KARYOTYPIC VARIATION IN THE GENUS *PLATEMYS* (TESTUDINES: PLEURODIRA)

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Made in United States of America
Reprinted from COPEIA
1985, No. 2

Copeia, 1985(2), pp. 445–449

Karyotypic Variation in the Genus *Platemys* (Testudines: Pleurodira)

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Standard karyotypes are reported for all currently recognized members of the South American genus *Platemys*. *Platemys platycephala* (2n = 64) has a karyotype distinct from the other four members of the genus (*P. macrocephala*, 2n = 48; *P. pallidipectoris*, *P. radiolata*, *P. spixii* 2n = 50). The range in diploid numbers within this genus is greater than all other members of the suborder Pleurodira. Such karyotypic variability is particularly uncommon among closely related turtles and supports generic separation of *P. platycephala* from the remaining four species. These four have karyotypes similar to Australian chelids, and, with *Chelus*, may represent a group intermediate between Australian and the other South American forms.

THE two families of living pleurodiran turtles are restricted to the southern hemisphere. The Pelomedusidae is comprised of three generic groups occurring in Africa, Madagascar and South America (Bull and Legler, 1980). The Chelidae is represented in South America by about 15 species and in Australia and New Guinea by about 18 species (Legler and Cann, 1980).

The karyology of the Pelomedusidae has been studied by Ayres et al. (1969), Killebrew (1975) and Rhodin et al. (1978), and the Chelidae by Barros et al. (1976), and Killebrew (1976). Bull and Legler (1980) provide an extensive review of pleurodiran karyology including new data on chelid and pelomedusid turtles. They divided chelid karyotypes into two groups on the basis of shared characters. One group with low diploid numbers (2n = 50–54) and few or no acrocentric chromsomes includes the Australian and New Guinea genera and possibly *Chelus* from South America. The second group has higher diploid numbers (2n = 58–64) and many or all acrocentric chromosomes and includes all the South American genera except *Chelus* (Table 1).

Standard karyotypic data are available for most currently recognized species of the Australian chelid genera *Chelodina*, *Elseya*, and *Emydura*. Karyotypes for most South American genera are known for only one or two species of most genera. We present here the standard karyotypes for all five species of the South American genus *Platemys*. Karyotypes of *Platemys platycephala* have been studied by other workers (Gorman, 1973; Barros et al., 1976; Bull and Legler, 1980), whereas those of *P. pallidipectoris*, *P. radiolata*, *P. spixii* and *P. macrocephala* previously have not been reported.

MATERIALS AND METHODS

Karyotypes were prepared from spleens of freshly killed animals, or from primary fibroblast tissue cultures established from heart muscle biopsies. Heart muscle tissue cultures were maintained at 32 C in Medium 199 fortified with 20% fetal calf serum. Methods of chromosome preparations from tissue cultures follow Sites et al. (1979b) with modifications by Baker et al. (1982). Actively dividing cell cultures were harvested by a mild trypsin treatment and placed in 0.075 M KCl hypotonic solution. Velban (0.05 ml of 0.005% solution) was added to the cell-hypotonic suspension as a mitotic inhibitor and the solution was incubated at 32 C for 40 min. The cells were then fixed (1 part acetic acid: 3 parts methanol), dropped onto clean slides, and air dried.

Chromosome preparations from spleen tissue were made as described by Bickham (1975) and modified by Baker et al. (1982). Phytohemagglutinin, a mitogen (0.1 ml/10 g body weight), was injected abdominally into each turtle 48 hr prior to sacrifice. Two hr prior to sacrifice 0.005% velban (0.1 ml/10 g body weight) was injected abdominally. Upon sacrifice, spleens were removed and minced with mortar and pestle in hypotonic solution (0.075 M KCl). All slides were stained for 5 min in 2% Geimsa in 0.01 M phosphate buffer, rinsed in distilled water, air dried and examined.

TABLE 1. DIPLOID (2n) AND FUNDAMENTAL NUMBERS (FN) OF CHELID TURTLES.

Species and locality	2n	FN	Reference
South America			
Chelus fimbriatus	50	66	Bull and Legler, 1980; Barros et al., 1976
Hydromedusa tectifera	58	62	Bull and Legler, 1980
Phrynops gibbus	50	—	Killebrew, 1976
	60	—	Barros et al., 1976
	58	64	This study
Phrynops dahli	58	66	Bull and Legler, 1980
Phrynops nasutus	58	64	Bull and Legler, 1980; Gorman, 1973
	50	—	Killebrew, 1976
Phrynops geoffroanus	58	64	Bull and Legler, 1980
Phrynops rufipes	58	64	This study
Platemys platycephala	96*	96	Bull and Legler, 1980
	68	—	Gorman, 1973
	64	64	Barros et al., 1976; This study
Platemys pallidipectoris	50	62	This study
Platemys radiolata	50	63	This study
Platemys spixii	50	62	This study
Platemys macrocephala	48	60	This study
Australia			
Chelodina expansa	54	72	Bull and Legler, 1980
Chelodina longicollis	54	72	Bull and Legler, 1980
Chelodina oblonga	54	72	Bull and Legler, 1980
Chelodina rugosa	54	72	Bull and Legler, 1980
Chelodina steindachneri	54	76	Bull and Legler, 1980
Elseya dentata	50	72	Bull and Legler, 1980
Elseya latisternum	50	72	Bull and Legler, 1980
Elseya sp.	50	72	Bull and Legler, 1980
Emydura australis	50	72	Bull and Legler, 1980
Emydura krefftii	50	72	Bull and Legler, 1980
Emydura macquarii	50	72	Bull and Legler, 1980; Killebrew, 1976
Emydura signata	50	72	Bull and Legler, 1980
Emydura sp.	50	72	Bull and Legler, 1980
Rheodytes leukops	50	72	Bull and Legler, 1980
Pseudemydura umbrina	50	68	Bull and Legler, 1980

* Presumably 3n.

Specimens examined.—*Platemys radiolata* ♂ Texas Cooperative Wildlife Collection (TCWC) 60727; *P. spixii* ♀ TCWC 60668; *P. pallidipectoris* ♀ TCWC 60667; *P. macrocephala* ♀ TCWC 60669; *P. platycephala* ♀ TCWC 61434.

RESULTS

Karyotypes are arranged by grouping chromosomes on the basis of size and centromere position following Bickham (1975). Metacentric and submetacentric macrochromosomes comprise group A, subtelocentric and acrocentric chromosomes comprise group B and microchromosomes in which centromere position is difficult to determine comprise group C. Group D is the presumed sex chromosome pair in *P. radiolata*. The number of chromosome pairs in groups A:B:C:D are given after the diploid number.

Platemys pallidipectoris 2n = 50, 5:5:15:0 (Fig. 1a). There are five pairs of group A chromosomes ranging from large to small, the largest is submetacentric, the remaining four pairs are metacentric. The third pair of chromosomes has secondary constrictions near the centromere. Group B is comprised of five pairs of medium to small acrocentric chromosomes, and group C contains 15 pairs of microchromosomes.

Platemys radiolata 2n = 50, 5:4:15:1 (Fig. 1b). The five pairs of group A chromosomes mostly are similar to those of *P. pallidipectoris*. However, the third pair in *P. radiolata* is much larger than its counterpart in *P. pallidipectoris*, and the secondary constrictions in pair three are not apparent in *P. radiolata*. Group B has only four pairs of medium to small acrocentric chromosomes. There are 15 pairs of microchromosomes. A fourth group, D, is established for the heteromorphic pair of one medium metacentric and one medium acrocentric chromosome.

Platemys spixii 2n = 50, 5:5:15:0 (Fig. 1c). The karyotype is essentially the same as *P. pallidipectoris*. Secondary constrictions are visible near the centromere of the third pair of group A chromosomes and near the centromere of one pair of group C chromosomes. In size, the third pair in group A is like the third pair of *P. pallidipectoris*.

Platemys macrocephala 2n = 48, 5:5:14:0 (Fig. 1d). There is one fewer pair of group C chromosomes than in *P. pallidipectoris*, *P. radiolata* and *P. spixii*. Otherwise, *P. macrocephala* has a karyotype apparently identical to *P. pallidipectoris* and *P. spixii*.

Platemys platycephala 2n = 64, 0:14:18:0 (Fig. 2). The karyotype of this species was first reported by Barros et al. (1976). The entire set

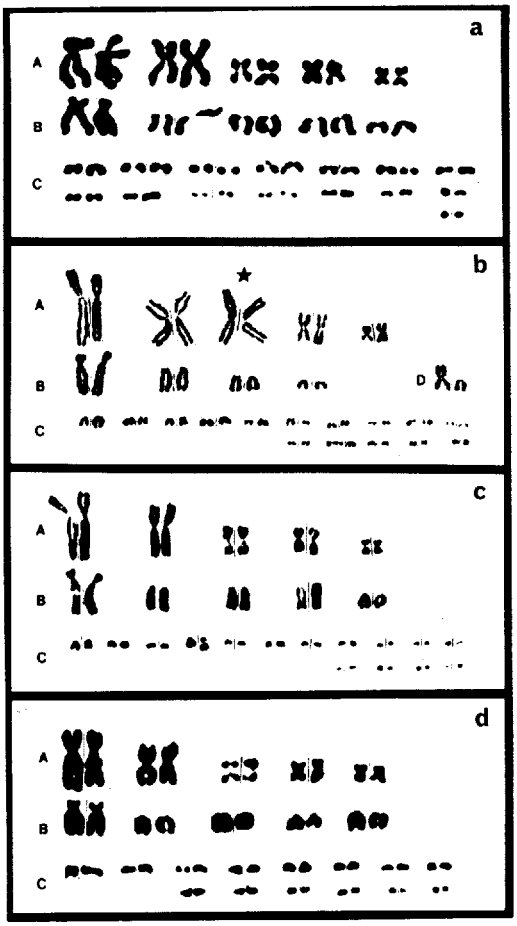

Fig. 1. Karyotypes of four species of *Platemys*; a) *Platemys pallidipectoris*, 2n = 50; b) *Platemys radiolata*, 2n = 50, star indicates enlarged group A chromosomes; c) *Platemys spixii*, 2n = 50; d) *Platemys macrocephala*, 2n = 48.

of chromosomes is acrocentric. There are 14 pairs of large to small macrochromosomes and 18 pairs of microchromosomes. The assignment of certain chromosomes to groups B or C is arbitrary because they grade without break in size from large to very small.

Discussion

The only member of the genus *Platemys* for which karyotypic data were available prior to this report was *P. platycephala*. Three different chromosome numers have been reported for this species: 68 (Gorman, 1973), 64 (Barros et al., 1976) and 96 (Bull and Legler, 1980). Bull and Legler (1980) hypothesized that the diploid

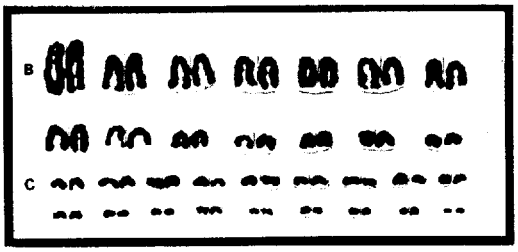

Fig. 2. Standard karyotype of *Platemys platycephala*, 2n = 64.

number of *P. platycephala* is, indeed, 2n = 64 and that the individuals they examined were triploids. We agree with their proposal, finding 2n = 64 as originally reported by Barros et al. (1976).

P. platycephala has one of the most unusual chromosomal complements known in Testudines. It has the highest known diploid number in pleurodiran turtles and it is the only turtle species with an entirely acrocentric chromosomal complement. Only the cryptodiran trionychoid turtles have higher diploid numbers (Bickham et al., 1983). The enigmatic karyotype of *P. platycephala* cannot be related to the karyotypes of its congeners or any other chelids without invoking at least six Robertsonian fission/fusion events, a feat uncharacteristic of the otherwise karyotypically conservative turtles (Bickham and Carr, 1983).

The other four species of *Platemys* (*P. pallidipectoris, P. macrocephala, P. radiolata, P. spixii*) are more similar to one another and to the other chelid genera in karyotypic patterns. The four *Platemys* are intermediate in general chromosome morphology to the two karyotypic groups proposed by Bull and Legler (1980): the low diploid numbers (2n = 48, 50) resemble the Australian chelids and the South American genus *Chelus*, whereas the number of biarmed chromosomes is intermediate between those of the Australian and South American chelids. These four species of *Platemys* and *Chelus* may represent a phylogenetic group intermediate between Australian and other South American chelids.

P. pallidipectoris, P. spixii (2n = 50) and *P. macrocephala* (2n = 48) are indistinguishable in standard karyology except for one fewer pair of microchromosomes in *P. macrocephala*. The absence of the pair of microchromosomes could be due to a fusion event or to the loss of a heterochromatic chromosome. *Platemys radiolata* differs from *P. pallidipectoris, P. macrocephala*

and *P. spixii* by having the second and third pairs of group A chromosomes equal in size, whereas in the other three the third pair is approximately half the size of the second pair. Heterochromatic additions or deletions could be responsible for this difference.

P. radiolata also has a pair of heteromorphic chromosomes not seen in any of the other species. The group D chromosomes have one medium-sized acrocentric chromosome and a metacentric chromosome about twice as large as the acrocentric one. The dimorphic group D pair may represent an XY, male heterogametic, sex chromosome pair. However, verification of this awaits the study of additional individuals of both sexes. Because *P. radiolata* is the only male we examined, sex chromosomes cannot be ruled out in *P. pallidipectoris, P. macrocephala* and *P. spixii. P. platycephala* does not have sex chromosomes (Barros et al., 1976; Bull and Legler, 1980). Sex chromosome heteromorphism has not been reported in any other member of Pleurodira and is known in Cryptodira only from *Staurotypus* (Bull et al., 1974; Sites et al., 1979a) and *Siebenrockiella* (Carr and Bickham, 1981).

P. spixii has been considered to be a subspecies of *P. radiolata* (Pritchard, 1979) and *P. macrocephala* was previously confused with *P. radiolata* (Rhodin et al., 1984) yet both show greater chromosomal similarity with *P. pallidipectoris* than with *P. radiolata.* The difference in diploid number of *P. macrocephala* and the distinct chromosomal morphology of *P. spixii* support specific status for both taxa.

Platemys, as currently recognized, shows a greater range of chromosomal variation than occurs in all the other members of this family or any other family of turtles. The karyotypic patterns elucidated in this report tentatively indicate removal of all species but *P. platycephala* from the genus *Platemys.* This separation is supported by morphological studies currently being conducted by Rhodin and Mittermeier which demonstrate the presence of several unique derived skeletal features in *P. platycephala.* The remaining members of the genus (*P. spixii, P. radiolata, P. pallidipectoris* and *P. macrocephala*) share a series of more primitive skeletal features and may represent a monophyletic unit closely related to the genus *Phrynops.*

Acknowledgments

We thank Roberto da Rocha e Silva, J. Robert McMorris, Klaus Lehmann and John M. Legler for aid in obtaining specimens, S. A. Smith for critically reading earlier manuscripts and C. M. Norton for assistance in preparation of drafts. AGJR was partly funded by the American Philosophical Society, RAM by World Wildlife Fund-US, KM by NIH-BRSG-55291 and JWB by NSF-DEB-7921519.

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Accepted 12 July 1984.