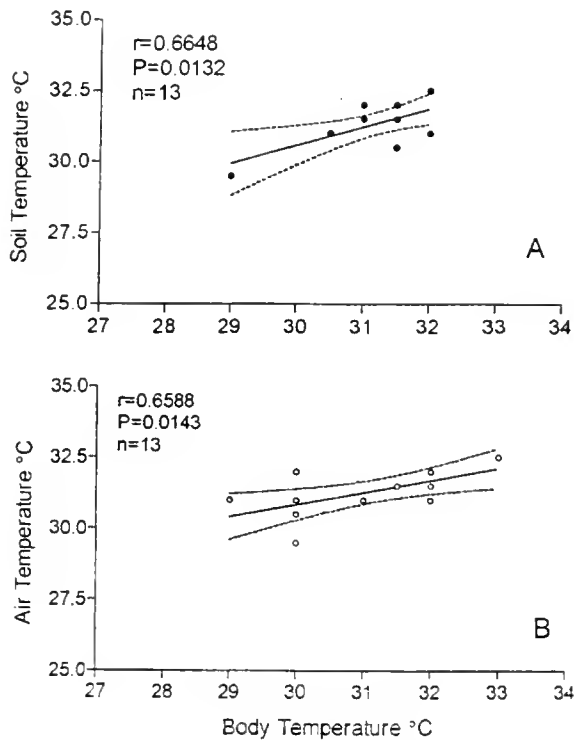


Figure 3. A. Relationship between body temperature (T_b) and soil temperature (T_s). B. Relationship between body temperature (T_b) and air temperature (T_a). (The regression line, significance value and sample size are as indicated).

can be easily differentiated between the tracks of other animals of the habitat.

The mean field body temperature for the active animals captured was 31.5°C (range $29.5 - 32.5^{\circ}\text{C}$; $n=13$). Regression analysis showed a significant difference between the $T_b - T_a$ and $T_b - T_s$ ($P<0.05$; Fig. 3). The mean selected temperatures in the gradient during the day and night were 26.4°C ($\text{SD} \pm 5.2$) and 26.3°C ($\text{SD} \pm 5.7$) respectively; the temperatures selected during day ranged between 15 and 38°C and between 14 and 38°C during the night (Fig.4). About ninety percent of *D. zarudnyi* selected temperatures between 23 and 36°C during the day and between 21 and 36°C during the night. Temperature selected did not differ significantly between day and night ($F=0.14$, $P=0.705$). However, individuals differed significantly in temperatures selected ($F=127.37$, $P=0.001$). Also, there was a highly significant interaction between individuals and the time of day ($F=7.72$, $P=0.001$). The average hourly temperature pattern of *D. zarudnyi* showed a gradual increase of body temperature (in the gradient) and reached its highest level just before the end of the dark period (Fig.5), and a

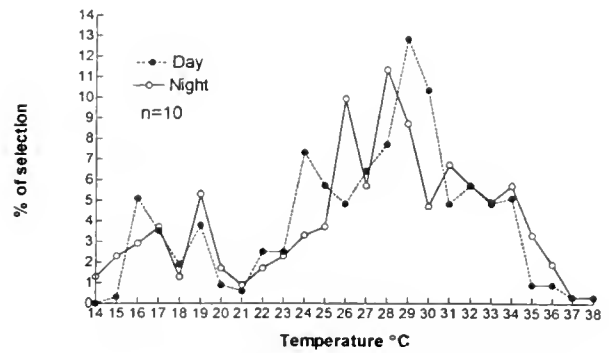


Figure 4. Representation of body temperature selection (%) by *Diplometopon zarudnyi* in the thermal gradient during day and night periods.

gradual decrease of body temperature to its lowest value by the end of the light period.

During the CTMax trials, animals below the soil surface emerged when the sand temperature reached 40°C . The CTMax was 47.6°C . During the CTMin trials, the animals lost the righting reflex at 9.4°C ($\text{SD}\pm 1$) and CTMin was 7°C ($\text{SD}\pm 0.6$). The second group of animals used in measuring CTMin lost the righting reflex at 11.3°C ($\text{SD}\pm 1.1$) and CTMin was reached at 9.5°C ($\text{SD}\pm 0.5$).

Discussion

D. zarudnyi come out of their refuges at night when the air temperature and the soil surface temperature are favorable. By employing two kinds of locomotion the task of foraging is accomplished. They expend vigorous efforts on their foraging activity, since they have to accomplish it in a short duration; multiple tracks crossing each other were observed for a single animal in a wide area, giving a false impression of many animals being active. This lead to many dead ends while following the leads of the tracks.

Previous reports (gradient studies) assumed that the body temperatures of amphisbaenians are equal to soil temperatures, since readings were obtained from the soil surrounding the animals. For a more accurate evaluation, here, we recorded body temperatures direct and continuously in the thermal gradient.

The selected mean temperature in the gradient (26°C) measured in this study explains the Metabolic rate-Temperature curve (M-T curve) reported for *D. zarudnyi*; in which both the adults (mean wt.6.34 g) and sub-adults (mean wt.3.15 g) showed stable O_2 consumption rates between $25-30^{\circ}\text{C}$ (Al-Sadoon, 1986).

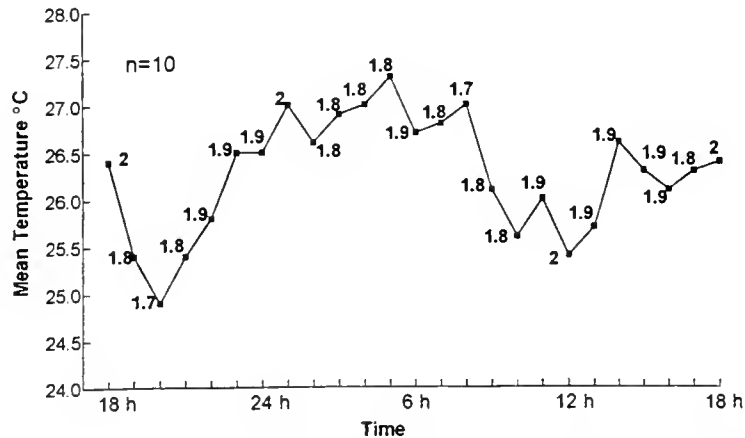


Figure 5. Mean selected body temperature pattern of *Diplometopon zarudnyi* in the thermal gradient during 24 hours. Numbers at each point indicate the SEM.

The body temperature for active *D. zarudnyi* are higher in the field than in the laboratory because the animals were deprived of food during the experiment, and also may be the result of confinement to a limited space in the gradient. Van Berkum (1980) demonstrated that the SBT is lower in lizards with decreased food consumption. It is recommended to measure metabolism during the post absorptive state of digestion, so as to minimize any contribution of specific dynamic action (Eckert and Randall, 1983).

As *D. zarudnyi* is fossorial, the low mean selected temperature (26°C) could extend activity and stabilize body metabolism in the sub-surface habitat. The present results supplement the previous published reports of lower temperature selection in amphisbaenians. Avery (1982) noted that many fossorial snakes, amphisbaenians, sea snakes, fresh water and marine turtles, appear not to thermoregulate and that many select low temperatures. Evidence of thermoregulation in *Amphisbaena mertensi* was reported by Abe (1984); mean field body temperature and preferred temperature in the gradient was 21.1°C and 21.4°C respectively. Also, Martin et. al. (1990) reported marked field thermoregulation in *Blanus cinereus* and it selected low field body temperatures. Gatten and McClung (1981) reported the low mean body temperature for *Trogonophis weigmanni* in a range of 21.7°C to 23.4°C. A field study of *Bipes biporus* demonstrated that this worm lizard can thermoregulate if necessary, by vertical and horizontal changes in substrate positioning (Papenfuss, 1982). More recently, Diaz-Paniagua et al., (1995) during a field study of seasonal and diel activity of *Blanus cinereus*, reported an activity range of ground temperatures between

13.4 -27.8°C which is low and as well clearly indicates thermoregulation.

The low temperature selection of *D. zarudnyi* is also in line with the observations made on other burrowing species of reptiles. Bury and Bolgooyen (1976) determined the thermal preferendum of the legless burrowing lizard *Anniella pulchra* to be 24-25°C. Clark (1968) reported five subterranean species of small snakes to select lower temperatures than surface dwelling species. Burrowing snakes of the family Uropeltidae also appear to select a low body temperatures between 18°C and 20°C (Gans, 1973). Low selected body temperatures were also reported for *Anguis fragilis* (Gregory, 1980).

A previous study showed that *D. zarudnyi* had no endogenous circadian rhythm and no periodicity in alternating light and darkness although it was more active at high temperatures (Cloudsley-Thompson, 1979). The finding in the present study wherein *Diplometopon* showed no significant difference between day and night time temperature selection is consistent with the lack of an endogenous circadian rhythm in temperatures selected in day and night. However, in the present results there is a highly significant difference among individuals. This is probably due to individual differences in body weight, health status, age and sex of the animals tested.

In the gradient *D. zarudnyi* elevated its body temperature slightly in the second half of the night, a time when they were seen to forage actively in the field. In contrast, *T. weigmanni* tested in a thermal gradient showed elevated temperatures in the afternoon and early evening (Gatten and McClung, 1981). Foraging activity at dawn and in early morning also is reported

for *Agamodon anguliceps* from Somalia; the animals were located in the top 2-3 inches of the soil at dawn, appearing to have moved upwards during the night. As temperatures rose, most animals disappeared from the top 6-12 inches of the ground (Gans and Pandit, 1965).

The CTMax value for *Diplometopon* is similar to that of certain terrestrial lizards of Central Arabia, but the CTMin was higher than that reported for most other sympatric lizards (Al-Johany, 1986). The relatively high CTMin might be related to the subterranean temperatures encountered by *Diplometopon* which seldom fall below freezing, unlike the surface temperatures which may drop below 0 °C. The 2.5 °C increase in the CTMin for animals that had experienced the CTMax might be attributed to a short-time heat-hardening effect (see Maness and Hutchison, 1980).

Prior to this study there was no data available on the field thermal ecology of *D. zarudnyi*. By the combined result of the field and laboratory study it is now confirmed that the animal is a thigmotherm but not in its strict sense. Since it was observed that the animal manages (probably by metabolic activity) to keep its body temperature a degree or half higher than the ground temperature.

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Description of a New Species of *Pseudorabdion* (Serpentes: Colubridae) from Panay Island, Philippines with a Revised Key to the Genus

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Abstract.— We describe a new species of snake in the colubrid genus *Pseudorabdion* from the western coastal mountain range of Panay island, Philippines. The new species appears to be related to the members of the *P. mcnamarae* species group (*P. mcnamarae* and *P. taylori* from the Philippines, *P. albonuchalis*, and *P. saravacensis* from Borneo, and *P. sarasinorum* from Sulawesi) but differs from each of these species by characters of scalation and color pattern. The new species is one of several other recently-discovered vertebrates from Panay island. Together, these discoveries suggest that diversity and endemism patterns of the Negros-Panay Pleistocene aggregate island platform (Negros, Panay, Cebu and Masbate islands) are more complex and interesting than previously thought.

Introduction

While engaged in a biological reconnaissance survey⁵ of the western coastal mountain range of Panay Island (Fig. 1), the senior author collected two specimens of what appeared at the time to be *Pseudorabdion mcnamarae*, a species already well known from Negros Island. On closer examination it was discovered that the Panay specimens differed in significant details from the Negros population while sharing features with *P. taylori*, known from Mindanao, and with *P. albonuchalis*, a species earlier known only from the type specimen (but see Inger and Leviton, 1966), which can no longer be located, and said to have come from Sarawak, Borneo. The observed differences among these related species leads us to believe that the Panay specimens represent a previously unrecognized, taxonomically distinct population of *Pseudorabdion* allied to the section of the genus *Pseudorabdion* whose members possess an elongate loreal (lori-ocular) that borders the orbit anteriorly.

Pseudorabdion talonuran new species (Figs. 2-3)

Holotype: PNM 2712 (Field no. PNM/CMNH 671), adult male, collected on western foothills of Mt. Madja-as (11°23' N, 122°09' E; elev. 1500 m), Barangay Allojipan, Municipality of Culasi, Antique Province, Panay Island, Philippines, 28 May 1992 by Rafe Brown and Roger Sison.

Paratype: CMNH 5076 (Field no. PNM/CMNH 670), young male, other data as for holotype except it was collected at 1410 m.

Etymology: The specific epithet is chosen from the Antique Province dialect Caray-a, and is derived from the words, "talon" (forest) and "uran" (rain), in reference to the high elevation rain forest habitat where the new species was collected on Mt. Madja-as.

Diagnosis: Elongate loreal (=lori-ocular) present, extending from the posterior border of the nasal to the orbit of the eye; ventrals (M) 139-146; subcaudals (M) 36-39; total of ventrals plus subcaudals (M) 175-185; scales of dorsum each edged with a mottling of brown pigment, the posterior and central portions of each scale pale, lacking a dense infusion of dark pigment, the lateral and latero-ventral scales with larger pale areas than those on the dorsum; no distinct nuchal collar but pattern of pale centers and dark rims characterizes nuchal scales and head shields, which are somewhat mottled dark and light.

Pseudorabdion talonuran belongs to the section of the genus *Pseudorabdion* characterized by the presence of a loreal (lori-ocular) shield. From the allied Philippine *P. mcnamarae*, it differs in lacking a pale nuchal color in adults and in having more than 30 subcaudals in both males and females; from *P. taylori* it differs in having the centers and apical tips of the dorsal scales nearly pigmentless whereas in *P. taylori* the centers are pale brown, and in having the hemipenes minutely

⁵This paper represents contribution number 23 to the results of the National Museum of the Philippines/ Cincinnati Museum of Natural History Philippine Biodiversity Inventory (PNM/CMNH PBI)

spinose (in *P. taylori* the apical tips are calyculate). Among the non-Philippine species having a lori-ocular, *P. talonuran* differs from *P. sarasinorum* in having the anterior chin shields in contact with the mental, from *P. albonuchalis* in having fewer subcaudals (36-39 vs. 43) and the frontal not border the eye, and from *P. saravacensis* in having a greater number of subcaudals (36-39 vs. fewer than 30) and fewer maxillary teeth (8 vs. 14). From the remaining species in the genus, it differs in having an elongate loreal (lori-ocular) shield that borders the eye.

Description of holotype: (Adult male) Rostral as high as wide, portion visible from above slightly greater than length of internasal suture; internasals small, greatest length about one-half greatest length of prefrontals, in contact with rostral, nasal, loreal and prefrontal; common suture between prefrontals about four-fifths length of frontal; prefrontal bordering eye between loreal and supraocular, also in contact with internasals; left prefrontal in contact with both internasals (because internasal suture offset from midline);

frontal two-thirds length of parietals, subtriangular, three-fourths as wide as long, in contact with prefrontals, supraocular and parietals, but not bordering eye; supraocular distinct, not fused to ocular brill; maximum length of parietals slightly greater than distance to tip of snout; nasal quadrangular, undivided, resting on first and in contact with second supralabials, nostril pierced in anterior lower quadrant; loreal elongate, resting on second and third supralabials, about twice as long as its distance to tip of snout, bordering eye; preocular absent (or more likely fused to form an elongate loreal [lori-ocular]); postocular, subquadrangular, about one half size of supraocular, in contact with fourth and fifth supralabials, not as high as eye, its lower border extending below level of eye and inserting between two supralabials; eye small, its diameter equal to its distance from mouth, pupil round; five supralabials and one large postsupralabial, supralabials three and four border eye, fifth largest and broadly in contact with parietals, followed by third, second, fourth, and first in descending order of

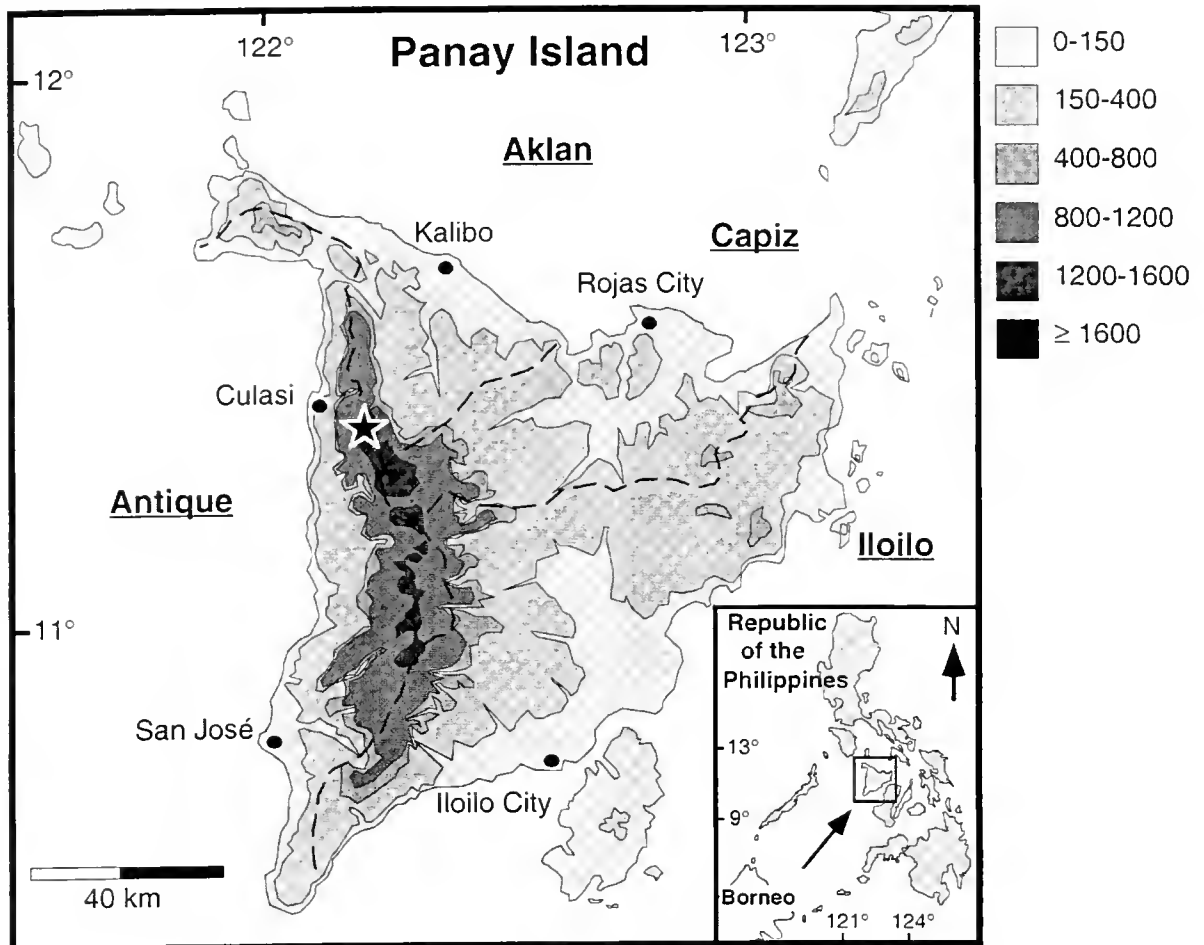


Fig. 1. Map of Panay Island, showing its position in the Philippines (inset), major cities (darkened circles), provinces (dashed lines; names underlined) and elevational topography (see key). The type locality (Mt. Madja-as) of *Pseudorabdion talonuran* is indicated with a star.

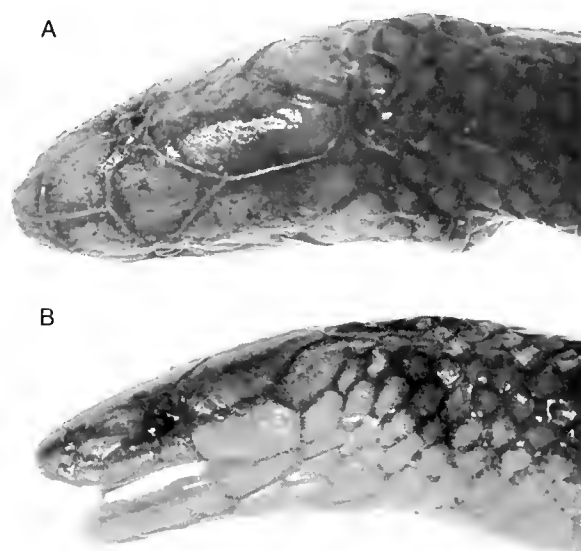


Fig. 2. Dorsal (A) and lateral (B) views of head of holotype of *Pseudorabdion talonuran*.

size; one large "posterior temporal" between postlabial and parietal; mental in contact with elongate anterior chin shields; infralabials five, first three in contact with anterior chin shields, third and fourth bordering posterior chin shields; posterior chin shields about three-fourths length of anterior pair, separated from one another in the midline for two-thirds their length by insertion of a gular scale, and about same size as bordering gular scales; maxillary teeth eight.

Scales smooth, without apical pits, in 15 longitudinal rows, not reducing posteriorly before vent; ventrals 146 mm; subcaudals 39 mm, paired; anal undivided.

Hemipenes extend *in situ* to 8th subcaudal plate, forked at the level of the 6th plate; apical ends with minute spines.

Total length 265 mm; tail 46 mm; head length (tip of snout to angle of jaws) 11.25 mm, (tip of snout to

posterior edge of parietals) 9.1 mm; diameter of eye 0.8 mm.

Color pattern (in alcohol): Dorsal scales each with irregular dark borders; apical end and centers pale, nearly pigmentless, pigmentless areas larger and more distinct laterally; ventrals unpigmented except for lateral edges; no nuchal collar.

Paratype: The paratype, a young male, differs from the holotype in the following particulars: ventrals 139; subcaudals 36; snout-vent length, 265 mm; tail length 47 mm; apical ends of hemipenes do not appear to be spinose (but the everted organ is poorly preserved and difficult to examine).

Ecological notes: The forest habitat of the new species on Mt. Madja-as (Fig. 4) has been classified by Whitmore (1984) as the transition zone between mixed dipterocarp (submontane) and mossy (upper montane) forests. The forest consists of two strata, a canopy of 10 m and subcanopy of 3-4 m with emergent trees as high as 18 m; an herb and shrub layer vegetation was also present. The forest near the collection site was mossy and contained high densities of epiphytic ferns and orchids. The topography was qualitatively characterized as steep, with numerous sheer rocky valleys and forest-covered ridges (see Ferner, et al., 1997 for more details.) Both holotype and paratype were found beneath logs.

Remarks: The section of the genus *Pseudorabdion* characterized by the presence of an elongate loreal (lori-ocular) that borders the orbit, termed here the "mcnamarae" section, includes three species in the Philippines, *P. mcnamarae*, *P. taylori*, and *P. talonuran*, and three non-Philippine species, *P. albonuchalis* and *P. saravacensis* from Sarawak, and *P. sarasinorum* from Gunung (Volcano) Soudara, Sulawesi.

Five species (*P. longiceps*, *P. ater*, *P. oxycephalum*, *P. eiselti*, and *P. montanum*) lack the loreal (lori-ocular); the prefrontals are in contact with the second and third upper labials. Of the forms lacking a distinct

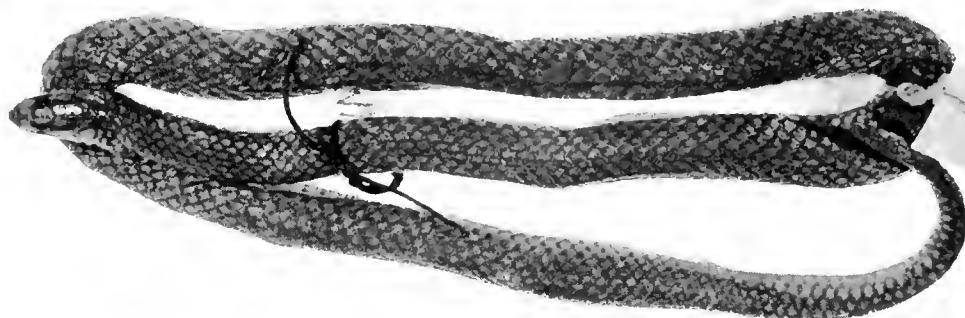


Fig. 3. Dorsal view of holotype (PNM 2712) of *Pseudorabdion talonuran*.



Fig. 4. Cloud forest habitat of *Pseudorabdion talonuran* at the type locality: Mt. Madja-as, Antique Province, Panay Island, Philippines.

loreal (or if a small scale is present in the loreal position, a rare occurrence among this group, it is neither elongate nor does it border the orbit), *P. oxycephalum*, *P. ater*, and *P. montanum* are confined to the Philippines. *P. eiselti* is known only from the type locality at Padang, Sumatra, and *P. longipes* has been collected at many localities on the islands of Borneo, Sulawesi, Nias, and Sumatra, from Singapore and elsewhere north along the Malay Peninsula, from the Riau (Riou) Archipelago, and as far north as Ban Gnara and Patani, in southern Thailand.

The presence of this new, endemic species of snake in the coastal mountains of western Panay island further bolsters our suspicion that the level of endemism on Panay is greater than previously thought. By virtue of the fact that Panay was intermittently connected to Negros and Cebu at various points during the Pleistocene (Heaney, 1986), biogeographers have justifiably expected that these islands to possess a high percentage of faunal elements in common (Leviton, 1963; Brown and Alcala, 1970). Nevertheless, recent discoveries of other vertebrates endemic to Panay (Gonzales and Kennedy, 1990, 1996; Brown et al., 1997; Ferner et al., 1997) suggest

that its high elevation montane regions warrant more intensive biodiversity survey efforts in the near future.

Key to the Species of *Pseudorabdion*

(Modified from Inger and Leviton, 1966)

- 1a. Lori-ocular (loreal) shield absent (if present, does not border orbit); prefrontal in contact with upper labials 2
- 1b. Lori-ocular shield present, very distinct, elongate, borders orbit; prefrontal not in contact with upper labials 7
- 2a. Preocular present; supraocular present; internasal not in contact with upper labials; maxillary teeth 11-12. *P. longipes*
- 2b. Preocular absent; supraocular present or absent; internasals almost always in contact with upper labial 3
- 3a. Supraocular absent; frontal borders orbit; nasal divided; maxillary teeth 10 or more 4
- 3b. Supraocular present; frontal does not border orbit; nasal undivided; maxillary teeth 10 or less 5
- 4a. Postocular absent; maxillary teeth 10. *P. ater*

- 4b. Postocular present; maxillary teeth 22-25
 *P. collaris*
- 5a. Postocular not fused to supraocular; ventrals 130;
 subcaudals 12 *P. eisehti*
- 5b. Postocular and supraocular fused; ventrals greater
 than 140; subcaudals greater than 15 6
- 6a. Each scale of outer row with dark centers and pale
 borders; ventrals uniformly dark brown except for
 extreme posterior outer edges which are pale; ocular
 shield usually fused to combined supra- and postocu-
 lars; subcaudals (M) 22-24, (F) 16- 17
 *P. oxycephalum*
- 6b. Each scale of outer row with pale centers; ventrals
 whitish with dark brown more or less confined to a
 broad median band; ocular shield not fused to com-
 bined supra- and postoculars; subcaudals (M) 28; (F)
 21-24. *P. montanum*
- 7a. Anterior chin shields not in contact with mental;
 nasal shield divided *P. sarasinorum*
- 7b. Anterior chin shields in contact with mental; nasal
 shield not divided 8
- 8a. Maxillary teeth greater than 15. 9
- 8b. Maxillary teeth fewer than 10 10
- 9a. Ventrals greater than 125; subcaudals greater than
 40 *P. albonuchalis*
- 9b. Ventrals fewer than 120; subcaudals fewer than 30
 *P. saravacensis*
- 10a. Pale nuchal collar usually present; subcaudals
 17-30; distal portion of hemipenes minutely spinose
 *P. mcnamarae*
- 10b. Pale nuchal collar absent in adults; subcaudals
 greater than 30 11
- 11a. Dorsal scales uniform pale brown, each scale
 thinly edged with pigmentless border; distal portion
 of hemipenes calyculate *P. taylori*
- 11b. Dorsal scales with pale, nearly pigmentless cen-
 ters and apical tips, anterior borders of each scale with
 brown mottling, larger areas of latero-ventral scales
 devoid of dark pigment; hemipenes minutely spinose
 *P. talonuran*

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Appendix 1. Specimens Examined

In addition to specimens listed in earlier publications by Leviton and Brown (1959) and by Inger and Leviton (1961 and 1966), the following new materials have been examined:

Pseudorabdion mcnamarae: Philippine Islands: Negros Island: Negros Occidental Prov.: CAS 185577 - Panakiyo, 22 km E Isabela, 16 April 1960 by Q. and L. Alcala. Negros Oriental Prov.: CAS 186052 - Cuernos de Negros, 3600-3800 ft., 24 December 1959 by Q. Alcala and R. Empeso.

P. talonuran: See Holotype and Paratype sections for this species.

P. albonuchalis: Malaysia: Sarawak (Fourth Division): CAS 101500 - Niah, Tangap, 6 December 1960 by T. Harrison.

P. oxycephalum Philippine Islands: Panay Island: Aklan Prov.: CAS 137643 - Nabas, Laserna Barrio, 12 May 1973 by L. Alcala. Negros Island: Negros Occidental Prov.: CAS 185453-185457 - Hinoba-an Town, barrios Alim and Asia, 9-10 June 1967 by L. Alcala and party; Barrio Asia, 30 May-3 June 1967 by A. Alcala, L. Pelingon and F. Pelingon. Negros Oriental Prov.: CAS 110974 - Camp Lookout, Valencia, 14 June 1967 by L. and F. Pelingon; 24-25 km NW of Bondo [about 10 km N of Siaton], 27-31 December 1958 by A. Alcala and party.

***Anguis melanostictus* Schneider, 1801, a Valid Species of *Barkudia* (Sauria: Scincidae) from Southeastern India**

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Abstract.— *Anguis melanostictus* Schneider, 1801, based on a watercolor in Russell (1796), from the Coromandel coast of India, is shown to be a species of *Barkudia*, nonconspecific with *B. insularis* Annandale, 1917, and is revived. *B. melanosticta*, is compared with the holotype and other specimens of *B. insularis* from Orissa State, and shown to be larger (SVL 161.0–164.9 mm, vs. 107.0–143.0 mm), in addition to differing in the following characteristics: palatal teeth present (vs. absent); anterior lobe of tongue distinctly narrowed (vs. not differentiated); and lobules around ear opening absent (vs. present). A neotype of *Barkudia melanosticta* (Schneider, 1801) is designated, based on an adult female from Visakhapatnam, Andhra Pradesh State, southeastern India (ZSI 20627).

Key words.— Sauria, Scincidae, *Anguis melanostictus*, *Barkudia insularis*, *Barkudia melanosticta*, neotype designation, Andhra Pradesh, southeastern India

Introduction

Patrick Russell (1726–1805), perhaps the first Western herpetologist in India, a medical doctor by training, was posted as naturalist by the British East India Company at Vizagapatnam (at present Visakhapatnam, Andhra Pradesh, southeastern India). Russell is best known for a two volume folio of watercolors of snakes, published in 1796 and 1801–1802 (finished between 1807–1810; see Adler, 1989; Zhao and Adler, 1993), that concentrated on the fauna of the region. Russell's books are unique in that he used local vernaculars of the species illustrated, but not their scientific or English names, and several leading herpetologists of the time have named new species on the basis of the watercolors in Russell. Accounts of the life of Patrick Russell can be found in Adler (1989) and Smith (1931). The only reptile that is not a snake described and illustrated in Russell (1796: 48; Pl. XLII), a blind worm snake (*Typhlops*)-like reptile, was named *Anguis melanostictus* by Schneider, 1801. Russell referred to the species only by the local vernacular name, Rondoo talooloo pam (an obvious corruption of 'renda talu pam', Telugu for two-headed snake), and referred the species to the genus *Anguis*. Subsequent workers (e.g., Gray, 1845; Günther, 1864) have assigned the species provisionally to the genus *Anguis*, the latter author crediting the name, in error, to Merrem (1820). The species is unlisted in the next several major works on the herpetology of the region, including Boulenger (1890) and Smith (1935).

Because the description was substantial, including details of scalations, coloration and scale counts, it is clear that the species illustrated by Russell and named as *Anguis melanostictus* by Schneider (1801) is a species of *Barkudia*, known to be endemic to the east coast of peninsular India (see Smith, 1935). Diagnostic features described by Russell (1796) matches only this genus amongst all other southern Asian species of scincids: ventrals 151; head and neck subequal; the forehead covered with "laminae of unusual shapes" (fide Günther, 1864); teeth small, numerous; eyes lateral, small; nostrils small; trunk cylindrical, of the small thickness throughout the body; body scales imbricate; each with a black dot, and eight to 10 parallel dotted lines forming a line that runs from the head to the end of the tail; length 10.5 inches; tail round, smooth, its tip blunt; tail length 4.5 inches; color reddish-brown; ventrals and subcaudals glossy white.

The genus *Barkudia* and its type species, *B. insularis*, was established on a single specimen of a legless scincid from Barkuda Island, Chilka Lake (19° 46'N; 85° 20'E), Ganjam District, Orissa State, Eastern India, by Annandale (1917). Smith (1935) provided a redescription of the species, expanding the original description based on a reexamination of the holotype at the Zoological Survey of India (ZSI). No further species of the genus has been described and Greer (1970), in his analysis of the phylogenetic relationships of scincid lizards, included the genus in the subfamily Scincinae. Although subsequent specimens have been found at the type locality (Annandale,



Figure 1. The neotype of *Barkudia melanosticta* (ZSI 20627). Bar = 20 mm.

1921; also ZSI 22540, collected from the type locality on 5 July, 1961), and from adjacent Nandan Kanan Biological Park (20° 13'N; 85° 50'E), Cuttack District, Orissa State (Biswas and Acharjyo, 1980), little is known of its biology (see Murthy, 1990a; 1990b).

Ganapati and Nayar (1952) reported *Barkudia insularis* from Waltair (17° 44'N; 83° 23'E; close to Visakhapatnam; 17° 42'N; 83° 18'E), Andhra Pradesh State, Southeastern India, at a distance of circa 300 km to the southwest of the type locality of *B. insularis*, and Ganapati and Rajyalakshmi (1955:279) noted that the type of the species was reported lost. Several subsequent publications (e.g., Murthy, 1990a; Pillai and Murthy, 1982; Sanyal, 1993; Sanyal *et al.*, 1993; Subba Rao, 1996) uncritically accepted the Waltair locality and listed the Andhra Pradesh locality for the species, although both Tikader and Sharma (1992) and Welch *et al.* (1990) omit this southern record. The recent rediscovery of the holotype of *B. insularis* by Das and Dattagupta (1997), permits an examination of this and additional material and a comparison with material from Waltair reveal that the Andhra Pradesh material is not conspecific with *B. insularis*. This paper redescribes the Southeast Indian material and designates a neotype.

Material and Methods

The following measurements were taken with dial vernier caliper (to the nearest 0.1 mm): snout-vent length (SVL; from tip of snout to vent), tail length (TL; from vent to tip of unregenerated tail), tail width (TW; measured at base of tail); head length (HL; distance between angle of jaws and snout-tip), head width (HW; measured at angle of jaws), head depth (HD; maximum height of head, from occiput to throat), body width (BW; greatest width of body), eye diameter (ED; greatest diameter of orbit), eye to nostril distance (E-N; distance between anteriormost point of eyes and nostrils), eye to snout distance (E-S; distance between anteriormost point of eyes and tip of snout), eye to ear distance (EE; distance from anterior edge of ear opening to posterior corner of eyes), inter-

narial distance (IN; distance between nares), and interorbital distance (IO; between orbits).

Comparative material of *Barkudia insularis* examined includes: ZSI 18075 (holotype of *Barkudia insularis* Annandale, 1917), Barkuda Island, Chilka Lake, Orissa, Eastern India; ZSI 22540; Barkuda Island, Chilka Lake, Orissa, Eastern India; ZSI 24086.1 and 24086.2 (Nandan Kanan Biological Park, Cuttack District, Orissa, Eastern India).

Systematic account

Barkudia melanosticta (Schneider, 1801) nov. comb. (Figs. 1-2)

Neotype. ZSI 20627 (adult female), Visakhapatnam (17° 42'N; 83° 18'E), Andhra Pradesh State, Southeastern India, 47.8 m above mean sea level, collected by P. N. Ganapati, 17 August 1954. The type locality is indicated in Fig. 3.

Other material. ZSI 25135 (adult female), collected by M. V. Subba Rao, 1984.

Diagnosis. A member of the genus *Barkudia* Annandale, 1917, *B. melanosticta* (Schneider, 1810), can be distinguished from *B. insularis* Annandale, 1917, as follows: larger size (SVL 161.0-164.9 mm, vs. 107.0-143.0 mm); palatal teeth present (vs. absent); anterior lobe of tongue distinctly narrowed (vs. not differentiated from the posterior lobe of tongue); and lobules around ear opening absent (vs. present).

Description of neotype. Adult female. Snout-vent length 164.9 mm; head elongated (HL/SVL ratio 0.05), narrow (HW/SVL ratio 0.04), depressed (HD/HL ratio 0.61), indistinct from neck; snout long (E-S/HW ratio 0.74), longer than the eye diameter (ED/E-S ratio 0.33), projecting beyond mandible; parietal eye absent; supraoculars three, supraoculars II and III largest; supraciliaries present; scales on snout and forehead smooth; rostral emarginate laterally, contacting supranasals posteriorly; rostral large, lacking rostral groove, wider than deep (rostral width = 3.0 mm; rostral depth = 1.7 mm; width/depth ratio 1.76), con-

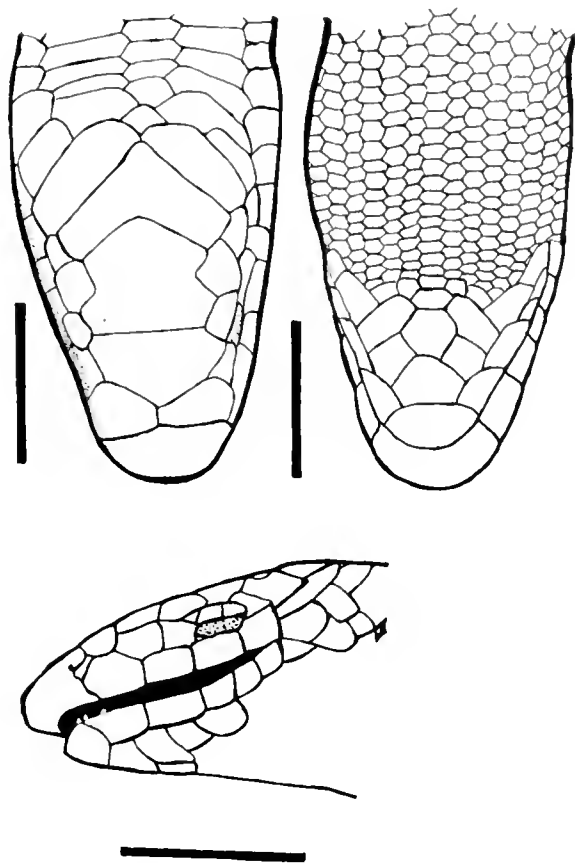


Figure 2. Head of the neotype of *Barkudia melanosticta* (ZSI 20627) in dorsal (top left), ventral (top right) and lateral (bottom) views. Bars = 5 mm, 20627). Bar = 20 mm.

tacted posteriorly by two nasals and two semicircular supranasals that are narrowly in contact. Posteroven- trally, rostral in contact with supralabial I. Nares slit- like, situated within nasals, oriented laterally; nasals in narrow contact with supralabial I. Supranasals contact supralabial I laterally and frontonasal posteriorly; frontonasal trapezoid, wider than long, contacting supranasals anteriorly and frontal posteriorly; frontal deeper than frontonasal, constricted laterally, where it contacts supraocular I; at its posterior end, frontonasal contacts a V-shaped interparietal, which is wider than frontal, a single pair of parietals contacts interparietal; a single preocular between loreal and orbit. Eye reduced (ED/HL ratio 0.18), orbit situated dorsolaterally; four supralabials (supralabial III in suborbital position), supralabial IV largest; supralabial followed by a single small scale; infralabials 4; upper eyelids undeveloped; lower eyelids scaly; two postoculars; a single anterior and two posterior temporals; ear opening minute, slitlike, measuring 0.05 mm; situated laterally at approximately the level of jaws; lobules

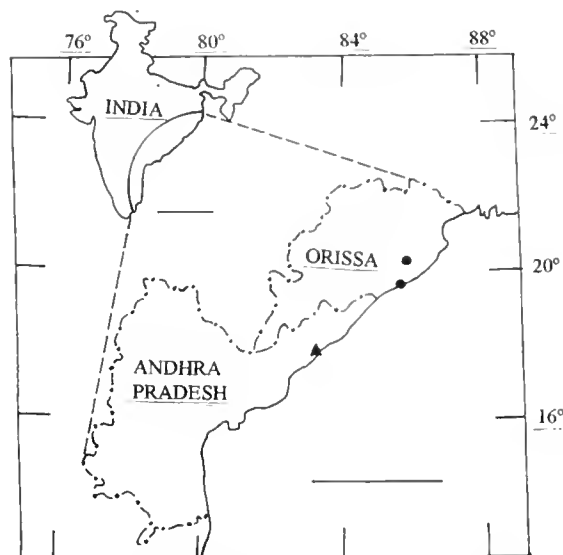


Figure 3. Map of India (marker = 800 km), showing (enlarged on right; marker = 400 km) eastern and southeastern India, and the distribution of the two species of *Barkudia*. Reference: Spots, *B. insularis* (both in Orissa State); triangle, *B. melanosticta* (in Andhra Pradesh State). See text for details.

around ear opening absent; eye-to-ear distance less than eye-to-nostril distance (E-E/E-N ratio 1.19). Inner rim of upper jaw smooth. Mental large, semicircular, wider than deep, single trapezoidal postmental, larger than mental, its width 0.29 per cent head width. Postmental contacts infralabial I, but fails to contact infralabial II, bounded posteriorly by a pair of smooth, rounded, juxtaposed chin scales that are separated by a single scale. Tongue narrowly elongate, distinctly narrowed distally, with a median cleft and scattered papillae on the dorsal surface. Palate with teeth arranged in a regular series: maxillary and mandibular teeth oriented towards the posterior, regularly arranged.

Body slender, elongate (SVL/BW ratio 0.04). Scales smooth, scale size subequal dorsally as well as ventrally. Anals three, smooth; preanal not enlarged, overlapped by the last ventral; two scales border anal laterally, exceeding its posterior level, over vent. Limbs absent. Tail short, tail length 67.5 mm, much shorter than snout-vent length (TL/SVL ratio 0.41), tail base slightly swollen and bluntly rounded at tip. Ventral surface of tail with smooth, undifferentiated subcaudals; scales on the postanal region and at the proximal part of the tail base smooth.

Coloration. (in alcohol) Dorsally yellowish-brown, turning chestnut brown towards the posterior half of tail; the tail tip (last 5 mm of tail) dark brown dorsally and ventrally, except for a pale yellow spot on the

ventrum. Ventrums of body uniformly yellow-cream. In life, these lizards are typically "glossy brown with a black spot in the middle of each scale" (Ganapati and Rajyalakshmi, 1955).

Measurements. (neotype, followed by ZSI 25135 [an adult female]; in mm) SVL 164.9 (161.0); TL 67.5- original unregenerated (32.2- partially regenerated); TW 4.7 (5.1); HL 8.7 (6.7); HW 6.6 (6.2); HD 5.3 (4.9); BW 6.8 (6.3); ED 1.6 (1.2); E-N 3.2 (3.1); E-S 4.9 (4.1); E-E 3.8 (3.3); IO 4.7 (5.0); and IN 3.3 (3.5).

Scutellation. (neotype, followed by ZSI 25135 in parentheses).- Ventrals (between postmental and preanal) 145 (143); subcaudals 78 (36+); supralabials 4 (4) (III in suborbital position in both types); infralabials 4 (4); and midbody scale rows 20 (20).

Variation. The non-type differs from the neotype in the following details: anal divided, lateral scales do not exceed level of anal; which bear fine keels and first scale following postmental contacts infralabial I. In the original description, the subcaudal count given (120) is significantly larger than that shown by the neotype- 78 (tail-tip regenerated in the non-type), but it is likely that adult males (of which no specimens have been examined) have longer tails and therefore, larger subcaudal counts. For instance, in a single male *Barkudia insularis* (ZSI 18075) examined, the subcaudal count was 108, as opposed to 82 in the only female (ZSI 24086) with an original tail.

Natural history. Several authors have provided information on the natural history of *Barkudia melanosticta*, including Ganapati and Nayar (1952), Ganapati and Rajyalakshmi (1955), Subba Rao (1996) and Subba Rao and Nageswara Rao (1998). The local protection given to the new species has precluded the collection of additional specimens.

Comparisons

The species being revived from obscurity is clearly a member of the genus *Barkudia* Annandale, 1917, due to the following features: fore and hind limbs absent, upper eyelids undeveloped, lower eyelids scaly, eyes vestigial, ear opening slitlike; nares situated in nasal, and body elongated. These features, in combination, separate members of the genus *Barkudia* from two other genera (both monotypic) of limbless scincids to which it is apparently closely related, including *Sepsophis* Beddome, 1870, containing *Sepsophis punctatus* Beddome, 1870, from the Eastern Ghats of Southeastern India and *Chalcidoseps* Boulenger, 1887, containing *Chalcidoseps thwaitesii* (Günther,

1872), from the Knuckles Range of Central Sri Lanka (see Smith, 1935, for diagnoses).

Barkudia melanosticta (Schneider, 1801) differs from *B. insularis* Annandale, 1917, in the following features: palatine teeth present (vs. absent); anterior lobe of tongue narrowed (vs. not differentiated); and lobules around ear opening absent (vs. present). The two specimens known are larger (SVL 161.0 and 164.9 mm) than the four (see Materials and Methods) examples of *B. insularis* (SVL 107.0- 143.0 mm) examined.

Barkudia melanosticta is known only from the Andhra University Campus at Visakhapatnam (northeastern Andhra Pradesh State, southeastern India), and is thus separated from the two known localities of *B. insularis* by a distance of circa 300 km to the southwest. Most of Russell's collections were presumably made in and around Visakhapatnam, ca. 5 km southeast of Waltair, the only known locality of *B. melanosticta*.

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The Dates of Publication of Amphibian and Reptile Names by Blanford and Stoliczka in the *Journal* and *Proceedings* of the Asiatic Society of Bengal

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Abstract.- The dates of publications of the *Proceedings* and the *Journal* of the Asiatic Society of Bengal are discussed. Several names of amphibians and reptiles were proposed, along with brief descriptions, by W. T. Blanford and F. Stoliczka in the monthly *Proceedings* before their intended formal description in the *Journal*, in some cases, a year before. These earlier publications constitute formal descriptions according to the Code of Zoological Nomenclature. A listing of two genera (one amphibian and one reptile) and 24 species (three amphibians and 21 reptiles) is appended; the type localities include Turkmenistan, Pakistan, India, Myanmar and Malaysia.

Key words.- Amphibians, reptiles, dates of publication, *Proceedings* and *Journal* of the Asiatic Society of Bengal.

Founded in 1784 by the Orientalist, Sir William Jones (1746-1794; see Cannon, 1960, for a biographic sketch), the Asiatic Society of Bengal, with its headquarters in Calcutta, has played, according to a report in *Nature* at the turn of the century, "...a leading part in the exploration of the natural history, philology, antiquities, and other branches of scientific inquiry connected with the East" (Anonymous, 1907). Although Jones himself was opposed to the collection of zoological specimens (Bose, 1885), examples of both plants and animals did start to arrive from various parts of the British Indian Empire, and occasionally from outside. Coupled with the expeditions organized or participated in subsequently by the staff of the Museum of the Society, the Asiatic Society of Bengal came to acquire one of the most important zoological reference collections in the world, which, after the passing of the Museum Act in 1866, came to the Indian Museum (Fermor, 1936) and is at present maintained by the Zoological Survey of India (Sewell, 1932; Das et al., 1998).

The periodicals of this two century old institution included the *Journal* and the *Proceedings*, which gradually replaced several leading oriental journals of the period, including the *Asiatick Researches* and the *Calcutta Journal of Natural History*. Because of delays in publishing the *Journal* (started in March 1832, the old series continuing until 1904, see Chaudhuri, 1956), the Society started the *Proceedings* in January 1865 (which were issued monthly till December 1904). The *Proceedings* was out "as soon as possible, after every monthly meeting", according

to the information on the cover page, as opposed to and separate from the more widely circulated *Journal*, which was published only once in two to three months (Mitra, 1885). As mentioned on an untitled page of the first issue, the separation of the *Journal* (which was issued in a "new series" between 1905 and 1934, when the *Proceedings* was reunited with the *Journal*) from the *Proceedings* was "In accordance with the announcement of the Council in the Annual Report read at the Annual General Meeting held on the 11th January, 1865" (Blanford and Heeley, 1865). Each fascicle of the *Proceedings* comprised 10-30 pages, and contained reports of the progress of the Society, including financial statements, additions of books to the library and coins to the Society's numismatic collection, exhibition notices, correspondence from its members and lists (and losses) of members, and also, "short notes, which were not deemed fit for introduction into the *Journal*" (Mitra, 1885).

Because the Society's *Proceedings* was less well known than the *Journal* and the ambiguity of descriptions in abstracts versus in "full papers", the dates of some of the descriptions of several genera and species of amphibians and reptiles from Asia have been assigned incorrectly in subsequent works (e.g., Smith, 1935; 1943) to the description published in the *Journal*, when, in fact, they were validly published earlier, in some cases, a year before, in the *Proceedings*. Some of the leading naturalists of the day read papers on faunistics, including the descriptions of new taxa, in the monthly meetings of the Society, which were reported as "abstracts" in the *Proceedings*. These

abstracts propose both new names and provide descriptions and diagnoses, thereby constituting a valid description according to the Code of Zoological Nomenclature. Since the aforementioned fascicles were generally issued monthly, and distributed to members through subscription, publication was rapid, leading to several names being available before their more complete description. In some of the cases, the titles of the papers published in the *Journal* and *Proceedings* were identical, in others, there were minor differences, such as the use of the more formal 'Reptilia' in Blanford (1879a) instead of 'reptiles' in Blanford (1879b). Regrettably, the type localities of some taxa are different in the two publications, the *Journal* tending to have a more precise type locality. In one instance, (Blanford, 1878a), the type locality ("Foot of Nawlabu hill, west of Tavoy") is different from that which appeared in the purported formal description (Blanford, 1878b: "Foot of Nawlabú Hill, east of Tavoy..."). If illustrations depicting the new taxon being described for the first time were provided, these appeared in the *Journal*. The months and, where available, dates of issue of each fascicle making available new zoological names are annotated with the references in Table 1. Only names published in the *Proceedings* that are at present attributed to the intended formal description in the *Journal* have been listed.

Names dealt with in this communication have been proposed either by William Blanford (1832-1905) of the Geological Survey of India, or Ferdinand Stoliczka (1838-1874), Secretary of the Natural History Department of the Asiatic Society of Bengal. Two genera (one amphibian and one reptile) and 24 species (three amphibians and 21 reptiles) were described as new by the aforementioned workers in the *Proceedings* before their intended formal publication in the *Journal*. Of these, one genus (an amphibian) and 16 species (three amphibians and 13 reptiles) are at present considered valid (see Table 1). The geographical coverage of the type localities includes Turkmenistan (five), Pakistan (five), India (five), Myanmar (six), and Malaysia (five).

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Table 1: List of names of herpetological taxa made available in the *Proceedings of the Asiatic Society of Bengal* before their intended formal publication in the *Journal of the Asiatic Society of Bengal*. References in the current allocation column refer to the most recent usage of the name, an important review of the taxon, the first time it was reallocated to a different genera, or inclusion in the first instance in the synonymy of an earlier name.

ORIGINAL NAME & TYPE LOCALITY	CURRENT ALLOCATION & REFERENCE	DATES OF PUBLICATION	
			<i>PROCEEDINGS JOURNAL</i>
* <i>Polypedates Hascheanus</i> Stoliczka, 1870a: 104 "Penang hill" (= Pulau Pinang, West Malaysia). "..island of Penang" in Stoliczka, 1870b	<i>Taylorana hascheana</i> (Stoliczka, 1870a) Dubois, 1992	April 1870	June 1870
* <i>Hylorana Nicobariensis</i> Stoliczka, 1870a: 104 "Nicobars" (in the Bay of Bengal, India)	<i>Rana nicobariensis</i> (Stoliczka, 1870) Dubois, 1992	April 1870	June 1870
* <i>Ansonia</i> Stoliczka, 1870: 104 (type species <i>Ansonia Penangensis</i> Stoliczka, 1870a, by monotypy)	<i>Ansonia</i> Stoliczka, 1870a Inger, 1960	April 1870	June 1870
* <i>Ansonia Penangensis</i> Stoliczka, 1870a: 104 "Penang hill" (= Pulau Pinang, West Malaysia). "Penang", including "...above Alexandra bath..." and "...halfway up the Penang hill.." in Stoliczka, 1870b	<i>Ansonia penangensis</i> Stoliczka, 1870a Berry, 1975	April 1870	June 1870
* <i>Cyrtodactylus affinis</i> Stoliczka, 1870a: 105 "Penang" (= Pulau Pinang, West Malaysia). "Penang hill" in Stoliczka, 1870c	<i>Cnemaspis affinis</i> (Stoliczka, 1870a) Dring, 1979	April 1870	September 1870
* <i>Tiliqua rugifera</i> Stoliczka, 1870a: 105 "Nicobars" (in the Bay of Bengal, India). "Camorta, (Nicobars)" in Stoliczka, 1879c	<i>Mabuia rugifera</i> (Stoliczka, 1870a) Greer, 1977	April 1870	September 1870
* <i>Mabouia Jerdoniana</i> Stoliczka, 1870a: 105 "Penang" (= Pulau Pinang, West Malaysia). "Pulo Tikos...north of Penang island" (= Pulau Tikus, West Malaysia) in Stoliczka, 1970c	<i>Enoia atrocostata</i> (Lesson, 1830) Brown, 1991	April 1870	September 1870
* <i>Riopa lineolata</i> Stoliczka, 1870a: 105 "Martaban" (= Mottama, southern Myanmar)	<i>Riopa (Lygosoma) lineolata</i> Stoliczka, 1870a See comments by Zhao and Adler. 1993	April 1870	September 1870

* <i>Ablabes Nicobariensis</i> Stoliczka, 1870a: 106 "Nicobars" (in the Bay of Bengal, India). "Nancowry haven on Camorta (Nicobars)" (in India) in Stoliczka, 1970c	<i>Liopeltis nicobariensis</i> (Stoliczka, 1870a) Wall, 1924	April 1870	September 1870
* <i>Cantoria Dayana</i> Stoliczka, 1870a: 107 "Amhers" (at the mouth of Moulmein [= Mawlamyine] River in Myanmar). ".near Amherst." in Stoliczka, 1870c	<i>Cantoria violaceus</i> Cantor, 1839 Smith, 1943	April 1870	September 1870
* <i>Trimeresurus mutabilis</i> Stoliczka, 1870a: 107 "Andamans and Nicobars" (in the Bay of Bengal, India)	<i>Trimeresurus labialis</i> (Fitzinger in Steindachner, 1867) Smith, 1943	April 1870	September 1870
* <i>Trimeresurus convictus</i> Stoliczka, 1870a: 108 "Penang" (= Pulau Pinang, West Malaysia). <i>Ovophis</i> Burger was made nomenclaturally available by Hoge and Romano-Hoge, "1978-1979" 1981: see Smith, 1989	<i>Ovophis convictus</i> (Stoliczka, 1870a) Burger was made nomenclaturally available by Hoge and Romano-Hoge, "1978-1979" 1981: see Smith, 1989	April 1870	September 1870
* <i>Stellio Stoliczkanus</i> Blanford, 1875a: 201 "..plains of Eastern Turkestan" and "..around Yárkand and Káshgar" (in southern Xinjiang Uyghur Autonomous Region, western China). "Plains of Eastern Turkestan" in Blanford, "1875"b (1876)	<i>Stellio stoliczkanus</i> Blanford, 1875 Moody, 1980	November 1875	January 1876
* <i>Gymnodactylus elongatus</i> Blanford, 1875a: 201 "..plains of Eastern Turkestan" and "..around Yárkand and Káshgar" (in southern Xinjiang Uyghur Autonomous Region, western China). "Yanghissar, Eastern Turkestan" in Blanford, "1875"b (1876)	<i>Cyrtopodion elongatus</i> (Blanford, 1875a) Kluge, 1993	November 1875	January 1876
* <i>Gymnodactylus micratis</i> Blanford, 1875a: 201 "plains of Eastern Turkestan" and "..around Yárkand and Káshgar" (in southern Xinjiang Uyghur Autonomous Region, western China). "Yanghissar, Eastern Turkestan" in Blanford, "1875"b (1876)	<i>Alsophylax przewalskii</i> Strauch, 1887 Kluge, 1993	November 1875	January 1876

* <i>Eremias Yarkandensis</i> Blanford, 1875a: 201 "..plains of Eastern Turkestan.." and "...around Yarkand and Káshgar.." (in southern Xinjiang Uygur Autonomous Region, western China). "Eastern Turkistán" in Blanford, "1875"b (1876)	<i>Eremias multiocellata</i> Günther, 1872 Nikol'skii, 1963	November 1875	January 1876
* <i>Eremias verruculata</i> Blanford, 1875a: 202 "..plains of Eastern Turkestan.." and "around Yarkand and Káshgar" (in southern Xinjiang Uygur Autonomous Region, western China). "Eastern Turkistán" in Blanford, "1875"b (1876). Restricted to "Yarkand (or Soche)" by Pope (1935)	<i>Eremias verruculata</i> Blanford, 1875a Pope, 1935	November 1875	January 1876
* <i>Pyodactylus homolepis</i> Blanford, 1875c: 232 "Khirthar range, west of Upper Sind" (= Kirthar Range, Sindh Province, Pakistan). "...near the Maki Nai in the lower portion of the Kirthar range.." in Blanford, "1875"e (1876).	<i>Pyodactylus homolepis</i> Blanford, "1875"e (1876). Kluge, 1993	December 1875	June 1876
* <i>Stenodactylus orientalis</i> Blanford, 1875c: 232 "...hills south of Rohri.." (in Sindh Province, eastern Pakistan). "South of Rohri in Upper Sind" in Blanford, "1875"e (1876)	<i>Crossobamon orientalis</i> (Blanford, 1875c) Kluge, 1993	December 1875	June 1876
* <i>Trapelus rubrigularis</i> Blanford, 1875c: 233 "Khirthar range, west of Upper Sind" (= Kirthar Range, Sindh Province, Pakistan). "...base of Khirthar hills in Western Sind" in Blanford, "1875"e (1876)	<i>Trapelus rubrigularis</i> Blanford, 1875c Moody, 1980	December 1875	June 1876
* <i>Draco major</i> Blanford, 1878a: 141 "..near Tavoy.." (in Myanmar). "forest east of Tavoy" including "from the foot of Nawlabú hill, a high ridge some eight miles east from Tavoy town" in Blanford, 1878b	<i>Draco blanfordi</i> Boulenger, 1885 Musters, 1983 and Henning, 1936. Not included in the synonymy of <i>Draco</i> spp. by Inger, 1983	June 1878	October 1878

* <i>Bronchocele burmana</i> Blanford, 1878a: 141 ".near Tavoy." (in Myanmar)	<i>Bronchocele cristatella</i> (Kuhl, 1820) Boulenger, 1885	June 1878	October 1878
* <i>Ulupe</i> Blanford, 1878a: 141 (type species <i>Ulupe davisoni</i> Blanford, 1878a, by monotypy)	<i>Dryocalamus Günther</i> , 1858 Boulenger, 1893	June 1878	October 1878
* <i>Ulupe davisoni</i> Blanford, 1878a: 141 "Foot of Nawlabu hill, west of Tavoy" (in Myanmar). ".the foot of Nawlabú Hill, east of Tavoy..at an elevation of about 1,500 feet above the sea" in Blanford, 1878b.	<i>Dryocalamus davisoni</i> (Blanford, 1878a) Boulenger, 1893 (incorrectly revised to <i>Dryocalamus davisonii</i>)	June 1878	October 1878
* <i>Ophites gammiei</i> Blanford, 1878a: 141 "British Sikkim" (= Sikkim State, eastern India), "South-eastern Sikkim" in Blanford, 1878b. Smith (1943) erroneously gave the type locality as "Cinchona plain, Darjeeling" (in West Bengal State, eastern India)	<i>Dinodon gammiei</i> (Blanford, 1878a) Wall, 1923	June 1878	October 1878
* <i>Hypsirhina maculata</i> Blanford, 1879a: 216 "Bassein in Pegu" (in Myanmar). "Pegu..in the neighbourhood of Bassein" in Blanford, 1879b.	<i>Enhydriis maculosa</i> (Blanford, 1881; replacement name for <i>Hypsirhina maculata</i> Blanford, 1879a, which is occupied by <i>Hypsirhina maculata</i> Duméril, Bibron and Duméril, 1854)	August 1879	November 1879

Size and Shape Description of Oviductal Eggs of *Draco obscurus formosus* (Squamata: Agamidae)

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Abstract.—Oviductal eggs were obtained by an abdominal dissection from a gravid *Draco obscurus formosus* which was captured near the northern limits of this species distribution during the Malaysian Heritage and Scientific Expedition to Belum, Temengor Forest Reserve, Ulu Perak, Peninsular Malaysia in 1993 and 1994. Egg size, shape, the species-specific egg bicone coefficient, clutch mass, and clutch volume are described. This is the first report that describes the egg and clutch characters of this species.

Key words.— Reptilia, Squamata, Agamidae, *Draco obscurus formosus*, Peninsular Malaysia, egg shape, bicone coefficient.



Introduction

Size and shape characteristics of eggs, as well as the ecological implications of egg parameters, have been extensively studied in birds (e.g., Preston, 1968, 1969, 1974; Paganelli et al., 1974; Hoyt, 1979; Smart, 1991) but rare in reptiles (Iverson & Ewert, 1991; Maritz and Douglas, 1994). One reason for this neglect lies in the fact that most squamate reptiles lay soft, flexible-



Figure 1. A. The adult female *Draco obscurus formosus* (ZRC.2.3693) from Sungei Halong, Temengor Rain Forest, Ulu Perak, Peninsular Malaysia. SVL = 87 mm. B. A clutch of four fully-shelled oviductal eggs of *Draco obscurus formosus*. Note the pinched projections at both ends of each egg.

shelled eggs. Length and width dimensions of such eggs change as they absorb or lose water through the soft parchment shell to produce changes in their size, shape, volume, and surface area. Consequently, soft-shelled eggs have to be measured in their oviducts or soon after they are oviposited. This is in contrast to rigid-shelled eggs of crocodylians, geckos, birds, and some chelonians which can be measured at any time after being laid.

Iverson and Ewert (1991) applied Preston's (1968) formulation to describe reptile eggs firstly. This formulation was developed to describe the shape of avian eggs and assumed that an egg was axis-symmetric and the revolved egg outline on the x-axis gives its volume of revolution. Recently, Maritz and Douglas (1994)

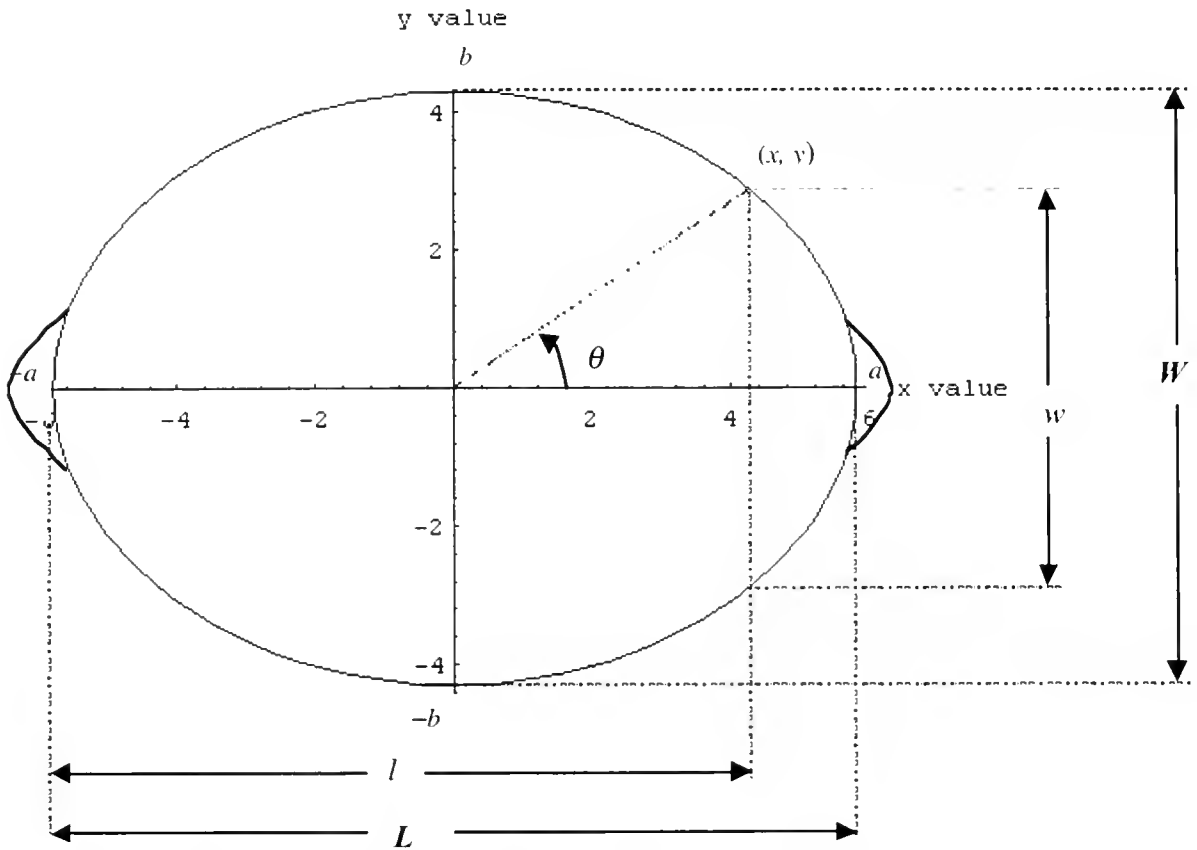


Figure 2. The coordinates and linear dimensions used to describe the shape of *Draco obscurus formosus* eggs; the outline of the egg shape, excluding the pinched ends, is generated by Mathematica software program. See text for details.

employed the Preston formulation to demonstrate the shape of reptile eggs from two linear measurements, egg length L , maximum width W , and the bicone coefficient c_2 . The bicone coefficient is a species-specific parameter which measures bluntness at both ends of an egg: c_2 is negative for eggs with pointed ends, positive for eggs with blunt ends, and zero when the shape is a perfect ellipse.

During the Malaysian Heritage and Science Expedition to the Belum Temengor Forest Reserves in 1993 and 1994, a gravid female of *Draco o. formosus*, a subspecies endemic to southern Peninsular Thailand and Peninsular Malaysia (Boulenger, 1903, 1908; Musters, 1983) was collected (Diong et al. 1995). This presented an opportunity to collect oviductal eggs for size and shape quantization for this subspecies.

Material and Methods

Material examined

One gravid female (Fig. 1a) was collected on 9 Dec 1994 from a trunk of a large tree along Sungai [river] Halong, about 2 km upstream of Camp Halong ($5^{\circ} 24' N$, $101^{\circ} 18' E$), Temengor Rain Forest Reserve, Ulu Perak, Malaysia, during the Belum Scientific Expedition (Diong et al. 1995). Specimen was measured and dissected the day after collection. Head and body shape, as well as mass of body and egg were measured to the nearest 0.01 mm and 0.01 g by digital calipers (Mitutoyo) and electronic balance (Ohaus), respectively. Voucher specimen was deposited in the Zoological Reference Collection, School of Biological Sciences, National University of Singapore.

Quantization of egg shape

Length (L) and maximum width (W) of each egg were measured to obtain elongation ($E = L/W$), bicone coefficient (c_2) and volume (V) using the method described by Maritz and Douglas (1994). The outline

of the egg was described parametrically using the Preston formulation, as follows :

$$x = \left(\frac{L}{2}\right)\cos(\theta),$$

$$y = \left(\frac{W}{2}\right)\sin\theta(1 + c_2\cos^2\theta),$$

$$0^\circ \leq \theta \leq 360^\circ$$

Mathematica (1993) software programme was used to generate the shape described by the parametric equations. A 35 mm slide of the clutch of eggs was projected on to a white cardboard mounted on a wall and three lines parallel to the y axis (c.a. 2/3 W) were drawn to the outline of each egg (Fig. 2) to obtain measurements of l, w, L, and W from the projected image; this procedure was repeated for the other half of the egg so that the bicone for each egg was an average of six estimations. The bicone coefficient was determined with the formula, $c_2 = \frac{1}{\alpha}\left(\frac{r}{\sqrt{1-\alpha}} - 1\right)$,

where $r = \frac{w}{W}$, $\alpha = \left[2\left(\frac{l}{L}\right) - 1\right]^2$, and was subsequently used to estimate egg volume, V (in cm³), as follows: $V = \frac{\pi}{6000}LW^2\left(1 + \frac{2}{5}c_2 + \frac{3}{35}c_2^2\right)$. Clutch volume was defined as the sum of individual egg volume in the clutch.

Results

Size of female lizard

Body mass 10.02 g, snout-vent length (SVL) 87 mm; tail length (TL) 167 mm; head length (HL) 16.78 mm; jaw length (JL) 16.86 mm, head width (HW) 10.59 mm; hind leg length (HLL) 270 mm; hind foot length (HFL) 170 mm; fore leg length (FLL) 270 mm; fore foot length (FFL) 9 mm.

Clutch size, egg size, and shape quantization of eggs

The clutch consisting of 4 oviductal eggs; 2 eggs in each oviduct; 2 yolked ova and 4 whitish ova in right ovary, 2 yolked ova and 5 whitish ova in left ovary; yolked ova, 5.07 - 5.15 mm in diameter, white undeveloped ova 1.50 - 1.72 mm in cross diameter. Eggs turgid, shelled with chalky-white parchment-like membrane. Egg shape ellipsoidal, ends blunt, but each with pinched cap at ends (Fig. 1b, 2). Pinched ends solid, thick, firm, flat and in same plane, less than half a semi-circle; outline of lip-like pinched structure

asymmetric. Mean \pm SD of L excluding pinched ends for 4 eggs 11.83 ± 0.53 mm (range: 10.94 - 12.30 mm); L including pinched ends 13.00 ± 0.26 mm (range: 12.14 - 13.40 mm); W 8.71 ± 0.20 mm (range: 8.40 - 8.91 mm); elongation L/W 1.36 ± 0.08 (range: 1.23 - 1.41); mean egg bicone coefficient c_2 -0.021 ± 0.008 (range: -0.032 to -0.010); egg volume 0.487 ± 0.022 cm³ (range: 0.460 - 0.510); egg mass 0.497 ± 0.022 g (range: 0.470 - 0.520 g); density 1.0205 ± 0.0009 (range: 1.0196 - 1.0217). Total clutch volume 1.86 cm³; total clutch mass 1.99 g; relative clutch mass [clutch mass divided by female (clutch plus body) mass] in percent, 19.86; relative clutch mass [clutch mass divided by female (body only, exclusive of clutch) mass] in percent, 24.78

Discussion

The present specimen has TL/SVL ratio equaled to 1.92 which is a middle size compared to Musters' (1983) report of 1.89 - 1.99 for females of this species. Musters (1983) while reporting a mean SVL of 82 ± 7 mm for four females of this lizard and a mean clutch size of 3.7 for three females of *Draco obscurus formosus*, made no reference to egg shape of this lizard. Hendrickson (1966) recorded a mean clutch size of 2 in *D. melanopogon* (n = 14) and a mean clutch size of 4 in *D. volans* (n = 20). This suggests that clutch sizes in *D. o. formosus* and *D. volans* may be similar, being larger than that in *D. melanopogon*. Among these gliding agamid lizards, clutch size is probably constrained by body size and clutch mass as a larger clutch mass is likely to reduce the horizontal displacement covered during gliding. In the present study, the total clutch mass accounted for almost 20 percent of female body mass.

The three *Draco* species, *D. obscurus formosus*, *D. melanopogon*, and *D. volans*, all have contrasting egg shapes. Causality of variation of egg shape in the genus are unknown. The eggs are distinctly pointed at both ends in *D. melanopogon* but are ellipsoidal, blunt, and smoothly rounded at both ends in *D. volans* (Hendrickson, 1966). Eggs of *D. obscurus formosus* is a nearly perfect ellipsoid as indicated by the species-specific bicone coefficient, c_2 -0.021, which is very close to $c_2 = 0$, the value for a perfect ellipse. Additionally, the eggs are much rounder, as indicated by its elongation factor of 1.36 which is smaller in eggs of *D. volans* ($E = 1.6$) and *D. melanopogon* ($E = 2.06$), as calculated from Hendrickson's (1966) L and W measurements of the eggs. The most unusual structure of *D. o. formosus* eggs is the flat projections pinched outwards from both ends. The significance of the cres-

cent-shaped pinched ends is unknown, but they may function as reserves of parchment shell materials to allow the stretching and expansion of the egg during development.

Acknowledgments

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Diel Activity of *Ranodon sibiricus* (Amphibia: Hynobiidae) in Relationship to Environment and Threats

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Abstract.—Field studies on the diel activity of *Ranodon sibiricus* were carried out in Dzhungaria, southeastern Kazakhstan, in August 1994 and June 1995. Counts showed that adults and juveniles were strongly nocturnal, which was also confirmed with the help of IR light/photocell equipment, whereas larvae were active both night and day. The daily activity period of adults and juveniles started near the 5-lux limit and peaked around 23-01 hrs. Its length followed that of the night and was accordingly shorter in June than in August. When the activity of aquatic adults and juveniles started in the early night, about 5% came out of the water and walked on land alongside the brook. Most ranodons, however, were relatively stationary sit-and-wait predators in the water. The nocturnal activity of *R. sibiricus* protects it against dehydration (terrestrial animals), strong UV radiation and diurnal predators. The most important task of the nocturnally and diurnally active larvae is to grow as fast as possible to survive the first, hazardous hibernation. The difference in diel activity between adults/juveniles and larvae reduces intraspecific competition and cannibalism within the population. Because of their secretive life, metamorphosed stages of *R. sibiricus* are not easily accessible to predators or Man. Hence, the most important threat is probably the large herds of cattle grazing in the area. Since adults and juveniles are well hidden by day, when the cattle wade the brooks, they are fairly safe, but cattle may pose a great threat to ranodon eggs and larvae.

Key words.—*Ranodon sibiricus*, Kazakhstan, diel activity, rare species, environmental adaptation, threats.

Introduction

A threatened, secretive species

The salamander *Ranodon sibiricus* Kessler, 1866 is distributed in the Dzhungarian mountains of southeastern Kazakhstan and the extreme west of Xinjiang province in China. Its range is very small and has undergone many man-made changes during the past. The species is therefore threatened and is on the red list of the former Soviet Union/Kazakhstan (Bannikov et al. 1978; Zhao and Adler 1993). Its habitat is cold, clear mountain brooks. Because of their quite secretive way of life, individuals are not always easy to find even where they may be locally abundant. Although Shnitnikov (1913) found it easily, Brushko and Narbaeva (1988) emphasized the difficulties of finding *R. sibiricus* in the mountain rivers. Its secretive life is also reflected in its nocturnal activity, an aspect briefly touched upon by several authors (Shnitnikov 1913; Paraskiv 1953; Kubykin 1986; Narbaeva and Brushko 1986; Wang 1990).

As part of a wider ecological study of *Ranodon sibiricus* in Dzhungaria, we investigated its activity, partly in 1993, but more thoroughly in 1994 and 1995, to learn the details of its diel activity rhythm and pos-

sible adaptations to the environment. Several other questions were also of interest. For instance, do the larvae behave differently from the metamorphosed animals to reduce intraspecific competition and cannibalism (as in *Triturus*; see Dolmen 1988)? Does the nocturnality of the species really make it almost safe from terrestrial predators and Man? To what extent can *R. sibiricus* avoid the heavy impact of some 150 cows which graze along and trample in the brooks when crossing the study area twice a day in summer (Kubykin et al. 1995; Dolmen et al. 1997)?

A preliminary report on these studies was presented by Dolmen et al. (1995). Hereafter, *Ranodon sibiricus* is mostly referred to as ranodon.

Principles of measuring activity

In principle, there are at least three ways of measuring the activity of an animal: 1) with an actograph or other technical device such as light beam/photocell equipment, 2) by interval trapping of active animals, and 3) by counting active animals. All three methods have been successfully used for *Triturus* species (Salamandridae) in Europe (e.g. Himstedt 1971; Dolmen 1983a,b; Griffiths 1985).

Preliminary studies

In June 1993, the trapping technique was tried out for ranodons in a small brook in the Borokhudzir valley in Dzhungaria. On 15 June, 10 simple funnel traps (1 L "Cola traps") and two more complicated traps with eight entrances each (constructed by Arne Haug) were placed in a 15 m long stretch of a small brooklet (Brook 3) in which six ranodons and 50 double clutches of eggs had been registered two days earlier. Alternate traps had their entrances pointing downstream and upstream. The traps were checked/emptied every third hour throughout a day and night, but no ranodons were caught. The water temperature varied between 3.8 °C and 7.6 °C. On 17 June, a similar set-up was tried on a 15 m stretch of another, somewhat larger brook (Brook 1) where, in addition to the ranodons living there naturally, 38 additional ones were released. A few were caught during the next day and night, but never more than two each time.

However, a study of a third brook (Brook 5) at night revealed that many ranodons were going onto land and wandering on the dew-wet banks beside the brook. On a 500 m stretch at 23.00-24.00 hrs. (standard time), four men with torches counted 14 ranodons that were active on wet land and 11 hidden under stones alongside the brook. Although aquatic individuals were also active, the main movements up and down the brook seemed to be taking place on land, not in the water. The trapping technique in water was accordingly unsuitable for measuring the activity of ranodons.

For 1994 and 1995, it was therefore decided to use the counting method for measuring the activity of ranodons, and for comparison, data from the use of light beam/photocell equipment could serve as a control.

Description of the area

The study area, on the southern slopes of the River Borokhudzir catchment in Dzhungaria at an altitude of approximately 2200 m, consists of grazing for cattle, bog, and juniper-covered hillsides. Two small tributary brooks to the River Borokhudzir (Brook 1 and Brook 5), approximately 200-300 m apart, were chosen for the study. Both were spring-fed, about 2-50 cm deep and 30-80 cm wide, and had gravelly or stony bottoms. In a few places, turf almost overgrew and covered the water. Boulders, where terrestrial ranodons hid, could also be found on land alongside the brooks. The velocity of the water at different sites along the two brooks varied from 0.0-0.2 and 0.0-1.0 m/s, respectively. Measurements were made in August 1994 when the flow was low.

The weather in both periods was mostly clear. The air temperatures in August 1994 and June 1995 varied between 5.7 °C and 17.1 °C, and 3.5 °C and 21.0 °C, respectively. The air humidity in August 1994 varied from less than 50 to 100% RH, with maximum humidity at night. The water temperature at the site investigated in Brook 1 varied in August 1994 between 5.5 °C in the early morning and 19.3 °C in the afternoon, and in June 1995 between 5.0 °C and 20.0 °C. In Brook 5, the water temperature varied between 6.2 °C and 14.8 °C in August 1994 and 4.8 °C and 15.6 °C in June 1995. A more detailed description of the area is given by Dolmen et al. (1997).

Material and Methods

Field investigations

The study took place in August 1994 and June 1995. Two persons equipped with good torches examined a total of 90 m and 190 m, respectively, along the two brooks (Brook 1 and Brook 5) every second hour for three days and nights. The walking speed was approximately 8 m/min. Deep pools and shallow slowly-flowing water were especially well examined, and also the nearest 2-3 m on land alongside the brooks. No stones were turned, however, and no ranodons were disturbed, except possibly by the torchlight. The two brooks had been divided into sections of 5 or 10 m, respectively, for statistical use, and the number of ranodons in each section were counted separately. The numbers in different main developmental stages were also kept apart: 0+ (young larvae, less than one year old), 1+ (old larvae and metamorphosed individuals from the previous year), larger juveniles, and adults.

Aquarium investigations

In December 1994, additional investigations of the activity of two ranodon individuals, an adult and a large juvenile, were carried out in Almaty. An experimental aquarium was made of transparent plexiglass shaped like a circular channel. The outer diameter was 36.5 cm, the inner diameter 15 cm and the height 10 cm. The aquarium was filled half full of water brought from Dzhungaria and some large stones acted as shelter for the animals and to enable them to climb up into the air. The aquarium was equipped with an infra-red light beam/photocell system (Visolux LS 4-GaAs). The beam was set to a diameter of about 2 mm and focused near the bottom of the aquarium, where the ranodons usually walk around. Interruptions of the beam were recorded by an electric counter (Visolux LU GaAs), which was read every second hour throughout eight days and nights. Food was not pro-

vided during the experiment. The aquarium was placed inside a building, close to a north-facing window. The light regime was therefore natural, i.e. approximately like outside. The temperature was constant and slightly lower than the usual room temperature.

The same equipment, connected to a 12 V car battery, was used for an adult and a large juvenile in Dzhungaria in June 1995. The aquarium was protected from disturbance and strong sunshine by being placed halfway under a car and partly covered with a blanket. The water was replenished (in daytime) once during the experiment, which ran for barely three days and nights. On the second evening, the onset of activity was studied in more detail at 5-minute intervals for two hours.

Environmental factors

Light intensities between 0 and 10 lux normally act as Zeitgeber for activity in plants and animals (cf. Dolmen 1983b). The 5-lux value is therefore often used in activity studies as a limit between day and night. In our study it was determined subjectively, evening and morning. The 5-lux value in Dzhungaria occurs roughly 10-15 minutes before (or after) the 0.1-0.5 lux values, which corresponds well with the light intensity required for reading a newspaper, for instance. This was confirmed with a Hartmann and Braun ECLX 4 luxmeter.

The times for reading the Visolux counter were set approximately symmetrically around the darkest time of night (ca. 24.00-00.30). All hours mentioned hereafter refer to standard time, not summer time. An important difference in the understanding of diagrams based on the two methods used for measuring activity here is that a) the counting method measures the activity at the time of counting, whereas b) the IR beam/photocell method measures the activity during the two hours preceding the time when the counter is read.

Statistics

Wilcoxon's matched-pairs signed-ranks test (two-tailed) was used to test any significance levels in the activity measured in the field, the number of ranodons within the different brook sections being counted for an hour and comparison being made with the situation two hours later.

The same test was used for the aquarium study, but it was based on the ranodon counts for all the days within a certain time interval combined, and these were compared with the corresponding counts in the next time interval.

The chi-square test was used to find out whether or not the activity patterns had a significant diurnal (06-18 hrs.) or nocturnal (18-06 hrs.) predominance, and the chi-square test with two variables without expected values was used to test any differences in activity pattern between, for example, adults and larvae.

Results

The field activity studies

The activity of *Ranodon sibiricus* was definitely nocturnal, but with some modifications with respect to the developmental stage and the month of the year. The animals in all stages were amazingly clever at hiding during parts of the day and night. Several were seen at night on a gravelly bottom along stretches of the brook which we had searched very thoroughly in daytime without finding any.

In August 1994, the diel activity showed basically the same patterns in both brooks, although the peak apparently varied slightly from day to day between 21 and 01 hrs. (Fig. 1).

Adults and juveniles had similar patterns, and seen together the rise in activity level from 19 to 21 hrs. was always statistically significant ($P < 0.002$; Wilcoxon's signed-ranks test, see Fig. 1). Likewise, the decrease in activity from 03 to 05 hrs. was always significant in Brook 5, where most animals were registered ($P < 0.002$). The variation in the time of peak activity from day to day was usually not significant.

In Brook 1, the larvae showed a similar rise in activity from 19 to 21 hrs. When 0+ and 1+ larvae are counted together, the increase is significant ($P < 0.002$ or $P < 0.02$) on two of three dates. The activity of 0+ larvae in Brook 5 was not as well defined as in Brook 1, being much more diurnal. In contrast to adults and juveniles, and also to the larvae of the other brook, which were all mainly nocturnal ($P < 0.001$; chi-square test), the larvae of Brook 5 showed a much higher degree of diurnality (nocturnality tested: $0.20 < P < 0.30$). Indeed, traces of diurnal activity can also be seen in the 0+ larvae of Brook 1.

The period of activity of this species fitted very well with the light and dark periods, i.e. activity was connected with the part of the 24-hour period when the light intensity was below 5 lux. This was approximately 20.00-05.30 hrs. at that time of year. Except for young larvae, any activity outside that period was negligible.

A pattern similar to that of August 1994 was revealed in June 1995. The period of darkness (< 5

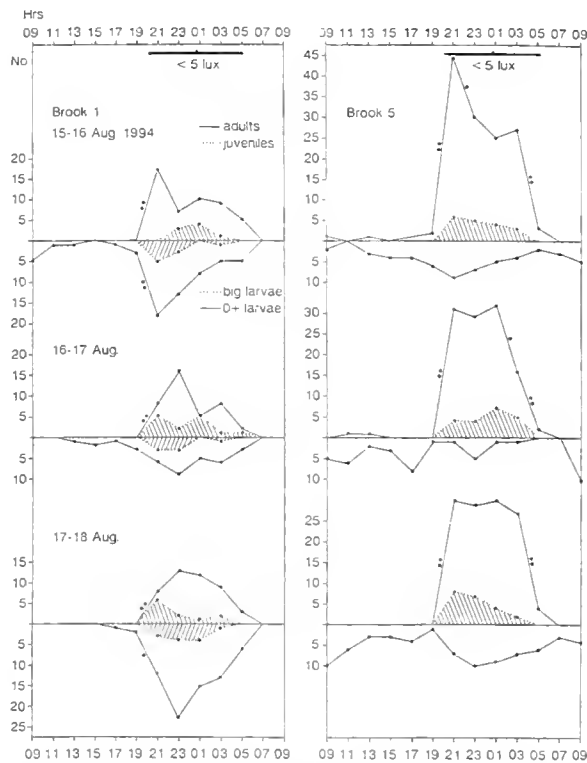


Figure 1. Diel activity of *Ranodon sibiricus*, based on counts in Brooks 1 and 5 in August 1994. Levels of statistical significance: * $P < 0.05$ or $P < 0.02$; ** $P < 0.01$ or $P < 0.002$.

lux) in June was shorter than in August, i.e. about 21.00-04.30 hrs., and the activity peaks of adults and large juveniles were narrower, at 23 (21) or 01 hrs. (Fig. 2).

The youngest stage (1+) showed activity peaks varying from 21 to 03 hrs. and also some diurnal activity. At least three times, larvae (one each time) were even registered directly exposed to sunshine, at 11, 13 and 17 hrs. Nevertheless, on the whole, the larvae definitely had a nocturnal pattern.

The activity period of the species again fitted very well with the (shorter) dark period. The rise in activity from 19 to 21 hrs. was always significant ($P < 0.002$ or 0.02) in Brook 5, both for adults/juveniles and 1+ larvae, as was the decline from 03 to 05 hrs ($P < 0.002$, 0.01 , 0.02 or 0.05), and usually even 01-03 hrs. for adults and juveniles in both brooks ($P < 0.002$, 0.02 or 0.05). The day-to-day variation in peak activity seen at midnight was not significant.

The aquarium studies

In the aquarium investigations in December 1994, the ranodons also showed a definite nocturnal activity, with peak activity at 20-22, 22-00 and/or 00-02 hrs. A cumulative curve for the whole period of investigation

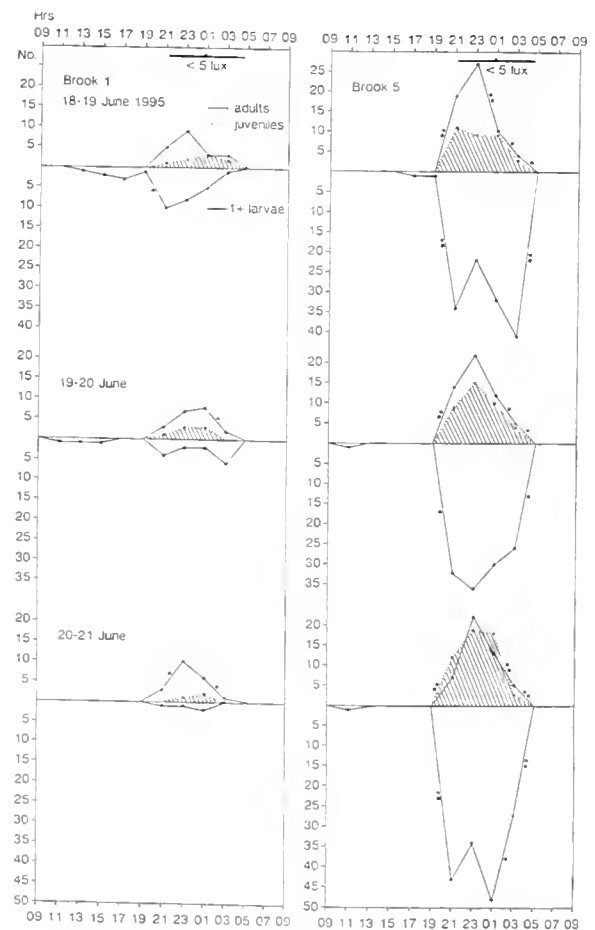


Figure 2. Diel activity of *Ranodon sibiricus* in Brooks 1 and 5 in June 1995. For explanation, see Fig. 1.

had a main peak at 02 hrs. (i.e. peak activity at 00-02 hrs.) and a minor peak at 22 hrs. (i.e. 20-22 hrs.), with little or no activity during the light hours. The rise in activity from 16-18 to 18-20 hrs. and from 22-00 to 00-02 hrs. was significant ($P < 0.05$), as was the decrease from 00-02 to 04-06 hrs. (two time intervals seen together) (Fig. 3).

The activity period of the animals at this time of year was clearly longer than the activity measured in summer, as was the period of darkness (< 5 lux), which lasted from approximately 17.30 to 08.00 hrs.

The aquarium study in June 1995 confirmed the results of the field investigations, although there were minor deviations, probably for methodological reasons. Activity was negligible until 21 hrs. when a rise in activity occurred resulting in a peak at 21-23, 23-01 or 01-03 hrs. (Fig. 4).

In the more detailed aquarium investigations on 20 June, the activity started about 15 minutes before the 5-lux limit was reached in the open. However, since the aquarium was partially covered (see Methods), the

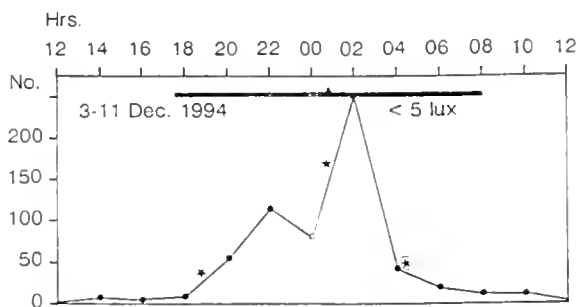


Figure 3. Diel activity of two *Ranodon sibiricus* specimens kept in an aquarium under natural lighting conditions for eight days and nights in December 1994, based on the IR light/photocell method. Level of statistical significance: * $P < 0.05$.

light intensity there was a bit lower and the activity probably started at a light value very close to 5 lux. It increased gradually over the next hour (Fig. 5).

Observations of aquatic and terrestrial ranodons

By far the majority of active ranodons were found in the water, not on land. The frequencies of finds made on land relative to the total number of ranodon counts were only 6.1% ($N=425$) and 3.6% ($N=279$) for August 1994 and June 1995, respectively. The number of ranodons counted on land alongside the brooks was thus relatively low, but sometimes increased when the number counted in the water increased. Animals walking on the moist grassy ground beside the brooks had a wet skin and were considered not to be really terrestrial, but aquatic animals which had only left the water temporarily for some reason.

Not all ranodons were active to the same degree at the same time, not even near midnight. In June 1993, when we counted ranodons on land at 23.00-24.00 hrs., i.e. when the activity was at its highest, 14 were seen walking along the bank, but another 11 really terrestrial specimens (with a dry skin) were still passive and very sluggish, hiding under stones.

Discussion

The activity pattern

Ranodon sibiricus proved to be highly nocturnal, with very little, if any, activity during the day. An exception was the young larvae (0+) in August, in part also the one-year old larvae (1+) in June, which showed a certain amount of diurnal as well as nocturnal activity. The 5-min. reading in the aquarium study showed in detail that the ranodons gradually started their activity near the 5-lux limit in the evening. When that limit

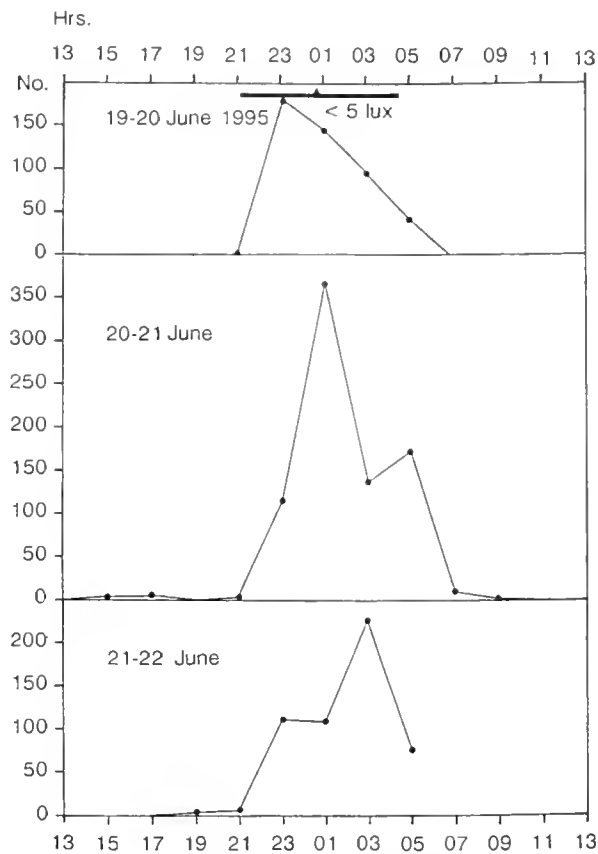


Figure 4. Diel activity of two *Ranodon sibiricus* specimens kept in an aquarium under natural lighting conditions for nearly three days and nights in June 1995, based on the IR light/photocell method.

was again reached in the morning, the activity had decreased to approximately zero. This means that even though the species is clearly nocturnal, it uses all the time it can get for activity, i.e. mainly hunting.

Most earlier authors have noted the extreme nocturnal habits of the ranodons and the more diurnal habits of the larvae (Shnitnikov 1913; Kubykin 1986; Narbaeva and Brushko 1986). Metamorphosed specimens, both in water and on land, hide under stones during the day, but come out at night and often move upstream or downstream for some distance. Wang (1990) stated that in western Xinjiang, China, ranodons also hide among plants and under stones during the day and is active between 24 and 06 hrs. The Xinjiang province, like the rest of China, follows Beijing time, which is three hours earlier than sun time in western Xinjiang. The true activity time of ranodons in western Xinjiang should therefore be around 21-03 hrs., i.e. similar to that found in our investigations in eastern Kazakhstan.

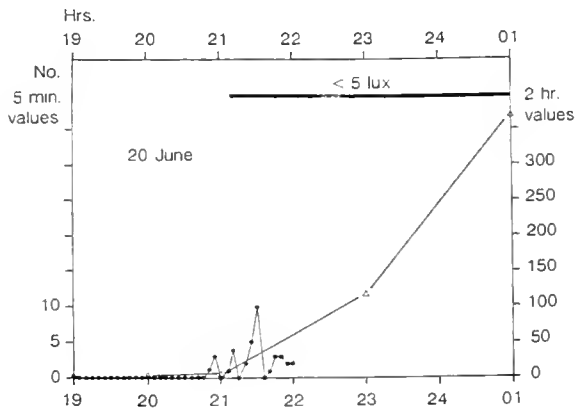


Figure 5. Details of the onset of activity of two *Ranodon sibiricus* specimens kept in an aquarium under natural lighting conditions on 20 June 1995, based on the IR light/photocell method. Activity values are shown as counts per 5 minutes (dots) and per 2 hours (triangles). The activity started near the 5-lux limit of light intensity (see text for further explanation).

We had no opportunity to investigate newly-hatched larvae. In 1995, in the most favourable brook (Brook 1), the first larvae hatched during the second half of June. According to Paraskiv (1953), newly-hatched larvae remain in calm, shallow water well illuminated by the sun, where they hide among stones on the bottom. However, they often come out and lie on the stones, and are active by day, not avoiding the sunshine (cf. Kubykin 1986). We found that at least part of this diurnal activity lasted throughout the summer - although the main activity was nocturnal in August - and even to some extent in the next year (June). A well-defined phase shift from diurnal to nocturnal activity, as described, for example, for *Triturus cristatus* larvae (see Dolmen 1983a,b), probably does not take place. Paraskiv (1953) mentions that older larvae, at the time of their metamorphosis when their gills are becoming resorbed and they are going onto land, avoid the sunshine and keep to shady places.

Water phase and land life

The metamorphosed ranodons stay in water or on land, or alternate more or less regularly between these two media. In daytime, but also sometimes at night, we found terrestrial individuals in damp hollows under stones and the like within a few metres of the water. We made no attempt to specifically measure the activity of such animals. The activity of ranodons in the brooks increased at night, however, and some also went up on to land. We are therefore dealing with two categories of ranodons on land: true terrestrial animals, which have a dry skin and which stay on land

for a day or more, and aquatic ranodons, which have a wet skin and which only temporarily come out of the water (Dolmen et al. 1997).

Shnitnikov (1913) and Paraskiv (1953) also noted that juvenile and adult ranodons leave the water or their terrestrial hiding places at twilight and in darkness, and are then active and are found on the banks of brooks, where they often move long distances (cf. Kubykin 1986; Narbaeva and Brushko 1986).

However, as these authors pointed out, not all ranodons leave the water at night, at least not at the same time. The majority, in fact, stay in the water and may only occasionally come up. We found that when the weather had been dry for many days (August 1994 and June 1995), relatively fewer ranodons were found on land, walking around at night or hiding under stones, than when the weather was wet (June 1993).

Environmental adaptations

The nocturnal/crepuscular activity pattern in amphibians is probably ultimately an adaptation to the night and day temperature and air humidity cycles. The vulnerability of the animals to dry conditions makes it necessary for them to hide during the driest and hottest hours, and even when, like the ranodons, they stay in water, this pattern is maintained. Most urodeles are thus primarily nocturnal, although their activity pattern may vary ontogenetically (e.g. Noble 1954; McDiarmid 1994). The activity is proximately controlled by the light/dark cycle. Different temperature regimes, even a rise in temperature of as much as 10-15 °C, seemed to have little, if any, influence on the activity pattern of *Ranodon sibiricus*. Relative humidity, or at least the moisture on the ground, could be important for whether or not amphibians will walk on land, however. All records of ranodons walking on land were made when the air humidity was above 80% RH, always between 21.00 and 03.00 hrs.

Another factor which could represent a threat to *Ranodon sibiricus*, living at high elevations (ca. 1700-2700 m), is the strong UV radiation. Except for a very few larvae of the youngest stages (0+ in August and 1+ in June), no animals were seen voluntarily exposing themselves directly to the sun. For cold-stenothermic animals like *R. sibiricus*, it is probably even more important to be nocturnally active than it is for most other amphibians. High temperatures, i.e. more than 22 °C, are reported to be lethal, at least to ranodon larvae (Brushko and Narbaeva 1988). Temperatures were as low as 6.0-11.5 °C in the air and 4.8-11.0 °C in the water during the peak aquatic and land-walking activity of ranodons at night. Maximum air temperatures during the day were 17.1-21 °C, and water tempera-

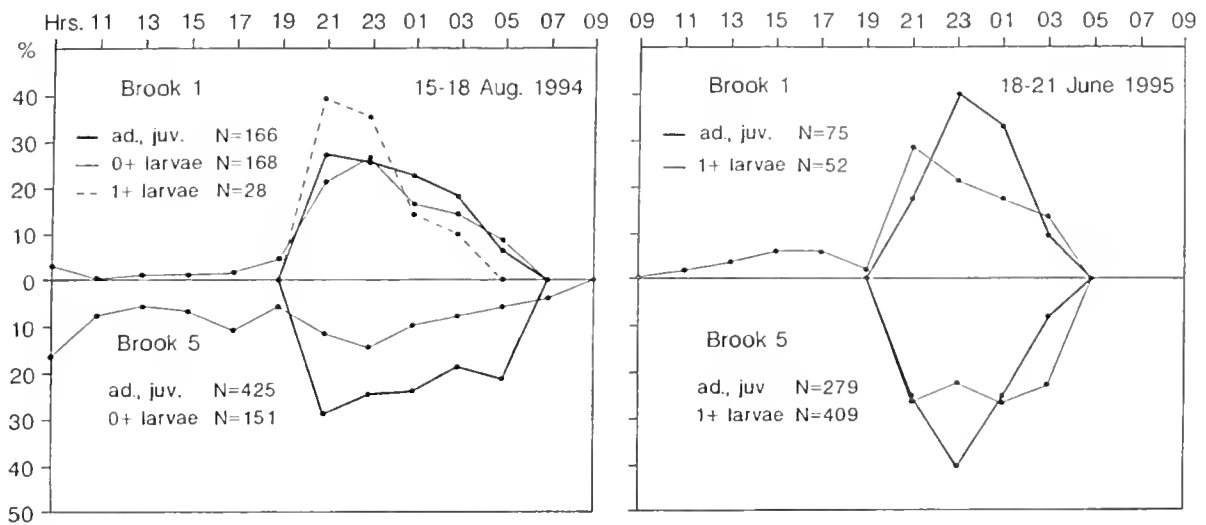


Figure 6. Relative degree of activity (%) of *Ranodon sibiricus* adults/juveniles and larvae in Brooks 1 and 5 (cf. Figs. 1-2). Young larvae (0+, and in part 1+) exhibit a markedly greater degree of diurnality than older life stages.

tures reached 19.3-20.0 °C. A few larvae were counted during the warmest periods, too.

We made no specific study of the possible influence of moonlight on the activity of ranodons. However, when we compared our data on activity with notes on the weather, etc. we found no trend of, for instance, decreasing activity when the half moon (at the most) was sometimes shining. The light regime under such circumstances was also always well below the 5-lux limit.

Ranodon sibiricus is clearly more nocturnal than, for instance, the *Triturus* species studied by Dolmen (1983a,b). It can also be seen that the activity period of *R. sibiricus* in June was shorter than in August, which is in conformity with the length of the night. A similar phenomenon was shown for the more crepuscular *Triturus vulgaris* and *T. cristatus* in Norway (Dolmen 1983a,b). Moreover, although normally two-peaked, in northern latitudes at midsummer the crepuscular peaks of the *Triturus* species fused, and the activity curve thus revealed only one, long midnight peak when real nights (<5 lux) disappeared. Especially *T. vulgaris*, being the more crepuscular species, takes advantage of these long periods of twilight in northern latitudes in that its hunting day thereby becomes longer and its growth better.

The activity period of the strictly nocturnal *Ranodon sibiricus*, however, becomes shorter at midsummer. It is advantageous for an animal to have the opportunity to seek food for as many hours as possible; the ranodons in fact spends the whole night. Its activity is limited, however, by the varying light regime through the season, i.e. the period per day and

night when light intensity is below 5 lux, which again is relatively short at midsummer. The need for protection against climatic factors, as described above, and possibly from (diurnal) predators thus seems to be greater than the need for extra growth in metamorphosed ranodons.

However, a marked difference could be seen in the diel activity of metamorphosed ranodons and larvae, especially 0+ larvae aged 1-2 months (Fig. 6). The youngest stages seem to be more or less active both day and night. Still younger larvae than those we have studied are possibly even more diurnally active (cf. Paraskiv 1953). A probable explanation of the prolonged activity of the larvae, compared to older stages, is their need to eat and grow as much as possible during the short frost-free season in the mountains. Being large may perhaps increase their chances of surviving their first, most hazardous, winter. Growing is therefore probably their most important task. A larger size also makes their migration up or down the brook to hibernation sites in late autumn easier and protects them from predatory fish (see below).

By staying during the summer in the lower, warmer part of small brooks, in shallow water, and alternating between shade and sunshine (Paraskiv 1953; Dolmen et al. 1997), the metabolism of the larvae is raised and kept at a near optimal level. There are, however, certain limits for a cold-stenothermic animal like *Ranodon sibiricus*. Small larvae are sensitive to warm weather, and 22 °C is said to be lethal (Brushko and Narbaeva 1988).

Such a difference in diel activity and microhabitat between adult urodeles and their larvae is not uncom-

mon (McDiarmid 1994). Dolmen (1983a,b; 1988) interpreted it as a means to reduce intraspecific competition between different developmental stages and as protection against cannibalism, which may be common in salamanders and which is also known to occur in *Ranodon sibiricus* (Kubykin 1986; Wang 1990; Kuzmin 1991a). In species whose adults and larvae live sympatrically, a behavioural mechanism which favours the avoidance of cannibalism could presumably be of adaptive value.

Hunting strategy

During our preliminary studies in June 1993, the method we used for trapping ranodons in water failed and we concluded that most translocations of *Ranodon sibiricus*, at least at that particular time of year, probably took place on land. However, only a small proportion of our many records of active ranodons in 1994 and 1995 were made on land. We therefore conclude that most of their activity, like hunting (not longer translocations), is nevertheless aquatic. Hence, ranodons are probably largely sit-and-wait hunters, as maintained by Paraskiv (1953), too; they do not move about much in the water. This contrasts with the European *Triturus* species (see Dolmen 1983a,b), which live in the stagnant water of ponds and small lakes. For a running-water species like *R. sibiricus*, it may be more economic to sit and wait for drifting prey than to spend energy looking for them.

Our preliminary results on food items showed that *Ranodon sibiricus* is an opportunist when it comes to choice of prey, and thus supports the results of stomach analyses made by Kuzmin (1991b). There was also a tendency for ranodon stomachs to be at their fullest after midnight. Based on the undigested stomach contents of a few specimens caught in daytime, Shnitnikov (1913) suggested that the ranodons feeds by day. However, apparently no ranodons caught by him at night were examined.

Diel activity and predators

During the day, metamorphosed ranodons cleverly hide under boulders and in earth cavities in the brook bed and are often difficult to catch. Because of their strict nocturnality and their ability to hide in daytime, night searches with good torches are the best way of looking for ranodons in a brook. Searches in daytime may be strenuous, and often negative if the population density is not large. At night, however, the animals are easy to see in small and medium-sized brooks.

It can be expected that the activity cycles of a predator and its prey will coincide. Wild boars *Sus scrofa* and grey herons

Ardea cinerea are said to be natural predators of ranodons (Brushko and Narbaeva 1988), likewise black storks *Ciconia nigra* (Shnitnikov 1913). We think that some of the many species of birds of prey which occur in great numbers in the mountains (e.g. buzzards *Buteo* spp.) will also occasionally take ranodons if they had the opportunity. However, of these terrestrial predators, only the wild boar is nocturnal. Adults and large juvenile ranodons therefore seem to have few, if any, really important predators. However, two kinds of fish (*Diptychus maculatus* and *D. dybowskii* (Cypriniformes)) are present in the River Borokhudzir, at least in the main river. Fish, which are usually nocturnal or crepuscular, may therefore be potential predators of ranodon larvae. However, since these fish are absent, or extremely rare, in the small, spring-fed brooks in which the ranodons breeds, they are hardly a threat to the ranodons there.

The influence of cattle

The negative influence of cattle on *Ranodon sibiricus* has been mentioned by several authors (Kubykin 1986; Narbaeva and Brushko 1986; Brushko and Narbaeva 1988; Wang 1990; Dolmen et al. 1997). Damage is mechanical when the cattle trample in the brooks, crushing ranodons and destroying eggs. Their excrements may also pose a threat to eggs and small larvae.

In 1993, up to about 150 cows, watched by shepherds on horseback, twice daily crossed the brooks of the study area. There were also flocks of horses, sheep and goats. Corresponding numbers of domestic animals were seen in 1994 and 1995. Most crossing of brooks took place in daylight at about 8 hrs. and 20 hrs., and at that time the adult and juvenile ranodons were still relatively safe in their hiding places. Nevertheless, we twice found dead ranodons which had been crushed under the feet of cows. On other occasions, we came across mechanically damaged eggs, presumably the victims of wading cattle. Larvae, too, may suffer from cattle. However, we believe that the damage to metamorphosed ranodons is not really large (cf. Kubykin 1986).

Conclusions

Ranodon sibiricus is a nocturnal species in the juvenile and adult stages, whereas the activity of young larvae is more evenly distributed through the day and night. The difference in diel activity between larvae and metamorphosed stages probably reduces intraspecific competition and cannibalism. In daytime, the ranodons hide cleverly in the brook bed or on land. This secretive life makes them difficult to find, both

for diurnal predators and people. At night, however, the ranodons are easy to see. Their nocturnality also makes adult and juvenile ranodons less vulnerable to mechanical damage from grazing cattle, but eggs and larvae may easily be harmed.

Acknowledgments

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The Hemipenes of Chinese Species of *Deinagkistrodon* and *Gloydius* (Serpentes: Crotalinae)

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Abstract.— The hemipenes of eight species of two genera, *Gloydius* and *Deinagkistrodon* are described in detail and illustrated. The results indicate that (1) the structures of hemipenes of *Gloydius* markedly different from that of *Deinagkistrodon*, (2) more similarities are present among species of *Gloydius*. At the end of the paper, the taxonomic relationships among species revealed by hemipenial structures are discussed.

Key words.— Serpentes, Crotalinae, *Deinagkistrodon*, *Gloydius*, Hemipenes.

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Introduction

Since Cope has firstly implied the characters of reptilian hemipenes to the classification in 1893, their important value in the classification and phylogeny was realized by more and more researchers around world (Pope, 1935; Smith, 1943; Dowling et al., 1960; Zhang et al., 1984; Zhang, 1986; Branch, 1988; Mao, 1989; Gloyd et al., 1990; Malnate, 1990). Although some of hemipenes of Chinese *Deinagkistrodon* and *Gloydius* were described (Pope, 1935; Smith, 1943; Mao, 1989; Gloyd et al., 1990; Malnate, 1990) previously, the authors didn't illustrate them except *Gloydius brevicaudus* and *Deinagkistrodon acutus*.

Prior to this paper, a comparative anatomy on skull of these two genera was done (in press.), we confirm that there are eight valid species of *Deinagkistrodon* and *Gloydius* in China. At the present work, the feathers of the hemipenes of these eight species will be described in detail, and the taxonomic relationships indicated by their structures will also be discussed, they will provide additional and further data for the classification and systematic of these species.

Material and Methods

The materials used in this study are based up formalin-preserved adult specimens; all of them are deposited in Chengdu Institute of Biology, the Chinese Academy of Sciences. The number and locality of examined and figured specimens are listed in Table 1.

In this paper, the descriptive terminology generally followed literature (Dowling et al., 1960; Zhang et al., 1984; Branch, 1986). In a majority of cases, the fully everted hemipenes were illustrated. In a few cases, the partly everted hemipenes were everted arti-

cially. The drawings were done under the binocular-dissecting microscope.

Results

***Gloydius ussuriensis* (Emelianov).** The divided hemipenis (Fig. 1) extends to eighth subcaudal plate and is forked opposite the third. While the Russia (Kebrovka, Primorskiy Kray) specimens described by Gloyd et al. (1990) and Malnate (1990) extend to the eleventh subcaudal and are forked for eight subcaudals. It is spinous proximally, calyculate distally. The transition from spines to calyces is inconspicuous. The calyculate area is about equal to that of spinous one. The calyculate edges are spinous. Over sixty spines, they reduce in size distally and away from sulcus. Base of organ, proximal to the large spinous, is beset with very minute spines. Centripetal sulcus distinct, forking near the crotch which is about the second subcaudal, it ends at the tips of the organ. The lips of sulcus are spinous and calyculate in the respective areas of ornamentation.

The *M. retractor penis magnus* ends in the twenty-third to twenty-fifth subcaudal, it is forked at the level of the eleventh subcaudal before its insertion the dorsal organ. The dorsal lobe is one subcaudal longer than the ventral lobe (*in situ*).

***G. brevicaudus* (Stejneger).** The hemipenes (Fig. 2) of the specimens from Anhui and Zhejiang extend to the fifth to eighth subcaudal plate and are forked at the level of the third to fourth subcaudal. Base of the organ nude. The proximally half of the organ is spinous and the other half calyculate. The calyculate ridges bears tiny spines. Gloyd et al. (1990) also described the hemipenes from Anhui, they were sure that the lips of top calyces were smooth. The demar-

Table 1. Material used in this study.

Species	Number	Locality	Date
<i>Gloydus ussuriensis</i>	4	Jiling	1979
<i>G. brevicaudus</i>	8	Anhui, Zhejiang, Liaoning	1972, 1974, 1979, 1991
<i>G. strauchii</i>	3	Sichuan	1975, 1978
<i>G. qinlingensis</i>	2	Shaanxi	1984
<i>G. intermedius</i>	5	Xinjiang	1976, 1977, 1980, 1987
<i>G. saxatilis</i>	3	Jiling, Liaoning	1978, 1979
<i>G. shedaensis</i>	4	Liaoning	1965, 1979
<i>Deinagkistrodon acutus</i>	3	Jiangxi, Fujian	1964, 1979

cation between the two areas is inconspicuous. In the spinous area, there are about twenty-eight to thirty spines, they become larger away from the sulcus. Sulcus prominent, it is forked at the second subcaudal and ends at the tips of the organ. The lips of the sulcus are spinous in the spinous area and calyculate in the calyculate area. The *M. retractor penis magnus* originates at the level of the twenty-fifth to twenty-fourth subcaudal, and is forked for the length of two to four subcaudals before its insertion the organ dorsal.

In this paper, three specimens from Liaoning, which are dominated *brevicaudus* by Gloyd (1972), were also examined, they are different from the formers slightly. The calyce is restrict to the tips of the lob, its extent is about fourth of the spinous one. The *M. retractor penis magnus*, which is about ten to fourteen in length, is shorter than the former one, it is forked for the length of two subcaudals. The hemipenes of two specimens examined extend to the seventh or eighth subcaudal, and are forked at the level of the fourth subcaudal, but the other one is much longer, it extends to the twelfth subcaudal and is forked at the level of the seventh subcaudal. Our observation is consistent with Gloyd *et al.* (1990) and Malnate (1990).

***G. intermedius* (Strauch).** The hemipenis (Fig.3) is seven to eleven subcaudals in length, forked for the length of four to five subcaudals. It is spinous proximally half, and the distally half of the organ is calyculate. The line of changing from spines to calyces is inconspicuous. The calyces small adjacent to the sulcus but larger away from it, their lips bearing tiny spines. About sixty spines are present in the spinous area, they reduce in size distally and near the sulcus. One or two strongly enlarged spines adjacent to the

sulcus at the crotch level. Small spines are found in the base of the organ. The sulcus is forked at the level of the second to the fourth subcaudal, it continues to the tips of the lobes. The sulcus prominent, its lips are spinous or calyculate in the respective area of ornamentation. *M. retractor penis magnus* originates at the level of the twenty-seventh to twenty-ninth subcaudal, it is forked for the length of two to four subcaudals before its insertion at the organ tips.

***G. saxatilis* (Emelianov).** The hemipenis (Fig. 4) of this species extends to the ninth to the eleventh subcaudal plate, forked at the level of the third or fourth subcaudal. It is spinous proximally and calyculate distally. The spinous and calyculate areas are about equal in length, the demarcation between two areas is inconspicuous. There are about sixty to seventy spines near the crotch, including six much larger spines, four of them appearing on the surface near the crotch, while the other two are on the opposite side. The spines reduce in size distally on the organ lobes. The margins of the calyces are spinous. The base of the organ is covered with tiny spines. While Gloyd *et al.* (1990) previously described the specimens collected from Korea (near Seoul), the organ base nude, the lips of the distal cups are smooth. The sulcus is bifurcated at the level of the second subcaudal and extends to the tips of the organ, their lips are prominent, bearing a few spines in the spinous area and calyces in the calyculate area. The *M. retractor penis magnus* originates at the level of the twenty-fourth to twenty-fifth subcaudal, and is forked for the length of two to three subcaudals before its insertion at the organ tips.

***G. shedaensis* (Zhao).** In this species, the hemipenis (Fig. 5) is eight to ten subcaudals in length, and forked for five to six subcaudals. The base of the

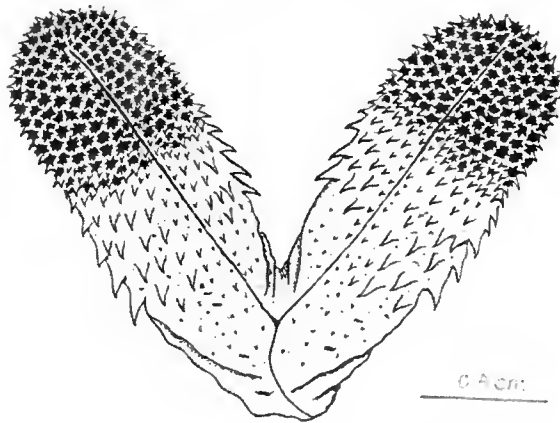


Figure 1. The right everted hemipenis of *G. ussuriensis* (CIB 785185, Panshi, Jilin)

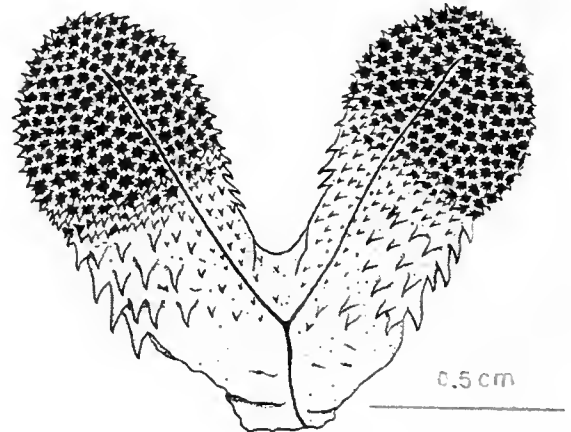


Figure 2. The right everted hemipenis of *G. brevicaudus* (CIB 726108, Taiping, Anhui)



Figure 3. The right everted hemipenis of *G. intermedius* (CIB 8010015, Xingyuan, Xinjiang)



Figure 4. The right everted hemipenis of *G. saxatilis* (CIB 7910090, R. Zhuang, Liaoning)

organ is covered with tiny spines. Spinose proximally, calyculate distally. The margins of calyces are tiny spines. The extent of calyces is about half of spines. Line of changing from spine to calyces is inconspicuous. There are ninety to one hundred spines in the spinous area, and six especially larger spines in each lobe. The sulcus prominent, forked at the level of the third subcaudal, and extends to the tips of the organ rami. Sulcus lips are spinous or calyculate in the respective areas of ornamentation. The *M. retractor penis magnus* originates at the level of the twenty-fourth to twenty-seventh subcaudal and is bifid for the length of two to three subcaudals before its insertion at the organ tips.

G. strauchii (Bedriaga). The hemipenis (Fig. 6) examined extends to the seventh to eleventh subcaudal and is forked for four to five subcaudals. It is spinous proximally half, and the distally half is calyculate. The line of changing from spines to calyces is inconspicuous. The calyces are small adjacent to the sulcus but larger away from it, their lips bearing tiny spines. About sixty spines are present in the spinous area, they reduced in size distally and near the sulcus. One or two strongly enlarged spines are adjacent to the sulcus at the crotch level. Small spines are present in the base of the organ. The sulcus forks at the level of the second to fifth subcaudal, and continues to the tips of the lobes. The lips of the sulcus prominent, they are spinous or calyculate in the respective areas of ornamentation. *M. retractor penis magnus* origi-

nates at the level of the twenty-seventh to twenty-

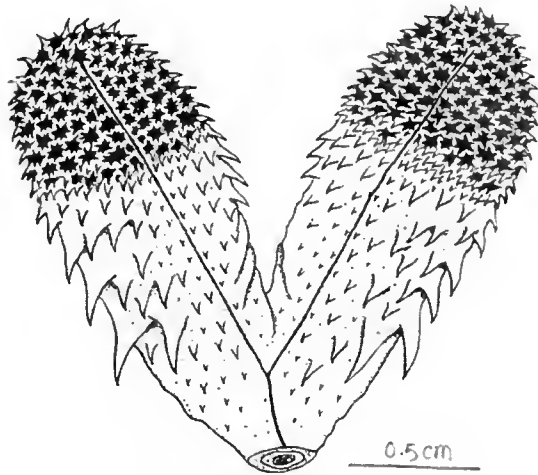


Figure 5. The right everted hemipenis of *G. shedaoensis* (CIB 79110006, Shedao, Liaonong)

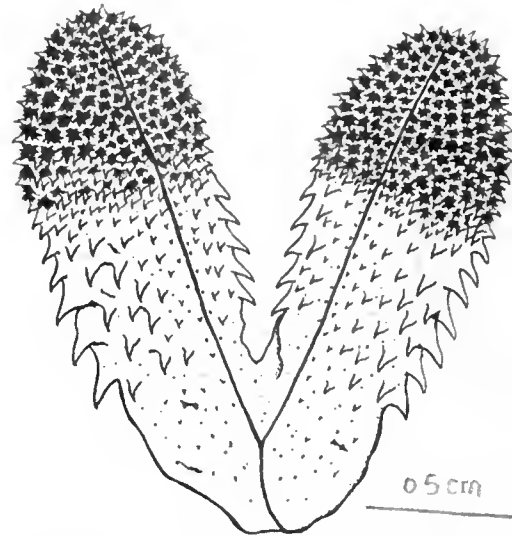


Figure 6. The right everted hemipenis of *G. strauchii* (CIB 755127, Hongyuan, Sichuan)



Figure 7. The right everted hemipenis of *G. qinlingensis* (840050, Qinling, Shaanxi)

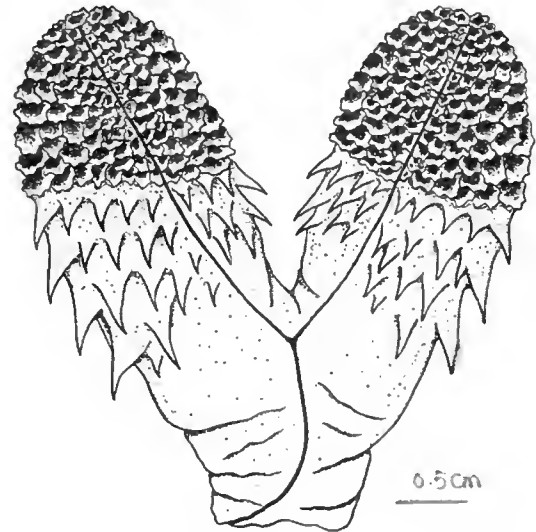


Figure 8. The right everted hemipenis of *D. acutus* (CIB 6416190, Fujian)

ninth subcaudal, and is forked for the length of two to

four subcaudals before its insertion at the organ tips.

***G. qinlingensis* (Song).** The hemipenis (Fig. 7) extends to the eleventh subcaudal plate in length and is forked at the level of the fifth subcaudal. Minute spines are present in the base of the organ. It is spinous proximally, calyculate distally, the spinous area is about twice to third times as the calyculate one. The line of demarcation between two areas is inconspicuous. The calyculate edges are spinous, there are approximately thirty to forty spines in the spinous region. The sulcus bifurcates the fourth to fifth subcaudal, terminates at the tips of the organ. Sulcus is distinct, bearing spines in spinous area and

calyces in the calyculate one. The length of *M. retractor penis magnus* extending is 14 subcaudals and it is forked for the length of two subcaudals before its insertion at the organ tips.

***Deinagkistrodon acutus* (Gunther).** The hemipenis (Fig. 8) extends to the eleventh to thirteenth subcaudal and is forked opposite the fourth subcaudal, base of organ nude. The proximally half of the organ is spinous, the distally half calyculate, the demarcation between these two areas is distinct. The calyces with smooth margins reduce in size distally and near the sulcus. The spines are about sixth to seventy, they are larger on the lateral surface of each lobe than those

near the sulcus. The sulcus forks at the level of the fourth subcaudal, it extends to the tips of the each lobe. The lips of sulcus prominent, they bearing calyces in the calyculate region and smooth in the spinous area. *M. retractor penis magnus* originates at the level of the twenty-sixth subcaudal and is bifid for the length of two to four subcaudals before its insertion at the tips of the organ.

Discussion

The hemipenes are deeply forked in eight Chinese species of pitvipers, the shape of the everted hemipenes are "Y". It is spinous proximally and calyculate distally. The centripetal sulcus is consistently bifurcated on the basal part of the organ in all of the species examined, and it extends to the tips of the lobes. These similarities reveal the taxonomic relationships between genus *Gloydius* and *Deinagkistrodon*. The deeply forking of the hemipenis and the bifurcation of the sulcus in the basal region of the organ are exactly like those of the other pitvipers (Pope, 1935; Smith, 1943).

D. acutus can be characterized by presence of the following hemipenial structures: (1) the margins of the calyces are smooth; (2) transition from spines to calyces is abrupt and conspicuous; (3) the sulcus lips are smooth in the spinous area; (4) the base of the organ is naked. These conditions further confirmed the validity of the independent generic status of *Deinagkistrodon*.

The seven species of *Gloydius* exhibit rather conservative and stable hemipenial structures, the interspecific variation is only limited to: (1) the length of the hemipenes and the depth of their forking; (2) the extent of the calyculate area on the lobes; (3) the length of the retractor muscle and the depth of their forking; (4) the manner of insertion of the retractor muscle onto the organ; (5) the number of enlarge spines. The results above indicated that the differentiation of the hemipenial structures in *Gloydius* is little.

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Catalogue of Type Specimens of Reptiles in the Herpetological Collections of Chengdu Institute of Biology, the Chinese Academy of Sciences

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Abstract.- There are nearly 15,000 specimens and 300 species of Chinese reptiles that are preserved in the herpetological collections of Chengdu Institute of Biology (CIB), the Chinese Academy of Sciences, these species are over 70% of the known species in China. Most of these specimens were collected by the scientific research personnel in the herpetology department of CIB in sixty years. In these specimens there are about 400 type series specimens (28 species or subspecies) which were selected by the scientific research personnel in the herpetology department of CIB when they described the new taxon. In order to reflect the achievements in scientific research on Chinese reptiles in CIB and provide the facility to scientific research and academic exchange, it is helpful to list all of the type specimens in the collections.

Key words.- Reptiles, type specimens, Chengdu Institute of Biology, China.

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LACERTILIA

AGAMIDAE:

Calotes medogensis Zhao and Li, 1984

Acta Herpetol. Sin., Chengdu, [new ser.], 3(4): 77.

Holotype: CIB 8370177, Male; Yarang, Medog Co., Xizang Autonomous Region; 910 m; Coll. S. Q. Li, 7. 23, 1983.

Japalura szechwanensis Hu and Zhao, 1966

Acta Zootaxon. Sin., Peking, 3(2): 158.

Holotype: CIB 613047, Male; Baishui He, Peng Co., Sichuan Prov.; 2000 m; Coll. C. C. Liu; 5. 9, 1961.

Allotype: CIB 6130460, Female; the same locality and time as holotype.

Paratype: CIB 625131, Male; Mt. Erlang, Tianquan Co., Sichuan Prov.; 1400 m; 8, 1962.

Laudakia papenfussi Zhao, 1998

Zoological Research, Kunming, 19(5): 401.

Holotype: CIB 775001, Male; Mayang River Valley between Mayang Village and Diya Village, Zanda Co., Xizang Autonomous Region; 3300 m; Coll. R. Z. Zhang; 7. 1, 1976.

Laudakia wui Zhao, 1998

Acta Zootaxon. Sin., Beijing, 23(4): 440.

Holotype: CIB 7315012, Male; Yi'ong, Bomi Co., Xizang Autonomous Region; 2350 m; Coll. Y. M. Jiang; 6. 11, 1973.

Allotype: CIB 7315014, Female; the same locality and date as the holotype.

Paratypes: CIB Nos. 7315011, 7315013, Females, CIB Nos. 7315008-10, Juveniles; the same locality and date as the holotype; CIB Nos. 7315037-38, Juveniles; between Tangmai and Yi'ong, Bomi Co.,

Xizang Autonomous Region; 2150 m; Coll. X. E. Wu; 7. 5, 1973.

***Phrynocephalus vlangalii hongyuanensis* Zhao, Jiang, and Huang, 1980**

Acta Zool. Sinica, Beijing, 26(2): 178.

Holotype: CIB 785795, Male; Coll. E. M. Zhao; 7. 25, 1978.

Allotype: CIB 785825, Female; Coll. Y. M. Jiang; 8. 2, 1978.

Paratypes: CIB Nos. 755111-12, 755114-15, 785793-94, 785796-824, 785826-27, 7 Males and 27 Females;

Coll. Y. Gao; 8, 1978.

Hongyuan Co., Sichuan Prov.; 3500 m.

=*Phrynocephalus hongyuanensis* Zhao, Jiang, and Huang, 1980

Ref: Y. Z. Wang and Y. M. Jiang, (1992), in: Y. M. Jiang (ed.), Coll. Papers Herpetol., Chengdu: 111.

***Phrynocephalus zetangensis* Wang, Zeng, and Wu, 1996**

Zoological Research, Kunming, 17(1): 27.

Holotype: CIB 0156, Male.

Allotype: CIB 0157, Female.

Paratypes: CIB 0151, Male; CIB Nos. 0154, 0158, Female; CIB 0155, Juvenile.

Zetang, Xizang Autonomous Region; 3950 m; Coll. Z. L. Fang and Z. J. Liu; 7. 5, 1993.

GEKKONIDAE:

***Tenuidactylus medogensis* Zhao and Li, 1987**

Acta Herpetol. Sin., Chengdu, [new ser.], 6(1): 48.

Holotype: CIB 8380188, Female.

Paratype: CIB 8380187, Juvenile (Male ?).

Kabu, Medog Co., Xizang Autonomous Region; 1250 m; Coll. S. Q. Li; 1983.

=*Cyrtodactylus medogensis* (Zhao and Li, 1987)

Ref: Zhao, E. M. and K. Adler, (1993), Herpetology of China, Oxford (Ohio), p: 179.

SCINCIDAE:

***Eumeces liui* Hikida and Zhao, 1989**

Copeia, Gainesville, (1): 111.

Allotype: CIB 645026, Male; Wuchang, Hubei Prov.; 3. 31, 1965.

***Scincella huanrenensis* Zhao and Huang, 1982**

Acta Herpetol. Sin., Chengdu, [new ser.], 1(1): 3.

Paratypes: CIB Nos. R810012, R810014, R810015-18, R810040-41; Huanren Co., Liaoning Prov.; 700 m;

Coll. K. C. Huang; 5. 8, 1981.

***Leiolopisma tsinlingensis* Hu and Zhao, 1966**

Acta Zool. Sin., Peking, 18(1): 82.

Holotype: CIB 627050, Male; 1800 m; Coll. G. F. Wu and E. M. Zhao; 6. 13, 1962.

Allotype: CIB 627039, Female; 1820 m; 6. 10, 1962.

Paratypes: Cib Nos. 627025-27, 627029, 627051-53, Males, CIB Nos. 627020-24, 627034-36, 627040-627049, 627069, Females, CIB Nos. 627028, 627030-32, 627054-62, 627070, Juveniles.

Mt. Tsinling [= Qinling], Chouchih [= Zhouzhi Co.], Shaanxi Prov..

=*Scincella tsinlingensis* (Hu and Zhao, 1966)

Ref: Y. Z. Wang and E. M. Zhao, (1986), Acta Herpetol. Sin., Chengdu, [new ser.], 5(4): 276.

SERPENTES

XENOPELTIDAE:

Xenopeltis hainanensis Hu and Zhao, 1972

Key Chin. Snakes [= Mater. Herpetol. Res., 1], Chengdu: 36.

Holotype: Cib 64III6016, Male; Dali, Mt. Diaoluo, Hainan Prov.: 200 m; 5. 15, 1964.

Allotype: CIB 64III6650, Female; Yacha Matou, Baisa Co., Hainan Prov.; 217 m; 9. 4, 1964.

=*Xenopeltis hainanensis hainanensis* Hu and Zhao, 1972

Ref: E. M. Zhao, (1995), Sichuan Jour. Zool., Chengdu, 14(3): 107.

COLUBRIDAE:

Achalinus meiguensis Hu and Zhao, 1966

Acta Zootaxon. Sin., Peking, 3(2): 162.

Holotype: CIB 639101, Female; Liang He Kou, Meigu Co., Sichuan Prov.: 2520 m; Coll. X. Y Tang; 5. 22, 1963.

Paratypes: CIB Kang076, Female; Tu Ba Kou, Baoxing Co., Sichuan Prov.; CIB Nos. 561836-37, 570080, Female, CIB Nos. 562154, 562174, 600030, Male; Mt. Omei, Sichuan Prov..

Natrix optata Hu and Zhao, 1966

Acta Zootaxon. Sin., Peking, 3(2): 160.

Holotype: CIB Chuan3624, Male; Liangfeng Gang, Mt. Omei, Sichuan Prov.: 700 m; Coll. C. C Liu; 8. 9, 1940.

Paratypes: CIB Nos. 561760, 562168, 562221, 570072, 600039, 645201, Males, Cib Nos. R001, 561764, 562167, 562231, Females, 2 Juveniles; Mt. Omei, Sichuan Prov.; 1956-1960.

=*Amphiesma optata* (Hu and Zhao, 1966)

Ref: E. M. Zhao and Y. M. Jiang, (1986), Acta Herpetol. Sin., Chengdu, [new ser.], 5(3): 239.

Dinodon rozozonatum Hu and Zhao, 1972

Key Chin. Snakes [= Mater. Herpetol. Res., 1], Chengdu: 36.

Holotype: CIB 64III6089, Male; Dali, Mt. Diaoluo, Hainan Prov.: 200 m; 5. 21, 1964.

Allotype: CIB 64III5246, Female; Mt. Wuzhi, Hainan Prov.; 540 m; 5. 11, 1964.

Paratypes: 3 Males and 5 Females; Mt. Diaoluo, Mt. Wuzhi and Haikou, Hainan Prov.; 80-580 m; 1964-1972.

Macropisthodon rudis multiprefrontalis Zhao and Jiang, 1981

Acta Herpetol. Sin., Chengdu, [old ser.], 5(7): 55.

Holotype: Cib 65I5143, Male; Xichang, Sichuan Prov.; 2650 m; Coll. C. C. Liu; 6. 8, 1965.

Allotype: CIB 65I5142, Female; Xichang, Sichuan Prov.; Xichang, Sichuan Prov.; 2650 m; Coll. G. F. Wu; 6. 8, 1965.

Paratypes: CIB Nos. 65I5001, 65I5148, Males, CIB Nos. 65I5034, 65I5012, Females, CIB 580960, Juvenile; Huili Co., Zhaojue Co., Yuexi Co., and Ganluo Co., Sichuan Prov.; 2000-2630 m; Coll. D. Y. Yang; 8, 1958 and 5-6, 1965.

Oligodon mulizonatum Zhao and Jiang, 1981

Acta Herpetol. Sin., Chengdu, [old ser.], 5(7): 54.

Holotype: CIB 80II0289, Male.

Allotype: CIB 80II0290, Male, CIB Nos. 80II0274, 80II0291, Juveniles.

Luding Co., Sichuan Prov.: 1400 m; 8. 17, 1980.

Opisthotropis guangxiensis Zhao, Jiang, and Huang, 1978

Mater. Herpetol. Res., Chengdu, 4: 21.

Holotype: CIB 602488, Male; Mt. Yao [= Dayao Shan], Guangxi Prov.; 6. 19, 1960.

Allotype: CIB 601588, Female; Mt. Yao [= Dayao Shan], Guangxi Prov.; 4. 28, 1960.

Paratype: CIB 603583, Female; Longsheng Co., Guangxi Prov.; 7. 2, 1960.

***Plagiopholis unipostocularis* Zhao, Jiang, and Huang, 1978**

Mater. Herpetol. Res. Chengdu, 4: 21.

Holotype: CIB 505121, Female; Yunnan Prov..

***Rhabdophis adleri* Zhao, 1997**

Asiat. Herpetol. Res., Berkeley, 7: 166

Holotype: CIB 64III5917, Male; Dali Village in Mt. Diaoluo, Lingshui Co., Hainan Prov.; 225 m; 6. 10, 1964.

Allotype: CIB 64III5228, Female; Mt. Wuzhi, Qiongzong Co., Hainan Prov.; 500 m; 5. 10, 1964.

Paratypes: CIB Nos. 64III5112, 5114-15, Females; 4. 24-25, 1964; CIB 64III5883, male, CIB 64III5441, Juvenile, 6. 1-9, 1964; Mt. Diaoluo, Lingshui Co., Hainan Prov.; 82-217 m; CIB 64III6612, Juvenile; Mt. Yinggeling, Baisha Co., Hainan Prov.; 670 m; 8. 25, 1964.

***Rhabdophis nuchalis pentasupralabralis* Jiang and Zhao, 1983**

Acta Herpetol. Sin., Chengdu, [new ser.], 2(1): 60.

Holotype: CIB 80II0040, Male; 2750 m; Coll. E. M. Zhao; 7. 26, 1980.

Allotype: CIB 80II0056, Female; 2750 m; Coll. Y. M. Jiang; 7. 26, 1980.

Paratypes: 95 Males, 48 Females, and 65 Juveniles; 2750-2850 m; 7. 24-28, 1980. Jiulong Co., Sichuan Prov..

=*Rhabdophis pentasupralabralis* Jiang and Zhao, 1983

Ref: E. M. Zhao, (1995), Jour. Suzhou Rail. Teach. Coll. (Nat. Sci.), Suzhou (Jiangsu), 12(2): 38.

***Sibynophis chinensis miyiensis* Zhao and Kou, 1987**

Chin. Herpetol. Res., Chengdu, No.1: 4.

Holotype: CIB 105027, Male; Miyi Co., Sichuan Prov.; 880 m; Coll. S. H. Kang; 6, 1986.

Allotype: CIB 105025, Female; Miyi Co., Sichuan Prov.; 880 m; Coll. S. H. Kang; 6, 1986.

Paratypes: CIB Nos. 105026, 105028, 105030-31, Males; CIB 105029, Female; Lugu Lake, Sichuan-Yunnan border; 2600 m; Coll. Z. T. Kou; 1985.

VIPERIDAE:

***Trimeresurus mangshanensis* Zhao, 1990**

Sichuan Jour. Zool., Chengdu, 9(1): 11.

Holotype: CIB ZS8901, Female.

Paratype: CIB ZS8902, Female.

Pingkeng, Mt. Mang, Yizhang Co., Hunan Prov.; 700-900 m; Coll. G. H. Chen and Y. L. Tao; 9, 1989.

=*Ermia mangshanensis* (Zhao, 1990)

Ref: Zhang, F. J., (1993), in: Zhao, E. M. *et al.* (eds.), Proceedings of the First Asian Herpetological Meeting, Beijing, p: 56.

***Agkistrodon shedaoensis* Zhao, 1979**

Acta Herpetol. Sin., Chengdu, [old ser.], 1(1): 4.

Holotype: CIB 79I0005, Male; Shedao, Liaoning Prov.; 5. 19, 1979.

=*Gloydus shedaoensis* (Zhao, 1979)

Ref: A. R. Hoge *et al.*, (1978/79), Poisonous Snakes of the World. Part I. Check list of the Pit Vipers Viperidae, Viperidae, Crotalinae. Mem. Inst. Butantan, Sao Paulo, 42/43. p: 194.

***Trimeresurus monticola zayuensis* Jiang, 1977**

Acta Zool. Sin., Beijing, 23(1): 67.

Holotype: CIB 73I5024, Male; 1800 m; 7. 22, 1973.

Allotype: CIB 73I5025, Female; 2070 m; 7. 30, 1973.

Paratype: CIB 73II5349, Male; 1965.

Zayu Co., Xizang Autonomous Region.

=*Ovophis zayuensis* (Jiang, 1977)

Ref: E. M. Zhao, (1995), Jour. Suzhou Rai. Teach. Coll. (Nat. Sci.), Suzhou (Jiangsu), 12(2): 37.

***Ovophis monticola zhaokentangi* Zhao, 1995**

Sichuan Jour. Zool., Chengdu, 14(3): 109.

Paratype: CIB 740003, Male; Pianma, Lushui Co., Yunan Prov.; 1980 m; 3. 17, 1974.

***Trimeresurus medoensis* Zhao, 1977**

Acta Zool. Sin., Peking, 23(1): 66.

Holotype: CIB 73II5208, Male; 1200 m; Coll. E. M. Zhao and Y. Gao.

Allotype: CIB 73II5209, Male; 1400 m; Coll. E. M. Zhao and X. E. Wu.

Near A-nie Bridge, Medog Co., Xizang Autonomous Region; 8. 3, 1973.

***Trimeresurus stejnegeri chenbihuii* Zhao, 1995**

Sichuan Jour. Zool., Chengdu, 14(3): 110.

Holotype: CIB 64III5599, Male; Mt. Diaoluo, Lingshui Co., Hainan Prov.; 250 m; 6. 6, 1966.

Allotype: CIB 64III5945, Female; Mt. Diaoluo, Lingshui Co., Hainan Prov.; 250 m; 6. 11, 1966.

Paratypes: CIB Nos. 64III5906, 64III5944, 64III5978-79, 64III6013, 64III6043-44, 64III6069, 64III6101, 64III6104, 64III6107, Males; 64III5600, 64III5735, 64III6014, Females; Mt. Diaoluo, Lingshui Co., Hainan Prov.; 225-290 m; 6. 6-15, 1964. CIB Nos. 64III5110, 64III5181, 64III5261-62, Males; Mt. Wuzhi, Qiongzong Co. Hainan Prov.; 500 m; 4. 23-5. 12, 1964.

***Trimeresurus xiangchengensis* Zhao, Jiang, and Huang, 1978**

Mater. Herpetol. Res., Chengdu, 4: 21.

Holotype: CIB 725050, Male; 3100 m; 10. 7, 1972.

Allotype: CIB 725049, Female; 3200 m.; 10.10, 1972.

Paratypes: CIB 725048, Male; CIB Nos. 725051-57, 5 Females and 2 Juveniles; Xiangcheng Co., Sichuan Prov.; 3000-3200 m; 10. 1-28, 1972.

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Correlations of Reproductive Parameters of Two Tropical Frogs from Malaysia

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Abstract.— A study on the relationships of reproductive parameters such as fecundity, egg size, clutch weight and body size of two frogs, *Rana cancrivora* (Gravenhorst) and *R. limnocharis* (Boie), from Malaysia was carried out in February 1992. The results showed that all the parameters quantified were greater in *R. cancrivora* than in *R. limnocharis*. No correlation was found between the spawned clutch size and egg size for both species. However, correlation analyses of unspawned clutch showed that there exist positive relationships between female size and clutch size, female size and clutch weight, female size and egg weight, clutch size and clutch weight, and clutch weight and egg weight in *R. cancrivora*. For *R. limnocharis* positive correlations were found between clutch size and clutch weight, and clutch weight and egg weight for the unspawned complement. The advantages, non-advantages, correlation and non-correlations of these parameters with respect to their reproductive strategies and success of these frogs are discussed and compared with the current information from the literature.

Key words.— Anura, Ranidae, *Rana cancrivora*, *R. limnocharis*, Malaysia, reproductive parameters, female size, clutch size, egg size, clutch weight, egg weight.

Introduction

The fecundity or clutch size in amphibians is highly variable depending on body size and reproductive mode (Salthe, 1969; Salthe and Duellman, 1973; Kaplan, 1980). As a general rule, large species produce more eggs than smaller ones and species that have generalized reproductive modes have larger clutches than those with specialised modes (Duellman and Trueb, 1986). The fecundity and hatching success of an amphibian population play an important role in the propagation and future survival of the population (Ibrahim and Ahmad, 1992). Reproductive success of a female depends upon the number of eggs produced and their quality, but are constrained by physiological capacities of the female and the trophic quality of the environment (Rafinska, 1991). The number of eggs deposited by amphibians varies greatly from species to species and this large disparity in fecundities is explained by differences among amphibians in sizes of eggs, patterns of development, sizes of females and reproductive behaviour (Porter, 1972; Salthe and Duellman, 1973; Kuramoto, 1978; Kaplan, 1980). Studies on the fecundity of temperate frogs have generated considerable data on this facet of amphibian reproduction (Collins, 1975; Kuramoto, 1978; Duellman and Trueb, 1986), but information on tropical oriental species are few and far between. Berry (1964) reported that seven anuran species in Singapore exhibited difference in fecundities between and within

species, while four species of frogs from Malaysia also showed differences in fecundities both between and within species (Ibrahim and Ahmad, 1992). Inger and Bacon (1968) found that four large ranids in Borneo have clutch sizes in excess of a thousand eggs, while Yorke (1983) reported that the average clutch size for a rhacophorid in Malaysia to be 225 eggs.

Ovum size is another important aspect of anuran reproductive biology. Large ovum size is generally associated with large female body size and thus directly hints of better fitness and survival value. Egg size variability in amphibians has been interpreted as adaptive since, where breeding habitats vary unpredictably, production of a wide range of sizes may enhance adult fitness more than does the output of a single 'optimal' egg size (Tejedo and Reques, 1992). The egg size of numerous temperate anurans are well documented in the literature (e.g. Salthe, 1969; Salthe and Duellman, 1973; Bell and Lawton, 1975; Collins, 1975; Kuramoto, 1978; Jorgensen, 1981; Duellman and Trueb, 1986; Reading, 1986; Rafinska, 1991; Tejedo and Reques, 1992), but again however, only a few reports concerning this parameter are available for tropical species (e.g. Liem, 1959; Alcalá, 1962; Inger and Bacon, 1968; Uchiyama et al., 1990).

Rana cancrivora (Gravenhorst) and *R. limnocharis* (Boie) are two species of anura that have a wide distribution across the oriental zoogeographical realm (Church, 1960; Alcalá, 1962; Berry 1964; Inger, 1966; Kuramoto, 1978). In spite of this only a

handful of information have been published concerning the fecundity and size of their eggs (Aleala, 1962; Berry, 1964; Kuramoto, 1978). It is also evident from the literature that there exist considerable variations in the relationships of body size, egg size and fecundity within the Amphibia (Salthe, 1969; Collins, 1975; Kuramoto, 1978; Jorgensen, 1981; Reading, 1986; Duellman and Trueb, 1986). This paper therefore determines the fecundity and egg size of both *R. cancrivora* and *R. limnocharis* and then examines the existing relationships in the reproductive parameters of both frogs.

The Study Site

The place chosen for the study is a rice growing region near the small town of Tanjung Karang (3° 20'N, 101° 10'E), in the state of Selangor, elevation about 3 m above mean sea level. It is situated about 100 km to the northwest of Kuala Lumpur, the Malaysian capital. Previously this area was a freshwater peat swamp forest which was opened up and converted to paddy fields by settlers in the 1920's. Presently due to the improved and upgraded irrigation and drainage system, the farmers are planting two rice crops a year.

Material and Methods

To determine the fecundity and egg size of spawned eggs, newly deposited egg clutches of both species were collected from recently inundated rice fields. Breeding choruses were visited, amplexant pairs of frogs were located and their positions marked with wooden pegs. Male frogs usually start calling as soon as dusk sets and individuals of both species usually initiated mating as soon as the males begin clasping the females. This usually occurs at around 0100hrs or 0200 hrs. The pairs will then move into the flooded rice paddy and egg laying will occur around 0500 hrs. The number of eggs in each clutch were then recorded and the eggs collected and preserved in 4% formalin and kept in labeled, airtight plastic 40 dram bottles. In the laboratory, 30 to 50 eggs from each clutch were then randomly chosen and their diameters measured under a stereo microscope with an ocular micrometer to determine the mean size of eggs of each species. All eggs measured were between Stages 3 and 9 of Gosner (1960).

To investigate the relationship between female body size and, (a) the number of unspawned eggs, (b) weight of unspawned egg clutch and (c) the weight of unspawned eggs, the snout-urostyle length of gravid females with large pigmented ova in the oviducts were measured, the unspawned egg complement were weighed (clutch weight) and counted (clutch size or

fecundity). This is a modified method of Kuramoto (1978). Inger and Bacon (1968) reported that in most Indomalayan frogs the eggs in the ovary are essentially in two size classes : an enlarged or developing set and a much smaller, white 'undeveloped set' ; the latter contributes no significant portion to the total volume of the ovary.

Results

Forty-two spawned batches of *R. cancrivora* eggs and thirty-four of *R. limnocharis* were collected from the field in February 1992. These were investigated for fecundity and size of spawned eggs. In addition thirty-two batches of *R. cancrivora* eggs and thirty of *R. limnocharis* were obtained from sacrificed gravid females.

The results showed that the average number of eggs laid by *R. cancrivora* females from Tanjung Karang is 1077.9 (sd = ± 238.97, range 662 to 1677). That for *R. limnocharis* 405.5 (sd = ± 92.45, range 233 to 657). Weighted t-test for unequal variance (Cochran and Cox, 1957) showed that the number of eggs oviposited by *R. cancrivora* is significantly greater than that laid by *R. limnocharis* ($t = 16.751$, $p < 0.001$, $df = 74$).

The mean diameter of *R. cancrivora* eggs is 1.35 mm (sd = ± 0.091, range 1.16 to 1.53) while the mean diameter of *R. limnocharis* eggs is 1.15 mm (sd = ± 0.027, range 1.10 to 1.21). Again a weighted t-test for unequal variance showed that the egg size for both species is significantly different ($t = 12.980$, $p < 0.001$, $df = 74$). However, no correlation was found between the spawned clutch size and egg size for both species.

For clutch complement of unspawned *R. cancrivora* eggs, the average clutch size was 2904.6 (sd = ± 1600.6, range 889 - 7573), the average clutch weight was 2.25 g (sd = ± 1.726, range 0.37 - 5.71) and the average egg weight was 0.75 mg (sd = ± 0.257, range 0.28 - 1.35). The same parameters for *R. limnocharis* was 702.6 (sd = ± 306.49, range 312 - 1659), 0.40 g (sd = ± 0.264, range 0.074 - 1.04) and 0.55 mg (sd = ± 0.249, range 0.237 - 1.01) respectively. Weighted t-tests for unequal variance on the clutch size, clutch weight and egg weight between the two species showed that these parameters were significantly different ($t = 7.634$, $p < 0.001$, $df = 60$; $t = 6.799$, $p < 0.001$, $df = 60$; and $t = 3.019$, $p < 0.05$, $df = 60$ respectively).

Correlation analyses carried out on the reproductive parameters of unspawned eggs for each species showed that for *R. cancrivora* there is a positive corre-

lation between female size and clutch size ($p < 0.001$, $r = 0.6333$), female size and clutch weight ($p < 0.001$, $r = 0.6867$), female size and egg weight ($p < 0.05$, $r = 0.4073$), clutch size and clutch weight ($p < 0.001$, $r = 0.8870$) and clutch weight and egg weight ($p < 0.001$, $r = 0.5983$). There was no statistically significant correlation between clutch size and egg weight. For *R. limnocharis* however, there exists positive correlation between clutch size and clutch weight ($p < 0.001$, $r = 0.7501$) and clutch weight and egg weight ($p < 0.001$, $r = 0.6818$) only, while no correlation was found between female size and clutch size, female size and clutch weight, female size and egg weight, and clutch size and egg weight.

Discussion

The number of eggs oviposited by breeding females of both species were lower than that found in their ovaries. This is not unexpected because presumably the breeding females were depositing the complement of eggs that were mature and retaining the unripe ones for later oviposition (Inger and Bacon, 1968; Telford and Dyson, 1990). It is likely that both females of *R. cancrivora* and *R. limnocharis* in Tanjung Karang may breed more than once during the breeding season. Wells (1976) concluded that one of the major advantages of a prolonged breeding period may be the capacity for females to breed twice or more in a single season. Hence it is possible that as a security measure the females only release a portion of eggs from their ovarian complement. The incidence of multiple clutches is not uncommon in amphibians and have been reported in a number of anuran species (Collins, 1975; Howard, 1978; Perril and Danial, 1983; Telford and Dyson, 1990; Rafinska, 1991).

Alcala (1962) and Uchiyama (1990) reported that the clutch size for *R. cancrivora* was probably more than 2000 and 1800 eggs respectively. Our study found a lower clutch size than these two. Berry (1964) presented data on the number of ovarian eggs in Singaporean *R. limnocharis*. Calculations based on her data gave the average clutch size for her samples as 576.4 eggs (range 266-1318). This is comparable to the results of this study. However, Kuramoto (1978) reported larger clutch size, clutch weight and egg weight for *R. limnocharis* from Japan. This discrepancy could be due to the clinal effect within the species (Duellman and Trueb, 1986) or even geographical differences, since the Japanese frog would be at the northernmost edge of its distribution.

The difference in egg size between different populations of amphibian species have been attributed to adaptation of the population to their par-

ticular environment (Pettus and Angleton, 1967; Salthe and Duellman, 1973; Jorgensen, 1981). There exists a positive correlation between egg size and hatchling size in frogs (Salthe and Duellman, 1973) and a large metamorphic size is important because of the interrelationship of body size, time of metamorphosis, age at first reproduction and fecundity (Wilbur, 1972; Wilbur and Collins, 1973; Doty, 1978). Thus the observed larger egg size of *R. cancrivora* over *R. limnocharis* could probably be assumed to confer reproductive advantage with respect to fecundity, and could probably also be assumed to do the same with respect to metamorphic size, post metamorphic growth and age at first reproduction.

The interspecific differences in the sizes of eggs in amphibians is well documented in the literature (Inger and Bacon, 1968; Collins, 1975; Kuramoto, 1978; Duellman and Trueb, 1986). For example, Collins (1975) found that there is significant difference in average egg diameter between *Hyla versicolor*, *R. sylvatica*, *Bufo americanus* and *R. catesbeiana*, while for *H. crucifer* and *Psuedoacris triseriata* there was no significant difference. Generally this reflects the great variety in amphibian species with respect to egg production, reproductive modes and other demographic parameters (Salthe and Duellman, 1973; Kuramoto, 1978; Kaplan, 1980; Duellman and Trueb, 1986) as well as the diversity of adaptations to the different array of environmental selection and pressure. Phenotypic plasticity related to female age, size, trophic conditions and genetic factors also contribute to the observed egg size variation in amphibians (Berven, 1982; Rafinska, 1991). Hence it is not surprising that there exists significant difference in egg size between *R. cancrivora* and *R. limnocharis* in this study since, as mentioned earlier, a number of workers have concluded that within a reproductive mode, the greater the female size, the larger will be the ovum size (Salthe and Duellman, 1973; Kuramoto, 1978; Kaplan, 1980).

For *R. cancrivora* in this study, there exist positive correlations between female size and clutch size, female size and clutch weight, clutch size and clutch weight, and clutch weight and egg weight. Since body size is an indication of body condition, the high correlation between body size and clutch size is expected (Reading, 1986). Other workers also found that there is a positive relationship between clutch size and female body size (Pettus and Angleton, 1967; Salthe and Duellman, 1973; Collins, 1975; Howard, 1978; Kuramoto, 1978; Banks and Beebe, 1986; Gibbons and McCarthy, 1986; Reading, 1986). The positive relationship between clutch size and clutch weight,

and clutch weight and egg weight in *R. cancrivora* is also anticipated because generally speaking, the greater the egg number, the greater will be the clutch weight, and the greater the egg weight, the higher the clutch weight. Kuramoto (1978) also found the same phenomenon in some of his Japanese anurans and similarly Reading (1986) found that there was a trend as a whole in the population of British *B. bufo* for egg weight, egg number and ovary weight to increase with female size.

Collins (1975) reported that there was positive correlation between egg diameter and female body size in *R. sylvatica*, *H. versicolor* and *B. americanus* from Michigan. Banks and Beebe (1986) found that snout-vent length is correlated with egg size in *B. calamita* from England and suggested that this was, in part, an adaptation to survive in habitats with erratic pond provisions. However, in this study no correlation could be found between female body length and egg size (as quantified by egg weight) in both *R. cancrivora* and *R. limnocharis*, nor could Kuramoto (1978) find a relationship between female body weight and egg weight in *R. limnocharis*, *R. tagoi*, *R. brevipoda*, *H. japonica* and *Rhacophorus schlegelii*. He attributed this event to the fact that all these species are lentic water-breeding frogs that generally produced their egg output, not in smaller number of large eggs but in larger number of small eggs. Thus no obvious advantage is procured in producing large eggs by large females. Also Jorgensen (1981) reported that egg size is not directly related to body size in *R. temporaria* in Denmark, while Howard (1978) wrote that the average egg size for *R. catesbeiana* from Michigan is not correlated to female size. Rafinska (1991) found no correlation between egg size and snout-vent length in European *Bombina bombina*, and Collins (1975) also found no statistically significant correlation between egg diameter and female body size in *P. triseriata*, *H. crucifer* and *R. catesbeiana*. Why some species exhibited correlation between female size and egg size while others do not lack complete explanation at this point, and even Howard (1978) was convinced that the underlying reasons for such relationships remain unclear.

It is thus evident that variations occurring in the relationships of reproductive parameters in various amphibian species are results of selective forces and environmental constraints that shape their life history traits such that a species will optimize reproductive energy into producing the optimum number of offsprings to ensure future survival and continuance of the species. And since anuran species occupy various and diverse types of habitats and have different types

of reproductive strategies, they are thus subjected to various differences in environmental pressures to which they must adapt in order to survive and prosper. This then manifests into the huge variability of amphibian reproductive demography as what we are witness to today.

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Utilization of Energy and Material in Eggs and Post-hatching Yolk in an Oviparous Snake, *Elaphe taeniura*

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Abstract.—The duration of incubation of *Elaphe taeniura* eggs at 30±0.3°C averaged 54.9 days. During incubation, pliable-shelled eggs of *E. taeniura* increased in wet mass. Dried shells of the freshly laid egg averaged 17.6% of the entire egg dry mass. Freshly laid eggs had significantly heavier shells than did hatched eggs with the same wet mass at oviposition. Dry mass conversion from egg contents of the freshly laid egg to hatchling averaged 84.5%. During incubation, approximately 74.6% of non-polar lipids and 80.8% of energy in egg contents of the freshly laid egg were transferred to the hatchling, with 25.4% of non-polar lipids and 19.2% of energy used for embryogenesis. Shells from freshly laid eggs had higher levels of calcium and magnesium than did shells from hatched eggs. Fully developed embryos could obtain almost all magnesium from the yolk but withdraw approximately 35.6% of their total calcium requirements from the eggshell. A few days after hatching, a decrease in post-hatching yolk mass was accompanied by an increase in carcass mass, indicating that post-hatching yolk could be used to support early growth of hatchlings.

Key words.—Reptilia; Squamata; *Elaphe taeniura*; Incubation; Egg; Post-hatching yolk; Hatchling

Introduction

As for reproductive investments in eggs and embryonic development, there are two patterns that seem to be common in oviparous reptiles. One pattern is that total energy and material stored in eggs generally exceed the needs for producing a complete hatchling. Hence, a portion of yolk, namely post-hatching yolk or residual yolk, may remain unutilized at the time of hatching. Post-hatching yolk represents a supply of energy and material for early activities of hatchlings (Kraemer and Bennett, 1981; Troyer, 1983, 1987; Wilhoft, 1986; Congdon and Gibbons, 1989); however, the exact function of this portion of resources allocated by the mother in eggs is not very clear. There is a growing evidence showing that resources in the post-hatching yolk can be transferred to the carcass (=total hatchling-yolk sac-fat bodies). The consequence of this transference is that carcass increases in mass during the first post-hatching days of hatchlings (Ji et al., 1997a). The other pattern is that embryos must mobilize minerals (e.g., calcium) from the eggshell to complete development (e.g., Bustard et al., 1969; Jenkins, 1975; Packard and Packard, 1984, 1989; Packard et al., 1984a, b; Shadrix et al., 1994; Ji et al., 1996, 1997a, b). As the consequence of this mobilization, eggshell decreases in mass and ash contents during incubation, particularly at the late stage of incubation (Ji et al., 1996, 1997a, b; Zhao et al.,

1997). In this paper, we present data on a colubrid snake, *Elaphe taeniura*. We address the following topics: (1) conversion of energy and material from egg to hatchling during incubation, (2) sources of calcium and magnesium during embryogenesis, and (3) post-hatching yolk and its contribution to early growth of newly emerged hatchlings.

Material and Methods

Elaphe taeniura is one of the most common snakes in our study areas in the Zhoushan Islands (29° 32'-31° 04' N, 121° 30'-123° 25' E), Zhejiang, eastern China. The distributional range of *E. taeniura* covers most provinces of China (including Taiwan and Hainan), India (Darjeeling and Assam), Indochina, and the northern half of Malay Peninsula (Zhao and Adler, 1993). For this species, many aspects of biology species have been previously examined, but little information on incubation and reproduction is available other than incidental notes (see Huang and Jin, 1990).

Four gravid *E. taeniura* [snout-vent length: 110.0-155.0 cm; body mass (excluding the clutch): 233.4-778.7 g] were obtained from a private collector in Baiquan, Dinghai, the Zhoushan Islands, in mid-June 1994. The snakes were individually maintained in our laboratory in 80 × 80 × 80 cm wire cages until oviposition (mean=16.3 days). We removed eggs from the

cages, measured and weighed them within 6 h of oviposition, and then randomly selected two eggs from each of the first three clutches and one egg from the last clutch to determine egg composition. Egg contents (embryo plus yolk) of the dissected freshly laid eggs were removed, placed in pre-weighed small glass dishes, and weighed to the nearest 0.1 mg. Shells from the freshly laid eggs were rinsed briefly, weighed to the nearest 0.1 mg, and then were saved for later analysis. All dissected freshly laid eggs contained a small embryo, which was too small and fragile to be sampled separately, and therefore was included with yolk.

Eighteen eggs, 1/3-buried in moistened substratum, were incubated in a constant temperature chamber at $30 \pm 0.3^\circ\text{C}$. The incubation medium consisted of sand to water in a ratio of 4:1, and water was added periodically to keep the initial water content. We measured and weighed the incubating eggs at weekly intervals before day 42, and daily intervals thereafter. Four eggs failed to hatch following incubation. Hatchlings were measured and weighed immediately after they left the eggs. Shells from hatched eggs were rinsed briefly, weighed to the nearest 0.1 mg, and then were frozen for later analysis. Nine hatchlings (2-3 from each clutch; hereafter 0-day hatchling) were frozen immediately after hatching. The remaining 5 hatchlings (1-2 from each clutch; hereafter 7-day hatchling) were fasted at room temperatures ($26\text{--}38^\circ\text{C}$) for 7 days, and then frozen. The preserved hatchlings were later thawed, dissected, and separated into the carcass, yolk sac, and fat bodies.

All samples for determinations of non-polar lipids, ash, calories, calcium, and magnesium were oven dried to constant mass at 65°C , weighed, and then ground in a mortar and pestle. Non-polar lipids were extracted from all samples of egg contents, carcass, post-hatching yolk, and fat bodies for a minimum of 5.5 h using absolute ether in a Soxhlet apparatus. The mass of non-polar lipids in each sample was calculated as the difference in sample dry mass before and after extraction.

Ash and calories of samples of egg contents, carcass, post-hatching yolk, and fat bodies were determined using a GR-2800 adiabatic bomb calorimeter (Changsha Instruments). Titrations were performed of the residue after calorimetry to correct for nitrogenous wastes. Samples of eggshells were burned in a muffle furnace at 550°C for 24 h to determine ash mass.

Samples for calcium and magnesium determinations were weighed out into glass tubes and digested completely in hot concentrated nitric acid. Digestates were brought to volume in volumetric glassware and

stored in a refrigerator until analysis for calcium and magnesium. Concentrations of the two elements in the digestates were determined using a WFX-1B model atomic absorption spectrophotometer (The 2nd Beijing Optical Instruments). To check if there were any differences in calcium and magnesium contents between shells from freshly laid eggs and hatched eggs, we took equal amount of sample from each shell, pooled separately the samples from the freshly laid eggs and hatched eggs, and treated them as two different samples.

All variables were tested for normality using Kolmogorov-Smirnov test and for homogeneity of variance using Bartlett's test prior to further statistical analysis, and arc-sine transformation was performed for percentage data. We used analyses of variance (ANOVA), analysis of covariance (ANCOVA), regression statistics, and partial correlation analysis to analyze our data. Significance level was set at $\alpha=0.05$. Prior to testing for differences in adjusted means, the homogeneity of slopes was checked. Throughout this paper, values are presented as mean \pm 1 standard error.

Results

Elaphe taeniura laid pliable-shelled eggs. Clutch size in our sample averaged 8.8 ± 0.9 (range=8-11, N=4). Freshly laid eggs averaged 26.8 ± 0.6 g (range=20.8-32.3, N=35) wet mass, 54.1 ± 0.9 mm (range=45.7-63.2, N=35) length, and 28.8 ± 0.4 mm (range=25.0-33.1, N=35) width. During incubation, eggs increased in wet mass and, one day prior to hatching, weighed $110.0 \pm 3.5\%$ (range=98.6-135.9, N=14) of egg wet mass at oviposition. The incubation time averaged 54.9 ± 0.2 days (range=54.1-55.7, N=14). Newly emerged young averaged 17.1 ± 0.7 g (range=13.2-21.8, N=14) wet mass, 381.4 ± 3.5 mm (range=357.0-405.0, N=14) SVL, and 88.5 ± 1.9 mm (range=77.0-103.0, N=14) tail length.

The data on components of the freshly laid eggs and 0-day hatchlings are given in Table 1. Egg contents averaged 74.0% water by mass; egg contents averaged 92.4% organic material, 7.6% ash, 31.5% non-polar lipid, 1.36% calcium, and 0.39% magnesium by dry mass (Table 1). Shells from the freshly laid eggs averaged 17.6% of total egg dry mass, and 81.7% organic material and 18.3% ash by shell dry mass (Table 1). Shells from freshly laid eggs had higher levels of calcium (8.21%) and magnesium (0.75%) than did shells from hatched eggs (calcium: 6.31%; magnesium: 0.61%).

0-day hatchlings averaged 70.6% water by mass. These hatchlings averaged 89.1% organic material,

Table 1. Components and F values of the ANCOVA for 7 *Elaphe taeniura* freshly laid eggs and nine 0-day hatchlings. Data are expressed as adjusted mean \pm 1SE with total egg wet mass at oviposition as the covariate. Symbols immediately after F values represent significant levels: NS P>0.05, * P<0.05, ** P<0.01, and *** P<0.001.

	Freshly laid egg	Hatched egg	F
	Egg contents	Total hatchling	
Wet mass (g)	25.4 \pm 0.1	18.8 \pm 0.3	306.58***
Dry mass (g)	6.64 \pm 0.14	5.61 \pm 0.14	19.68***
Water (g)	18.8 \pm 0.2	13.3 \pm 0.2	402.18***
Organic mass (g)	6.14 \pm 0.13	5.00 \pm 0.13	28.25***
Ash mass (mg)	502.4 \pm 24.2	604.7 \pm 15.7	10.31**
Non-polar lipid(g)	2.09 \pm 0.07	1.56 \pm 0.05	30.99***
Calcium(mg)	90.1 \pm 4.2	139.9 \pm 5.4	31.96***
Magnesium(mg)	26.0 \pm 1.2	27.1 \pm 1.0	0.84 ^{NS}
Energy(Kcal)	39.6 \pm 0.8	32.0 \pm 0.8	33.41***
	Eggshell	Eggshell	
Dry mass(g)	1.42 \pm 0.04	1.21 \pm 0.05	7.82 [*]
Organic mass(mg)	1.16 \pm 0.03	1.03 \pm 0.04	4.13 ^{NS}
Ash mass (mg)	259.8 \pm 10.5	180.8 \pm 8.2	22.86***

10.8% ash, 27.8% non-polar lipids, 2.49% calcium, and 0.48% magnesium by dry mass (Table 1). Shells from hatched eggs averaged 85.1% organic material and 14.9% ash by dry mass (Table 1).

0-day hatchlings contained significantly lower quantities of total dry mass, organic mass, non-polar lipids, and energy, but significantly higher quantities of calcium and ash mass than did egg contents (Table 1). There was no significant difference in the quantity of magnesium between egg contents and 0-day hatchlings (Table 1). Shells from hatched eggs contained lower quantities of total dry mass and ash mass than did shells from the freshly laid eggs. No significant difference in organic mass was found between shells from the freshly laid eggs and hatched eggs (Table 1).

During incubation, approximately 84.5% of dry mass, 74.6% of non-polar lipids, and 80.8% of energy in egg contents of the freshly laid egg were transferred to the

hatchling, with 15.5% of dry mass, 25.4% of non-polar lipids, and 19.2% of energy used for embryogenesis (Table 1). Fully developed embryos could obtain almost all magnesium from the yolk, but should withdraw 35.6% of their total calcium requirements from sources other than yolk (Table 1).

Egg contents (1.36 \pm 0.07%, range=1.14-1.62%, N=7) of the freshly laid egg had a higher level of calcium than did post-hatching yolk (0.99 \pm 0.10%, range=0.64-1.45%, N=9) (ANOVA: $F_{1,14}$ =8.38, P<0.05). There was no significant difference in the level of magnesium between egg contents (0.39 \pm 0.03%, range=0.32-0.45%, N=7) and post-hatching yolk (0.42 \pm 0.05%, range=0.27-0.62%, N=9) (ANOVA: $F_{1,14}$ =0.54, P>0.05).

Dry masses of carcass (r^2 =0.96, $F_{1,7}$ =157.76, P<0.001) and fat bodies (r^2 =0.88, $F_{1,7}$ =55.29,

Table 2. A comparison between nine 0-day and five 7-day hatchlings of *Elaphe taeniura*. Data are expressed as mean \pm 1 SE; all mass units are in grams.

	0-day hatchling	7-day hatchling
Hatchling wet mass at hatching	18.8 \pm 0.9	15.7 \pm 0.3
Hatchling wet mass 7 days after hatching	—	15.0 \pm 0.2
Decrease in wet mass	—	0.70 \pm 0.22
Hatchling dry mass	5.28 \pm 0.31	4.00 \pm 0.06
Carcass	3.51 \pm 0.20	3.18 \pm 0.05
Yolk sac	0.66 \pm 0.05	0.09 \pm 0.01
Fat bodies	1.11 \pm 0.09	0.73 \pm 0.02
% water of hatchling	70.6 \pm 0.5	73.2 \pm 0.3

$P < 0.001$) of the 0-day hatchlings were both correlated with total hatchling dry mass. In the 7-day hatchlings, we only found a positive correlation between carcass dry mass and total hatchling dry mass ($r^2 = 0.88$, $F_{1,3} = 22.34$, $P < 0.05$). 7-day hatchlings had significantly heavier carcasses than did 0-day hatchlings with the same wet mass at hatching (ANCOVA: $F_{1,11} = 16.38$, $P < 0.01$) (Table 2). There was a negative correlation between post-hatching yolk dry mass and carcass dry mass when holding total hatchling dry mass and fatbody dry mass constant ($r = -0.90$, $t = 6.53$, $df = 10$, $P < 0.001$). There was no significant correlation between post-hatching yolk dry mass and fatbody dry mass when holding total hatchling dry mass and carcass dry mass constant ($r = 0.43$, $t = 1.50$, $df = 10$, $P > 0.05$). There was no significant correlation between carcass dry mass and fatbody dry mass when holding total hatchling dry mass and post-hatching yolk dry mass constant ($r = -0.20$, $t = 0.65$, $df = 10$, $P > 0.05$). 7-day hatchlings (23.2 \pm 0.4%, range=22.1-24.1%, $N = 5$) had significant lower levels of non-polar lipids than did 0-day hatchlings (27.4 \pm 0.6%, range=25.3-29.5%, $N = 9$) ($F_{1,12} = 32.1$, $P < 0.001$).

Discussion

Similar to that reported for pliable-shelled eggs of other reptiles (e.g., Fitch, 1954; Fitch and Fitch, 1967; Andrews and Sexton, 1981; Vitt and Cooper, 1986; Vleck, 1991; Ji et al., 1996, 1997a, b), eggs of *E. taeniura* overall increased in wet mass and swelled during incubation due to a net gain of water absorbed

from the substrate on which the eggs were incubated. However, water uptake seemed not to be obligate for *E. taeniura* eggs, because some eggs whose final mass was less than initial mass also hatched successfully.

Small *E. taeniura* embryos were present in all freshly laid eggs, but they, relative to the large egg size, were too small to be considered as an important part of the egg at oviposition. Therefore, the transference of energy and material from egg to hatchling during incubation was approximately equal to the transference overall. This makes it possible to compare our data with those for other oviparous reptiles whose freshly laid eggs also contain small embryos and embryonic stage is near the oviparous end in the oviparity-viviparity continuum (Shine, 1983). *Elaphe taeniura* exhibited high conversion efficiencies of energy and material from egg to hatchling. The values in Table 3 show that the conversion efficiencies of energy and material recorded in *E. taeniura* were higher than those reported for any other studied reptiles. The values in the Table also show that the conversion efficiencies vary considerably among species; however, the explanations to these differences are unknown at this time. It has been known that costs of embryonic development vary considerably among reptiles (Dmi'el, 1970; Black et al., 1984), parental investment in each offspring should be related to its survivorship (Congdon and Gibbons, 1989; Fischer et al., 1991), and incubation environments may influence embryonic development (Gutzke and Pachard, 1987). So, further studies in a wider field covering parental reproductive investment, embryonic metabo-

Table 3. Comparison of conversion efficiencies of dry mass, non-polar lipids, and energy between *Elaphe taeniura* and other oviparous reptiles.

Species	Conversion efficiency (%)			Data resources
	Dry mass	Non-polar lipids	Energy	
Lizards				
<i>Podarcis muralis</i>	75	46	61	Ji & Braña, submitted
<i>Eumeces chinensis</i>	66	44	62	Ji et al., 1996
Snakes				
<i>Elaphe taeniura</i>	85	75	81	This study
<i>Elaphe carinata</i>	81	64	72	Ji et al., 1997a
<i>Dinodon rufozonatum</i>	81	70	79	Ji et al., submitted
<i>Ptyas korros</i>	77	54	69	Ji et al., submitted
<i>Xenochrophis piscator</i>	74	52	66	Ji et al., submitted
<i>Rhabdophis tigrinus lateralis</i>	70	37	61	Zhao et al., 1997
<i>Zaocys dhumnades</i>	76	63	70	Ji, unpubl. data
<i>Naja naja atra</i>	75	64	69	Ji et al., 1997b
Turtles				
<i>Chelydra serpentina</i>	—	60	60	Wilhoft, 1986
<i>Deirochelys reticularia</i>	72	—	—	Congdon et al., 1983
<i>Chrysemys picta</i>	72	—	—	Ewert, 1979
Alligator				
<i>Alligator mississippiensis</i>	79	74	—	Fischer et al., 1991

lism, and ecology of neonates will be very important for our giving reliable explanations.

As in other oviparous squamates (Packard et al., 1984a, 1985; 1988; Ji et al., 1996, 1997a, b), turtles (Packard et al., 1984b; Packard and Packard 1986), and the American alligator (Packard and Packard, 1989), *Elaphe taeniura* embryos use eggshell as a secondary source of calcium. The result that eggshells decreased in mass and calcium content during incubation supports this interpretation. The level (35.6%) of calcium withdrawn by *E. taeniura* embryos from the eggshell was much lower than the values reported for crocodylians and turtles (50-80%; Bustard et al., 1969;

Jenkins, 1975; Packard and Packard, 1984, 1989). In squamates, the level was slightly lower than that reported for *Eumeces fasciatus* (39%; Shadrix et al., 1994) but higher than those reported for *Eumeces chinensis* (19%; Ji et al., 1996), *Coluber constrictor* (20%; Packard et al., 1984a), *Elaphe carinata* (31%; Ji et al., 1997a), and *Naja naja atra* (14%; Ji et al., 1997b). These differences presumably reflect the interspecific differences in eggshell structure and allocation of minerals between eggshell and yolk.

The level of calcium in *E. taeniura* post-hatching yolk was significantly lower than that in egg contents of the freshly laid egg. This suggests that *E. taeniura*

embryos deplete the yolk of almost of its calcium before hatching and none of the calcium withdrawn from the eggshell is deposited in the yolk. This pattern of mobilization and deposition of calcium is similar to that observed in other non-crocodilian reptiles (e.g., Packard et al., 1984b, 1985, 1987; Packard and Packard, 1986; Packard and Packard, 1988; Shadrix et al., 1994).

The estimated amount of magnesium in egg contents of the freshly laid egg (95.9% of total magnesium in the hatchling) was slightly less than that in the hatchling. We are presently not very certain that *E. taeniura* embryos use the eggshell as an additional source of magnesium, because any slightly biased estimation might account for the remaining 4.1% of magnesium. However, the fact that shells from hatched eggs were lighter in mass and lower in the level of magnesium seemed to imply that *E. taeniura* embryos should withdraw a small portion of magnesium from the eggshell. Since studies of embryonic magnesium metabolism have been unfortunately extremely limited, we cannot discuss this problem in detail. In the American alligator (Packard and Packard, 1989) and other species of snakes that have studied by us, embryos apparently obtain all magnesium necessary for development from the yolk.

One interesting finding in this study was that a decrease in post-hatching yolk mass was accompanied by an increase in carcass mass a few days after hatching. This finding quantitatively confirms that post-hatching yolk can be used to support early growth of hatchlings. Compared with post-hatching yolk, fat bodies were used mainly for hatchling maintenance. An obvious decrease in the level of non-polar lipids in the 7-day hatchlings supports this interpretation.

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Redescription and Generic Redesignation of the Ladakhian Gecko *Gymnodactylus stoliczkai* Steindachner, 1867

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Abstract.— *Gymnodactylus stoliczkai* is redescribed on the basis of recently collected topotypes, and is redesignated to the genus the *Cyrtodactylus*. It is found to be morphologically distinct from *C. walli* and *C. yarkandensis* with which species it has long been synonymized. Reproduction, ecology and distribution of this highland gecko is discussed.

Key words.— *Cyrtodactylus stoliczkai*, taxonomy, reproduction, ecology, zoogeography.

Introduction

In 1866 Dr. Ferdinand Stoliczka deposited a collection of animals, collected from different parts of the Indo-Pak subcontinent, in Naturhistorisches Museum Wien, Austria (NMW). The collection contained a gecko from Karoo, north of Dras, Kashmir, apparently collected during geological survey of western Himalayas, which was described by Steindachner in 1867 as a new taxon, *Gymnodactylus stoliczkai*, honoring its collector. The specimen is still available in the museum under registry number NMW 16756 (Tiedemann et al., 1994). Ever since the description of this taxon, it has become a habit with herpetologists working on collections from northern Pakistan, to try to place almost every angular-toed gecko encountered, in the synonymy of *G. stoliczkai* without going into details of morphological comparisons: Smith, 1935 (*Gymnodactylus walli* Ingoldby, 1922); Minton, 1966 (*Cyrtodactylus mintoni* Golubev and Szczerbak, 1981) and Mertens, 1969 (*Cyrtodactylus dattanensis* Khan, 1980), creating taxonomic chaos. Recent collections from circum-Himalayan region has shown that *G. stoliczkai* does not belong to the fauna of Pakistan, moreover, all the geckos placed in its synonymy are themselves valid independent taxa (Khan, 1992, 1993, 1994; Khan and Baig, 1992).

A long, detailed morphological redescription of *Gymnodactylus stoliczkai* is urgently due, to make intertaxal comparisons possible and to come out of “*stoliczkai* myth”. Due to lack of working material the present project has considerably been delayed. The type specimen (NMW 16756) and syntype in the Museum of Comparative Zoology, Cambridge (MCZ 7132) are not allowed to be loaned for study. Therefore the data for present study is drawn from several sources: one of us (HR) has studied series of 14 topo-

types of this species in Zoologische Staatssammlung Munchen (ZSM: Table 1; collected by Gruber, 1981), while Dr. G. R. Zug (National Museum of Natural History, Washington) has kindly taken data on topotype MCZ 7132 for us. Moreover, present redescription is further supplemented with the data available in literature (Steindachner, 1867:15; Boulenger, 1890:63; Smith, 1935:57; Constable, 1949:84; Szczerbak and Golubev, 1986:205). Photographs of topotypes (Fig. 1: ZSM 124.77) and (Fig. 2: ZSM 45.77), and type NMW 16756 (Khan, 1994; Fig. 2) have helped us immensely to understand morphology of this important taxon.

Taxonomic Notes

Sprix's (1825:17) genus *Gymnodactylus* included all the then known non dilated angular-digited geckos (Boulenger, 1885:22, 1890:59; Annandale, 1913:309; Smith, 1935:37). Until Underwood (1954) restricted this genus to South American angular-digited geckos, placing all south Asian geckos in the genus *Cyrtodactylus* Gray 1827. Most of the subsequent workers on the herpetology of Pakistan have followed Underwood's view point (Minton, 1966; Khan, 1980; Khan and Mirza, 1977). However, Mertens (1969) is orthodox and a bit cautious by retaining *Gymnodactylus* as a genus and placing Pakistani geckos in the subgenus *Cyrtodactylus*.

A recent break through towards a solution comes from Szczerbak and Golubev (1984, 1986): the genus *Tenuidactylus* is erected to include Palearctic angular-digited geckos. It is divided in three subgenera to accommodate the rest of the southeast Asian gekkota: subgenus *Tenuidactylus* includes two Pakistani species *T. montiumsalsorum* and *T. kohsulainanai*, and the group of Tibeto-Himalayan species: *T. tibetanus*,

Table 1. Range of scale counts and measurements of topotype series of *Tenuidactylus stoliczkai* in Zoologische Staatssammlung Munchen (ZSM) collection (data from juvenile ZSM 119/77 not taken into account).

Character	Range
Supralabials	9-11
Infralabials	7-9
Nasals	3-3
Internasals	1-3
Postmentals	2-2
Loreals	10-15
Scales between eye-ear	15-20
Interorbitals	17-19
Tubercle rows across body middorsum	8-12
Sale rows across midbelly	25-30
Midventrals	110-133
Lamellae under first toe	11-14
Lamellae under fourth toe	22-27
Cloacal spines	2-3
Granular scale rosette around dorsal tubercle	7-9
SVL	27-49.4 mm
TL	25-49.7 mm
Head length	8-11.5 mm
Head width	5-10.7 mm
Head height	3-6.1 mm
Nostril-eye distance	2-4.4 m
Eye-ear distance	2-4.1 mm
Body length	10-23.6 mm
Eye diameter	2-2.8 mm
Ear diameter	0.4-0.9 mm
Head length/head width	0.99-1.43 mm
SVL/body length	2.09-2.64 mm
Head length/head width	0.99-1.43 mm
Distance nostril-eye/eye-ear	0.98-1.29 mm
Eye/ear diameter	2.33-5 mm



Figure 1. *Cyrtodactylus stoliczkai* (ZSM 124.77), adult female, with unregenerated tail.

T. mintoni, *T. chitralensis*, *T. stoliczkai* and *T. kirmanensis*; the genus *Cyrtopodion* Fitzinger, 1843 is resurrected as a second subgenus to include four Pakistani forms: *agamuroides*, *scaber*, *watsoni* and *kachhensis*. A third subgenus *Mediodactylus* is left floating (Golubev, pers. comm., 1996).

Undoubtedly southeast Asian cyrtodactylid geckos are morphologically distinct from Palearctic tenuidactylids (Leviton and Anderson, 1970; Khan, 1988, 1989, 1991; Khan and Tasnim, 1990). Khan (1993) maintains the genus *Cyrtodactylus* Gray, 1827, to include Tibeto-Himalayan species group and all the southeast Asian species (Smith, 1935). The southeast Asian cyrtodactylids are a very heterogeneous assemblage of closely allied species. Their shared characters are: smooth tubercular granular scales with scattered round-oval smooth or slightly keeled tubercles on head and body dorsum; more than 25 heterogeneous interorbitals; subcylindrical and subequal body and tail; dorsal vivid pattern; subdigital lamellae about twice as broad as high with a pair of lateral row of granular scales, lamellae not swollen at the digital angles; 2-10 preanal pores in male, rare femoral pores; small blunt caudal tubercles, subcaudals small rarely broad. In the past there have been several attempts to arrange them in a logical array (Annandale, 1913; Smith, 1935; Khan, 1993). Considering morpho-ecogeography of these geckos, we distinguish two lineages:

Circum-oceanic group: tropical, scattered along sub continental coastal strip and oceanic islands, confined between lat. 7-32° N, long 75-105° E; dorsal pattern of vivid cross bars or spots, dorsal granular scales mixed with larger rounded, smooth or slightly keeled tubercles, tail and body cylindrical, tail often longer

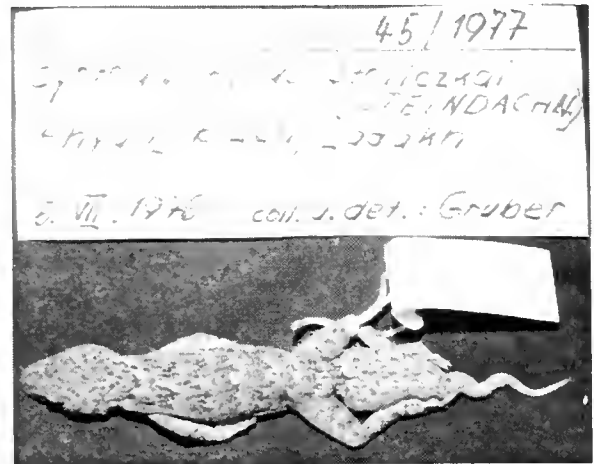


Figure 2. *Cyrtodactylus stoliczkai* (ZSM 45.77), adult female, with regenerated tail.

than body: *Cyrtodactylus pulchellus*, *intermedius*, *consobrinoides*, *angularis*, *khasiensis*, *rubidus*, *tri-drus*, *nebulosus*, *collegalensis*, *dekkanensis*, *albofasciatus* and *jayporensis*.

Circum-Himalayan group: subtropical, highland forms, mainly extending between lat. 34-4° N, long 75° 50-50'E, tubercular, beady, scarcely imbricate granular dorsal scales, interspersed by 2-3 times larger oval keeled or keelless tubercles arranged in more or less in 12-13 longitudinal rows; body and tail subequal and subcylindrical, subcaudals small in several rows, inconspicuous dorsal pattern of transverse bands, spots or reticulation.

Further distinguished in three subgroups:

stoliczkai subgroup: body and tail rather flat in cross section, caudal tubercles flat, smooth; anterior half of tail segmented, segments laterally lobulated in older animals, regenerated tail flattened and abnormally swollen, no preanal and femoral pores. Includes highland species: *Cyrtodactylus lawderanus*, *C. stoliczkai*, *C. yarkendensis* and *C. baturensis*.

tibetinus subgroup: Body and tail round in cross section, tail segments not distinct, 4-10 preanal pores, feebly keeled caudal tubercles, regenerated tail not flattened. Dorsal pattern of vivid cross bars, spots or reticulation. Includes Tibeto-Himalayan low altitude submountain geckos: *Cyrtodactylus tibetinus*, *C. himalayanus*, *C. mintoni*, *C. dattanensis* and *C. battalensis*.

walli subgroup: Body flatter, tail quadrangular in cross section, distinctly segmented, caudal tubercles large slightly keeled, median row of subcaudals transversely enlarged; 4-6 preanal pores. Species included are: *Cyrtodactylus kirmanensis* and *C. walli*.

Table 2. Comparison of morphology of 1= *Tenuidactylus yarkandensis* (J. Anderson, 1872) with its closest congeners: 2= *T. stoliczkai* (Steindachner, 1867), 3= *T. walli* (Ingoldby, 1922) and 4= *T. baturensis* Khan and Baig, 1992; vl=vertical, (measurements in mm).

Character	1	2	3	4
Snout vent length	47.15	46-48	46-54	44-53
Tail length	? (lost)	49	78	55-57
Supralabials	11/10	10-13	9-11	9-11
Infralabials	8/8	7-8	8-10	7-9
Subdigital lamellae under 4th finger	18	22	19-20	20-21
4th toe	25	25	23-25	24-27
Interorbitals	15-16	17-20	20-21	17-20
Scale rows across midbelly	29-30	26-27	38-40	26-30
Midventral scales	138-140	142-149	160-172	158-171
Prenal pores	?	?	4	?
Head length	11.5	10.8	11-14	13.3
height	5.3	5.2	5-5.75	5.9
breadth	9.3	9.6	9-10.5	10.1
Ear diameter	0.8 (vi)	1.1	1-2	1.5
Eye diameter	2.4	2.9	2-3	3.3
Number of cross bands on body dorsum	7	8	8-9	8

Redescription

Cyrtodactylus stoliczkai (Steindachner, 1867)

Gymnodactylus stoliczkai Steindachner, 1867, *Reise Novara, Zool.*, 1:15 Rept. 1, Plate 2, Fig. 2, 2a.

Cyrtodactylus stoliczkai Underwood, 1954, *Proc. Zool. Soc. London*, 124:475.

Type locality: near Karoo, north of Dras, Kashmir.

Holotype: NMW 16756 (Fig. 1), female, near Karoo, north of Dras, 3200 m, Kashmir (34° 28' N, 75° 46' E), donated by Stoliczka in 1866.

Paratypes: MCZ 7132, female, pholidosis counts and measurements by Dr. Zug, (per comm., 1989) Ladakh, 3100 m, Stoliczka, 1908.

Topotypes (14, examined): ZSM 45.77 (1, 2, 3, 4, 5) and 149.77 (1, 2, 3, 4, 5), 2 males, 5 females, three

juveniles, Phyang Ghompa, Ladakh, 3600 m, 5.7.77 and 19.8.77; ZSM 46.77, and 124.77, 2 females, Hemis, Ladakh, 3700 m, 6.7.77 and 17.8.77; ZSM 119.77, juvenile, Kargiul, Ladakh, 2750 m, 12.8.77; ZSM 121.77, female, Saspool, Ladakh, 3100 m, 16.8.77, all collected by Ulrich Gruber (1981).

Diagnosis: Body and tail moderately depressed, tail a little longer than body, caudal segments of unregenerated tail deeply sected on lateral sides to anterior half of the tail, three tubercles, and several series of small subcaudals arranged in four transverse rows; body dorsum with flat, mostly juxtaposed granular scales, distinctly arranged in transverse rows, interspersed with large flat smooth oval tubercles, about three times larger than granular scales, more or less arranged in 9-10 transverse rows across middorsum and 19-20 along paravertebral line; 16-20 interorbital tubercular scales; 27-32 scales across midabdomen, 117-150 midventrals; both preanal and femoral pores not indicated; dorsal pattern of M-shaped transverse

dark bands with heavier posterior margin, broader than interspaces.

Description of holotype NMW 16756, Fig. 1, (statements in parenthesis are from paratype and topotypes adding up to the original description of holotype NMW 16756 by Steindachner, 1867:15): (habitus depressed); rostral scale big (7-angular, broader than deep), slightly convex at the upper edge, and forked in the middle reaching the anterior end of the snout (median dorsal rostral longitudinal furrow narrowly misses anterior border of the rostral scale); nasal opening (small, dorsolateral) bordered in front by rostral plate, (ventrally by) second upper lip shield (first supralabial. Note rostral shield is regarded as first supralabial in original description), posteriorly by three small nasal shields, of which upper most is the largest (separated from that of other side by a pair of granular scales, rostral area with heterogeneous tubercular scales mostly arranged in longitudinal rows, 11-12 tubercular loreal scales between posterior nasals and anterior rim of orbit; head with heterogeneous granular tubercular scales, 19-20 between orbits arranged in longitudinal rows, smaller on eye bulgings; a series of sharp supraciliary scales jutting out from posterior half of the upper eyelid border followed by a row of large tubercular round scales running along the eyelid; temporals and neck with small tubercular granular scales; ear opening round, much larger than largest dorsal tubercle).

19-21 upper lip shields (10-11 supralabials, first three of the same size; suture between first supralabial and rostral scale almost equals former's length along oral orifice); 13 lower labial shields (7-9 infralabials, second narrowest while fourth the longest; 2-3 rows of sublabials). The anterior most lower lip shield (mental scale) is very large, triangular (about twice deep as broad, extends deep between first pair of postmentals), three pairs of chin shields (first pair, largest, narrowly in contact with each other behind posterior tip of mental scale, second pair smaller, less than half the size of first not in contact with each other), third pair (smallest) almost fully separated from lower labials (on right while in contact on left).

Body dorsum clearly granulated (with flat, juxtaposed, rarely slightly imbricate granular scales, arranged in transverse rows) with numerous, only little bigger fully rounded tubercles (interspersed with 2-3 times large, flat, smooth, round or oval tubercles, scattered evenly on body dorsum, more or less arranged in longitudinal rows, separated from each other by 3-4 granular scales, a rosette of 7-9 granules around a tubercle; tubercles on sides slightly conical).

(Gular scales flat, juxtaposed or slightly imbricate, mostly arranged in transverse rows interspersed with larger scattered tubercles, becoming flatter and pentagonal on chest, at abdomen flat, hexagonal, broader than long, slightly imbricate rarely juxtaposed, arranged in transverse rows, 30-32 scales across midabdomen; slightly marked lateral abdominal folds, 5-6 rows of lateral abdominal scales differ little from dorsals, however clearly marked from abdominals; femoral and preanal pores absent (even their site not marked by distinct scales; 142-149 midventral scales between first pair of submental and anterior anal lip. A pair of lateral cloacal tubercles, no postanal bulge).

(Limbs medium) sized, covered with imbricate smooth scales arranged in transverse and longitudinal rows, without tubercles; subfemorals in 5-6 transverse rows, as large as abdominals; median-subtibial scales largest, imbricate, arranged in transverse rows; granular postfemorals extend on to the sides of preanals, with no tubercles; tips of finger-claws extend to anterior of eye, when forelimbs are adpressed forward, those of toes a little anterior of axilla; subdigital lamellae 23-24 under fourth toe, equally broad throughout, not enlarged under basal and angular parts of digits).

Tail, as it appears in the specimen examined by us, is regenerated in the anterior, somewhat longer than the body. Lined on each (dorso-lateral) side by three rows of large, spike shaped raised tubercles, of which those of lower most row are the biggest and conical. No large transverse plates under side of the tail (regenerated?, this parenthesis is by Steindachner), posterior half of the tail is with uniform scales (morphologically tail of the holotype reminds original tail of older specimens of *Hemidactylus flaviviridis*. Doubtless the type specimen has original tail which is normal for older animals of the species as in ZSM 124.77. The tail is moderately depressed, its stump is less than half the width of body and the segmented part almost equals body's width, a median dorsal and ventral longitudinal slight furrow runs along its length. Its anterior half is divided in 10 distinct laterally lobulated segments, while posterior unsegmented half gradually narrows to sharp terminal tip; dorsolaterally caudal tubercles are given from the middle of the segments, four on 1st to 5th segment, three on 6th to 10th. The dorsal tubercles are small roundish about 2-3 times smaller than laterals which are elongated conical blunt, in contact with each other, are gradually reduced in size, until almost indistinct in posterior half of the tail. Dorsally 6-7 transverse rows of heterogeneous, slightly imbricate, smooth, tubercular scales cover anterior caudal segments while 4 transverse

rows of small imbricate subcaudals are present on ventral side of segment. In the posterior quarter dorsal and ventral caudal scales become indistinct from each other, get longer, flatter, pointed at tips and are strongly imbricate).

Color: Body dorsum light blue grey, with pink edged transverse bands with denser wavy posterior edges, broader than interspaces, three on nape, six on body and 13 on tail (in preserved specimens bands are dark and does not extend on subcaudals). (Head), labials and tail plates with fine grey dots, limbs and digits barred, ventrum light.

Measurements (in mm): Snout vent length 48, tail length 52, trunk length 24, head length 11.5, head width 10, head height 7, eye diameter, not including bony orbit 2.5; snout length 5; oculo-orbital space 5.

Variation: Table 1 presents the measurements and pholidotic counts in ZSM series which fall within the range of the type specimen NMW 16756 and paratype MCZ 7132, differing in minor details of pholidotic morphology: rostral scale is protruberant in most of the specimens, however, in some it is flat, the median rostral groove in some specimens extend to middle, while in other it narrowly misses the anterior end of the scale; the supraciliary pointed scales vary in their pointedness; dorsal tubercles uniformly scattered on dorsum, 2-3 times larger than dorsal granular scales, separated from each other by 2-4 granular scales, surrounded by a roset of 6-9 granular scales; lateral abdominal folds strongly or poorly indicated.

Study of tail morphology of MCZ 7132 and specimens in ZSM series indicates that the tail of the holotype NMW 16756 is undoubtedly original. Steindachner (1867) himself was doubtful about its being "(regenerated?)". In young specimens with unregenerated tail, the tail is uniformly broad from basal stump till mid-tail, where it gradually tapers to its tip (ZSM 45.77:5, 119.77, 124.77, 149.77: 1, 2, 4, 5). As animals get older (NMW 16756 and ZSM 124.77), the anterior half of the original tail becomes broader, flatter and deeply sected on sides so much so it appears laterally lobulated at segments. From lateral lobes caudal tubercles strongly jet out. While in regenerated tail, MCZ 7132 and ZSM 49.77, the tail swells up almost round at the base, with no indication of segmentation, lobulations and tubercles, while posteriorly it abruptly tapers. Moreover, instead of transverse bands of original tail, the regenerated tail is spotted with longitudinal spots, scattered all over it. Tail in ZSM 45.77, 49.77:3, 45.77, 121.77 and ZSM 49.77 represent different stages of tail regeneration in *Tenuidactylus stoliczkai*.

Dorsal pattern of bands is vividly distinct in juveniles (ZSM 119.77), but is gradually lost as the animal grows older.

Sex: Though dissection is the sure way of sex determination of geckos, however, presence of preanal and femoral pores in males and their absence in females are almost universal sex indicators in these animals. In the type specimen of *Tenuidactylus stoliczkai* NMW 16756, paratype MCZ 7132 and ZSM series the preanal and femoral are absent, moreover their position is not indicated by distinctiveness of scales in the area. On the other hand, swollen postanal sacs are usually distinct in male less so in female geckos (Smith, 1933, 1935; Khan and Baig, 1992). There is no indication of postanal sacs in type nor in paratype, however, are well indicated in two specimens in ZSM series which are males. Constable (1949:84) designated MCZ 7132 as a male specimen, which on dissection is proven to be an adult female with well developed vitellogen follicles (Zug, personal communications, 1989).

Ecology: Ladakh lies around 3000 m, above timberline. It is completely dry snow desert, with sparse vegetation of herbs, shrubs and grasses. The area is highly arid with sub-tropical continental highlands cold climate. Heavily snowy winters, getting rain in winter and spring. Maximum summer July temperature is 24.7° C, minimum 10.2°, while maximum winter temperature in January is -1.4° C, dropping to minimum -13.3°. Maximum rain fall, 15.0 mm, is received during August, minimum, 1.0 mm during November (Ahmad, 1951).

Gruber (1981) collected *T. Stoliczkai* from rocky habitat, where this gecko prefers desert, bare, dry situations in the non-irrigated areas without or with very sparse vegetation, apparently avoiding direct neighborhood of human settlements. Other reptiles collected from the area are *Phrynocephalus theobaldi*, *Laudakia himalayana*, and *Scincella ladacensis*, while *Bufo latastii* is the only amphibian recorded from waters of the area.

Comparison with congeners: Absence of trihedral tubercles, preanal and femoral pores, broader subdigital lamellae and peculiar tail morphology differentiate *Tenuidactylus stoliczkai* from Palearctic group of Pakistani tenuidactylid *Cyrtopodion* geckos: *C. scaber* (Heyden, 1827), *C. watsoni* (Murray, 1892), *C. kachhensis* (Stoliczka, 1872), *C. montiumsalsorum* (Annandale, 1913) and *C. kohsulaimanai* (Khan, 1991). While Pakistani members of Tibeto-Himalayan group of cyrtodactylid geckos: *C. mintoni* (Golubev and Szerbak, 1981), *C. dattanensis* (Khan, 1980) *C. battalensis* Khan, 1993 are similar to *T. stoliczkai* in the morphology of subdigital lamellae, body configu-

ration, larger number of interorbitals, subabdominals, however, they differ markedly from it because of their plump rounded body and tail, tail morphology, dorsal tuberculation and pattern which extends on the tail ventrum, presence of preanal pores in male.

The tenuidactylid group of Pakistani geckos: *T. indusoani* (Khan, 1988), *T. rohtasfortai* Khan and Tasnim, 1990 and *T. fortmunroi* Khan, 1993 are distinguished from the nominated species by their different body configuration, smaller number of interorbitals, subabdominal pholidotic counts, dorsal scalation and pattern, caudal morphology with keeled large tubercles, single row of subcaudals, presence of both preanal and femoral pores.

Following Annandale (1913:316), Smith (1935:39) grouped *T. stoliczkai* with low altitude *T. lawderanus* (Stoliczka, 1972), despite very obvious differences from the former: more flattened body and tail, few and feebly developed dorsal tubercles, inconspicuous dorsal pattern, single pair of nasal scales, presence of preanal pores in male, tail morphology (compare Fig. 18, Smith, 1935).

C. yarkandensis (J. Anderson, 1872) and *C. walli* (Ingoldby 1922), have long been synonymized with *C. stoliczkai* and *C. chitralensis* (Smith, 1935), mostly due to their similar dorsal pattern, body configuration, dorsal tuberculation and absence of pores (Boulenger, 1885; Blanford, 1878; Annandale, 1913; Smith, 1935; Minton, 1966; Mertens, 1969; Szczerbak and Golubev, 1986). *C. chitralensis* has been found to be conspecific with *C. walli* (Khan, 1992).

Cyrtodactylus stoliczkai is defined by the following combination of characters: dorsal granular scales smooth, round, beady, juxtaposed, interspersed with oblong, smooth beady tubercles, arranged roughly in longitudinal rows; normal tail in juveniles uniformly broad from basal stump till mid-tail, where gradually tapering to tip, as animal gets old, the anterior half of the original tail becomes broader, flatter and deeply sected on sides in lateral lobules at segments, regenerated tail much swollen and rounded; subcaudals small, in several rows; caudal tubercles thick and blunt.

Geographical Distribution

All evidence goes in favor of the idea that the family Gekkonidae evolved in southeast Asia and dispersed westward through southwestern Asia into Indo-Pakistan and Africa (Kluge, 1967). Ranges of several circum-Indian oceanic cyrtodactylids overlap, however circum-himalayan highland geckos are widely distributed and the ranges of none of them are known to overlap: the northern most extralimital, *Cyrtodactylus*

yarkandensis, is known from lat. 38° 40'N, long 77° 50'E, along the western border of China's Xinjiang Province (Khan, 1994) and *Cyrtodactylus stoliczkai* is confined to Ladak, between lat. 34°-35° 45' N, long 75° 50'-76° 70'E (Annandale, 1913; Schmidt, 1922; Gruber, 1981). A highland Pakistani gecko, *C. baturensis*, is reported from Gilgit Agency at lat. 36° 20'N, long 74° 50' E (Khan and Baig, 1992). The western most Pakistani form, *C. walli*, occurs between lat. 35-36° N, long 71-72° E, while the western extralimital Iranian gecko, *C. kirmanensis*, is reported from lat. 30° N, long 58° E (Szczerbak and Golubev, 1986).

Worldwide distribution of some geckos is largely known to be due to human transportation (Darlington, 1957). The circum-oceanic coastal forms are characteristically carried by sailors from coast to coast, however, some of the species penetrated deep inland. Similarly, westward dispersal of *Cyrtodactylus* in the lower Himalayas appears largely due to westward migrations of Buddhist peoples and seasonal nomadic high-low-high altitude migrations which played important role in the distribution of cyrtodactylid geckos of the area. Massive pagoda buildings and temples are still the frequent haunts of these geckos. In the valley-complex of the sub Himalayan system they evolved into closely allied forms: *C. mintoni*, *C. dattanensis* and *C. battalensis*. A parallel example of human transportation of a gecko is presented by the recent record of Palearctic geckos, *Hemidactylus persicus*, from Rohtas Fort, Jhelum valley, Punjab, Pakistan (Khan and Tasnim, 1990). The fort was built from 1542 to 1550 with rock-blocks transported from Balochistan. *H. persicus* is the dominant house-gecko in Balochistan and has never been reported from Punjab (Khan 1987; Khan and Ahmed, 1987). The present disjunct population most probably descended from few individuals so transported. Presently the few observed individuals are in severe competition with the common indigenous house gecko, *H. flaviviridis*, which is dominant throughout the building. The few individuals of *H. persicus* are still holding on in remote recesses of the fort.

Acknowledgments

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Appendix 1. Abbreviations used

BMNH= British Museum, Natural History, London; CAS= California Academy of Sciences, California, USA; MSK= Herp laboratory, 15/6 Darul Saddar North, Rabwah 35460, Pakistan (author's personal collection); NMW= Naturhistorisches Museum Wien, Austria; SR= Institute of Zoology, Academy of Sciences, Kiev-30, Ukraine; UF= Florida State Museum, Gainesville, USA; USNM= National Museum of Natural History, Washington, D.C.

Appendix 2. Additional material examined

Cyrtodactylus battalensis BMNH 1990.2; *C. collegalensis* BMNH 82.4.14.28-29; *C. fasciolatus* BMNH 1913.11.11.2; *C. nebulosus* BMNH 82.4.14.32-33; *C. oldhami* BMNH 1916.6.22.4; *C. pulchellus* BMNH 1916.3.27.1-2; *C. triedrus* BMNH 68.3.17.11-12; *C. dattanensis* MSK 0056.78; *C. yarkandensis* BMNH 72.3.22.4; *C. tibetinus* CAS 196850, CAS 196854; *Gymnodactylus walli* BMNH 1910.7.12.1; *G. chitralensis* BMNH 1946.8.23.19; *Tenuidactylus baturrensis* BMNH 1990.3; *T. longipes* CAS 115944, SR 307:3267-68; *T. longipes voraginosus* CAS 130323; *T. montiumsalsorum* BMNH 1904.11.19.1 and MSK 014.86; *T. indusoani* MSK 0467.86; *T. rohtasfortai* USNM 284133; *Gymnodactylus stoliczkai* (photograph) NMW 16756; *Tenuidactylus fedtschenkoi* SR 1078:8837-8; *T. caspius* SR 2546:16713-14; *T. turcmenicus* SR 961:8016-17.

Distribution and Natural History of the Lidless Skinks, *Asymblepharus alaicus* and *Ablepharus deserti* (Sauria: Scincidae) in the Aksu-Djabagly Nature Reserve (Western Tian-Shan Mountains), Kazakstan

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Abstract.— The data of 8 years observations on two species of the Lidless Skinks, *Asymblepharus alaicus* and *Ablepharus deserti* from the Aksu-Djabagly Nature Reserve (Western Tian-Shan Mountains) are analysed with special attention to geographical distribution and to some aspects of the natural history of these lizards. It is noted that in spite of altitude contiguity the two species form few sympatric zones with low population density. Both species have prolonged seasonal activity, ranging from March–April to October–November. The appearance of the skinks after winter hibernation, their activity and the beginning of breeding season depend on climatic conditions, slope exposition and altitude of the site.

Key words.— Reptilia, Squamata, Scincidae, *Asymblepharus alaicus*, *Ablepharus deserti*, Kazakstan, distribution, activity, breeding.

Introduction

The Aksu-Djabagly Reserve is one of the unique natural reserves of Kazakstan. Its flora and fauna are of a great interest because of the presence a number of endemic and localized species, as well as of species adapted to extreme environmental conditions. The Aksu-Djabagly Reserve occupies the western part of the Talas Alatau Range in the Western Tian-Shan Mountains between 1100–4200 m s. l. Permanent snow level is at about 3000 m. The climate is very continental with an average monthly temperature of -4.9 C for the coldest month (January) and of +21.6 C for the hottest month (July). Four basic altitudinal zones are present in the Reserve (Karmisheva, 1973; Kovschar, Ivashchenko, 1990). These are mountain foothill zone with a low mountain dry steppe and xerofitic plant association; the steppe meadows, with scattered tree-like *Juniperus* forests; subalpine zone and alpine high mountain meadows. The relatively small territory of the Aksu-Djabagly Reserve (7400 ha) is inhabited by many species typical for the zoogeographical provinces of the Europe, North Africa and Middle Asia. Among reptiles (total number 10 species) there are two species of the skinks, the Alpine Lidless Skink (*Asymblepharus alaicus*) and the Desert Lidless Skink (*Ablepharus deserti*). The first species (Fig. 1) is widespread in the Reserve, the other is rare species of the region.

Asymblepharus alaicus and *Ablepharus deserti* are of an interest from few points. The taxonomic position *Asymblepharus alaicus* and *Ablepharus deserti*

was cleared only recently after detailed revision by Eremchenko and Shzherbak (Eremchenko, 1981; Eremchenko a. Shzherbak, 1986) who separated the genus *Asymblepharus* and suggested an independent evolution for the two lineages of ablepharid lizards. Data on morphology and biology of the Alpine and the Desert Lidless Skinks are not numerous and scattered through a number of works (Atayev, 1985; Bannikov et al., 1977; Bogdanov, 1960; 1965; Bruscho, 1995; Said-Aliyev, 1979; Shammakov, 1981; Shnitnikov, 1929; Terentjev a. Chernov, 1949; Yakovleva, 1964). The monograph by Eremchenko and Shzherbak (1986) deserves special attention because it contains all known data on the morphology, distribution and biology of the ablepharid lizards of the former USSR.

The distribution and biology of two skink species remains poorly studied. The present paper deals mainly with the distribution of the Alpine and the Desert Lidless Skinks in the Aksu-Djabagly Nature Reserve and presents data on their natural history.

Material and Methods

Field observations together with the description of live and museum material carried out on *Asymblepharus alaicus* and *Ablepharus deserti* over a period of 8 years (1988–1996) and served as a basis for the present paper. We observed and collected skinks from 22 localities of the Aksu-Djabagly Nature Reserve (Fig. 2). We identified the species in the field basing on their external morphology and altitude distribution.

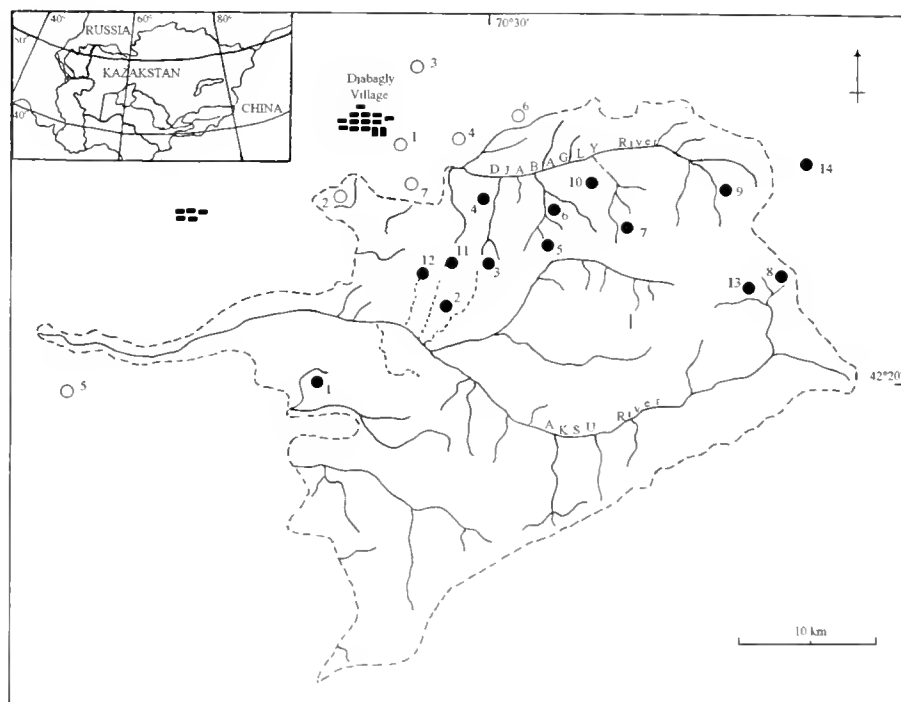


Figure 1. The map of the localities of the Lidless Skink in the Aksu-Djabagly Nature Reserve (Western Tian-Shan Mountains). *Asymblepharus alaicus* and *Ablepharus deserti* have independent enumeration: open circles represent the localities of *Asymblepharus alaicus*; closed circles represent the localities of *Ablepharus deserti*. *Asymblepharus alaicus*: 1) Chuuldak (2000 m); 2) Minzhilky Ravine (2600 m); 3) Kshy-Kaindy Pass (2200 m); 4) Kshy-Kaindy Ravine (1850 m); 5) Ulken-Kaindy Pass (2900 m); 6) Ulken-Kaindy Ravine (2000 m); 7) Kaskabulak (2600-3300m); 8) Aksay Pass (2900 m); 9) the Upper of the Djabagly River (3000 m); 10) Kizolgenkol Lake (2200 m); 11) the upper of the Middle Karasay River (3000-3100 m); 12) Low Karasay Ravine (3000-3100 m); 13) south-western slope of Kokseky Ravine (3100-3200 m); 14) eastern slope of the Aksay Ravine (2600 m). *Ablepharus deserti*: 1) western mountain foothill of the Talas Alatau Range (1300 m); 2) Taldibulak Ravine (1100 m); 3) northern mountain foothills of the Talas Alatau Range near Djabagly Village (1200m); 4) Djabaglitau Range (1300 m); 5) valley between Aksu and Baldabrek Rivers near the Kizilblek Village (1400m); 6) eastern slope of the Baldabrek Ravine (1600 m); 7) mountain foothills near the Ergaly Ravine (1200 m).

Additionally we used museum material fixed in 10 % neutralized formalin and preserved in 70 ethanol. The confirmation of species identification was mainly based on the peculiarities of the scalation around the eyes (see Eremchenko a. Shzherbak, 1986).

We also described character of the skink habitats together with plant composition and visually classified the dominant substrate of the sites. Altitudes and slope exposition were also taken into consideration. All colonlected and museum specimens were measured in mm according to Eremchenko and Shzherbak (1986); their life history stages were recorded as juvenile, subadult, adult. All the skinks captured for the present study were returned to the sampling sites.

Skink populaton density inferred by counting the numbers of lizards active on the ground surface along

a transect and by turning over stones which were refuges over certain small square areas.

Results and Discussion

Distribution, habitats and density of the populations-Figure 2 shows 22 localities of two species examined. All the records (including museum material) were made by the authors themselves.

In the Aksu-Djabagly Nature Reserve the Alpine Lidless Skink mostly inhabits the subalpine and alpine zones between 2500-3000 m where the density of the lizards is highest, particularly just on passes and on mountain ridges. For example, on 5 August 1988 we recorded 47 specimens of *Asymblepharus alaicus* along the main ridge of the Kazanchukur Range (3100 m) over an area of 50 x 50 m and over turning more than 150 stones. During a sunny midday



Fig. 2. *Asymblepharus alaicus*.

in July 1995 we observed 16 adult specimens in an area of 200 x 30 m in the course of half of hour on the Ulken-Kaindy Pass (2900 m). Transect accounting on the way down to 2400 m revealed 8 specimens over an in area of 40 x 4 m. Using secondary ranges and lateral splinters of the main ridge the skinks can come down to a slope as low as 1500 m (extremely rare to 1200 m) where they have very low population density.

The habitats of *Asymblepharus alaicus* in the Aksu-Djabagly are very variable. There are alpine meadows scattered with stones (Fig. 3), rocky slopes with *Juniperus* brush (Fig. 4) and scree slopes. There is practically no Alpine Lidless Skink in sparse *Juniperus* forests. A combination of the slopes with different exposition is the first important condition for *Asymblepharus* habitats because of the possibility of using them during different times of the day. The second essential condition is in the presence of suitable refuges represented by bushes, screes or stone rubbles.

Typical habitats for the Desert Lidless Skink in the Aksu-Djabagly Reserve are dry-south exposed slopes of no more than 2000 m in altitude (Fig. 5). As a rule, the plant community includes *Ferula* plant xerofitic steppe, with bushes of *Rosa*, *Honeysuckle*, *Cotoneaster*, *Spirea* scattered juniper trees (Fig. 6). However, the density of *Ablepharus deserti* population in the Reserve is not high in general. Only in rare cases we observed the lizards near human settlements.

The Aksu and Djabagly rivers with their banks of southern and northern exposure are the natural boundaries separating the habitats of the Alpine and the Desert Lidless Skinks in the reserve. The two species were found as sympatric in few areas only. This occurs in the valleys of rivers flowing down a slopes



Fig. 3. Subalpic meadow at 3000 m of altitude is a typical locality of *Asymblepharus alaicus* in the Aksu-Djabagly Reserve.



Fig. 4. The locality of *Asymblepharus alaicus* in the Ulken-Kaindy Valley (2100 m).

having northern exposition. In this situation around at 1200-1500 m *Asymblepharus alaicus* occupies, as a rule, the river banks and *Ablepharus deserti* inhabits the slopes having an eastern and western exposition (Fig. 7). However, both species have here very low density.



Fig. 5. The locality of *Ablepharus deserti* in the mouth of the Djabagly River (1200 m).

Daily and seasonal activity

Eremchenko and Shzherbak (1986) noted that the appearance of the skinks after winter hibernation depends on climatic conditions, slope exposition and altitude. According to these authors, the earliest appearance of *Asymblepharus alaicus* in northern Kirgizstan was registered on 26 March 1977 in the Kirgiz Range (1600 m). In the mountains bordering the eastern coast of the Issik-Kul Lake (2400 m) and on the slopes of the San-Kul-Too Range having western exposition (2700 m) the lizards appear by late April. Yakovleva (1964) noted an earlier activity of males in comparison with females.

Our observations on the Alpine Lidless Skink in the Aksu-Djabagly show that lizards appear after winter hibernation in mid April and, as a rule, are active until late October-early November. On 29 October 1992 we observed some active adult specimens on the northexposed slopes at 1900 m. On 1 November 1995 skinks were registered on northern slope at 1300 m. For southern slopes, active lizards were recorded later. Based on data of Eremchenko and Shzherbak (1986) the last active lizards in the Kirgiz Range (up to 3000 m) were registered on 3 November 1974. It seems the skinks have very prolonged seasonal activity. According to Veventzev (1978) who studied *Asymblepharus alaicus* in the Almaty Nature Reserve (Northern Tian-Shan Mountains) some individuals were occasionally found active during sunny days even in January-February when small areas of ground get free of snow. On 5 August 1988 at altitude 3100 m we recorded the beginning of morning activity of the lizards about 11:00 hrs; air temperature +13C. That low temperature be enough for primary skink activity may shed some light on the prolonged yearly activity of the lizards.

The daily activity of the Alpine Skink from the Aksu-Djabagly doesn't visibly differ from that of the



Fig. 6. View on southern slopes of the Djabaglitau Range (1500 m). The locality of *Ablepharus deserti* is in the background, the locality of *Asymblepharus alaicus* in the foreground.

Alpine skink, described by Eremchenko and Shzherbak (1986) for the Kirgiz Range. These authors wrote that in spring (April - first half of May) skinks were active between 11:00-12:00 hrs and 19:00-20:00 hrs. According to our data for summer period, these lizards appear on ground surface earlier and are active until 20:00-21:00 hrs. In autumn their activity shifts to the second half of the day.

As a rule, *Ablepharus deserti* appears after winter hibernation earlier than *Asymblepharus alaicus* because of lower altitude of its habitats and the exclusively southern slope expositions. In the Aksu-Djabagly the earliest record for the beginning of *Ablepharus deserti* spring activity was noted on 8 March 1989 at 1300 m. Most of the population, however, emerges from hibernation in mid March. Kaluzhina (1951) reported that also in the Turkmenistan the Desert Skink appears after hibernation in first half of March. According to Yakovleva (1964), in Kirgizstan lizards of this species come to the ground surface around mid March. Paraskiv (1956) studied the Desert Skink in southern Kazakstan also noted the first half of March as the time for the beginning of lizard activity.

In the Aksu-Djabagly the Desert Lidless Skink is active until late October-early November. The latest record here belongs to 3 November 1988. In other regions of southern Kazakstan the skinks have the same activity pattern (Paraskiv, 1956). Yakovleva (1964) for Kirgizia and Said-Aliyev (1979) for Tad-jikistan reported late September - mid October as the period for winter leaving of *Ablepharus deserti*. The daily activity pattern of *Ablepharus deserti* in the Aksu-Djabagly Reserve doesn't differ markedly from that of the Desert Skink previously described by other authors from the surrounding territories. A visibly variable daily activity of *A. deserti* was observed by Bogdanov (1960) in Uzbekistan. For two months in

the year (March and September) skinks are active for most of the day (from 10:00-11:00 hrs to 18:00-19:00 hrs), whereas in summer they have a two-peak day activity. The first peak occupies the time between 09:00 and 12:00 hrs. The second peak is between 17:00 and 19:00 hrs. In February and October their activity is maximal after the midday. A two-peak activity is also typical for the Desert Skink from Kirgizia (Yakovleva, 1964).

All previous authors noted that juveniles and sub-adults appear after winter hibernation earlier than adult specimens and return later to their winter refuges.

Breeding

As was first observed by Shnitnikov (1928), viviparity is a typical feature of *Ablepharus* (= *Asymblepharus*) *alaicus*. According to Yakovleva (1964), period of breeding activity of *Asymblepharus alaicus* in Kirgizstan occupies May-June, although some specimens copulate in July as well. Such data contradict to Eremchenko and Shzherbak' (1986) who registered frequent copulation lizards in Kirgizstan in late March-early April. However, this contradiction could be a consequence of different climatic conditions in different years. According to Said-Aliyev' (1979) in the southern regions of Tadzhikistan the Alpine Skink copulates in late March-April, in the northern regions in late May-first decade of June. A single female pregnant with the eggs of 8.2 x 5.1 mm; 10.2 x 5.0 mm and 10.1 x 5.0 mm in diameter was found on 3 July 1954.

In the Aksu-Djabagly Reserve, we have found 5 gravid females on 5 August 1988 at an altitude of 3100 m. One of these delivered 3 young on the next day. Some pregnant females approaching delivery we have also observed 20 July 1995 at the Ulken-Kaindy Pass (2900 m). On the next day the new born lizards were met at 1900m. Based on our own data and Yakovleva's (1964) notes over a two-month period of embryonic development of *Asymblepharus alaicus* we consider mid May - mid June as a period of copulating activity of the Alpine Skink in the Aksu-Djabagly Reserve.

As noted by previous authors, for *Ablepharus deserti* the egg-laying period varies depending on the geographical location of the population. In Kirgizstan, this period occupies the first decade of June (Yakovleva, 1964). In Uzbekistan, the Desert Skink lays its eggs after mid May. Paraskiv (1956) recorded mid May - early June as optimal time for egg-laying by *A. deserti* in the Betpak-Dala Desert (southern Kazak-

stan) and in the northern coastal territory of the Aral Sea.

Unfortunately, we have no data on the breeding season or on the clutch size of *Ablepharus deserti* in the Aksu-Djabagly Reserve. However, some information on the breeding activity of the Desert Skink in Kazakstan are present in the work by Bruscho (1995), who notes that the beginning of the breeding season depends on altitude. In the Borolday Mountains females with eggs at the last stage of the development were found from mid May to the second half of June. For Northern Aral Sea region Paraskiv (1956) recorded the beginning of egg-laying by *A. deserti* in second the half of May. As Eremchenko and Shzherbak (1986) wrote, in Kirgizstan the clutch size of the Desert Skink varies from 1 to 5 eggs and rarely reaches 11 per female. Yakovleva (1964) recorded 2-8 eggs per female for the Kirgizstan populations. Said-Aliyev (1979) has found 10 July 1959 one female having 3 eggs in every oviduct, the sizes of which varied from 9,1 x 3,2 mm to 11,1 x 4,1 mm in diameter. Shammakov (1981) notes 3-5 eggs in the clutch of the Desert Skink from Turkmenistan.

The lizards of both species become sexually mature in the second year (Eremchenko a. Shzherbak, 1986; Said-Aliyev, 1979).

Enemies

According to literature data (Eremchenko a. Shzherbak, 1986; Said-Aliyev, 1979; Yakovleva, 1964) and to our own observation, the Halys Pit Viper *Agkistrodon halys*, the Mountain Raser *Coluber ravergeri*, the Dione Snake *Elaphe dione*, the Steppe Ribbon Snake *Psammophis lineolatum*, the Steppe Viper *Vipera ursini* and the among birds the Legger Grey Shrike *Lanius cristatus*, the Long-tailed Shrike *L. schach* and the Rock Thrush *Monticola saxalitis* are the main enemies of both the Alpine and the Desert Skinks in nature. We also found skink remnants in the nests of the Black-billed Magpie (*Pica pica*). Kuzmina (1970) observed the Himalayan Ruby Throat (*Calliope pectoralis*) feeding on the Alpine Skink in the Almaty Nature Reserve.

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Seasonal Variations of Testicular and Epididymal Structure and Plasma Levels of Testosterone in the Soft-shelled Turtle (*Pelodiscus sinensis*)

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Abstract.- For purpose to consider the annual cycle of testis in soft-shelled turtle, *Pelodiscus sinensis*, the testes and epididymides were examined histologically, and the plasma levels of testosterone was measured by radioimmunoassay (RIA) though the year. Gradual increase in testicular weight from May to August was followed by a decrease in degrees. Spermatocytogenesis was first observed from late April to early May which remained active to August. Spermatogonial division became to decline in September and stopped by the end of November. The epididymal weights rose from August to November and remained heavy during hibernation with a rapid decline of about 53.76% in the mating period of next year. From July to September, the epithelial cells of epididymal ductus grew and increased in height especially. There were a great number of secretory granules in the cytoplasm with active synthesis. The plasma concentration of testosterone started to rise in April, which then fell in May and June. In July, it rose rapidly to peak levels and then declined to minimum. We suggested that it's more important that the peripheral testosterone promotes reproductive behaviour and stimulates secretion synthesis in epithelial cells of epididymis.

Key words.- *Pelodiscus sinensis*, testosterone.

Introduction

Only scattered and equivocal literature can be found concerning the reptilian gametogenetic cycle, the endocrinological function of testis and pituitary gonad interrelationships. The information is almost exclusively derived from investigations on squamate species (Bartholomew, 1953; Lofts, 1971; Courty, 1980). One does find, however, a few studies on such reptiles as turtles, for example: *Chrysemys picta* (Callard, 1976); *Pelodiscus sinensis* (Lofts and Tsui, 1977); *Chrysemys dorsibigni* (Silva, 1984). The authors of those studies hold identical views that the testicular structure and function of reptiles change in annual cycle, and the epithelium of seminiferous tubule has spermatogenetic ability in several months while in other months it is in a static condition (Lofts, 1987).

The aquatic soft-shelled turtle, *Pelodiscus sinensis*, lives in rivers, lakes and ponds. The available literature on its reproduction relates only to its gonads. Hu Zeng-Gao (1988) reported that the testis of *Pelodiscus sinensis* produced sperms in reproductive period, which were used for copulation in same period. Lofts and Tsui (1977) in Hong Kong reported the results of their studies on the histological and histochemical changes in the testis. They were of the opinion that *Pelodiscus sinensis* has a postnuptial pattern which the spermatozoa are produced soon after mating period. The spermatozoa are stored in the epi-

didymal canals from October until their discharge in the following April. Seasonal changes in size and histology of the testis and the accessory sexual organs (epididymis) are well known in a lot of reptile species (Lofts, 1969; Courty, 1979), and correlated variations in androgen levels are documented but still poorly detailed.

It has been shown by metabolic conversion of precursors that testosterone is the principal androgen in some reptiles as in mammals (Callard, 1967; Hews and Kime, 1978; Courty, 1979). Seasonal changes in the level of circulating testosterone have been reported in a few species of reptiles but data tend to be differ with different species (Bourne and Seamark, 1975; Callard, 1976; Silva, 1984). Some researchers suggested that the spermatogenesis correlated with the plasma testosterone levels in turtles (Kuchling, 1981; Silva, 1984). In other references we found that the highest plasma testosterone levels did not appear in spermatogenesis period, but in mating period (Callard, 1976). Lofts and Tsui (1977) considered that the interstitial tissue was active in steroids synthesis during mating period, but inactive during spermatogenesis period. The purpose of our study, therefore, was to determine if the variations in the plasma testosterone levels during the annual cycle in *Pelodiscus sinensis* are correlated with histological variations of the germinal epithelium and epididymal canals.

Material and Methods

Animals and samples collection

The samples used in our experiments were provided by Chang Zhou Breeding Center of Soft-Shell Turtle (near Nangjing in Jiangsu Province in east China). The breeding environment was similar to their natural living condition. Except December and January every month we obtained blood plasma, testes and epididymides that those were from six adult male soft-shelled turtles, *Pelodiscus sinensis* (there body weights were about 650-1000g) in one year. The blood plasma samples were stored at -24° until the testosterone analysis was performed. The tissue samples of testes and epididymides were weighted and fixed in Bouin fluid.

Histological procedures

For the histological examination, a piece of testis and a piece of epididymis (about 25mm^3) were fixed in Bouin fluid for 24 hours. After discolored, the samples were embedded in paraffin, cut in $5\mu\text{m}$ sections, and then stained with haematoxylin and eosin.

Testosterone assay

We determined the plasma concentration of testosterone by radioimmunoassay (RIA) using a technique provided by WHO. The kits used in our experiments were from Hua Mei Biological Engineering Co. (a Sino-US joint venture). Briefly, the hormone was extracted from plasma samples (250-750l)

twice with 5 ml of anhydrous ether, with an average recovery of 96%. The extract was dried at 40° by constant temperature bath, and dissolved in 2 ml phosphate-buffered saline containing gelatin (GPBS), we placed 0.5 ml of this solution in duplicate assay tubes and incubated with radio labeled testosterone ($^3\text{H-T}$ 10,000 cpm) and anti-testosterone antiserum (0.1 ml) for 18-24 h at 4°C . After removing the unbound fraction with dextran-coated chorale, the samples were placed in scintillation fluid (TP-POPOP-toluene) and the radioactivity (cpm) was detected on a liquid scintillation counter. RIA data were analyzed with a program utilizing a weighted logic-log regression analysis on an IBM PC. All data are as the mean \pm SEM.

Results

Weight and histology

The seasonal weight variations in testes and epididymides of *Pelodiscus sinensis* are shown as unit body weight in Figure 1. From March (shortly after emergence from hibernation) to April, testicular weights remained low (particularly in April). The seminiferous tubules remained atrophied and spermatogenetically inactive. The germinal epithelium contained Sertoli cells and spermatogonia only (Fig. 2A). In April there was a significant reduction (by about 53.76%) in the epididymal weights as spermatozoa were evacuated from the epididymal canals. During May and June the epididymal weights declined continuously (by about 19.65%, Fig. 1).

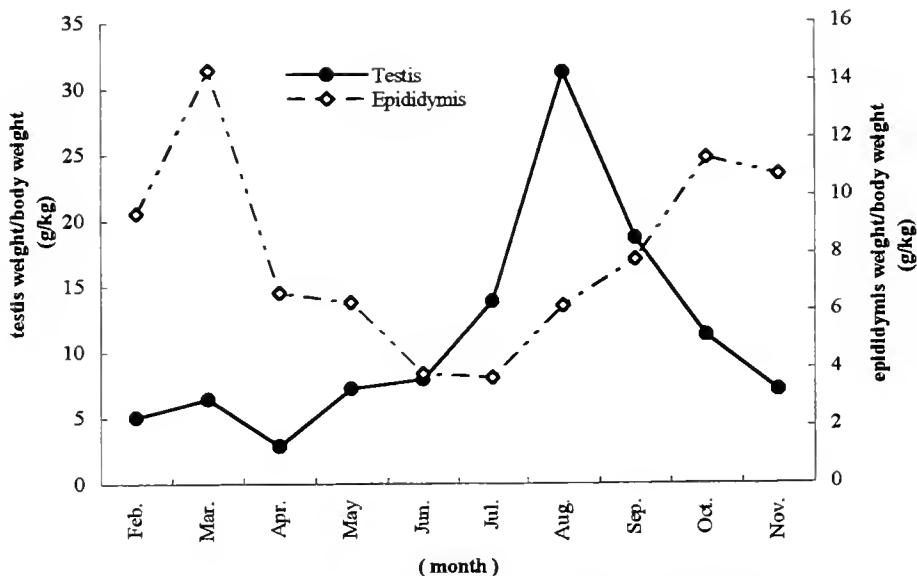


Figure 1. Seasonal variations in testis and epididymis weight in the soft-shelled turtle, *Pelodiscus sinensis*. Each point represents mean of six values.

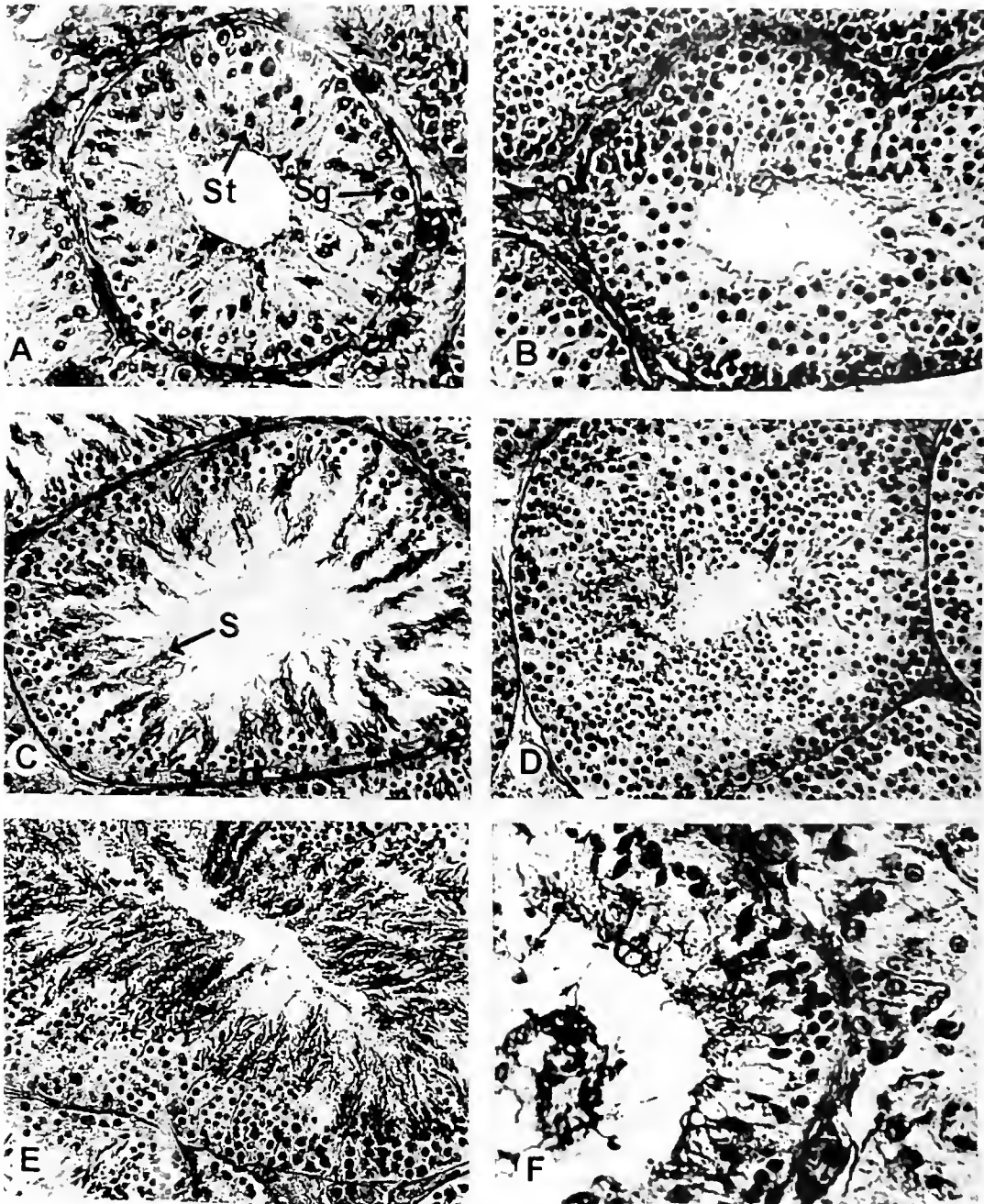


Figure 2. Microscopic figures of testis in the soft-shelled turtle, *Pelodiscus sinensis*. A. The seminiferous tubules contracted and contained Sertoli cells (St) and spermatogonia (Sg) in April 200. B. The germinal epithelium contained dividing spermatogonia, spermatocytes and spermatids in May 200. C. Spermatozoa (S) were released from germinal epithelium in June 132. D. The thickness of spermatogenic epithelium increased but spermatozoa decreased in July 132. E: Spermatozoa occluded the lumen of the seminiferous tubules in August 132. F: The germinal epithelium atrophied in February 200.

Spermatocytogenesis started from late April to early May, and the spermatocyte layers increased progressively in germinal epithelium. The testicular weights began to increase in May. By the end of the month, the germinal epithelium contained dividing spermatogonia, spermatocytes and a few spermatides (Fig.

2B). In June, spermatozoa were released from germinal epithelium (Fig. 2C) without accompanying apparent increase in epididymal weights. The active spermatogonia division progressed until July. The thickness of spermatogenic epithelium increased and the diameter of seminiferous tubules reached

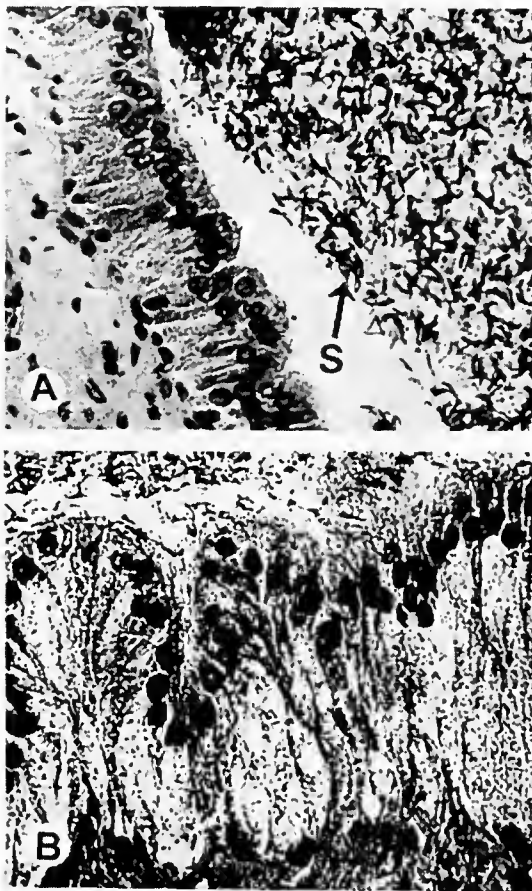


Figure 3. Microscopic figures of epididymis in the soft-shelled turtle, *Pelodiscus sinensis*. **A.** There were great amount of spermatozoa (S) in the ductus, The simple columna was thin in March 200. **B.** The heights of epithelial cells were at maximum, great amount of spermatozoa could be seen in the ductus in August 200.

maximum (Fig. 2D). By the end of August, when the testicular weights were at their top values, a large amount of spermatozoa gathered in the lumen of the distended seminiferous tubules (Fig. 2E) and passed into the epididymal canals with the epididymal weights increasing rapidly.

In September, spermatogonial division started to decline with less frequent mitotic figures. The germinal epithelium contained mainly spermatides and spermatozoa. At that time, testicular weights began to decrease. By the end of November, testis atrophied highly. And there were only a few spermatozoa remaining in the seminiferous tubules. During hibernation, the testis remained atrophied, and the germinal epithelium was composed of Sertoli cells as well as spermatogonia only and was heavily lipoidal (Fig. 2F). Often, there were necrotic cells in the lumen of the seminiferous tubules.

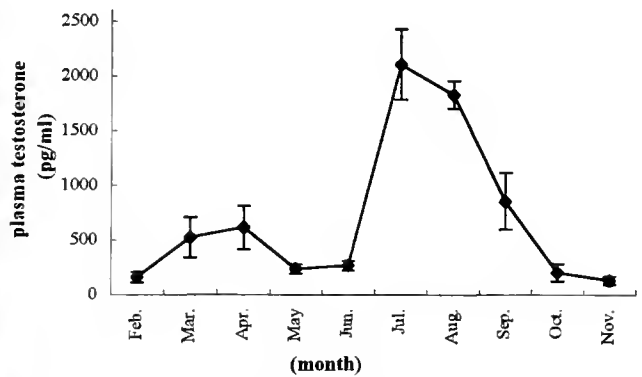


Figure 4. Seasonal variations in plasma testosterone concentration in soft-shelled turtle, *Pelodiscus sinensis*. Each point represents mean SEM of six values

In the ductus of the epididymis, spermatozoa could be recognized in great number in most months. Although in March soon after emergence from hibernation epididymal weights were high and their ductus were full of numerous spermatozoa (Figs. 1, 3A). Only in June (after mating period) were there markedly less spermatozoa present in the ductus of the epididymis. At this time the simple columnar epithelium of epididymis became thin and the nuclei moved toward the basal region of the cells. From the end of July, we could see that the spermatozoa were transported from the testis into the ductus of epididymis, and their epithelial cells increased their heights and secretory granules in cytoplasm. In August, their heights reached the maximum (Fig. 3B).

Plasma testosterone concentration

The variations of the plasma testosterone concentrations are shown in Figure 4. In March and April (spring), when the testis still remained atrophied, the mean values of plasma testosterone concentration started to rise from 160.87 pg/ml to 613.79 pg/ml. After that, there was a fall in plasma testosterone concentrations which coincided with the progressive atrophy of the epididymides. The plasma content remained relatively low in June, but rapidly increased to the highest levels (2104.97 pg/ml) in July. It did not drop significantly until September. After that, the plasma values fell sharply to their baseline levels in October. It could be seen that in an annual cycle of the plasma testosterone concentration had two peaks. The lower peak occurred during the spring and the higher one was in summer. The higher peak coincided with the maximal testicular weight and the hypertrophy of the germinal epithelial cells as well as the full growth of epithelial cells at the epididymal ductus.

Discussion

The present study has demonstrated that the spermatogenetic activity of germinal epithelium changes with season in *Pelodiscus sinensis*. Resembling that of most turtles reported (Lofts, 1987), its spermatogenesis resumes relatively soon after the mating period, so that advanced germinal stage is completed before the onset of the colder winter months. Spermatozoa are stored in the epididymal canals until the breeding season of next year. This result is consistent with that reported by Lofts and Tsui (1977) in *Pelodiscus sinensis*. We have pointed out that spermatogenetic peak that we observed is in July and August, which is somewhat later than that observed by Lofts and Tsui. We suggest that this discrepancy might be due to the different temperatures and photoperiods at different latitudes.

The measurement of plasma testosterone in the male *Pelodiscus sinensis* indicates that an annual increase in peripheral levels begins in spring after emergence from hibernation period and is interrupted from the end of May through June. In July and August, the plasma testosterone rises to its maximum level. This finding is identical to that in tortoise, *Testudo h. hermanni*, reported by Kuchling (1981). In fresh water turtle, *Chrysemys picta*, the highest levels were in spring and the lowest were in summer, while in autumn the testosterone levels increased significantly (Callard, 1976). Although in *Chrysemys dorsibigni* there was only one peak of plasma testosterone levels, the increase started in mating period and reached the maximum value in autumn during the principal period of spermatogenesis (Silva, 1984). It is similar to that in *Pelodiscus sinensis*.

Lofts (1968) described lipid accumulation in the interstitial cells of the testis in *Pelodiscus sinensis* which occurred prior to regression of the cells themselves, this indicated the cessation of steroidogenesis. Lofts and Tsui (1977) reported that they detected 3-HSD (⁵3-hydroxysteroid dehydrogenase) activity in *Pelodiscus sinensis*. It showed that the interstitial cells had a positive reaction for 3-HSD and were considerably depleted of lipoidal droplets in the mating period (March-April) and in the period of enhanced epididymal weight (August-October), but in active spermatogenesis period the interstitial cells were negative to the tests. And they said that the seminiferous tubules gave same histochemical feature of high testosterone secretory activity at the onset of spermatogenesis. As well known, the secretory activity of interstitial cells coincides with the variations of plasma testosterone levels, because the Leydig cells are the main origin of

the plasma testosterone. Therefore, there is a discrepancy about the annual cycle of the secretory activity of interstitial cells between the histochemical study reported by Lofts and Tsui (1977) with the direct determination of plasma testosterone levels in our study. We also found that the seminiferous tubules have the ability to secrete testosterone (unpublished result).

In the turtle *Chrysemys dorsibigni* and the tortoise *Testudo hermanni*, the maximum value of plasma testosterone acts on the secondary sexual organs in autumn, preparing them for the storage of spermatozoa which occurs in winter (Kuchling, 1981; Silva, 1984). This situation which can also be observed in our study. The decline of plasma testosterone levels in *Pelodiscus sinensis* in May and June is associated with the decrease in epididymal weights and the atrophy of the epididymal epithelium. The epididymal epithelial cells enlarge in July and August, when the plasma levels are highest. They increase secretory granules progressively. During this period, the epididymal weights increase significantly accompanying with the spermatogenesis. The spermatozoa are stored and matured in the epididymis until the following mating period.

In our study the seminiferous tubules are spermatogenetically inactive when the plasma testosterone increases in the mating period. However, when plasma levels are low in May, the spermatogenesis begins. It implied that the germinal epithelium is sensitive to testosterone when environmental temperature rises. Our conclusion is that the peripheral testosterone in *Pelodiscus sinensis* is more important to support reproductive behavior and stimulate the actions of epididymis.

Acknowledgments

We wish to express our deep gratitude to Mr. Chang Hue for assistance in our experiment. We are grateful to Professor Ning-Zheng Sun and Mr. Shu-Yu Gu for graphics technical assistance. This work was supported by a grant of Education Commission of Jingsu Province, China.

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Food Consumption and Growth of Juvenile Chinese Soft-shelled Turtles (*Pelodiscus sinensis*) in Relation to Body Weight and Water Temperature

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Abstract.— Food consumption and growth of juvenile Chinese soft-shelled turtles (*Pelodiscus sinensis*) with different body weight were measured in the laboratory at 22, 26, 28, 30, 32 and 35 °C. At 30 °C, the daily maximum consumption C_{\max} (J/ind. · day) showed the following relationship with body weight W (g): $C_{\max} = 47.58 W^{0.7907}$. The correlation between the maximum daily food consumption rate R_{\max} (J/g · day) of a turtle with a standard body weight of 40 g, and temperature could be described as: $\log R_{\max} = -0.0136T^2 + 0.8372T - 11.42$. At ± 31 °C, a turtle with a standard body weight of 40 g has the biggest daily consumption rate of 447 J/g · day. The daily growth rate GR (g/day) had a functional relationship with body weight at 30 °C: $GR = 0.04W^{0.8248}$. For a turtle with a standard body weight of 40 g, GR showed the following relationship with water temperature: $GR = 30.138 - 3.671T + 0.147T^2 - 0.002T^3$, and at 30 °C, turtles had the biggest daily growth rate of 1.06 g/day. Gross conversion rate of the juvenile turtles (9–109 g) did not vary with body weight, but temperature had a distinct effect on it.

Key words.— Reptilia, Testudines, Trionychidae, *Pelodiscus*, China, bioenergetics.

Introduction

The Chinese soft-shelled turtle (*Pelodiscus sinensis*) is an aquatic chelonian of great commercial importance, widely distributed in China. Resource of this species has decreased sharply in recent years. Therefore, its aquaculture has become more and more important. Basic information on the biology of *P. sinensis* is very important in developing the technology of its cultivation. Bioenergetics of this species is one aspect that has not been well-studied yet (Niu et al. 1994). The purpose of this study was to investigate the effects of body weight and water temperature on food consumption and growth of juvenile soft-shelled turtles.

Material and Methods

The experimental animals were juveniles with a live weight ranging from 7 g to 112 g reared in the laboratory. The diet used in the experiment contained 50% crude protein, 3% crude fat, 9.5% carbohydrate and 15% ash. Energy value of the diet was 14.64 KJ/g.

Rearing conditions

Each experimental animal was housed in a 4.6 l glass aquarium filled with clean tap water. 6–8 aquariums were placed in a water bath with water temperature controlled to an accuracy of ± 0.5 °C. The animals were tested at six different temperatures (22, 26, 28,

30, 32 and 35 °C). The photoperiod was 12L:12D. Turtles were fed to satiation twice a day at 8:00 and 16:00. Each turtle was acclimated at the test temperature for at least one week before the feeding experiment started.

Experimental process

Before the start of the feeding experiment, turtles were not offered food for 48 hours and weighted. In the experiment, turtles were fed to satiation with a pre-weighted amount of food. The uneaten portion and feces were collected with a decompression concentration equipment and dried with control diet at 65 °C to a constant weight. Food consumed by a turtle was the difference between the dry weight of pre-weighted amount of food and the uneaten portion. The feeding regime lasted for a month followed by two days of fasting. After the experiment, fresh weight was measured again and some of the experimental animals were sacrificed and dried to a constant weight on 65 °C. Caloric value of the dried diets, feces, and turtles were all measured with a Schimdu CA-4P caloric meter.

Data analysis

The daily maximum consumption, C_{\max} is an average value during the whole experimental period. $R_{\max} = C_{\max}/W$. W is the mean value of the initial and final weight of the turtle. The daily growth rate in wet

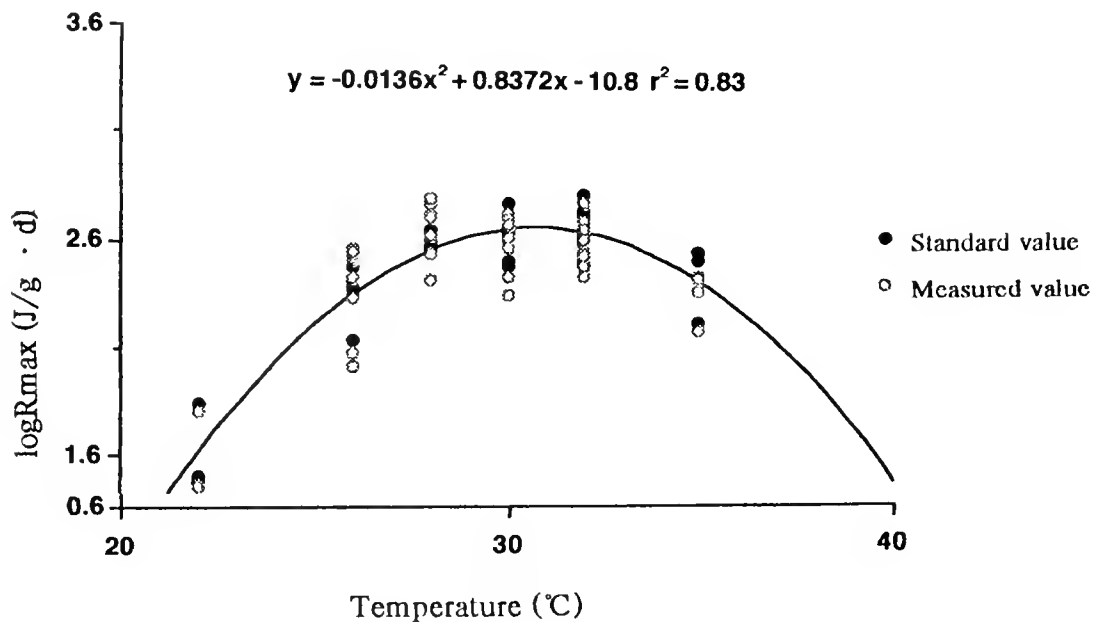


Figure 1. Relationship between the daily maximum consumption rate, R_{\max} (J/g · day) of a turtle with a standard body weight of 40 g (average value of all experimental animals) and water temperature.

weight, $GR = (W_1 - W_0)/t$, where W_1 and W_0 is the body weight at the end and the beginning of the experiment, respectively, and t is the experimental period. The gross conversion efficiency, CE — named as the percent of energy used for body growth to consumed energy — was calculated. All data were analysed with SPSS/PC statistical software.

Results

Analysis of covariance showed that the maximum food consumption of a turtle was greatly affected by body weight and water temperature. Multiple regression analysis showed the following relationships among the daily maximum consumption, C_{\max} (J/ind. · day), wet body weight, W (g) and water temperature, T (°C): $\log C_{\max} = 0.7338 \log W + 0.8354 \log T - 0.0136T^2 - 11.00$; ($r^2 = 0.98$, $n = 45$, $P < 0.01$). At 30 °C, C_{\max} (J/ind. · day) showed the following relationship with body weight W (g): $C_{\max} = 47.58W^{0.7907}$; ($r^2 = 0.92$, $n = 30$).

Figure 1 shows the relationship between the daily maximum consumption rate, R_{\max} (J/g · day) of a turtle with a standard body weight of 40 g (average value of all experimental animals) and water temperature. Their relationship could be described as: $\log R_{\max} = -0.0136T^2 + 0.8372T - 11.42$ ($r^2 = 0.83$, $n = 45$). At ± 31 °C, a turtle with a standard body weight of 40 g has

the biggest daily food consumption rate of 447 J/g · day. The daily growth rate, GR (g/day) also had a close relationship with body weight and water temperature, which could be showed as the following: $GR = 0.0165W + 0.9142T - 0.0156T^2 - 13.16$ ($r^2 = 0.89$, $n = 45$, $P < 0.01$).

Figure 2 shows the relationship between the daily growth rate, GR (g/day) of a turtle with a standard body weight of 40 g and water temperature. At 30 °C, a turtle with a standard body weight of 40 g has the biggest daily growth rate of 1.06 g/day. At 30 °C, GR had a functional relationship with body weight as: $GR = 0.04W^{0.8248}$ ($r^2 = 0.78$, $n = 39$). At five different body weight groups ranging from 9–109 g, no distinct differences were found among their gross conversion efficiency, CE ($F_{4,20} = 0.96$, $P > 0.05$), but the temperature had a distinct influence on CE ($F_{5,20} = 4.21$, $P < 0.01$). At 22 °C, CE was about 59%. At 26–32 °C, CE ranged from 21–27%. At 35 °C, CE declined to 12%.

Discussion

According to the present work, temperature has a positive effect on the daily maximum food consumption and growth under 31 °C, but above 31 °C, the effect becomes negative. Temperature for maximum growth rate is slightly lower than the temperature for maximum food consumption. This phenomenon has also been observed in similar studies of the lizards *Uta*

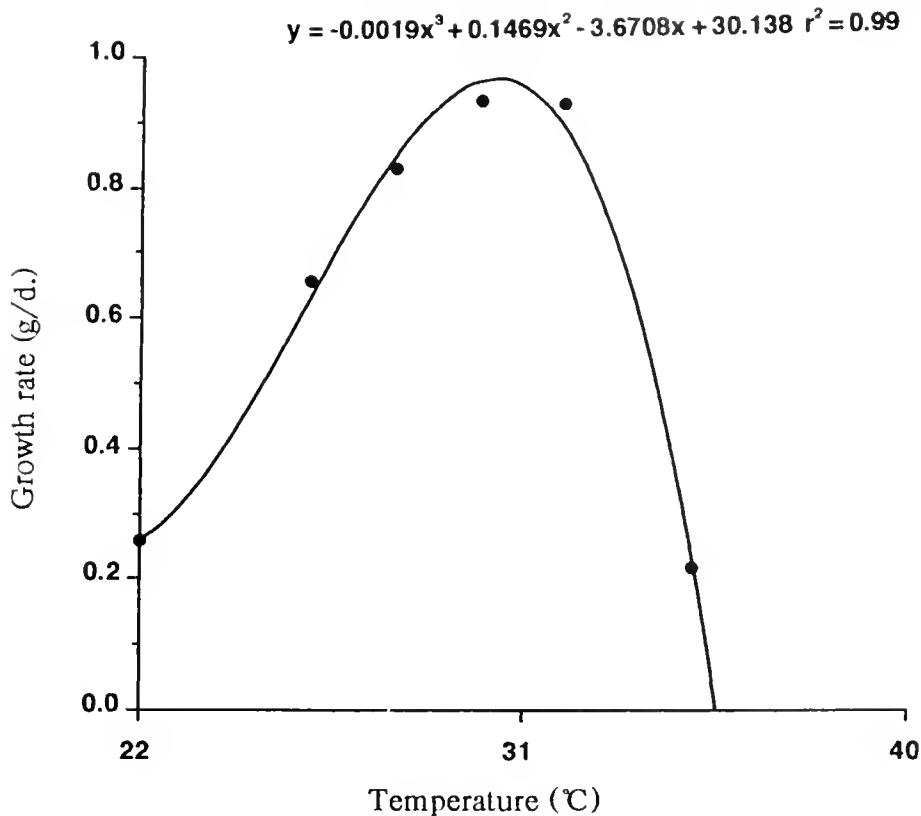


Figure 2. Relationship between the daily growth rate, GR (g/day) of a turtle with a standard body weight of 40 g and water temperature.

stansburiana (Waldschmidt et al. 1986) and *Takydromus septentrionalis* (Ji et al. 1993), and the southern catfish (*Silurus meridionalis*) (Xie and Sun 1992). This study indicates that effects of temperature on food consumption and growth of soft-shelled turtles resemble that of most fishes (Brett and Groves 1979) and some lizards. More comparative studies should be conducted for other reptile species.

The body weight has a double logarithm relationship with C_{max} , and the slope of the regression line is 0.79. This value is very similar to the exponent b value 0.75 in the metabolic rate to body weight regression line for turtles (Bennett and Dawson 1976). This phenomenon suggests that food consumption is related to metabolic rate of the turtle. As energy loss through metabolism is a large component in the energy budget, metabolism level may affect energy intake to remain an effective budget. Studies on the relationship of food consumption and metabolic rate are recommended. Perhaps the maximum consumption rate can be defined as an index of daily metabolism level of the animals.

The gross conversion rate, CE is a reflection of the consumed energy allotted to body growth. Our work

showed that under 109 g body weight, CE values of the juvenile turtles were similar. Temperature has a distinct effect on CE. The relatively high CE (59%) at 22 °C may be explained by the distinctly low metabolic cost at a low temperature. From 26–32 °C, CE was relatively similar, but declined at 35 °C. Smith et al. (1981) found that for juvenile walleye pollock (*Theragra chalcogramma*), CE was higher at colder temperatures. Cui et al. (1995) showed that water temperature had no effect on CE in the grass carp (*Ctenopharyngodon idella*). Perhaps the relationship of temperature with CE varies with different species.

In this work we present mathematical models relating maximum food consumption and growth rate to water temperature and body weight. The results can be utilized for estimating daily amount of food needed by juvenile turtles according to water temperature and body weight in a turtle-culture farm.

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First Record of the Lacertid *Acanthodactylus boskianus* (Sauria: Lacertidae) for Iran

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Abstract.- The first record of the lacertid lizard *Acanthodactylus boskianus* for Iran is presented based on material collected by the author in 1995 and 1996 from 2 km west of Harsin, Kermanshah province, western Iran, on the *Astragalus*-covered sandy hills at about 1420 m elevation. Systematics and distribution of this lizard are discussed and its conventional known subspecies are questioned.

Key words.- *Acanthodactylus boskianus*, Lacertidae, Subspecies, New record, Western Iran, Kermanshah province, Harsin, Systematics, Distribution.

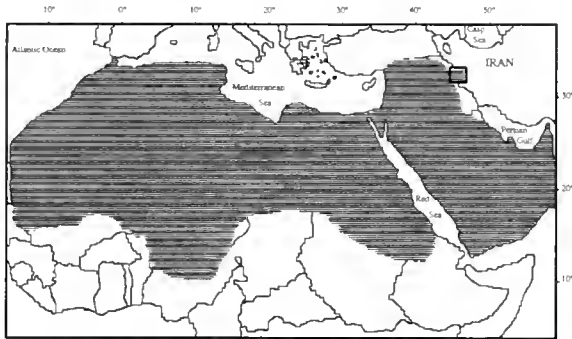


Figure 1. The distribution of *Acanthodactylus boskianus* in north Africa and the Middle East.

Introduction

The lacertid genus *Acanthodactylus* Wiegman, 1834 consists of about 30 species, distributed from Spain and Portugal across the Sahara desert and its periphery to the Red Sea, over most of Arabia and as far north as Cyprus and the Syrian-Turkish border; it also extends through Iraq, southern, and eastern Iran, southern Afghanistan, Pakistan and northwestern India (Arnold, 1983).

Apart from the present record, four additional species of this genus occur in Iran, mainly in southern and eastern parts of the country (*A. micropholis*, *A. grandis*, *A. schmidtii*, and *A. blanfordi*) (Anderson, 1974; Anderson, in press; Salvador, 1982). As well, *A. ophiodurus* Arnold, may occur in lowland southwestern Iran (Anderson, in press). The genus is Saharo-Sindian in its affinities and distribution and does not penetrate to a great extent into the Iranian Plateau and only two species go beyond the plateau as far east as Afghanistan, Pakistan, and northwestern India (*A. cantoris* and *A. micropholis*) (Clark, 1990;

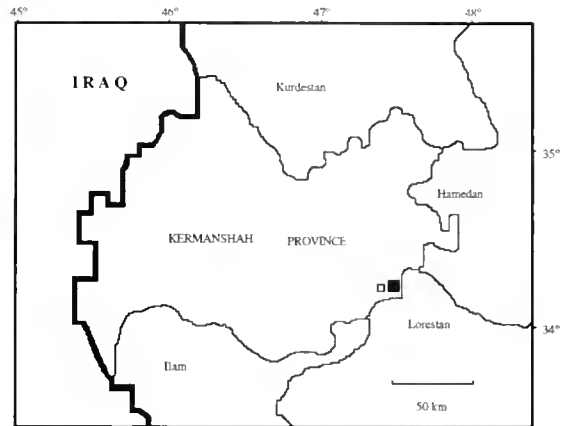


Figure 2. The locality of *Acanthodactylus boskianus* specimens collected by the author from Harsin, southeast of Kermanshah Province, western Iran. (■) = Harsin, (□) = Locality of *A. boskianus* specimens.

Salvador, 1982). This genus has recently been revised by Salvador (1982) and Arnold (1983) who divide it into 9 species groups. Among these groups is the "*A. boskianus* and *A. schreiberi*" group defined by several distinguishing characters (Arnold, 1983: 315).

So far, there is no record in the literature for the occurrence of *A. boskianus* in Iran. In this paper, I report this taxon for the first time inside Iranian territory based on three specimens (two adults and one juvenile) collected from Kermanshah province, western Iran during my two long-term excursions on the Iranian Plateau in 1995 and 1996.

Acanthodactylus boskianus (Daudin, 1802)

Lacerta boskiana Daudin, 1802, 3: 188, Pl. 36, Fig. 2 (type locality: Egypt).

Bosc's fringe-toed lizard.

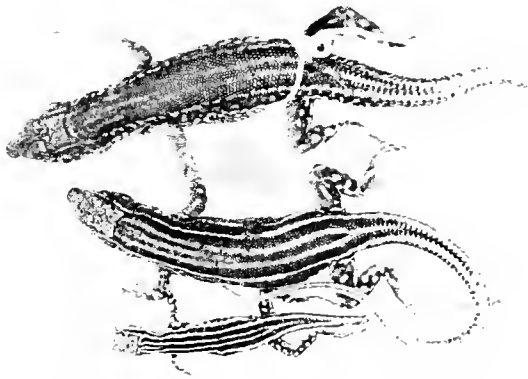


Figure 3. *Acanthodactylus boskianus*, top to bottom: male, female, juvenile.

Definition: Usually 4 entire supraoculars, occasionally the 1st divided; anterior border of ear pectinate; temporal scales more or less keeled; eyelids slightly denticulated; conspicuous gular fold; 3 series of scales around fingers; ventrals usually in 10 (and sometimes 12) straight longitudinal rows; usually large, keeled, imbricate dorsals (sometimes small, slightly keeled and imbricate); granular scales on flanks; moderate to strong fringe on 4th toe; upper surface of tail with large, imbricate, sharply keeled scales.

Distribution: *A. boskianus* is the most widespread species of the genus and occupies a large area from north Africa (Mauritania, Morocco, Algeria, Mali, Niger, Tunisia, Libya, Chad, Nigeria, Sudan, Ethiopia, Egypt) eastward into the Middle East (Israel, Lebanon, Jordan, Iraq, Syria, Turkey, and Arabian peninsula) (Salvador, 1982). It is also reported for the first time from Iran [(present paper) (Figs. 1-2)].

Collecting of *A. boskianus* specimens

I conducted two long-term excursions and field work in various parts of the Iranian plateau in 1995 and 1996. On July 8, 1995, I surveyed the area around Harsin, a small town in southeastern Kermanshah province, western Iran to collect *Trapelus ruderatus* and *Laudakia nupta* for ongoing research. In 2 km west of Harsin (34° 17' N, 47° 24' E) on the sandy hills covered by various species of *Astragalus*, I collected a lacertid specimen incidentally. It was an adult male of *A. boskianus* (GNHM Re. ex. 5142). I surveyed this locality several times in 1995, but I could not find more specimens of *A. boskianus*. Again, on August 29, 1996, and during my second field trip to Iran, I surveyed the same area and collected two other specimens of *A. boskianus*, an adult female and a juvenile (GNHM Re. ex. 5143-4) (Fig. 3). I collected all specimens on an *Astragalus*-covered sandy hill,



Figure 4. *Acanthodactylus boskianus* on the top of an *Astragalus* bush, 2 km west of Harsin, Kermanshah Province, western Iran.

among or under the *Astragalus* bushes. It seems that they have acquired special adaptations for living among and on the spiny bushes of *Astragalus* (Fig. 4).

Remarks

The three specimens which I collected 2 km west of Harsin, Kermanshah province, western Iran, have the general characteristics of most other *A. boskianus* specimens except that the dorsal scales are relatively small, as is the case in the Iraqi and Syrian populations. The main characteristics of these specimens are as follows:

Maximum SVL (Snout-Vent Length) = 65.5 mm; maximum TL (Tail Length) = 134 mm; scales across mid-dorsal region 43-48; ventral plates in 10 longitudinal rows with an extra row of smaller plates on each side; four entire supraoculars; temporal region slightly keeled, subocular does not reach the mouth and is wedged between the 4th and 5th supralabial; 7 / 7 upper and 7 / 7 lower labials; temporal scales feebly keeled; anterior edge of tympanum feebly serrated; feeble to moderate pectination of eyelids; 5 / 5 supraciliaries; 22-25 femoral pores; dorsal scales relatively small and feebly keeled; 3 rows of scales around fingers; 8-9 collar plates; distinct gular fold; 25-27 scales in a longitudinal row from symphysis of chin shields to collar; 21-23 keeled lamellae under the forth toe which is not exceptionally pectinate; ventral plates in 25-30 transverse rows; 13-15 scales between hindlegs; 25-26 scales on the 5th caudal whorl behind the vent.

Coloration and color pattern: In the male specimen dorsum sandy grey with two light dorsolateral stripes on each side enclosing a broad brown band with light reticulations, also a weakly visible dark-brown vertebral stripe present; base of tail with two light lateral stripes, distal 4 / 5 of tail uniformly grey dorsally, upper surface of limbs greyish-brown with numerous light spots; upper surface of head olive-brown; all of the ventral surfaces whitish.

In the female specimen dorsum is dark brown with 7 narrow, light stripes, the two dorsolateral ones on each side being lighter and in strong contrast with the dark-brown pattern of back, the three vertebral and paravertebral ones duller, proximal 1/4 ventral part of tail whitish, distal 3/4 pink or bright-red, other ventral surfaces whitish.

In the juvenile specimen, upper surface of head is light olive, dorsum dark-brownish-black with 6 strongly contrasting light lines, vertebral stripe whit-

ish on neck, disappears towards the posterior part of back.

Systematic account

As pointed out before, *Acanthodactylus boskianus* is the most widespread species of its genus. It occurs throughout a wide range which is extended from north Africa into the Middle East. It is a polytypic taxon and very well represented in most museum collections and shows obvious geographic variation in different parts of its range. Traditionally, *A. boskianus* has been divided into three subspecies; *A. b. boskianus* (Daudin, 1802) in the Nile delta and some parts of Sinai, *A. b. euphraticus* Boulenger 1919, from Ramadieh (central Iraq), and *A. b. asper* (Audouin, 1829) which covers almost the whole of the species range (Arnold, 1983; Boulenger, 1919, 1921; Salvador, 1982).

Boulenger (1919, 1921) divided the populations of *A. boskianus* into three varieties (subspecies):

The first subspecies, *A. b. boskianus* (forma typica), characterized by the lack of subocular contact with the lip, the common division of the first supraocular, and the small and numerous dorsal scales (34-52). The second subspecies, *A. b. asper*, characterized by a subocular which does not border the lip, an undivided first supraocular, and the large, relatively few dorsal scales (23-38). And the third subspecies, which I have examined the syntypes, *A. b. euphraticus* (described based on 8 specimens collected at Ramadieh on the Euphrates Front, central Iraq, in 1918 by Boulenger's son Capt. C. L. Boulenger), is characterized by a subocular which is usually bordering the mouth (in 7 out of 8 specimens = 87.5%), 38-43 scales across middle of body, 14-16 scales between hind limbs, and 23-37 femoral pores on each side.

This simple tripartite division is not satisfactory, for some of the supposedly distinctive features of *A. b. euphraticus* are not consistent and there is some differentiation within the populations assigned to *A. b. asper* (Arnold, 1983).

As the separation or contact of subocular with the lip is not a fixed character, it can not be considered of taxonomic value. As well, scale counts which were once thought typical for subspecies proved to be clinal and are therefore not apt to discriminate subspecies (Schleich et al., 1996).

On the other hand, Salvador (1982) divided various populations of *A. boskianus* into four groups; north African populations, Egyptian populations, Arabian populations, and Iraq, Syria, and Jordan populations. According to this author, the populations of

north Africa are relatively uniform, suggesting a recent invasion to this region. There is a progressive degree of variation towards the eastern part of the range, in the Middle East. As the position of the subocular greatly varies in individuals throughout this species entire area of distribution (this being especially so in Iraq), the taxonomic value of this character is greatly reduced (Salvador, 1982). According to Arnold (1983), over most of north Africa, the number of dorsal scales in a transverse row at mid-body varies from 26-41. I have examined specimens of *A. boskianus* from Sudan, Morocco, and Libya (see under material examined). All of these specimens have 35-37 scales across mid-body and, apparently, belong to *A. b. asper*. As well, Arnold (1980) regards all populations of this lizard in the Arabian peninsula as *A. b. asper*. In the Nile delta and north Sinai, populations assigned to *A. b. boskianus* have high dorsal scale counts (34-52). Populations with high dorsal counts (38-48) also occur in northeast of Jordan, northern and central Iraq, east Syria and adjoining Turkey. In one specimen from Jordan (GNM.Re. ex. 4799) which I have examined, there are 40-43 scales across middle of dorsum. Arnold (1983) believes that geographic variation in *A. boskianus* reflects differences in niche across its range. This species is often associated with dense vegetation and large dorsal scales may well be protective when shrubs are rigid and spiny. The fine-scaled populations occur in relatively mesic areas where vegetation is less damaging than in more arid regions. Reed and Marx (1959) reported *A. schreiberi*, based on having numerous dorsal scales, from Jarmu, Kirkuk Liwa, northern Iraq, in an isolated area far from the nearest known localities for this taxon in Lebanon. Salvador (1982) examined these specimens and attributed them to *A. boskianus*. Due to scarcity of material, the presence of *A. schreiberi* in Iraq needs more confirmation. I have studied three specimens of *A. schreiberi* (GNM.Re.ex. 4646 [1-3]) from Cyprus. They have 51-59 small, rather granular, smooth, or weakly keeled, scales across widest part of dorsum and 12 ventral plates across the widest part of venter. As well, the temporal scales are smooth and the anterior edge of tympanum is not serrated or very weakly so. According to Khalaf (1959), both *A. b. asper* and *A. b. euphraticus* occur in Iraq without specifying their exact localities. Also, Leviton et al. (1992) regard *A. b. asper* as the subspecies found in Iraq.

Based on the fore mentioned discussion, and with respect to characteristics of specimens collected by the author, it is evident that these specimens belong to a form with fine and numerous dorsal scales which are

weakly keeled, intact first supraocular, and lack of subocular contact with the mouth. These specimens do not entirely fit into the Boulenger's tripartite key (Boulenger, 1919, 1921). Since there is no more material at hand, it may be difficult to say if they represent the fourth form of *A. boskianus* or not. An adequate intraspecific treatment of *A. boskianus* is beyond the scope of this paper. Thus, I have chosen not to use a subspecific name for my own material, pending a thorough and knowledgeable revisionary work on this widespread and polytypic taxon. In spite of the considerable variation which occurs in *A. boskianus*, as mentioned above, there is as yet no definitive evidence that it consists of more than one species. It appears that *A. schreiberi* has originated as an isolate of *A. boskianus* (Arnold, 1983).

Material examined

Acanthodactylus boskianus (n = 3): GNHM. Re. ex. 5142-44, from Harsin (34° 17' N, 47° 24' E), Kermanshah province, western Iran.

Acanthodactylus boskianus euphraticus (n = 8): BMNH 1946. 8. 4. 83-90, from Ramadieh, Iraq (33° 25' N, 43° 17' E).

Acanthodactylus boskianus asper (n = 4): GNHM. Re. ex. 3333, 3346, 4799, 4937, from Sudan, Morocco, Libya, and Gaza respectively.

Acanthodactylus schreiberi (n = 3): GNHM. Re. ex. 4646 (1-3), from Cyprus.

Abbreviations

BMNH = British Museum (Natural History); GNHM. Re. ex. = Gothenburg Natural History Museum, Reptilia exotica.

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Two New Subspecies of *Trapelus agilis* Complex (Sauria: Agamidae) From Lowland Southwestern Iran and Southeastern Pakistan

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Abstract.— Based on conducting long-term excursions and carrying out extensive field work in various parts of the Iranian Plateau and studying preserved (museum) material including the syntypes of Olivier's *Agama agilis*, and paralectotypes of Boulenger's *Agama isolepis*, two new subspecies of the wide-ranging Asian ground agamid *Trapelus agilis* complex are described from the lowland southwestern Iran and southeastern Pakistan (and adjoining northwestern India) respectively. The former of the new subspecies has conventionally been considered as belonging to *T. a. agilis*, and the latter to *T. a. isolepis*. They are distinguishable from the other subspecies of *T. agilis* complex by having several distinctive morphological characteristics. The distinctiveness of both subspecies is confirmed according to the author's previous extensive studies on this complex using uni- and multivariate analyses of morphological characters. Both subspecies mainly occur as geographical isolates in the periphery of the main range of the complex, and both have mainly been separated from the central continuum (= *T. a. agilis*) by eco-geographical barriers and are almost entirely restricted in distribution to the lowlands, desert and semi-desert regions with high annual temperature. A taxonomic and biogeographic account as well as a key to subspecies of *T. agilis* complex are presented.

Key words.— *Trapelus agilis* complex, *T. a. khuzistanensis*, *T. a. pakistanensis*, New subspecies, Agamidae, Lowland southwestern Iran, Southeastern Pakistan, Systematics, Distribution, Biogeography.

Introduction

The, taxonomically, controversial ground agamid *Trapelus agilis* is a wide-ranging species complex distributed from extreme southwestern Iran (ca 31° N, 47° E) to eastern Kazakhstan and western China (ca 48° N, 83° E) (Fig. 1), encompassing numerous local populations (Rastegar-Pouyani, 1998). Traditionally, this complex has been divided into three subspecies: *Trapelus agilis agilis* (Olivier, 1804), *T. a. sanguinolentus* (Pallas, 1814), and *T. a. isolepis* (Boulenger, 1885) (e.g., Anderson, 1974; Welch, 1983; Wettstein, 1951). The latter two subspecies are sometimes regarded as full species by some authors (e.g., Ananjeva, 1981; Ananjeva and Tsaruk, 1987; Boulenger, 1885; Moody, 1980; Nikolsky, 1915; Zhao and Adler, 1993). On the other hand, Anderson (in press) places all different forms of *T. agilis* complex under the inclusive name “*agilis*” and believes that without a firm statistical ground, it is not advisable to divide the complex into separate taxonomic entities.

In a series of studies, using uni- and multivariate statistics, the author analysed geographic variation in *T. agilis* complex, synonymized *T. a. isolepis* (Boulenger) with *T. a. agilis* (Olivier), designated a new type locality as “*terra typica designata*” (central Iranian Plateau, about 110 km southeast of Esfahan city)

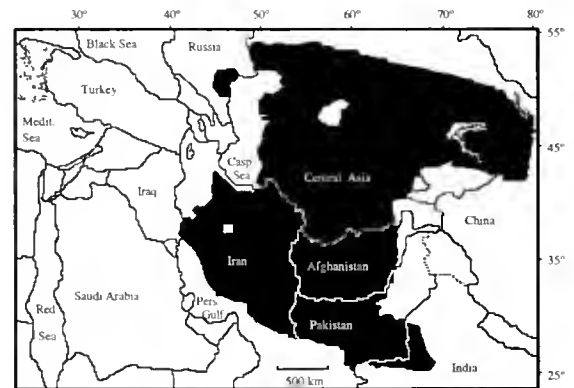


Figure 1. Geographic distribution of *Trapelus agilis* complex. (□) = *Terra typica designata* (central Iranian Plateau, about 110 km southeast of Esfahan city)

(Fig. 1), and showed that *T. agilis* complex consists of four distinct taxonomic entities (subspecies) and that the traditional tri-partite division of the complex, to a great extent, does not portray the actual phenetic patterns of geographic variation (Rastegar-Pouyani, in press, unpublished manuscript). The four distinct subspecies identified are as follows:

T. a. agilis (Olivier, 1804) is distributed in the central Iranian Plateau, central and southern Afghanistan, and southwestern Pakistan. Populations of this

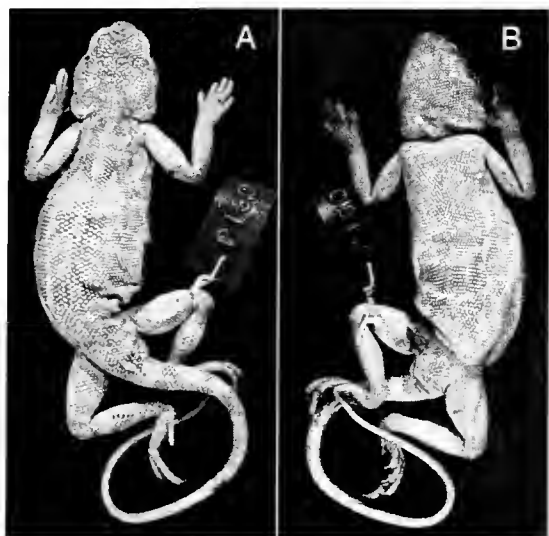


Figure 2. *Trapelus agilis khuzistanensis*, holotype (GNHM Re. ex. 5424); A- dorsal view, B- ventral view. Note that almost all dorsal scales are distinctly small and only slightly keeled.

form (as the central core of the complex) are morphologically most similar to the syntypes of Olivier's *Agama agilis*.

T.a. sanguinolentus (Pallas, 1814) is the northern representative of the complex, distributed over a wide area from northeastern Iran into the Central Asian countries as far east as western China (Anderson, in press; Rastegar-Pouyani, 1998; Zhao and Adler, 1993). The two other taxa, described in this paper as new subspecies, are geographically isolated, occurring in the southwestern and southeastern margins of the main range of *T. agilis*, and mainly separated from the central continuum (*T. a. agilis*) by eco-geographical barriers. Both new subspecies are morphologically different from the syntypes of Olivier's *Agama agilis* based on several distinguishing characters (Table 1).

Since the subspecific name "*isolepis*" has been synonymized with "*agilis*" and is no longer available (Rastegar-Pouyani, in press), new taxonomic names are designated for these two new subspecies.

So, the main objective of this work is to describe and introduce these two new taxonomic entities based on the study of distinguishing morphological characters which make them recognizable from the other subspecies of *T. agilis* complex.

Material and Methods

I conducted three long-term excursions and carried out extensive field work in various parts of the Iranian Plateau in 1995, 1996, and 1998, collected hundreds of specimens of *Trapelus agilis* and noted the pattern

of adaptation of different populations to the local conditions as well as presence of eco-geographical barriers which have been involved in differentiation and subsequent evolution of all subspecies of this complex. Also I studied preserved materials including the syntypes of *Agama agilis* Olivier, 1804 and paralectotypes of *Agama isolepis* Boulenger, 1885, borrowed from various museum collections around the world (see under "Appendix I, Abbreviations, and Acknowledgments").

In my previous studies, based on examination of about 1000 specimens of *T. agilis* all over the range, I employed uni- and multivariate statistical techniques and explored the patterns of geographic variation in morphological characters within this complex (Rastegar-Pouyani, in press, unpublished manuscript).

Indeed, the present paper is the continuation (and part of the results) of my previous studies concerning taxonomy and geographic variation in *T. agilis* complex according to which both the lowland southwestern Iranian as well as southeastern Pakistani populations do warrant taxonomic recognition.

Subspecies accounts

Trapelus agilis khuzistanensis ssp. nov.1 (Figs. 2-6)

Khuzistan Ground Agama

Holotype and type locality: adult female, GNHM Re. ex. 5224, collected by the author on 27 July 1996 from Iran, Khuzistan Province, 5 km northwest of Haft-Gel on the road to Shushtar.

Paratypes: 14 specimens (ten males and four females) have been designated as paratypes as follows: GNHM. Re. ex. 5225, same information as the holotype; GNHM. Re. ex. 5226, collected by the author on July 28, 1996 from Iran, Khuzistan Province, 38 km south of Masjid-e-Suleiman, Golgir village; CAS 86342, 86346, 86390, Iran, Khuzistan Province, Tul-i-Bazum [31° 55' N, 49° 25' E], about 500 m elevation, collected by S. C. Anderson on 17 April and 22 May 1958; CAS 86403-6, Iran, Khuzistan Province, along road to lake east of Haft-Gel, about 300 m elevation, collected by S. C. Anderson on 23 May 1958; CAS 86418-19, Iran, Khuzistan Province, along road between Haft-Gel and Masjid-Suleiman, by S. C. Anderson on 23 May 1958; CAS 86556, Iran, Khuzistan Province, Haft-Gel (on golf course) [31° 28' N, 49° 30' E], about 300 m elevation, by S. C. Anderson on 5 October 1958; CAS 86464, Iran, Khuzistan Province, along road south of Shushtar, by S. C. Anderson on 18 July 1958; FMNH 170936, Iran, Khuzistan Province, 85 km southeast of

Table 1. legend. Main morphological differences between *T. a. khuzistanensis*, *T. a. pakistanensis*, and the Olivier's syntypes (*T. a. agilis*).

Characters	<i>T. a. agilis</i> (n =2) (Olivier's syntypes)	<i>T. a. khuzistanensis</i> (n =97) (ssp. nov.1)	<i>T. a. pakistanensis</i> (n =32) (ssp. nov. 2)
- Reverse imbrication of head and neck scales	—	+	—
- Upper head scales	smooth	rugose and keeled	smooth or slightly rugose
- Dorsal scalation	subequal to homogeneous	subequal to heterogeneous	subequal to homogeneous
- Dorsal scales shape	distinctly keeled and mucronate	weakly to moderately keeled and mucronate	distinctly keeled and mucronate
- Ventrals	weakly keeled	smooth or weakly keeled	distinctly keeled in males
- Body	moderately depressed	moderately depressed	sometimes spindle-shaped and compressed in males
- Head	subcordiform	often roundish	subcordiform and more pointed
- Tail	round or weakly compressed	often distinctly compressed in males	strongly compressed in males
- Preanal callose scales	two well developed rows	often one row (in the case of two, the second undeveloped)	almost always one row in males, absent in females
- Nuchal crest	absent	absent	often present
- Background coloration	olive-grey	yellowish-grey-cream	often sandy-grey
- Scales around body	79-88	80-97	67-83
- Supralabials	17-19	15-18	13-16
- Infralabials	17-20	14-19	12-16
- Subdigital lamellae under fourth toe	24-26	16-20	22-28

Ahwaz, Meshrageh, collected by D. Womochel and A. De Blase on 20 October 1968.

Diagnosis: *Trapelus agilis khuzistanensis* differs from all other subspecies of *T. agilis* in its higher number of scales around body (80-97); subequal and almost heterogeneous dorsal scalation with distinctly small dorsals and ventrals; a shorter head and neck; significantly lower mean number of subdigital lamellae under the fourth toe; reverse imbrication of the posterior head and anterior neck scales; keeled or rugose upper head scales; usually one, and sometimes two rows of callose preanal scales (absent or slightly developed in females); strongly compressed tail in males of most populations; and an exclusive dorsal coloration (yellowish-grey-cream with weak or without reddish-brown cross bars).

Description of holotype: an adult female, preserved in 70% ethyl alcohol in good condition; head short but longer than broad with very convex forehead, its length 0.26 of body length and 0.19 of tail length; canthus rostralis more or less continued as a supraciliary ridge; nostril on, or, barely, above the canthus, pierced in a flat shield and posteriorly directed; 3 internasals in a transverse row; upper head scales heterogeneous, keeled or rugose, imbricate and subimbricate; supraciliary ridge strongly developed, composed of 9 scales on each side; 17-18 upper- and



Figure 3. *T. a. khuzistanensis*, holotype (GNHM Re. ex. 5424); neck and head regions with reversally-imbricated scales.

17-17 lower labials; tympanum horizontally elliptical, smaller than orbit, partly covered above by 4-5 small spinose scales; scales of posterior part of head and anterior part of neck distinctive in that their imbrication is reversed (i.e., towards the head) and the posterior border of reversally-imbricated scales is defined by a single large and pointed scale; gular pouch moderately developed; gular region covered by small, imbricate, slightly keeled or smooth scales; gular fold and a fold in front of shoulder strongly developed; body and head moderately depressed; limbs rather slender; dorsal scales subequal to unequal, small, imbricate, slightly keeled and mucronate; median dor-

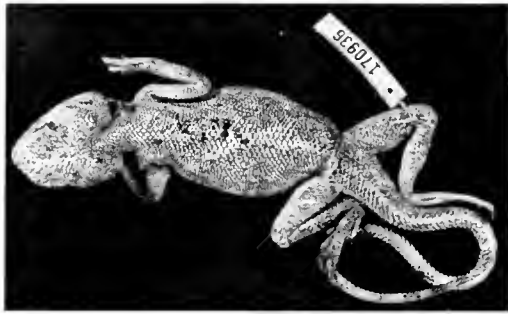


Figure 4. *T. a. khuzistanensis*, one of the male paratypes (FMNH 170936). Note the presence of a strongly compressed tail and relatively heterogeneous dorsal scalation.



Figure 5. *T. a. khuzistanensis*, posterior ventral region of a male paratype (GNHM. Re. ex. 5426) with one row of callose preanal scales.



Figure 6. Habitat and type locality of *T. a. khuzistanensis*, 5 km northwest of Haft- Gel on the road to Shush-tar, Khuzistan province, southwestern Iran. In the foreground the author is attempting to capture the holotype from inside of its underground hole.

sals relatively larger, grading into distinctly smaller scales of dorsolateral region which are only slightly keeled and mucronate; 94-95 scales round middle of body; scales of upper surface of limbs larger than median dorsals, distinctly keeled, slightly mucronate; lower surface of digits covered by bi- or tri carinate lamellae, 18-19 under the fourth toe; ventral scales almost as large as median dorsals, imbricate, very slightly keeled or smooth, 84-85 scales in a single row from gular fold to the anterior edge of anus; callose preanal scales in one row, slightly developed, consisting 10 scales; caudal scales larger than median dorsals, strongly keeled, distinctly mucronate, 34-36 around base of tail just behind vent; tail weakly compressed at base, distinctly so towards the tip, its length 1.39 of body length.

Coloration and color pattern: upper surface of head and limbs uniformly yellowish-grey, ground color of dorsum yellowish-grey-cream with 5 large, broad, light-brown cross bars from nape to sacrum, interrupted by a series of light, large vertebral ocelli and

two paravertebral rows of smaller ones, proximal upper caudal region with distinct dark-grey bars; ventral surfaces uniformly whitish.

Measurements (mm): Total length = 206; Snout-vent length (SVL) = 86; Tail length (TL) = 120; Head length (HL) = 23; Head width (HW) = 19; Head depth (HD) = 12.8; Length of forelimb = 45; Length of hindlimb = 62.

Variation of the type series: all paratypes closely approximate the holotype both in morphology and meristics.

The range of the number of scales around body for the whole series ($n = 14$) is 82-97 and the mean 88.5; in all paratypes the number of subdigital lamellae under the fourth toe varies between 16-19; dorsal scales are subequal or heterogeneous (especially in males); there are some differences, however, between male and female paratypes.

- **Male paratypes:** all male paratypes ($n = 10$) are either uniformly yellowish-grey-cream dorsally or with a weakly developed pattern of dark cross bars; as well, the ventral pattern is either uniformly whitish or with distinct pattern of bluish-brown on the gular region, chest and flanks; tail strongly compressed in almost all males; preanal callose scales in one or two rows, in the latter case the second row weakly developed, their number varies from 10-21; dorsal scales more keeled and mucronate and distinctly heterogeneous, approaching *T. persicus* in this respect; ventrals weakly keeled, gular sac distinctly developed; reverse imbrication of the posterior head and anterior neck scales relatively more pronounced and the large scale on posterior end of these reversally-imbricated scales more distinctive and mucronate than in females.

- **Female paratypes:** all female paratypes ($n = 4$) resemble the holotype in almost all pertinent details; the preanal pores are only in one row, weakly devel-

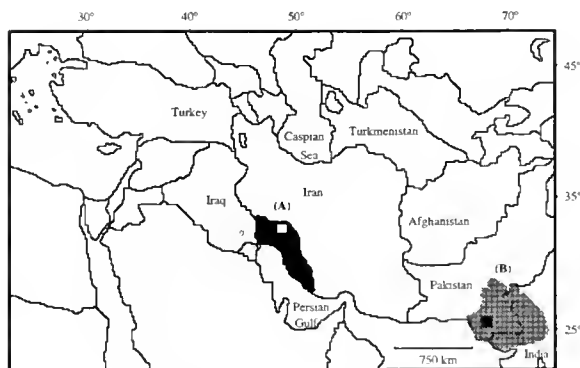


Figure 7. Geographic distribution of *T. a. khuzistanensis* (A) and *T. a. pakistanensis* (B). Squares represent the type localities.

oped, each occupying the tip of a scale, their number varies from 8 to 11; all the dorsal scales weakly keeled and mucronate; tail round or weakly compressed; slight differences in dorsal pattern occur (in some specimens the dorsal cross bars are more intense).

Habitat: in lowland southwestern Iran and in the western foothills of the Zagros Mountains, this subspecies mainly occurs on sand dunes, alluvial soils, open plains, low hills, and on dry stream channels (Anderson, 1966a, in press: personal observations). The type locality is specified by numerous low sand dunes as well as gypseous hills imminented with an open plain. The vegetation is sparse, mainly *Artemisia*, *Alhagi*, *Zygophyllum*, and *Euphorbia* association. The climate conditions being harsh, with hot and long summers (about 40°- 50°C) and mild, short winters. It seems that this lizard is active throughout the year but Anderson first observed it in the western foothills of the Zagros in early March, becoming numerous by mid-April; both newly hatched and half-grown juveniles were observed in late October and early November (Anderson, 1963: 446).

The holotype and one of the paratypes were collected about 5 km northwest of Haft-Gel on the road to Shushtar. They were active when air temperature was 45°C and the substrate was 49.5°C. When alarmed, unlike most of the central Plateau populations which usually retreat into the base of dense bushes, they retreated into the underground holes: this may be due to vegetation scarcity. Both specimens were captured inside their underground holes (Fig. 6). The third specimen collected near the mouth of an old well in the vicinity of the village of Golgir (38 km south of Masjid-e-Suleiman) while trying to retreat into the well.

Distribution: the main distributional range of this subspecies is the Khuzistan Plain which is an extension of the Mesopotamian lowlands (Fig. 7). Also, it penetrates into the western foothills of the Zagros Mountains up to 900 m elevation. It is the westernmost representative of the wide-ranging *T. agilis* species complex. The Zagros Mountains serve as a strong barrier to its further eastward distribution, so it has almost no contact with the central Iranian Plateau nominal subspecies (*T. a. agilis*), except in the southeastern regions of Bushehr province, southern Iran where the two taxa occur as parapatric (Rastegar-Pouyani, Manuscript, a). Its occurrence in the lowland southeastern Iraq is almost unlikely and, so far, there is no proper record inside the Iraqi territory [except the Olivier's original record (1804) which is strongly doubtful]. If it occurs in southeastern lowland regions of Iraq, then the Tigris might have served as an effective barrier to its further westward distribution.

In some areas of lowland southwestern Iran (e.g., 85 km southeast of Ahvaz) it occurs as sympatric with *Trapelus persicus* (Blanford, 1881) but there is no proper record of intergradation, if any, between the two taxa.

Etymology: *Trapelus agilis khuzistanensis* is so named as it is mainly restricted in distribution to the lowlands of southwestern Iran, Khuzistan province.

Trapelus agilis pakistanensis ssp. nov.2 (Figs. 8-9)

Pakistan Ground Agama

Holotype and type locality: adult male, SMF 63258, collected by M. G. Konieczny on 31 March 1957 from Gaj-River, Kirthar Range, southeastern Pakistan.

Paratypes (7 specimens): SMF 63259, 63279, same information as the holotype; SMF 63256, collected by R. Mertens on 1st December 1952 from Sonda, Distr. Thatta, southeastern Pakistan; SMF 63243-4, 63286-7, collected by M. G. Konieczny on 26 April 1961 from Old Airport of Karachi, southeastern Pakistan.

Diagnosis: *Trapelus agilis pakistanensis* differs from the other subspecies of *T. agilis* complex by having a combination of distinctive characters; body and head sometimes compressed (not depressed) in males; males almost always with one row of callose preanals (rarely a second undeveloped row may be present); females without callose preanals; dorsal scales relatively flat, subequal to homogeneous, distinctly keeled throughout and mucronate, grading into small dorso-laterals rather abruptly (especially in males), 67-83 around body; ventral scales also often distinctly keeled in males; body and limbs often strongly slender and head distinctly pointed (in adult males); tail

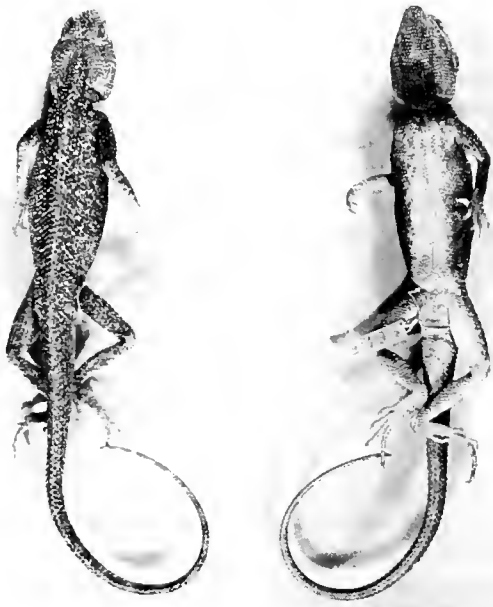


Figure 8. *T. a. pakistanensis*, holotype (SMF 63258); A- dorsal region, B- ventral region. Note the presence of a strongly compressed tail, and pointed and slender body and head.

often strongly compressed in adult males, its length more than 1.55 of body length; the mean number of supra- and infralabials significantly lower than those of the other subspecies; a rudimentary nuchal crest often present.

Description of holotype: an adult male, preserved in 70% ethyl alcohol in good condition; head pointed with a slightly convex forehead, its length 1.36 of its width and 0.27 of body length and 0.15 of tail length; canthus rostralis continued as a supraciliary ridge which is rather strongly developed, composed of 7 scales on each side; nostril as a horizontal slit, slightly above canthus rostralis, pierced in a triangle-shaped scale, posteriorly directed; 4 internasals in a single transverse row; upper head scales subequal, smooth, or slightly rugose, imbricate and subimbricate; 14-14 upper- and 13-13 lower labials; tympanum horizontally elliptical, smaller than orbit, partly covered above by 6-8 spinose scales; no reversal imbrication of scales on the posterior part of head and anterior part of neck; a rudimentary nuchal crest composed of 6-7 spinose scales; gular sac moderately developed and rather pointed posteriorly; gular region covered by large and small, imbricate, and slightly keeled scales intermixed; gular fold and a fold in front of shoulder strongly developed; body slender and distinctly compressed laterally, a prominent vertebral ridge throughout the dorsum; limbs distinctly slender; dorsal scales subequal to homogeneous, rather large,



Figure 9. *T. a. pakistanensis*, holotype (SMF 63258); posterior ventral region. Note the presence of only one row of callose preanal scales and distinctly slender hindlimbs.

imbricate, almost entire individual scale distinctly keeled (unlike the eastern populations of *T. a. agilis* in which only the proximal part of scales are keeled) and mucronate; median dorsals larger, grading into distinctly smaller scales of dorso-lateral region which are only slightly keeled and mucronate; 72-73 scales round the widest part of body; ventral scales rather distinctly keeled, imbricate, large, but slightly smaller than median dorsals, 70-71 in a single longitudinal row from gular fold to the anterior edge of anus; scales of upper surface of limbs almost as large as median dorsals, strongly keeled and mucronate; lower surface of digits covered by bi- and tri-carinate keeled lamellae, 24-25 under the fourth toe; preanal callose scales only in one row, not exceptionally developed, encompassing 8 scales; all caudal scales distinctly keeled and larger than median dorsals, 33-35 around base of tail, just behind the vent; tail long and strongly compressed throughout, its length 1.81 times of body length.

Coloration and color pattern: upper surface of head yellowish-grey; dorsum uniformly sandy-grey with numerous light scales scattered throughout; upper surfaces of limbs olive-brown; upper caudal region as dorsum in coloration with barely distinct dark-brown rings; gular region, chest, and flanks heavily suffused by lavender-blue; other ventral surfaces yellowish-white; no black patch on the shoulder fold.

Measurements (mm): Snout-vent length (SVL) = 101.6, Tail length (TL) = 184, Head length (HL) = 28.2, Head width (HW) = 20.7, Head depth (HD) = 13.5, Length of forelimb = 50, Length of hindlimb = 73.

Variation of the type series: all paratypes are similar to the holotype both in morphology and meristics. In all specimens nostril is almost above the canthus rostralis and a rudimentary nuchal crest, more or less,

developed. The range of scale counts around body for the whole series ($n = 7$) is 69-81 (mean 74.42). The range of ventral scales from gular fold to the anterior edge of anus (in a single longitudinal row) varies from 67-77 (mean 74.57). In all paratypes the number of subdigital lamellae under the fourth toe varies from 22-26 (mean 23.85). The number of supra- and infra labials varies from 13-16 (mean 14.55) and 12-16 (mean 14.25) respectively. Dorsal scales being homogeneous or subequal and, more or less, set off from the small dorsolaterals.

However, there are some differences in morphology between males and females:

- **Male paratypes:** all male paratypes ($n = 4$) resemble the holotype in almost all relevant details; the mean SVL = 90.5 mm, TL = 155 mm; mean TL 1.71 times as mean SVL; the preanal callose scales almost only in one row (in the case of two, the second row rarely developed), encompassing 10-11 scales (mean 10.4); median dorsal scales enlarged, homogeneous, distinctly keeled throughout and mucronate, forming distinct ridges along dorsum, grading, rather abruptly, into small dorsolaterals which are weakly keeled and mucronate; all ventral scales, as in holotype, more or less keeled, rather large, but smaller than median dorsals; tail strongly keeled in almost all specimens, although body not as strongly compressed and pointed as in the holotype; color pattern almost as in holotype.

- **Female paratypes:** in females ($n = 3$), the body and tail are normal (neither distinctly compressed nor slender), the dorsal scales are moderately keeled and mucronate and median dorsals are not clearly set off from the small dorsolaterals; as well, ventrals are slightly keeled and distinctly smaller than the median dorsals; callose preanal scales absent; gular sac not as well developed as in the holotype; in color pattern some of them are rather different from the holotype in the presence of, more or less pronounced, dark cross bars and a series of vertebral light ocelli and in the absence of ornamentation in the lower parts of body; ventral surfaces being uniformly whitish.

Habitat: the habitat of this subspecies is characterised by flat alluvial plains as well as some high slopes. The vegetation consists of some grass, herbs, and stunted shrubs. Some populations occur on the lowlands around Karachi, and the eastern part of the range is a typical desert known as Thar Desert in eastern Sind extending into adjacent northwestern India (the great Indian Desert). This desert mainly consists of sandy hills which vary from small dunes to hills with 100-150 m elevation. In summer, dust storms are the main feature of the area (Khan, 1980).

Distribution: *Trapelus agilis pakistanensis*, as the southeasternmost subspecies of *T. agilis* complex, is restricted in distribution to the lowland and semi-desert regions of Sind province, southern Punjab, and some regions of eastern Baluchistan (southern and southeastern Pakistan), from around Hab River in the west through Karachi and Thatta to the vicinity of Hyderabad and Mirpur Khas eastward into the Indian Desert (Fig. 7). Biswas and Sanyal (1977) recorded this lizard inside the Indian territory (from Jaisalmir, Kolayat, Pugal, Phalodi and some other localities in Rajasthan Desert, northwestern India); to the north, it is distributed along the Kirthar Range up to the areas south of Khuzdar (south-central Pakistan). Apparently the Hab River serves as a barrier for further distribution of this taxon towards the west; however it is parapatric with its *T. a. agilis* in the eastern regions of Baluchistan province. In the east, it goes up to the Nagaur District, Rajasthan, northwestern India.

Etymology: *Trapelus agilis pakistanensis* is so named as it is restricted in distribution to the lowland and semi-desert regions of southeastern Pakistan and adjacent northwestern India.

Taxonomic and biogeographic account

Detailed discussion concerning systematics and patterns of geographic variation in *Trapelus agilis* complex is presented elsewhere (Rastegar-Pouyani, Manuscript a-b) and here is not dealt with in details. As a brief account, however, it can be mentioned that I classified all populations of *Trapelus agilis* complex throughout the range into four distinctive taxonomic entities (and as the most parsimonious definition = subspecies): *T. a. agilis* (Olivier, 1804), *T. a. sanguinolentus* (Pallas, 1814), *T. a. khuzistanensis* ssp.nov.1, and *T. a. pakistanensis* ssp.nov.2.

An ANOVA-based pairwise comparison showed that in most metric and meristic characters these four taxonomic entities are significantly different ($P < 0.05$) (Rastegar-Pouyani, Manuscript, a). Also employing multivariate statistical techniques (principal component analysis, canonical variate analysis, and cluster analysis), to a great extent, showed the objectivity of these four distinct groups within *T. agilis* complex and re-confirmed my previous taxonomic decisions (Rastegar-Pouyani, Manuscript, b). Of the four subspecies of *T. agilis*, the nominal form (*T. a. agilis*) occurs in a wide range of habitats, shows higher degree of morphological variability, occupies the central and southern parts of the species range (these regions might have served as a refugia during intervals of unfavorable and cold climatic conditions in the Tertiary and Quaternary), and consists of two western and eastern

groups of populations (clines) (Rastegar-Pouyani, in press, unpublished manuscript). With regard to these factors, it could be logical if we consider this form as the central core of the complex and as the parental population from which the other subspecies have been derived. So, preliminarily, I propose the following scenario for origination and subsequent evolution of the three marginal subspecies which occur in the periphery of the main range of the complex being parapatric with the central continuum (= *T. a. agilis*):

It seems that *T. a. sanguinolentus*, in spite of being distributed over a very wide area, is the most recently-evolved group, originated from the parental populations in the southern parts of the range (apparently) in the very Late Pliocene and Pleistocene (2-1.2 MYBP =millions of years before present), invading towards the northern and northeastern regions during intervals of favorable climatic periods. The very low degree of variability observed in this subspecies is indicative of its recent history. *Trapelus a. pakistanensis* separated from the central continuum in the southeastern parts of the range (probably by dispersal or, less likely, due to a vicariant event) distributed towards the east and reached as far east as northwestern Indian desert. This invasion might have taken place in the Pliocene (5-1.7 MYBP). In the southwestern part of the range, a drastic vicariant event (the huge orogeny of the Zagros) separated the southwesternmost populations from rest of the complex and from the parental populations. These isolated populations served as founders and, with further divergence, gave rise to *T. a. khuzistanensis*. Apparently, this vicariant event has taken place in the Late Miocene or early Pliocene (7-4.5 MYBP). Therefore, we can say that both dispersal and vicariance have been involved in radiation and subsequent evolution of various subspecies of *Trapelus agilis* complex though the role of dispersal in evolution of *T. a. sanguinolentus* and *T. a. pakistanensis*, and vicariance in evolution of *T. a. khuzistanensis* are more prominent.

Trapelus agilis (Olivier, 1804) is the easternmost representative of an essentially homogeneous and similar group of about five species complexes which also include *T. persicus* (Blanford, 1881) in the Mesopotamian Plain and lowland southwestern Iran, *T. flavimaculatus* Rüppell, 1835 in Saudi Arabia, *T. savignii* (Dumeril and Bibron, 1837) in Israel and eastern Egypt, and *T. turnevillei* (Lataste, 1880) in north Africa (north of Sahara). Although the monophyly of *Trapelus* has been shown by Moody (1980) with a morphological approach and by Joger (1991) using molecules (neither Moody nor Joger studied all the species of *Trapelus*) but, so far, no comprehensive

revisionary study has been done on all species of this genus and this is mainly because of political instability of the region and difficulties in collecting proper material throughout the range. Furthermore, even in a few studies done on a limited number of *Trapelus* species, the results obtained by morphological and the other approaches (e.g., immunological) were contradictory (e.g., Anderson, in press; Joger and Arano, 1987; Rastegar-Pouyani, in press, unpublished manuscript).

Trapelus is mainly Saharo-Sindian in distribution, often associated to the lowlands, desert, and semi-desert regions with high annual temperature.

Key to the subspecies of *Trapelus agilis* complex

- 1a. Tail almost always round, two (or more) rows of callose preanal scales 2
- 1b. Tail often compressed, often only one row of callose preanal scales, in the case of two, the second undeveloped (preanals absent or weakly developed in females). 3
- 2a. Body size variable: 65-91 scales around body; dorsal scales subequal, weakly to moderately keeled, often strongly mucronate; ventral scales smooth or weakly keeled; usually 2, sometimes 3 (rarely 4-5) rows of callose preanals; background coloration variable; central Iranian Plateau, central and southern Afghanistan, southwestern Pakistan. . . *Trapelus agilis agilis* (Olivier, 1804).
- 2b. Body stout: 52-73 scales around body; all dorsal, ventral, and gular scales larger in size, homogeneous, strongly keeled and mucronate; almost always 2 (rarely 3) rows of callose preanals; background coloration of males often dark sandy-grey; northeastern Iran, northern Afghanistan, Central Asian Republics, western coast of the Caspian Sea (Daghestan), western China. . . . *Trapelus agilis sanguinolentus* (Pallas, 1814).
- 3a. Body and limbs smaller than those of the other subspecies and sometimes relatively slender, not compressed; head and neck distinctly short; all body scales smaller than those of the other subspecies; dorsal scales subequal to unequal (heterogeneous), weakly to moderately keeled, weakly mucronate; 80-97 scales around body; ventral scales slightly keeled; scales of posterior part of head and anterior part of neck reversally imbricated; upper head scales keeled or rugose; often 14-19 upper- and lower labials; background coloration often yellowish grey-cream; lowland southwestern Iran (0-900 m elevation)
 *Trapelus agilis khuzistanensis*, ssp. nov.1

3b. Body and limbs often distinctly slender, sometimes compressed in males; dorsal scales subequal to homogeneous, distinctly keeled and mucronate, usually clearly set off from small dorsolaterals; 67-83 scales around body; ventrals distinctly keeled in adult males; no reversal imbrication of head and neck scales; upper head scales often smooth; 12-16 upper- and lower labials; background coloration often sandy-grey; southeastern Pakistan and adjoining northwestern India . . . *Trapelus agilis pakistanensis*, ssp. nov.2

Abbreviations

BMNH = British Museum (Natural History; (London, UK), CAS = California Academy of Sciences (San Francisco, USA), FMNH = Field Museum of Natural History (Chicago, USA), GNMH = Göteborg Natural History Museum (Göteborg, Sweden), MNHN = Museum National d'Histoire Naturelle (Paris, France), MZLS = Museo Zoologico de "La Specola" (Firenze, Italy), NMW = Naturhistorisches Museum Wien (Vienna, Austria), SMF = Museum und Forschungsinstitut Senckenberg (Frankfurt, Germany), SMNH = Swedish Museum of Natural History (Stockholm, Sweden), ZFMK = Zoologisches Forschungsinstitut und Museum Alexander Koenig (Bonn, Germany); ZISP = Zoological Institute St. Petersburg (St. Petersburg, Russia), ZMUC = Zoological Museum University of Copenhagen (Copenhagen, Denmark).

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Appendix 1. Material examined

Trapelus agilis agilis (n =541)

MNHN 5708 (2) = 1994. 1178 (2) (Olivier's syntypes):

- 170987, FMNH 170988: Iran, Gilan prov. 4.2 mi N of Tukestan (Zanjan prov. Takestan?). NMW 33166-1, Iran: Fars prov. 9 km N of Abadeh. NMW 33167, NMW 33085-2: Iran, Hormozgan prov. 14km E Bandar-Abbas. NMW 7276-1, NMW 7276-2: Iran: Kerman prov.? Sabzewaran. NMW 7276-3: Iran: Kerman prov.? Rigmati, 150 km SE of Sabzewaran. NMW 7276-4: Iran, Esfahan prov. 135 km N of Esfahan. NMW 33159-3, NMW 33159-4, NMW 33159-5, NMW 33165-1: Iran, Esfahan prov. Murcheh-Khurt, N of Esfahan. NMW 33165-2, NMW 33165-3, NMW 33085-1: Iran: Esfahan prov. 4 km SE Robat-Tork (165 km NW of Esfahan). NMW 33085-3: Iran, Zanjan prov. 26 km SE Ghazwin. NMW 33083-1, NMW 33083-2, NMW 33083-3: Iran, Zanjan prov. 41 km SE Ghazwin. Elev: 1380 m. NMW 33164: Iran, Tehran prov. 60 km S of Tehran. NMW 33084-3: Iran, East Azarbaijan prov. 42 km SE Mianeh. Elev: 1200 m. NMW 33166-1: Iran, Kerman prov. 15 km SE Kerman. SMNH 3139-1, SMNH 3139-2, SMNH 3139-3, SMNH 3139-4, SMNH 3139-5. BMNH 85. 5. 27. 14. BMNH 85. 5. 27. 15, BMNH 85. 5. 27. 16, BMNH 85. 5. 27. 17, BMNH 85. 5. 27. 18, BMNH 85. 5. 27. 19: Iran: Tehran (exact locality?). GNHM. Re.ex. 4437, GNHM. Re.ex. 3324, GNHM. Re.ex. 4395-1: Iran, Gilan prov. 65 km SSW of Rasht. GNHM Re.ex. 4395-2: Iran, Tehran prov. 150 km S of Tehran, Siah-Kuh, Shah-Abbas post. GNHM Re.ex. 4396-1, GNHM. Re.ex. 4396-2: Iran, Mazandaran prov. 150 km E of Gorgan, Golestan Park. La Specola 30584, La Specola 30585: Iran, Tehran prov. 50 km S Qum? BMNH 1920. 3. 20. 1: Iran, Hamadan prov. Hamadan, Jinjan? BMNH 1936. 10. 12. 1: Iran, Kerman prov. N. Of Kerman, Sekonj, 7000 ft. BMNH 1934. 12. 16. 3, BMNH 1936. 12. 16. 4: Iran, Sheakuh, seat (Salt?) desert. BMNH 1912. 3. 26. 13, BMNH 1912. 3. 26. 14, BMNH 1912. 3. 26. 15, BMNH 1906. 8. 10. 25, BMNH 1900. 5. 9. 9, BMNH 1900. 5. 9. 10, BMNH 1900. 5. 9. 11, BMNH 1900. 5. 9. 12, BMNH 74. 11. 23. 104, BMNH 74. 11. 23. 105, BMNH 74. 11. 23. 106, BMNH 74. 11. 23. 107, BMNH 74. 11. 23. 108, BMNH 74. 11. 23. 109, BMNH 74. 11. 23. 110, BMNH 74. 11. 23. 111, BMNH 74. 11. 23. 112, BMNH 74. 11. 23. 113: Iran, Sistan-Baluchestan prov. BMNH 1951. 1. 6. 50, BMNH 1951. 1. 6. 51-2, BMNH 94. 11. 13. 4, BMNH 94. 11.13. 5: Iran, Hormozgan prov. BMNH 87. 12. 20. 1: Iran, Hormozgan prov. Kishim (=Gheshm?) Island, Persian Gulf. BMNH 79. 8. 15. 10-15: Iran, Fars prov. Dehbid, north of Shiraz. BMNH 1903. 3. 14. 1: Iran, Bushehr prov. Bushehr. BMNH 1933. 4. 1. 20-22: Pakistan, Waziristan, N. W. F. P. BMNH 86. 9. 21. 17, BMNH 86. 9. 21. 18, BMNH 86. 9. 21. 23, BMNH 86. 9. 21. 24, BMNH 86. 9. 21. 25: Afghanistan: Helmand. ZMUC R36133, ZMUC R36149-54, ZMUC R36160, ZMUC R36204-5: Afghanistan: Seistan prov. Faisabad (south of Afghanistan), ZMUC R36155-9, ZMUC R36145-8, Afghanistan, Seistan prov. Baqrabad. ZMUC R36161: Afghanistan, Seistan prov. Faisabad-Farah. ZMUC R36206: Afghanistan, Kandahar prov. Przadah, W of Kandahar. ZMUC R36208-9: Afghanistan, Kabul prov. near Kabul? ZISP 7361: Afghanistan: Harat. NMW 33170-12: IRAN, East Azarbaijan, 42 km SE Mianeh. NMW 33173-1-2: IRAN, Zanjan prov. 26 km SE Qazwin. NMW 33170-1: IRAN, Tehran prov. 95 km S Tehran. NMW 33148-1-10: IRAN: Tehran prov. Zaveyeh, 80 km SW Tehran. NMW 7276-5-6: IRAN, Kerman prov. Sabzewaran. NMW 33175-5, NMW 33170-5, NMW 33170-6, NMW 33170-7: Iran, Esfahan prov. NMW 33172-3-4: IRAN: Kerman prov. 138 km S Rafsanjan. NMW 33172-5: IRAN: Kerman prov. 29 km SE Sirjan. NMW 33165-4: IRAN, Kerman prov. 110 km SW Kerman. NMW 33172-2, NMW 33175-6. NMW 33175-7: Iran: Yazd prov. 200km SE Yazd. NMW 33174-1-8: IRAN, Khorasan prov. 15-25 km S Qayen. NMW 33163: Pakistan: Nepandgur (where?). NMW 33162: Pakistan: Delbandin. NMW 33161-1-2: Iran, Shirgesht Beiteabas. NMW 33161-3: Iran, eastern Iran, Ozbak-Kuh. NMW 33172-6: Pakistan, Baluchestan prov. 50 km W Nushki. NMW 33150-9: Iran W Sangbast. NMW 24763-1-3: Iran, N Persian, Kuh Dasehteh (Taj-abad). NMW 24767: Iraq, Baghad? (Paris Museum). CAS 120280-1: Afghanistan, 10km NE Darweshan (Central Afg.). CAS 84642-45: Afghanistan, 35 mil. down stream from Girishk. Dasht-e-Margo area, Chah-e-Angir, (Central Afg.). CAS 120242-4 Afghanistan, 12 km S Lashkargah (near Girishk), 2700ft. CAS 97990: Afghanistan, 20 mil. SE Kandahar, 31 23 N, 65 53 E, 3800 ft. CAS 90762-9: Afghanistan, Sharisafa, 60 km NE Kandahar. 1400m. CAS 90777: Afghanistan, Tarnak river, 75 km NE Kandahar. 1405m. CAS 120276-9: Afghanistan, 30km S Ghazni-Qalat 7100ft. FMNH 20987-1-10: Iran, Esfahan prov. Yazd-e-Khast .FMNH 20985-90: Iran, Esfahan prov. FMNH 20988-1-2: Iran, Daria-Masila. FMNH 245507-10: Pakistan, Baluchistan prov. SMF 63226-37, SMF 63262, SMF 63285, SMF 63255: W-Pakistan, Siah-Kuh, S Delbandin. BMNH 1951.1.6.54: Iran, Bandar-e-Lengeh. BMNH 1919.5.2.2-3: Iran, Fars prov. Abadeh. BMNH 1936.10.12.3: Iran, Fars prov. between Quatru-Chah Salz to Neiriz road. BMNH 1966-355-57: Iran, Kerman prov. 148 km E Neiriz on Zaidabad road to Sirjan. BMNH 1951.1.2.20-22: Iran, Kerman prov. 20 mil. S of Kerman. Jupar. BMNH 1951.1.6.45-48: Iran, Sistan prov. Khash, SE Iran. BMNH 1940.3.1.19-24: Afghanistan, Ghazni, (E Afgh.). CAS 141028: Iran, Kerman prov. 17km SSE Minab on inland road to Jask. CAS 141020: Iran, Kerman prov. 13 km E of Eastern edge of city of Kerman. CAS 141051: Iran, Kerman prov. 19km SE Shagu on road to Minab. CAS 141027: Iran, Kerman prov. 21 km N Rudan on road to Jiroft. CAS 141097: Iran, Sistan prov. 10km SW Hirmand, abandoned village, SE of road from Zabol to Dust-e-Mohammad Khan. CAS 141065: Iran, Baluchistan prov. 13km southerly of Zahedan on road to Khash. CAS 102484-90: Iran: Sistan prov. 15 mi SW Zabol. CAS 102491-2: Iran, Fars prov. Ahram. CAS 141149: Iran, Fars prov. 5km northerly from Dalaky on road to Shiraz where foothills begin. CAS 96270: Iran, Khorasan prov. Tayebat, about 10 mi from Afghanistan border. NMW 33150-1-6: Iran, Khorasan pro. 5 km N Taybad. NMW 33166-1: Iran: Kerman prov. 15 km SE Kerman.
- Trapelus agilis sanguinolentus (n =238)***
GNHM Re.ex. 4396 (1-2): Iran: Mazandaran prov. 150 km E of Gorgan, Golestan Park. GFN 40: Turkmenistan, Lowland steppe, East Kopet Dagh. GNHM. Re. ex. GFN 41: Turk-

menistan, Murgab River, 150 km S of Marv (Mary). GNHM. Re. ex. GFN 42-50: Turkmenistan, Sahra-Bairam-Ali, 45km NW of Marv, Kara kum desert. CAS185104-9, CAS 185134-5: Turkmenistan, Ashgabad region [38 00 N, 58 00 E]. CAS 184570-6: Turkmenistan: Krasnovodsk region [39 45 N, 54 33 E]. GNHM. Re. ex. GFN 35: Turkmenistan, Archenjan village. GNHM. Re. ex. GFN 36: Turkmenistan, Krasnovodsk region [39 45 N, 54 33 E]. GNHM. Re. ex. GFN 43 (1-2): Turkmenistan, Kaka. GNHM Re. Ex 10622-3: Kazakhstan, at the lake (artificial) at Illi River, near village Bokter [43 54 N, 77 16 E]. ZISP 5109, 13701-15 Kazakhstan, E. Kazakhstan, Illi River, near border of China. ZISP 5796: Kazakhstan, Tardski, Dshungaria, between Kazakhstan-China. ZISP 20298 (7745-79): Kazakhstan, near Ilisk city, Illi River, E. Kazakhstan. ZISP 15143, 17329, 13695, 1168: Kazakhstan, near Ilisk city, Illi River, E. Kazakhstan. ZISP 11070 (1-12): Kazakhstan, Illi River, E. Kazakhstan, close to lake Balkhash. ZISP 19115 (7643-84): Uzbekistan, Vicinity of Nucus city, Caracal, near Aral sea. ZISP 19398 (7428-48): Uzbekistan-Tajikestan. Fergan Valley. ZISP 15803 (1-15): Tajikistan, Fergan Valley. ZISP 20097(371-87), 10715 (3-13): Tajikestan-Afghanistan border. Termez. ZISP 13588, 13590 (1-12), 13592, 6914 (1-10): Kazakhstan-Uzbekistan, between Aral Lake and Caspian Sea. ZISP 19392 (1-24): Russia (west of Caspian Sea), Daghestan. ZISP 15753: Iran, Astarabad (= Gorgan?). CAS 183032-39: Russia, Chechen-Ingush Autonomous Republic, the lowland between Terek-Kuma Rivers [43 21 N, 46 06 E]. CAS 120249-50: Afghanistan, 30-70 km E of Herat 3700-5350 ft. CAS 115922-3: Afghanistan, Maimana, 35 54 N, 64 43 E. 884m. CAS 120280-1: Afghanistan, 10km NE Darweshan (Central Afg.). CAS 120275: Afghanistan, 25 km E Khanabad (between Mazare-Sharif-Faizabad), 2400ft. CAS 115920: Afghanistan, northeastern Afghanistan, 64 mi by road E Faizabad, 37 05 N, 70 40 E. CAS 115921: Afghanistan, Paghman Vicinity, 34 36 N, 68 56 E. 2440m. CAS 120251: Afghanistan, 25km NW Pul-e-Khumri, 2400ft. (near Mazar-sharif?). CAS 120255: Afghanistan, 20 km E Mazar-e-Sharif. CAS 120253: Afghanistan, 10 km W Tashkurgan, Near Mazar-Sharif? CAS 120256-8: Afghanistan, 20-50 km E Mazar-e-Sharif. CAS 120259-61: Afghanistan, 45 km W Mazar-e-Sharif, 1500ft. CAS 120273: Afghanistan, 50 km W Mazar-e-Sharif. FMNH 161197-9: Afghanistan, Maimana, 35 54 N, 64 43 E 884 m. FMNH 161133: Afghanistan, 64mil E Faizabad, 35 05 N, 70 40 E. FMNH 161191-2: Afghanistan, Paghman vicinity, 34 36 N, 68 56 E. 244m. FMNH 141399: Iran, Mazandaran prov. 1 mi N of Pahlavi Dezh.

Trapelus agilis khuzistanensis (n =97)

GNHM. Re. ex. 5424 (holotype): Iran, 5 km NW Haft-Gel on the road to Shushtar. GNHM. Re. ex. 5425 (paratype): Iran, Khuzistan prov.. 5 km NW of Haft-Gel on road to Shushtar. GNHM. Re. ex. 5426 (paratype) : Iran, Khuzistan

prov., 38 km S of Masjid-e-Sulaiman, Golgir village. CAS 86403-6, 86418-19 (paratypes), : Iran, Khuzistan prov. along road to lake east of Haft-Kel, CAS 86464 (paratype): Iran, Khuzistan an prov. along road south of Shushtar, CAS 86556 (paratype): Iran, Khuzistan prov. Haft-Kel (on golf course) [31 28 N, 49 30 E]. CAS 86342, 86346, 86390 (paratypes): Iran, Khuzistan prov. Tuli-Bazum road [31 55 N, 49 25 E]. FMNH 170936 (paratype): Iran, Khuzistan prov. 53 mi SE Ahwaz , Mashraheh. CAS 86341, 86343-72, 86374-89: Iran, Khuzistan prov. Tuli-Bazum road [31 55 N, 49 25 E]. CAS 86320, 86323-28: Iran, Khuzistan prov. Masjid-Suleiman [31 57,N, 49 16,E]. CAS 86322: Iran. Binak, near Persian Gulf at foot of Kuh-e-Bang [29 44,N, 50 19,E]. CAS 86338-51: Iran, Khuzistan prov. along old Masjid-Suleiman and Ahvaz road [32 N, 49 11,E]. CAS 86251: Iran, Khuzistan prov. Masjid-i-Suleiman [31 57 N, 49 16 E]. CAS 86373: Iran. Khuzistan prov. road south of Shushtar. CAS 102491-2: Iran. Fars prov. Ahram. CAS 86625: Iran, Khuzistan an prov. Binak, on Persian Gulf at foot of Kuh-i-Bang, north of Ganaweh. [29 44 N, 50 19 E]. CAS 86487: Iran, Fars prov. Agha Jari. [29 48 N, 49 46 E]. FMNH 141392-3, 141395-6, 141398: Iran, Fars prov. Ahram. ZISP 10335: Iran, Kochrud, Irak-Adschemi (exact locality?) (Nikolsky's Type of *Agama kermanensis* , var. *brevicauda*). ZISP 9321: Iran, Kochrud, Irak-Adschemi (exact locality?) (Nikolsky's Type of *Agama kermanensis*). ZISP 9889: Iran (exact locality ?).

Trapelus agilis pakistanensis (n =32)

SMF 63258 (holotype), 63259, 63279 (paratypes): Pakistan, Gaj-River, Kirthar Range. SMF 63256 (paratype): Pakistan, Sonda Distr. Thatta. SMF 63236: Pakistan, Old Airport of Karachi. 63243-4 and 63286-7 (paratypes): Pakistan. Old Airport of Karachi. SMF 63239, 63264: Pakistan, Hab River at Goth Mauladad. SMF 63263: Pakistan, Jati, Sind. SMF 63276: Pakistan, Karangee at Karachi. SMF 63265: Pakistan, Karachi. SMF 63267: Pakistan, Karachi area. FMNH 224946: Pakistan, Sind Prov. Dadu dist. Ranicot. FMNH 244977: Pakistan, Sind Prov. Karachi Dist. Malir, contonment. FMNH 244948: Pakistan, Sind Prov. Karachi Dist. Malir, contonment. BMNH 1933.7.8.23: Pakistan, Salt Range, Punjab. BMNH 74. 4. 29. 1432. 80. 11. 10. 25: Pakistan. Sind area. BMNH 98, 12, 22, 7-8: Pakistan: Kurrachee (karachi). BMNH 1964-271: Pakistan, Mirpur Khas. BMNH 1933.12.7.1: Pakistan, Sind area.

Rhacophorus leucomystax in Vietnam with Acoustic Analyses of Courtship and Territorial Calls

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Abstract.— Acoustic parameters of both courtship and territorial calls were analysed for two populations of Vietnamese treefrogs, genus *Rhacophorus*. These treefrogs from Ba Be National Park and nearby Na Hang Nature Reserve were identified as *R. leucomystax* as opposed to *R. megacephalus*, based on an acoustic analysis of courtship calls. They represent the first confirmed record of this species outside of Borneo and suggest that the species may also occur in China. The courtship call, composed of a long note of similar pulses and pitch, has a mean dominant frequency of 1940 ± 154 Hz at Ba Be National Park and 1950 ± 70 Hz at Na Hang Nature Reserve. The territorial call, a two to three note call, has a mean dominant frequency of 1940 ± 124 Hz at Ba Be National Park and 2190 ± 256 at Na Hang Nature Reserve. No significant differences were found between the call characters of these two populations of Vietnamese *R. leucomystax*. There was no significant association between snout-vent length and dominant frequencies.

Key words.— Amphibia, Anura, call analysis, Rhacophoridae, *Rhacophorus leucomystax*, Vietnam, vocalizations.

Introduction

Until recently, the amphibian fauna of Vietnam has been based on the works of Bourret (1942), and accounts from neighbouring China (e.g., Yang, 1991; Zhao and Adler, 1993; Zhao, 1995). The Chinese occurrence of one species of treefrog, *Rhacophorus leucomystax*, was recently placed into doubt by Matsui et al. (1986) based on chromosomal, morphological and acoustic evaluations. These authors reported that *R. leucomystax* was restricted in distribution to Borneo whereas a similar species, *R. megacephalus*, occurred in Taiwan. They stated that mainland Chinese populations of these frogs required further study, but in the interim, mainland Chinese populations should be referred to as *R. megacephalus*. Zhao and Adler (1993) reported that *R. leucomystax* did not occur in China, but rather that *R. megacephalus* occurred throughout the country. Our recent field work in northern Vietnam revealed the presence of two apparent forms of treefrogs similar in appearance to *R. megacephalus* and/or *R. leucomystax*. We successfully recorded vocalizations of one form from two allopatric populations. Herein, we report our analysis of the acoustic parameters of both courtship and territorial calls, including the dominant frequency, which is species specific.

Anurans are capable of different types of vocalizations which serve different functions. The courtship

call, also known as the mating or breeding call, is emitted by males and has two functions: the attraction of conspecific females and the announcement of an occupied territory to other males of the same or different species. There are three types of courtship calls: the courtship call produced by males in attempt to attract a conspecific female, a territorial call produced by a resident male in response to an courtship call received above a critical threshold of intensity, and an encounter call evoked during close range agnostic interactions between males (Duellman and Trueb, 1994). The courtship call acts as a courtship isolating mechanism (Duellman and Pyles, 1983). A second type of call, the territorial call, is comprised of acoustic signals and is accompanied by corporal vibrations produced by a male or an unreceptive female in response to amplexus. Other types of calls include the reciprocation call which is given by a receptive female, and the distress call which is delivered in response to an enemy or predator for defence.

Signals are found to vary between and within individuals and between geographically separated populations of a single species (Cocroft and Ryan, 1995). Anurans use these differences in call characteristics to identify individuals of the same or different species and to signal intent. Frost and Platz (1983) proposed that these interspecific and intraspecific differences in fundamental call characteristics such as pulse rate, duration of call, dominant frequency, or a combina-

tion of these parameters, permits females to reduce the likelihood of error in mate choice. Anurans recognize individuals of the same and different species by their dominant frequencies.

Duellman and Pyles (1983) discovered that smaller frogs tend to call at higher frequencies and have a reduced auditory sensitivity compared with larger frogs. They concluded that an upper limit of approximately 5000 Hz is most common. Furthermore, the reception of acoustic signals is affected by habitat and interference from synchronously calling species. In rainforest habitats, which are complex habitats, anurans were found to produce sounds at lower frequencies because the dense vegetation attenuates sound waves, notably those at higher frequencies. Therefore, transmission frequencies of less than 4000 Hz would be most effective for anuran communication in such complex habitats. Partitioning of the acoustic community can be affected by several factors such as type of call produced, oviposition site, environment and the onset of the breeding season (Duellman and Pyles, 1983). But within any given community, the available acoustic environment is partitioned distinctly based on its particular ecological and geographic assemblage of anurans. Although selection may operate to maximize acoustic properties for species recognition, acoustic interference from factors of the physical habitat and from community members also operates to minimize acoustic variability. This implies that members of the same breeding fauna have voices with common characteristics (Duellman and Pyles, 1983).

Material and Methods

Frog calls were recorded and calling specimens collected from two different localities in northern Vietnam. Approximately 24 frogs of the *R. leucomystax* complex were recorded and collected in Ba Be National Park (22°24'N 105°37'E) from 15 to 29 May 1995. Five *R. leucomystax* were recorded and collected in the village of Pac Ban (22°21'N 105°23'E) located in Na Hang Nature Reserve, from the 23 May to 3 June, 1996. In both locations, frogs were selected at random and a sufficient call sample was recorded using a Realistic™ unidirectional microphone and a Marantz™ PMD 201 portable cassette recorder. Air temperatures were recorded in degrees Celsius at Ba Be National Park at the time of capture, using a thermometer; each specimen was collected during evening hours from a concrete pool adjacent to the Park's research centre. In Pac Ban, frogs were collected from various locations, including in rice patties, on trees, and in ponds. All specimens were eutha-

nised, preserved and deposited in the herpetology collections of the Royal Ontario Museum, Toronto, Canada. Calls were transferred from the cassette recorder to a Macintosh computer using a Macrorecorder™ digitizer. Sound Edit Pro™ version 2.0.5 for Macintosh computer systems was used for call analyses. Note duration, dominant frequency, fundamental frequency, notes per call, pulses per note and time between segments were measured. Descriptive analyses were performed for all courtship and territorial calls, using Excel™ 5.0 (Microsoft). Snout-vent length (SVL) was measured for all specimens to the nearest 0.1 mm using digital calipers.

Results

Of the 24 specimens recorded and digitized at Ba Be National Park, 42 courtship calls were analysed from 22 individuals, and 33 territorial calls from 18 individuals. Table 1 summarizes measurements of call parameters for all mating calls.

In most cases, the fundamental frequencies overlapped the dominant frequencies, rendering it impossible to obtain exact measurements. However, in those that were observed, the fundamental frequency appeared to be approximately one half the value of the dominant frequency (900-1000 Hz). Air temperatures did not vary significantly, with a mean of 26.2°C for territorial and courtship calls, thereby having little effect on call characteristics. All courtship calls had one note, and pulses ranged from 2 to 17 per note. In contrast, all territorial calls were either 2 or 3 notes per call, the most common being the former, with variability in numbers of pulses in all three notes (Figure 1).

Of the 5 specimens recorded and digitized in Pac Ban, 2 courtship calls were analysed from 2 individuals, and 21 territorial calls from 5 individuals. Call parameters are summarized in Table 1. Fundamental frequencies were obtained from the sonograms, where values were noted as approximately one half the value of the dominant frequency (800-900 Hz). All courtship calls had one note, and pulses ranged from 4 to 16 notes per call. In contrast, all territorial calls were either 2 or 3 notes per call, the most common being the former, with variability in numbers of pulses in all three notes.

The courtship call resembled the sound of a trill, or long sequence of pulses of similar pitch, whereas the territorial calls were more similar to the sound of clicks. Dominant frequencies were consistent within all calls; mean dominant frequency in courtship calls were 1940 Hz for the Ba Be population and 1950 Hz

Table 1. Call measurement summary of note duration, notes/call, pulses/note and dominant frequencies for *Rhacophorus leucomystax* in Ba Be National Park and Pac Ban in Na Hang Nature Reserve.

	Ba Be National Park		Na Hang Nature Reserve	
	Courtship	Territorial	Courtship	Territorial
Note duration (secs)	0.33±0.24	0.08±0.02	0.64±0.54	0.08±0.02
Notes/call (range)	1	2-3	1	2-3
Pulses/note (range)	2-17	1-3, 1-5, 2	4-16	1-3, 2-4, 1-4
Dominant frequency (Hz)	1940±154	1940±125	1950±70	2190±256

for the Pac Ban population. For territorial calls, the mean dominant frequency was also 1940 Hz in Ba Be and 2190 Hz in Pac Ban. Therefore, *R. leucomystax* has a species-specific dominant frequency of approximately 2000 Hz. A consistency between segments or pulses was observed for the territorial calls. The mean time between segments was 0.02 seconds (standard deviation \pm 0.003).

Snout-vent length was measured for all specimens and correlated with dominant frequency. Our regression analyses failed to confirm a significant association ($F=1.948$, $p=0.176$, $r^2=0.08$) between SVL and dominant frequency.

Discussion

Call duration and pulse rate are temperature dependent (Platz, 1989). Regression analysis of call characteristics and temperatures achieve the highest correlation, where a correction function is generated. The correlation function is then used to correct data for temperature differences revealing little variation. For example, a few obvious differences were discovered in the values of the call characteristics as shown in the sonograms for the eastern and western samples of *Rana pipiens* when differences in temperature between the localities and variation within the localities were taken into account (Dunlap and Platz, 1981). However, because temperatures were not significantly variable, temperature values were not used to account for variation in our data. Furthermore, our regression analyses revealed that snout-vent length did not account for variation in our acoustic data, as it does in some other anuran species (Duellman and Pyles, 1983).

Because dominant frequency is characteristic of species, we compared our data with those of Matsui et al. (1986). The dominant frequency of *Rhacophorus leucomystax* ranged from 2250-2550 Hz whereas it

ranged from 1040-1070 Hz in *R. megacephalus*. Further, Matsui et al. (1986) found that *R. leucomystax* had one note/call whereas in *R. megacephalus* it ranged from 2-4 notes/call. Given that the mating calls of our frogs from northern Vietnam had a dominant frequency of 1.94-2.49 kHz (Table 1) and consisted of a single note/call, we conclude that our specimens are best identified as *R. leucomystax* pending additional morphological evaluations (in preparation). Our study sites at Ba Be National Park and Na Hang Nature Reserve are located only 125 km south of Yunnan Province, China. Consequently, given that *R. leucomystax* ranges from at least Borneo to northern Vietnam, the species likely also occurs in China and elsewhere in Asia. Further, it seems likely that it occurs sympatrically with *R. megacephalus*, at least in Vietnam.

Disturbance to the environment may have negative impact on anuran populations. As a result of increased accessibility to Vietnam, tourists are beginning to exploit the relatively primitive country. Such exploitation will likely contribute to increased deforestation and wildlife loss. Human population growth continues to rise in Vietnam at an alarming rate (ca. 2.3%/yr) and deforestation is increasing in quest for construction materials and agriculture for both in-country needs and cash-crop export, especially coffee. Species mining is occurring for export of animals to foreign countries, regardless of species type, number or Vietnamese efforts to protect the species. Littlejohn and Roberts (1975) remarked that the environment of north central Victoria, Australia, had been greatly modified over the last 130 years by increased land clearing for agriculture, timber cutting for building and mining, and by the establishment of extensive irrigation and drainage systems. They suggested that these alterations have influenced the position, extent, and nature of the main zones of intergradation, which may further increase dispersal due to global warming

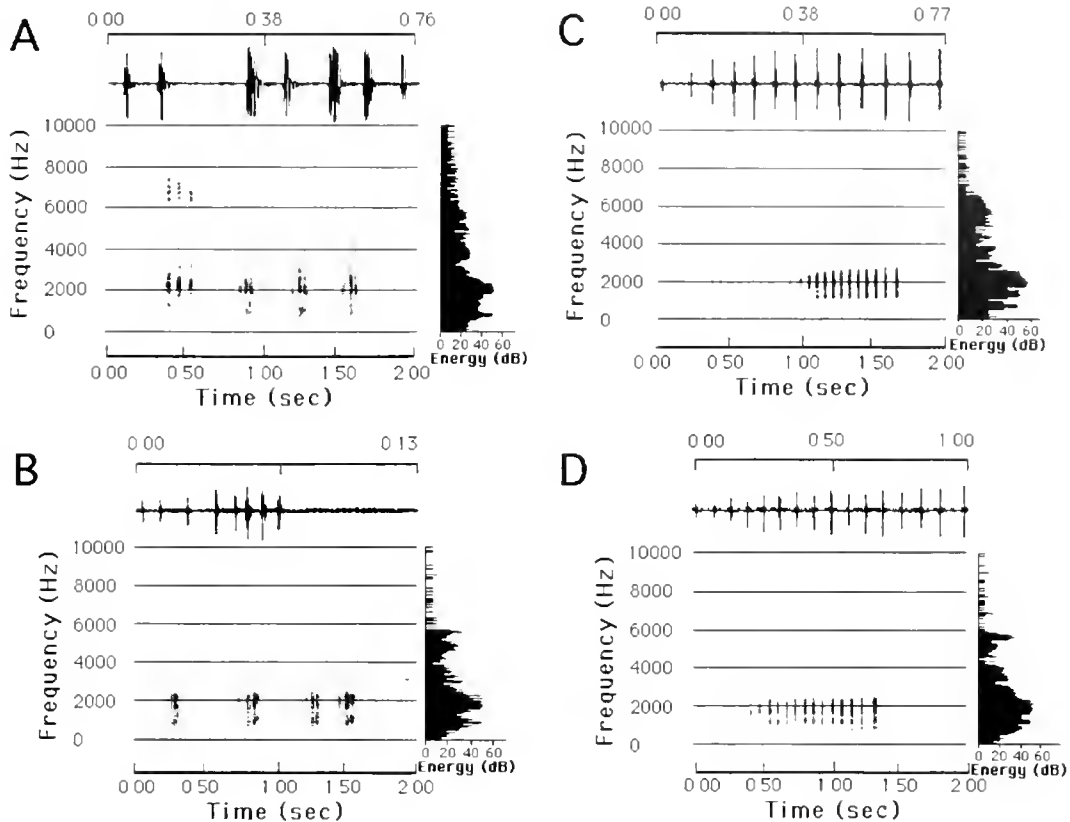


Figure 1. Call characteristics for *Rhacophorus leucomystax* as illustrated through audiospectrograms (frequency vs. energy) and sonograms (frequency vs. time). An expanded oscillogram (above) shows finer details of the call. Territorial calls from A) Babe (ROM 19334); B) Pac Ban (ROM 6514), and courtship calls from C) Babe (ROM 19486); and D) Pac Ban (ROM 6514).

and the concomitant northward movement of anuran populations. The possibility for divergence in mating call structure could then result increasing the frequency of mismatching and nonviable offspring. Like Victoria, it is not known what the future beholds for the relatively fragile, undiscovered and endemic species of Vietnam. Therefore, identification of species for many areas of study, such as behavioural, ecological and evolutionary fields, will be critical in the next few years so that as many species as possible can be studied, identified and conserved.

Acknowledgments

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Immuno-chemical Study of TSV-PA, a Specific Plasminogen Activator from the Venom of *Trimeresurus stejnegeri*

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Abstract.- Rabbit antibodies were prepared against purified TSV-PA, a specific plasminogen activator from the venom of *Trimeresurus stejnegeri*. They strongly cross-reacted with *Crotalinae* snake venoms like *Trimeresurus stejnegeri*, *Trimeresurus mucrosquamatus*, *Agkistrodon halys* and *Agkistrodon acutus*. In contrast, immunological cross-reactions with *Elapidae* snake venoms, *Ophiophagus hannah*, *Naja naja atra* were relatively lower and no cross-reactions with *Bungarus fasciatus*, *Bungarus multicinctus* venoms. On the other hand, enzymatic assays only revealed the existence of plasminogen activation activity in the venom of *Trimeresurus stejnegeri*. Except trypsin, anti-TSV-PA sera and antibodies did not cross-react with other serine proteases, such as physiological urokinase-type plasminogen activator (u-PA) and tissue-type plasminogen activator (t-PA). Anti-TSV-PA antibodies inhibited both the amidolytic activity and plasminogen activation activity of TSV-PA, but they did not inhibit the plasminogen activation activity of u-PA and t-PA.

Key words.- Snake venoms, antibodies, serine proteases

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Introduction

TSV-PA, a specific plasminogen activator from the venom of *Trimeresurus stejnegeri*, is a serine protease which specifically cuts the Arg560-Val561 bond of plasminogen and forms plasmin (Zhang et al., 1995). Sequence analysis indicates that TSV-PA is a typical snake venom serine protease which show great homology with other snake venom serine proteases, such as thrombin-like enzymes (Itoh et al., 1987; Shieh et al., 1988), protein C activator (McMullen et al., 1989), and factor V activator (Tokunaga et al., 1988). Serine proteases are widely distributed in snake venoms especially in *Crotalinae* snake venoms (Stocker, 1990). TSV-PA is the first specific plasminogen activator found in snake venoms. In this investigation, we reported the immuno-chemical study of TSV-PA.

Material and Methods

Snake Venoms were from the stock of Kunming Institute of Zoology, Chinese Academy of Sciences. TSV-PA from *Trimeresurus stejnegeri* venom is purified as described previously (Zhang et al., 1995). TSV-PA was denatured by adding β -mecaptoethanol to 1% final concentration and boiled at 100 °C for 5 min. Native TSV-PA and denatured TSV-PA were emulsified with 50% Freund adjuvant and were administered

s.c. at 2 week intervals. Boosts, which were performed when the serum titers were decreasing, were achieved by injecting s.c. the same components, in the presence of incomplete Freund adjuvant. The immunization protocol was performed by the dose of administered antigen 100 μ g per rabbit.

Microtitration plates (96 wells) were coated in phosphate-buffered saline (PBS) by over night incubation of antigen (5 μ g/ml) and saturation was carried out with 3% BSA in PBS. Plates were washed with PBS containing 0.1% Tween-20. The solutions to be tested (100 μ l/well), diluted in PBS containing 3% BSA, were incubated 1 hour at 37 °C, then washed. Peroxidase-labelled goat antibodies anti-rabbit immunoglobulins (Biosys, Compiegne) were added at a 1:2000 dilutions, incubated 1 hour at 37 °C and washed. Substrate for peroxidase (O-phenylenediamine dihydrochloride in 10 mM sodium phosphate, pH 7.3, 0.01% H₂O₂) were added and the absorbance was recorded at 405 nm with a Dynatech microplate reader.

Antibodies (IgG) were purified from the sera by double ammonium sulfate precipitations (35% final concentration), and then dialyzed against PBS. Inhibition of enzymatic activity was carried out by preincubating for 30 min at 37 °C a fixed concentration of the enzyme (5 μ g/ml) with variable amounts of antibodies

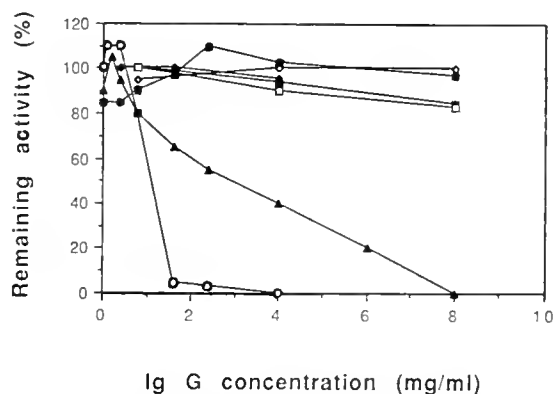


Figure 1. Inhibition of plasminogen activation by IgG against native and denatured TSV-PA. Fixed concentrations of TSV-PA, human two chains lower-molecular weight urokinase (u-PA) or human two chains tissue type plasminogen activator (t-PA) were incubated with various concentrations of antibodies against TSV-PA for 30 min in 37 °C. Then the remaining plasminogen activation activity was determined and expressed as the percentage of the original values. (○) TSV-PA test, (●) TSV-PA control, (Δ) TSV-PA test with anti-denatured TSV-PA antibodies, (◆) u-PA test, (◇) u-PA control, (■) t-PA test, (□) t-PA control.

and by testing the residual enzymatic activity. Plasminogen activation activity and amidolytic activity of the enzymes were assayed as described by Zhang et al. (1995).

Results

We immunized rabbits with purified TSV-PA, both in its native and denatured forms. Table 1 shows that the antiserum directed against native TSV-PA has a much higher ELISA titer than that directed against denatured TSV-PA, showing that native TSV-PA is much more antigenic than denatured TSV-PA. On the other hand, antiserum directed against native TSV-PA has a much lower ELISA titer when coating the plate with denatured TSV-PA, indicating that large parts of the antibodies raised against native TSV-PA are against conformational epitopes of the protein.

In the first series of experiments, we examined the immunological cross-reactions by ELISA. The anti-TSV-PA antibodies (from here, if not specially noted, anti-TSV-PA serum and antibodies means those raised against native TSV-PA) cross-reacted with TSV-PA and *Trimeresurus stejnegeri* venom. In the mean time, they cross-reacted with several other *Crotalinae* snake venoms in which we did not find the activity of plasminogen activation. From the ELISA titers, we can see, first, the immunological cross-reaction with venoms from *Crotalinae* snakes are much higher than with those from *Elapidae* snake venoms. Second,

Table 1. The titers of the antibodies against TSV-PA (a specific plasminogen activator from the venom of *Trimeresurus stejnegeri*)

Antigen	Serum	Immunoglobulins
TSV-PA	10 ⁶	200 ng/ml
denatured TSV-PA(1)	6400	2 μg/ml
denatured TSV-PA	800	25 μg/ml
TSV-PA(1)	12800	1 μg/ml
<i>Trimeresurus stejnegeri</i>	10 ⁶	200 ng/ml
<i>Trimeresurus mucrosquamatus</i>	5X10 ⁴	2 μg/ml
<i>Agkistrodon acutus</i>	5X10 ³	2 μg/ml
<i>Agkistrodon halys</i>	5X10 ⁴	2 μg/ml
<i>Ophiophagus hannah</i>	10 ²	100 μg/ml
<i>Vipera russelli</i>	5X10 ³	10 μg/ml
<i>Naja naja atra</i>	10 ²	200 μg/ml
<i>Bungarus fasciatus</i>	<10	>20 mg/ml
<i>Bungarus multicinctus</i>	<10	>20 mg/ml
batroxobin	10 ⁴	2 μg/ml
trypsin	640	31 μg/ml
thrombin	<10	>20 mg/ml
u-PA	<10	>20 mg/ml
streptokinase	<10	>20 mg/ml
t-PA	<10	>20 mg/ml
plasmin	<10	>20 mg/ml

ELISA titers were defined as the serum dilution or immunoglobulin concentration that produced half of the maximal response. The indicated values are the means of three independent experiments, standard deviations being 10%. (1) means immuno-cross reactions with anti-denatured TSV-PA antibodies.

even though we did not find plasminogen activity in other venoms, but in these venoms such as *Trimeresurus mucrosquamatus*, *Agkistrodon halys* and *Agkistrodon acutus* there are serine proteases which are very similar with TSV-PA in structure. This observation is also coincident with the sequence comparison of TSV-PA with other snake venom serine proteases like thrombin-like enzymes, protein C activator and factor V activator (Zhang et al., 1995). The sequence of TSV-PA shows 64% homology with thrombin-like enzyme from *Trimeresurus flavoviridis*.

We further analyzed the immunological cross-reactions of anti-TSV-PA antibodies with other serine proteases. They strongly cross-reacted with batroxobin, a thrombin-like enzyme from *Bothrops atrox* venom. In addition, they only slightly cross-reacted with trypsin (Table 1). Even TSV-PA shares the same biological activity (plasminogen activation) with physiological activators, u-PA and t-PA, there is no immunological cross-reactions among them. These results is in agreement with that the venom serine proteases which have been well studied like thrombin-like enzymes belong to trypsin-kallikrein subfamily (Itoh et al., 1987; 1988, McMullen et al., 1989).

	1	20	40
TSV-PA	VFGGDECNINEHRSLVVLFN----	SNG--FLCGGTLINQDWVVTA	AHC-----
batroxobin	VIGGDECDINEHPFLAFMY--	SPR--YFCGMTLINQEWVLTAA	AHC-----
trypsin I	IVGGYTCPEHSVPYQVSL-----	NSGYHFCGGSLINDQWVVSAA	AHC-----
t-PA	IKGGLFADIASHPWQAAIFAKH	RSPGERFLCGGILISSCWILSA	AHC FQERFP
u-PA	IIGGEFTTIENQPWFAAIYRR	RH-RGSVTYVCGGSLISPCWVI	SATHCFID-YP
	60	80	
TSV-PA	DSNNFQLLFGVHSHKILNEDE	QTRDPKEKFFCPNRKKDDEV--	DKDIMLIKLDS
batroxobin	NRRFMRIHLGKHAGSVANYDE	VVRYPKKFCIPNKKKNVIT--	DKDIMLIRLDR
trypsin I	YKSRIQVRLGEHNINVLEGEQ	F-INAAKIIKHPNYSSWTL--	NNDIMLIKLS
t-PA	P-HHLTVILG-RTYRVVPGEEE	QKFEVEKYIVHKEFDDDT--	YDNDIALLQLKS
u-PA	KKEDYIVYLG-RSRLNSNTQ	GEMKFEVENLILHKDYSADTL	AHHNDIALLKIRS
	100	120	140
TSV-PA	SVSNSEH----IAPLSLPSSP---	PSVGSVCRIMGWGKTIPTKEI	YDPVPHCAN
batroxobin	PVKNSEH----IAPLSLPSNP---	PSVGSVCRIMGWGAIITSEDT	YPDPVPHCAN
trypsin I	PVKLNAR----VAPVALPSAC---	APAGTQCLISGWGNTLSNGV	NNPDLQCV
t-PA	DSSRCAQESSVVRTVCLPPADL	QLPDW-TECELSGYGKHEALS	PFYSERLKEAH
u-PA	KEGRCAQPSRTIQTICLPSM-	YNDPQFGTSCFITGFGKENST	DYLYPEQLKMTV
	160	180	
TSV-PA	INILDHAVCRTA-YSWRQVANT	TLCAGILQGG-----	RDTCHFDSGGPLICNG
batroxobin	INLFNNTVCREA-Y--NGLPAK	TLCAGVLQGG-----	IDTCGGDSGGPLICNG
trypsin I	APVLSQADCEAA-YP-GEITSS	MCVGFLEGG-----	KDSCQGDSGGPVVCNG
t-PA	VRLYPSSRCTSQHLLNRTVT	DNMLCAGDTRSGGPQANLH	DACQGDSGGPLVCLN
u-PA	VKLISHRECQQPHYYGSEVTT	KMLCAADPQWKT-----	DSCQGDSGGPLVCSL
	200	220	234
TSV-PA	I----FQGIVSWGGHPCGQP	GPGEVYTKVFDYLDWIKSII	IAGNKDATCPP
batroxobin	Q----FQGILSWGSDPCAEP	KPAFYTKVFDYLPWIIQSI	IAGNKATATCP
trypsin I	Q----LQGIVSWG-YGCALPD	NPVYTKVCNFGVGIQDTIA	AN
t-PA	DGRMTLVGIIISWG-LGCGQ	KDVPVYTKVTNYLDWIRD	NMRP
u-PA	QGRMTLTGIVSWG-RGCAL	KDKPGVYTRVSHFLPWIR	SHTKEENGLAL

Figure 2. Amino acid sequence comparison of TSV-PA with batroxobin, rat trypsin, human t-PA and u-PA.

The sequence comparison was performed with a Clustal V software package in a computer. Sequences were from the following sources: TSV-PA, Zhang et al. (1995); batroxobin, Itoh et al. (1987); rat trypsin, MacDonald et al. (1982); human t-PA and u-PA protease domains, Pennica et al. (1983) and Steffens et al. (1982).

In second series of experiments, we tested the inhibitory effect of antibodies directed against TSV-PA on the enzymatic activities of TSV-PA, u-PA, t-PA and other proteases. Figure 1 shows the plasminogen activation inhibition by antibodies against both native and denatured TSV-PA. In agreement with the results above, anti-TSV-PA antibodies did not inhibit the plasminogen activation activity of u-PA and t-PA. On the contrast, both anti-native-TSV-PA and anti-denatured-TSV-PA antibodies inhibited plasminogen activation by TSV-PA. Further experiments expressed that only anti-native-TSV-PA antibodies inhibited the amidolytic activity of TSV-PA, but anti-denatured-TSV-PA antibodies did not. Combining the results of

figure 1 and the inhibition of amidolytic activity of TSV-PA, we could find that anti-native-TSV-PA antibodies inhibited both amidolytic and plasminogen activation activities of TSV-PA, on the other hand, anti-denatured-TSV-PA antibodies did not inhibit amidolytic activity of TSV-PA, but inhibited plasminogen activation by TSV-PA. These results indicate that the inhibition of plasminogen activation by anti-denatured-TSV-PA antibodies is not caused by the block of catalytic centre but by the block of the substrate binding site. Probably, this is caused by a piece of peptide in the enzyme which involves in the binding of the substrate, especially for large protein substrate binding, and which appears as a sequential

epitope. Both anti-native and anti-denatured TSV-PA antibodies did not inhibit the thrombin-like activity of *Trimeresurus stejnegeri* and *Agkistrodon acutus* venoms.

Figure 2 shows the sequence comparison among TSV-PA, batroxobin, rat trypsin, human t-PA and u-PA. TSV-PA shares 63% sequence identity with batroxobin (a thrombin-like enzyme), 42% with rat trypsin, but only 23% with human u-PA and 21% with human t-PA. The sequence comparison results are in agreement with the immuno-chemical studies above.

Discussion

Snake venoms contain numerous different proteases which act on blood cascade (Stocker, 1990). Biochemically, two main classes of these protease are recognized: serine proteases and metalloproteinases. For one group of venom serine proteases, they share a trypsin homologous catalytic domain and the molecular weights are usually around 25 kDa-35 kDa depending on the carbohydrate content of the enzyme. Even the sequence similarity among them are around 65-70%, for example thrombin-like enzymes (Itoh et al., 1987; Shieh et al., 1988), protein C activator (McMullen et al., 1989), factor V activator (Tokunaga et al., 1988), kallikrein-like enzyme (Komori et al., 1988) and newly determined plasminogen activator (Zhang et al., 1995), this group of venom serine protease is characterized by their highly divergent substrate specificity. Unlike their trypsin homologous, they are usually highly specific. The explanation of their highly homology primary sequence verse their highly divergent substrate specificity (for protein substrate) and the determination of their substrate binding site is extremely significant in our understanding of protein structure-function relationship and further the protein reconstruction for medical purpose in future.

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A New Locality for *Cuora pani* Song 1984 with Comments on its Known Range

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Abstract.- The discovery of *Cuora pani* Song 1984 in northern Sichuan Province, China lends credence to the type locality in southern Shaanxi Province. The localities reported from southern Yunnan Province (the type region of *C. "chriskarannarum"*) require verification.

Key words.- Reptilia, Chelonia, Bataguridae, *Cuora pani*, *C. chriskarannarum*, *C. yunnanensis*, *C. zhoui*, China, Sichuan, Yunnan, distribution, biogeography

Cuora pani Song, 1984 is the first of many new Chinese chelonians to be described over the past fifteen years. The original description was based on two specimens from the Qin Ling Mountains (Pingli County, southern Shaanxi Province). Later, *C. pani* was inadvertently redescribed from southern Yunnan Province as *C. chriskarannarum* by Ernst and McCord (1987) based on pet-trade specimens. De Bruin (1988) first suggested the synonymy of *C. chriskarannarum* with *C. pani*. Subsequently, other authors (e.g., Phillipen pers. comm. in Stubbs, 1989; Pritchard, 1990; Zhao, 1990; McCord and Iverson, 1991) have either independently come to the same conclusion or at least agree with it. McCord and Iverson (1991) recognize that this synonymy requires that *C. pani* is reported from two areas which are separated by nearly 1200 km (Fig. 1). Given this unlikely distribution, they raise the possibility that one of the localities (if not both) are erroneous, but withhold further speculation since the distributions of all *Cuora* are very poorly understood. In this paper we report a new locality for *C. pani* which may shed some light on this issue.

In April of 1994 one of us (DL) heard reports of a turtle with a moveable plastron from people living in northern Sichuan. Due to the scarcity of these turtles, it took five years to acquire the first specimens. Three individuals were purchased in Guangyuan City by DL and were reportedly captured in the nearby tributaries of the Jialing River (105° 40' E, 32° 30' N; Fig. 1). Two of the turtles are females; the largest is 180 mm in straight-line carapace length and weighs 625 grams. The smaller female is 110 mm in carapace length and 130g¹. The only male is a juvenile (Fig. 2), 98 mm in carapace length and weighing 95g. All three turtles match descriptions of *Cuora pani* (Song, 1984; McCord and Iverson, 1991). Diagnostic characters

include an olive head, olive-brown carapace, and, most importantly, a plastron with dark pigment associated with the seams in the form of rectangular bars.

The presence of these turtles in northern Sichuan Province lends credence to the type locality of *Cuora pani* in southern Shaanxi Province. The new locality also closes the distributional gap between the southern Shaanxi locality and the type region of *C. "chriskarannarum"* from nearly 1200 km to about 1050 km. Nevertheless, we feel that the Sichuan locality, by verifying the presence of *C. pani* in the northern tributaries of the Yangtze, casts doubt on the validity of the *C. "chriskarannarum"* localities. The latter localities become even more suspicious when the distribution of other species of *Cuora* are taken into account. For example, the type locality of *Cuora aurocapitata* (Luo and Zong, 1988) is in the eastern Yangtze drainage. *Cuora aurocapitata* is a close relative of *C. pani*, and may prove to be conspecific (McCord and Iverson, 1991). The close relation of *C. pani* and *C. aurocapitata* is reflected by their biogeographical distribution since both taxa inhabit the Yangtze River or its northern tributaries.

The type region of *C. "chriskarannarum"* is in the drainage of the Red River. Two divides separate these localities from the type region of *C. pani* (in southern Yunnan the Pearl River drainage lies between the Red River and the southwestern tributaries of the Yangtze). We can not rule out a corridor between the Red River and the Yangtze in northern Yunnan, but note that, in addition to a mountainous divide, other species of *Cuora* historically inhabited this area. *Cuora zhoui*

¹ This specimen has been accessioned into the Chengdu Institute of Biology herpetological collection as CIB-9910544

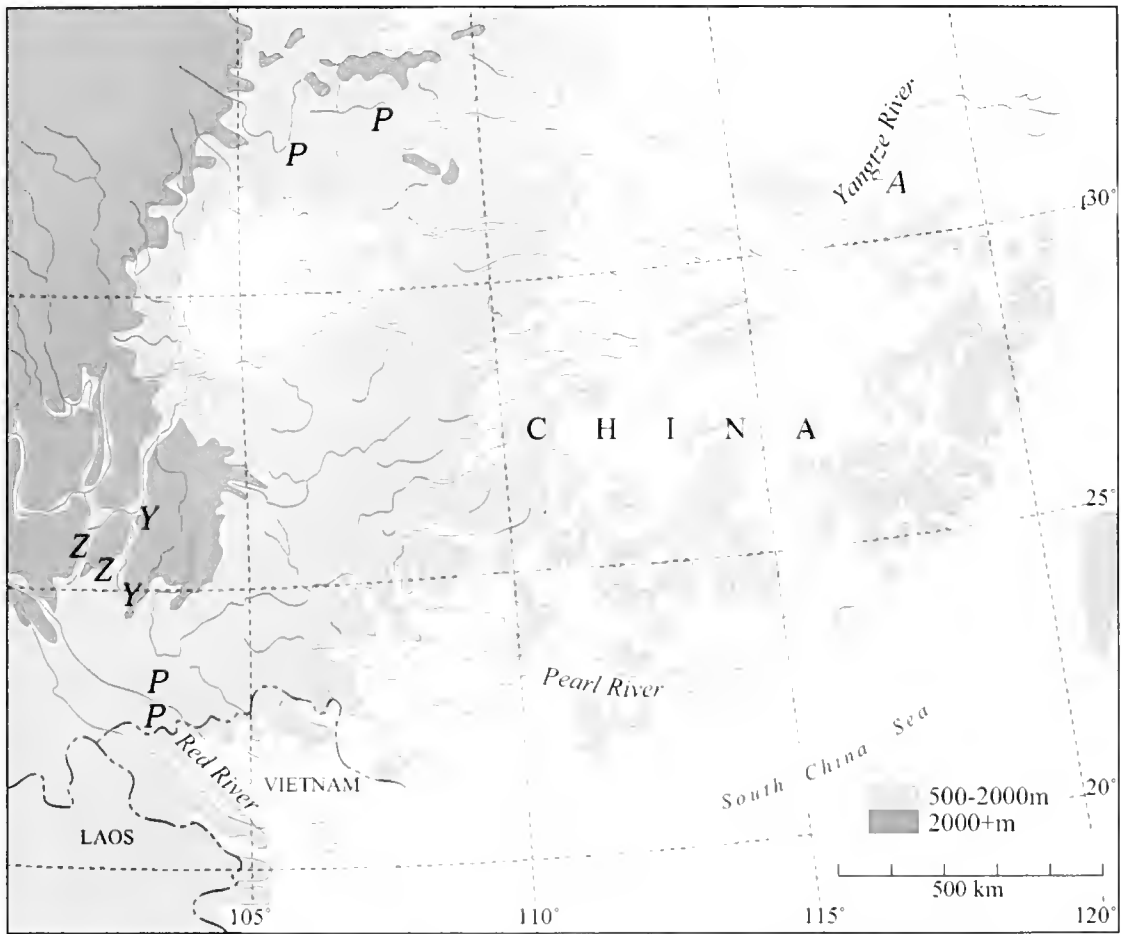


Figure 1. Map of China showing known localities of select aquatic *Cuora* species. A= *C. aurocapitata*, P= *C. pani*, Y= *C. yunnanensis*, Z= *C. zhoui*. Localities were taken from original descriptions, McCord and Iverson (1991), and Iverson (1992).

Zhao, 1990 (= *Cuora pallidicephala* McCord and Iverson, 1991) and *Cuora yunnanensis* (Boulenger, 1906) are both reported from tributaries of the Yangtze in northern Yunnan (Fig. 1). Like *C. "chriskarannarum"*, the Yunnan records for *C. zhoui* (the type series of *C. "pallidicephala"*) are derived from the pet trade (McCord and Iverson, 1991) and are from a different province than the type description (Zhao, 1990). Unfortunately, the type series of *C. zhoui* itself was purchased from a market so the exact provenience of this species remains uncertain. The localities for *C. yunnanensis*, however, are probably valid since they predate the increased turtle trade associated with recent economic reforms in China. The presence of at least one, and possibly two, congeners within the reported range of *C. pani* is strange since aquatic *Cuora* are generally allopatric.

A historic lowland connection between the southern Yunnan and northern Yangtze *Cuora pani* localities would require even greater geographical

separation. Furthermore, this hypothetical connection would pass through the known distribution of *C. trifasciata* (McCord and Iverson, 1991), a close relative of *C. pani* and *C. aurocapitata* (de Bruin, 1988; Buskirk, 1989). In short, if the Yunnan Province *C. pani* localities are real, the biogeographical history of these turtles is very complex.

It is important to note that many of the localities for Chinese turtles, including the one reported here, are based upon market-bought or else pet trade animals. Pet trade localities, such as the southern Yunnan *Cuora pani* localities, should be treated with caution. The demand for rare species by turtle fanciers greatly increases their value and, therefore, there is incentive to hide the true locality as a "trade secret". Direct cooperation with people from turtle-bearing provinces, however, can help determine the distributions of these rare turtles.

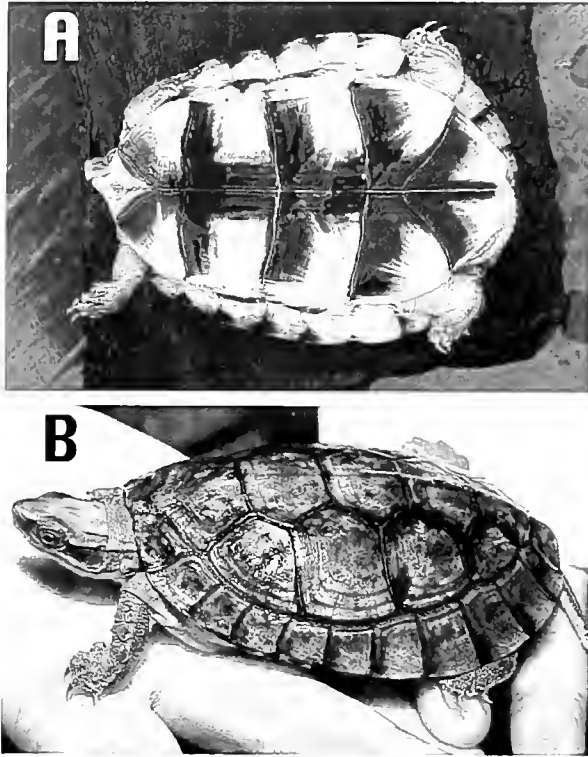


Figure 2. A) Ventral view of *Cuora pani* (male) from northern Sichuan Province (photo by Ermi Zhao). B) Lateral view of same individual (Photo by JFP).

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Guidelines for Manuscript Preparation and Submission

Summary

Manuscripts must:

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- 2) be of letter quality (laser printed or typewritten on bond paper).
- 3) include camera ready figures (if any).
- 4) include complete and accurate literature citations.
- 5) include complete and accurate localities with latitude and longitude.
- 6) include a camera ready map illustrating regions discussed (when applicable).

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12.1 <tab><tab><tab>	91020.4<tab><tab>	0.6<tab><tab><tab><tab>	0.02

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The names and addresses of all authors must be complete enough to allow postal correspondence. Please include email and World Wide Web addresses if applicable.

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Key words provide an index for the filing of articles. Key words provide the following information (when applicable): 1) Taxonomy (e.g. Reptilia, Squamata, Gekkonidae, *Gekko gecko*). 2) Geography (e.g. China, Thailand). 3) Subject (e.g. taxonomic validity, ecology, biogeography). The order of taxonomy, geography, and subject should be observed.

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Manuscripts must be in English and spelling must be correct and consistent. Use *Webster's New International Dictionary* for reference. For clarity, use active voice whenever possible. For example, the following sentences in active voice are preferable to those in passive voice.

Active voice: "Lizards were extremely common on the site." and "I examined three female snakes."

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Accurate and standard references are a crucial part of any article. This is especially important when dealing with publications from many different countries. The reader must be able to precisely identify any literature cited. References in the text must be checked for consistency with references in the literature cited section. All references cited in the text must be in the literature cited section. The literature cited section may not contain any references not mentioned in the text. Articles containing inaccurate or inconsistent literature citations will be returned for correction.

References in Text. 1) References to articles by one or two authors must include both surnames in the order they appear in the original publication. References to articles by more than two authors must include the first author's surname, followed by "et al." 2) The year of article follows the authors, separated only by a space. 3) References with the same author and year are distinguished by the lower case characters "a, b, c, . . ." 4) References cited in text are listed in alphabetical order by first author.

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Journal article

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Gatten, R. E. Jr. 1974. Effect of nutritional state on the preferred body temperatures of turtles. *Copeia* 1974(4):912-917.

Journal article, title translated, article not in English

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Book

Pratt, A. E. 1892. To the snows of Tibet through China. Longmans, Green, and Co., London. 268 pp.

Article in book

Huey, R. B. 1982. Temperature, physiology, and the ecology of reptiles. Pp. 25-91. In C. Gans and F. H. Pough (eds.), *Biology of the Reptilia*, Vol. 12, *Physiological Ecology*. Academic Press, New York.

Government publication

United States Environmental Data Service. 1968. Climatic Atlas of the United States. Environmental Data Service, Washington, D. C.

Abstract of oral presentation

Arnold, S. J. 1982. Are scale counts used in snake systematics heritable? SSAR/HL Annual Meeting. Raleigh, North Carolina. [Abstr].

Thesis or dissertation

Moody, S. 1980. Phylogenetic and historical biogeographical relationships of the genera in the Agamidae (Reptilia: Lacertilia). Ph.D. Thesis. University of Michigan. 373 pp.

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