Karvotype Information on some Toad Agamas of the *Phrynocephalus* guttatus Species Group (Sauria, Agamidae) of the former USSR.

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Abstract. -Karyotypes of several toad agamas of the Phrynocephalus guitatus species group (sensu lato) were investigated in specimens from a variety of localities of the former USSR. Differences in the diakinetic stage of meiosis have been observed, permitting distinctions among three groups of species. The forms guttatus, moltschanovi, kushackewitschii, and alpherakii comprise Group I; P. guttatus salenskyi represents the second group; and P. versicolor hispida represents Group III.

Key words: Reptilia, Sauria, Agamidae, Phrynocephalus guttatus, Kazakhstan, Middle Asia, Precaucasus, karyology.

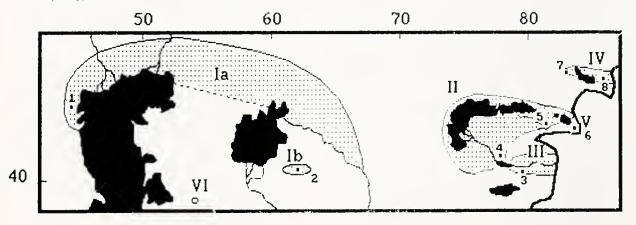


FIG. I. Scheme of distribution of forms of P. guttatus species group of the former USSR fauna: Ia- P. g. guttatus; Ib- P. g. moltschanovi; II- P. g. kushackewitschii; III- P. g. alpherakii; IV- P. g. salenskyi; V-P. versicolor hispida; VI-P. guttatus spp. (the numbering of populations is in accordance with the data in table I).

Introduction

The first and only extensive karyological investigation of the agamid lizard genus Phrynocephalus Kaup is the work of Sokolovsky (Sokolovsky, 1974; 1977). Karyotype characteristics permitted the recognition of five groups. The "guttatus" group included two species, P. guttatus (Gmel.) and P. versicolor Str. These species have a diploid number of 46, all chromosomes are telocentric. karyotypes could be divided into 12 pairs of macrochromosomes and 11 pairs of microchromosomes. Approximately 50% of the metaphase plates in P. guttatus contained satellite chromosomes on the first pair of chromosomes, but these were never observed in the *P. versicolor* karyotype.

The specimens examined came from Daghestan (P. guttatus) and Central Gobi, Mongolia (P. versicolor) and were believed to represent the nominative forms of both species.

The systematics of the *P. guttatus* group based on external morphological characteristics is extremely complicated and remains unclear. At various times the forms alpherakii Bedr., moltschanovi Nik.. kushackewitschii Bedr., salenskyi Bedr., etc. have either been included in P. guttatus, sensu stricto or treated as related species. The forms bogdanowi Bedr., hispida Bedr., and paraskiwi Semenov et al. have been assigned to P. versicolor. (Bedriaga, 1909; Nikolsky, 1915; Terentjev and Chernov, 1949; Peters,



FIG. 2 The karyogramme of Phrynocephalus guttatus salenskyi.

1984; Semenov et al. 1987). Golubev (1989) suggested that *P. guttatus* and *P. versicolor* from Kazakhstan are eonspecific. Karyotype details of the forms listed above have never before been examined. The purpose of this study is to determine whether karyotype information will aid in our understanding of the systematics and evolution of *Phrynocephalus*.

Methods

Between 1989-1991 we collected specimens of nearly all listed forms of both species of *Phrynocephalus* inhabiting the territory of the former USSR with the exception of P. v. bogdanowi from the extreme south of Tuva (Central Asia) and P. guttatus ssp. from Turkmenistan (Fig. 1 and Table 1). Chromosome samples were prepared from cellular suspension of bone marrow, blood, and testis. We used a smear method and a method known as "digging out" in conformity with procedures described by Ford and Hamerton (1956) and McGregor and Varley (1986) as partially modified by Manilo (1986). Chromosomal staining was

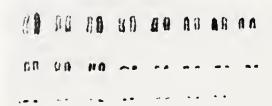


FIG 3. Diakinetic stage of meiosis of five forms of P. guttatus species group. s. Iato: I— all elements are ring- or stick-shaped; (guttatus, moltschanovi, kushackewitschii, alpherakii); II— one or two elements are cross-shaped (salenskyi); III— two and more (up to four) elements are cross-shaped (hispida).

performed by Giemsa stain (2% solution) in 0.01 M sodium-phosphate buffer (pH 6.8) for 20-30 minutes. After washing in distilled water, the preparations were passed through alcohols and xylols (orthoxylol) and subsequently embedded in Canadian Balsam. In excess of 30 metaphase plates from each form were investigated using a Biolam 1-212 microscope.

Metaphase plates of spermatogonial division, spermatocyte I (diakinesis) and spermatocyte II (metaphase II) bivalents were investigated in testis preparations. Chromosome morphology is described according to the classification proposed by Levan et al. (1964).

Results and Discussion

Our data support the findings of Sokolovsky (1974, 1977). The diploid number is uniformly 46 and the Fundamental Number (NF) is 46. In several forms (guttatus, moltschanovi, kushackewitschii) we noted satellite ehromosomes on several plates; whereas in other forms (alpherakii, salenskyi) we saw no evidence of satellites. The revelation of this structure largely depends on the degree of spiralization. It is possible that satellites will be found in the latter forms with further investigation and more extensive material.

TABLE 1. Localities, sample sizes, and taxa of *Phrynocephalus guttatus* s. lato populations collected and investigated in this study (numbering of populations is given in accordance with the data shown in Fig. 1.

No	Taxa	Locality	Sample size, sex
		N. Transcaucasus: N. Daghestan:	
1 _	P. g. guttatus	sands on right bank of Kuma River	2 male
		N. Kysylkum in Karakalpakia:	
2	P. g. moltschanovi	Kostruba Well	2 male
		E. Kazakhstan:	
3	P. g. alpherakii	Alma Ata District; Near Karakuldek	3 male
		E. Kazakhstan: Taldy-Kurghan District:	
4	P. g. kushackewitschii	NW bank of Kapchagay Reservior	3 male; 2 female
5	P. g. kushackewitschii	Near Andreevka (left bank of Chyndjaly River)	9 male
6	P. versicolor hispida	E. Kazakhstan: Djungar Gate	4 male
		E. Kazakhstan: Zaissan Depression:	
7	P. g. salenskyi	Left bank of Bukhtarma Reservior: Kkuludjunsky Sands	3 male
8	P. g. salenskyi	Left bank of Black Irtysh near Karatal	3 male

Sokolovsky described all chromosomes as telocentric. We cannot confirm this with confidence. Second arms are clearly visible on metaphasic plates with premetaphasic (elongated) chromosomes on several pairs of large elements. Such chromosomes could be acro- or even subtelocentric. The karyotype of *salenskyi* is an example (Fig. 2). This characteristic is not peculiar to any one form or group of forms of the *guttatus* group and cannot be used to distinguish a subordinate group.

We also observed distinct peculiarities of chromosome morphology in meiosis in the diakinetic stage. The chromosome bivalents of the various taxa differ in the number of ring-shaped and cross-shaped bivalents. Based on this difference in pairing, it is possible that the taxa of the toad agamas of the "guttatus" group might be grouped in the following way:

I: guttatus*, moltschanovi, kushackewitschii, alpherakii all diakinetic bivalents are ring or stick-shaped.

II: salenskyi one or two of the elements are cross-shaped (Fig. 3), the remainder as in Group I.

III: hispida from two to four cross-shaped elements (Fig. 3), the remainder as in Group I.

This grouping by cross-shaped elements in diakinesis is a continuum. In this system, P. v. hispida is closer to P. g. salenskyi from the Zaissan Depression [sometimes attributed to P. versicolor (Paraskiv, 1953; Bannikov et al., 1977)] than to other forms of P. guttatus sensu stricto. However, it may be important that the toad agama from the Zaissan Depression occupies an intermediate position between Groups I and III. It is interesting to note the absence of chromosomal differences in the I-st. group, while its members, as mentioned above, are attributed by a number of authors to different species.

The data may be interpreted to suggest uniformity in the species of the *guttatus* group from Kazakhstan, Middle Asia, and the Precaucasus (Golubev, 1989) as well as a close relationship between *salenskyi* from Zaissan Depression and *hispida* from Djungar Gate and northern Djungaria (Golubev, 1992).

* On several testis preparations of the nominative form we observed a picture similar to that of the preparations in Group II.

Literature Cited

BANNIKOV, A. G., I. S. DAREVSKY, V. G. ISCHENKO, A. K. RUSTAMOV AND N. N.

- SHCHERBAK. 1977. [Field guide of the USSR amphibians and reptiles]. Prosveschenje Publishing House, Moscow. 369 pp. (in Russian).
- BEDRIAGA, YA. 1909. Amphibien und Reptilien.
 Pp. 73-502. *In:* Wissenschaftliche Resultate der Reisen N. M. Przewalskijs durch Zentralasien. Zoologische Teil. Band 3. Part 1. Lacertilia. Sankt-Petersbourgh. (In Russian/German).
- FORD, C. E., AND J. L. HAMERTON. 1956. A colchicine hipotonic citrate squash sequence for mammal's chromosomes. Staining Technology 31:247-251.
- GOLUBEV, M. L. 1989. [Phrynocephalus guttatus (Gmel.) or P. versicolor Str. (Reptilia, Agamidae): which Phrynocephalus species occurs in Kazakhstan?]. Zoological News, Kiev (5):38-46. (In Russian).
- GOLUBEV, M. L. 1992. [Variegated toad agama *Phrynocephalus versicolor* (Reptilia: Agamidae) of the Djungar Gate (Eastern Kazakhstan) with notes on systematics of the species]. Zoological News, Kiev no. 2:31-38. (In Russian).
- LEVAN, A., K. FREDGA, AND A. A. SANDBERG. 1964. Nomenclature for centromeric position on chromosomes. Hereditas 52:201-220.
- MACGREGOR, H. C., AND J. M. VARLEY. 1986. [Working with animal chromosomes]. Mir Publishing House, Moscow. 272 pp. (In Russian).
- MANILO, V. V. 1986. [Karyotypes of gecko genera *Alsophylax* and *Crossobamon*]. Zoological News, Kiev (5):46-54. (In Russian).

- NIKOLSKY, A. M. 1915. [Fauna of Russia and adjacent countries. Reptiles. Vol. I. Chelonia and Sauria]. Imperial Academy of Sciences, Petrograd. 532 pp. (In Russian).
- PARASKIV, K. P. 1956. [Reptiles of Kazakhstan]. Kazakh Academy of Sciences Press, Alma Ata. 228 pp. (In Russian).
- PETERS, G. 1984. Die krotenkopfagamen Zentralasiens (Agamidae: *Phrynocephalus*). Mitteilungen aus dem Zoologischen Museum in Berlin. Akademie Verlag, Berlin 60(1):23-67.
- SEMENOV, D. V., Z. K. BRUSHKO, R. A. KUBYKIN, AND G. I. SHENBROT. 1987. [Taxonomic position and protective status of the round-headed lizard (Reptilia, Agamidae) in the territory of the USSR]. Zoological Journal, Moscow 68(12):79-87. (In Russian).
- SOKOLOVSKY, V. V. 1974. [A comparative karyological study of lizards of the family Agamidae. 1. Chromosome complements of 8 species of the genus *Phrynocephalus* (Reptilia, Agamidae)]. Cytology, Moscow 16(7):920-925. (In Russian).
- SOLOLOVSKY, V. V. 1977. [Systematic relations in the family Agamidae based on karyological data]. P. 195 *In:* Questions of herpetology abstracts of the report at the Fourth All Union Conference of Herpetology, Nauka, Leningrad. (In Russian).
- TERENTYEV, P. V. AND S. A. CHERNOV. 1949. [Guide to reptiles and amphibians]. Soviet Sciences Publishing, Moscow. 315 pp. (In Russian).