

Um die Wirksamkeit der Attrappen genauer festzustellen und die Versuchssituation zu verschärfen, wurden Einzeltieren von *T. nilotica* in Simultanwahlversuchen 2 verschiedenen grosse Attrappen geboten. Das Tier wurde dabei 10 min in einem Glaszylinder von 5 cm \varnothing belassen, während links und rechts vom Zylinder in einem Abstand von etwa 12 cm je eine Attrappe bewegt wurde (wie oben). Dann wurde der Glaszylinder hochgezogen, so dass sich das Tier jetzt frei schwimmend einer der beiden Attrappen zuwenden konnte.

Verwendet wurden die Attrappenkombinationen 8,0 + 6,0, 2,5 + 3,0, 6,0 + 5,0, 3,0 + 3,5, 5,0 + 4,0, 3,5 + 4,0, 5,0 + 3,0 cm \varnothing mit 45 Tieren in insgesamt 45 Einzelversuchen.

Die Reaktion in den ersten 10 min verlief in den meisten Fällen wie folgt: Das Tier schwamm in der Regel zuerst die grössere der beiden Attrappen an, pendelte dann kurze Zeit zwischen ihnen hin und her und blieb schliesslich bei der Grösse, die der Attrappe mit 4,0 cm \varnothing am nächsten lag (5,0 cm \varnothing stärker als 3,0 cm \varnothing !). Auf diese Attrappe entfielen auch die meisten Kontakte.

Wie schon PETERS³ feststellte, reagiert *H. multicolor* als Einzeltier sehr schlecht auf Attrappen, so dass Simultanwahlversuche in dieser Form hier nicht möglich waren. In einem Simultanwahlversuch mit einer Gruppe von 10 Tieren an der Attrappenkombination 1,0 + 1,5 cm \varnothing wurde eindeutig die Attrappe mit 1,0 cm \varnothing bevorzugt.

Die Grösse geschlechtsreifer *T. nilotica* variiert sehr stark. Die Totallänge von Tieren des Instituts lag zwischen 10,1 und 32,5 cm (Weibchen!), die Kopfhöhe (in der Mitte über den Augen) zwischen 2,0 und 7,2 cm, die Kopfbreite (unmittelbar unter den Augen) zwischen 1,2 und 4,8 cm. Bei *H. multicolor* variierte die Totallänge zwischen 4,05 und 6,7 cm, die Kopfhöhe zwischen 0,85 und 1,5 cm und die Kopfbreite zwischen 0,5 und 0,95 cm.

Unter natürlichen Bedingungen dürfte *T. nilotica* kaum jemals bei so geringer Grösse zur Fortpflanzung gelangen

wie im Aquarium. Auf Grund verstreuter Angaben in der Literatur ist ferner damit zu rechnen, dass das mitgeteilte Maximum in der Natur gelegentlich erheblich überschritten wird. Bezüglich *H. multicolor* sei noch vermerkt, dass das grösste Exemplar in der Sammlung des British Museum (Natural History) eine Länge von 8,5 cm aufweist⁵.

Zusammenfassend kann gefolgert werden, dass die Reaktionsbereitschaft der Jungfische von *T. nilotica* und *H. multicolor* an die artspezifische Grösse der Elternfische, und vielleicht auch an deren unterschiedliche Grössenvarianz, angepasst ist⁶.

Summary. Female mouth-brooding Cichlid fishes pick up the young fry again when disturbed. The young, for their part, collect about the head of the mother fish and, contacting it, try to enter the mouth cavity. This 'contact behaviour' is based upon an inborn readiness to react on the visual stimulus offered by the mother. Mother fishes in *Tilapia nilotica* are much larger than those of *Hemihaplochromis multicolor*. By comparative experimental studies with models of various size (black balls of different diameters) it is shown that the young of the 2 species strictly prefer different sizes of the model, corresponding with the different sizes of the parent fishes.

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⁵ Die Messungen wurden freundlicherweise von Herrn Dr. W. HEINRICH ausgeführt.

⁶ Im Rahmen eines von der Deutschen Forschungsgemeinschaft unterstützten Forschungsprogramms (H. M. PETERS).

Hypotheses on the Phylogeny of the Salientia, Based on Karyological Data

None of the more modern theories on the phylogeny of the Salientia¹⁻³ takes account of the karyological data collected about this order of Amphibia. A brief report on the results of several years of karyological observations on primitive Anura are given and certain hypotheses on the phylogeny of the families treated, which are prompted or supported by the present study.

The families of Anura considered to be the most primitive are: Ascaphidae, Discoglossidae, Rhinophrynidae, Pelobatidae and Microhylidae; species of all these families have been studied, with the exception of the Rhinophrynidae.

The Ascaphidae, with the 2 genera *Ascaphus* and *Leiopelma*, are generally considered the most primitive of the Anura. 2 females of the rare *Leiopelma hochstetteri* have been studied: one specimen has $2n = 34$ (Figure 1); the other has $2n = 23$. The latter specimen lacks 11 of the 12 smaller acrocentric chromosomes (microchromosomes) present in the karyotype of the former; in a separate work⁴, this difference is discussed. In both specimens, numerous chromosomes of the set are acrocentric (24 in the former and 13 in the latter).

Ascaphus truei has about 44 chromosomes in the diploid set; as many as 32 of these are acrocentric⁵. The

fact that the 2 genera of Ascaphidae possess numerous microchromosomes and acrocentric chromosomes – the latter normally being considered a characteristic of karyological primitiveness⁶ – constitutes a further confirmation of the data already noted regarding the primitive nature of the family.

As regards the Discoglossidae, species have been studied of 3 of the 4 living genera: *Alytes obstetricans* has $2n = 38$; *Discoglossus pictus* has $2n = 28$; *Bombina orientalis* and *B. variegata* have $2n = 24$. In *Alytes*, microchromosomes are present; in *Alytes* and *Discoglossus*, pairs of acrocentric chromosomes are present⁷.

On a first analysis, the karyotypes of the Ascaphidae and the Discoglossidae appear to be morphologically

¹ I. GRIFFITHS, Biol. Rev. 38, 241 (1963).

² M. K. HECHT, Syst. Zool. 12, 20 (1963).

³ R. F. INGER, Evolution 21, 369 (1967).

⁴ A. MORESCALCHI, Caryologia, in press.

⁵ A. MORESCALCHI, Atti Soc. pelorit. Sc. fis. mat. nat. 13, 23 (1967).

⁶ R. MATTHEY, Les chromosomes des Vertébrés (F. Rouge, Lausanne 1949).

⁷ A. MORESCALCHI, Riv. Biol. 59, 3 (1966).

inter-related (*Alytes* has a karyotype of a clearly 'ascaphid' morphology). It seems possible to assume the existence of mechanisms of chromosome evolution through centric fusions ('Robertson's law') (cf. ⁶) between Ascaphidae and Discoglossidae; of the 2 families, the Ascaphidae appear more primitive, karyologically. Furthermore, these 2 families differ from the remaining Anura (with the possible exception of the American Pipidae) since, in the male germ line, they show a long and well-defined diplotene stage and a short diakinesis; in these stages, the bivalents normally show various interstitial chiasmata (the large ones) or a single interstitial chiasma (the small ones) (Figure 2); the terminalization of the chiasmata in metaphase is rarely total, as in the higher Anura. The other Anura, on the other hand, have, in the male line, a short and ill-defined diplotene stage and a long diakinesis; the large and small bivalents show the precocious presence of 2 chiasmata only, generally terminal⁸, which give them a typical ring-form (Figure 3): among the

species studied, the only exceptions are *Pipa parva* and also – but only partially and in few meiotic figures – certain Hyperolidae and African Ranidae (data not published).

Also among the Pipidae species have been studied of 3 of the 4 living genera⁹. *Xenopus laevis* has $2n = 36$; *Hymenochirus boettgeri* has $2n = 24$; both the species have meta- and submetacentric chromosomes and, in the male line, show a meiosis typical of most of the Anura. *P. parva* has $2n = 30$ and – a unique feature in the Anura – the chromosomes of this species are all acrocentric; in the male line, *P. parva* has bivalents provided with a single procentric chiasma¹⁰.

Among the Pelobatidae (s.l.), *Pelobates cultripes* ($2n = 26$), *Pelodytes punctatus* ($2n = 24$)¹¹ and *Scaphiopus couchi* ($2n = 26$, Figure 4) have been studied. The karyotype of the Pelobatidae does not show acrocentric chromosomes; the meiosis of the male line has a morphology typical of most of the Anura. Various authors consider the Pelobatidae as similar to the Discoglossidae: the karyological indications do not confirm this hypothesis: they rather seem to be associated with certain families of higher Anura, particularly with the Leptodactylidae¹².

The few species of Microhylidae (s.l.) which were studied (*Gastrophryne carolinensis*: $2n = 22$ (unpublished), *Breviceps gibbosus*: $2n = 24$; *Kaloula pulchra*: $2n = 28$; *Phrynomerus bifasciatus*: $2n = 26$)¹³ are karyologically very different from one another but, apart from this, they do not show any marked characteristics of karyological primitiveness in the karyotype or in the meiosis of the male (Figure 3). From the morphology of the chromosome set, certain specialized Microhylidae (*Phrynomerus*) may be related to the Ranidae¹³.

The data so far assembled regarding the primitive Anura allow certain working hypotheses to be formulated about the phyletic relationships between the various families of the order; on the basis of these hypotheses, the Anura studied may be divided into 4 groups of families:

(1) Ascaphidae and Discoglossidae, closely related to each other from the karyological point of view, show no relationships with other families of Anura, with the possible exception of the American Pipidae.

(2) The Pipidae are extremely heterogeneous from the karyological point of view. Among the American species, *P. parva* has all acrocentric chromosomes, though they are not very numerous. *P. pipa* seems to have $2n = 22$, with 4 pairs of large metacentric, 3 pairs of large acrocentric and 4 pairs of small acrocentric chromosomes¹⁴: this karyotype may have some resemblance to that of the Ascaphidae. The bivalents of the male of *P. parva* have interstitial (procentric) chiasmata: the American Pipidae therefore may have some karyological features in common with Ascaphidae and Discoglossidae.

On the other hand, the African Pipidae have the karyological characteristics of the higher Anura, from the morphology of the meiosis of the male line and that of the chromosomes of the diploid set, all meta- or submetacentric. The African Pipidae have a high number of

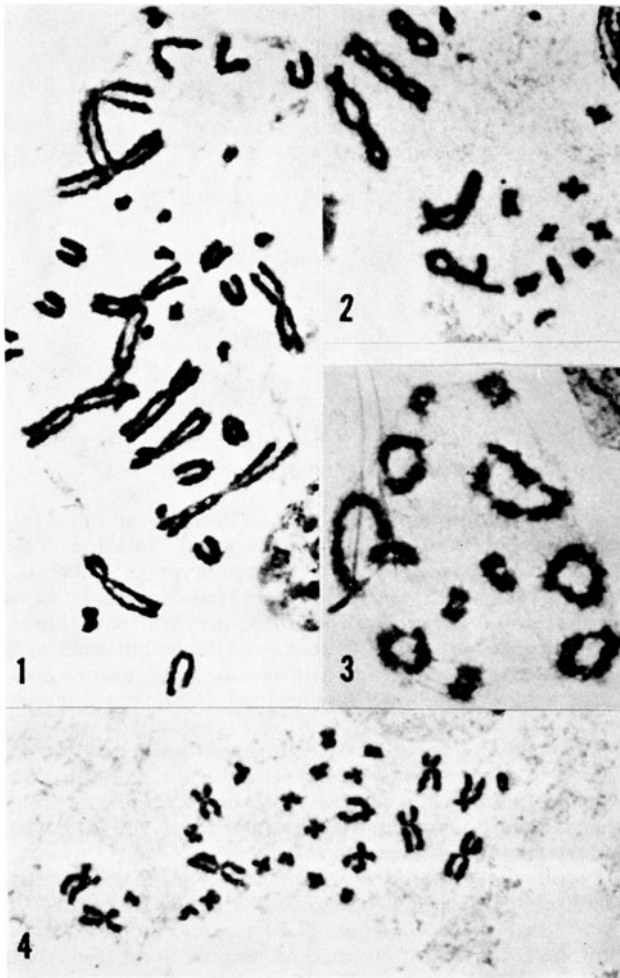


Fig. 1. Intestinal metaphase plate of *Leiopelma hochstetteri* (♀) (Ascaphidae). $\times 2000$.

Fig. 2. Spermatocyte bivalent chromosomes of *Alytes obstetricans* (Discoglossidae). $\times 2000$.

Fig. 3. Spermatocyte bivalent chromosomes of *Breviceps gibbosus* (Microhylidae). $\times 2000$.

Fig. 4. Spermatogonial metaphase of *Scaphiopus couchi* (Pelobatidae). $\times 2000$.

⁸ M. GALGANO, *Archo ital. Anat. Embriol.* 32, 171 (1933). – M. GALGANO and A. MORESCALCHI, *Rc. Accad. Sci. fis. mat., Napoli* 33, 79 (1966).

⁹ E. R. DUNN, *Am. Mus. Novit.* 1384, 1 (1948).

¹⁰ A. MORESCALCHI, *Experientia* 24, 81 (1968).

¹¹ A. MORESCALCHI, *Rc. Accad. Sci. fis. mat., Napoli* 31, 326 (1964).

¹² A. MORESCALCHI, *Experientia* 23, 1071 (1967).

¹³ A. MORESCALCHI, *Experientia* 24, 280 (1968).

¹⁴ T. WICKBOM, *Hereditas* 36, 363 (1950).

large chromosomes (18 pairs in *Xenopus* and 9 in *Hymenochirus*): if it is true, as regards the Amphibia, that evolution is generally accompanied by a reduction of the chromosome material^{6,15}, this fact may constitute a characteristic of karyological primitiveness with respect to the Microhylidae, the Pelobatidae and the higher Anura, which have, at the most, 7 pairs of large homologues in the more primitive forms and a smaller number in the higher forms.

(3) As previously stated, the Pelobatidae show various affinities with the Leptodactylidae; compared with the latter, they do not appear more primitive. What seems more interesting is that certain Leptodactylidae, in their turn, show karyological relationships, on the one hand with the Hylidae and on the other hand (certain Australian Leptodactylidae) perhaps with the Bufonidae and the Atelopodidae^{16,17}. All these families therefore constitute a group of karyologically associated forms, perhaps converging in a leptodactiloid (or pelobatoid) stock; they form a natural group, also from the anatomo-comparative point of view, because they comprise the higher Anura having an arciferal shoulder girdle¹.

(4) The Microhylidae, karyologically heterogeneous but not obviously primitive, may be related to the Ranidae, which are related to the Hyperolidae¹⁸. This vast group of forms, comprising the firmisternal Anura¹, is contrasted, from its extent and the number of species, with the group discussed in section (3), together with which it comprises all the higher Anura.

The fact that the Bufonidae, considered 'a pedomorphic offshoot' of the primitive Australian Leptodactyli-

dae¹, approach the karyotype of the families of group 4, may signify that (perhaps among the Leptodactylidae) there may exist forms of which the karyotype has characteristics intermediate between these of the 2 large groups of families (cf. ¹⁸)¹⁹.

Riassunto. Lo studio carilogico di specie di Anuri appartenenti alle famiglie più primitive dell'ordine conduce a formulare le seguenti ipotesi: (1) Ascaphidae e Discoglossidae sono strettamente collegati fra loro, e si differenziano da quasi tutti gli altri Anuri; (2) i Pipidae africani e quelli americani sono molto diversi fra loro; (3) i Pelobatidae hanno varie affinità carilogiche con i Leptodactylidae, e questi con gli Anuri superiori prevalentemente arciferi; (4) i Microhylidae hanno rapporti carilogici con i Ranidae.

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¹⁵ A. E. MIRSKY and H. RIS, J. gen. Physiol. 34, 451 (1951).

¹⁶ A. MORESCALCHI and G. GARGIULO, R. Accad. Sci. fis. mat., Napoli, in press.

¹⁷ F. H. ULLERICH, Chromosoma 21, 345 (1967).

¹⁸ R. LAURENT, Bull. Mus. r. Hist. nat. Belg. 18, 1 (1942). — O. A. REIG, Act. Trab. prim. Congr. sudam. Zool. 7, 271 (1961).

¹⁹ Research carried out through a contribution from the C.N.R. (Genetics Enterprise).

The Mitotic Chromosomes of the Lamprey *Mordacia mordax* (Agnatha: Petromyzonidae)

As representatives of the most primitive group of living vertebrates, the lampreys are of considerable phylogenetic interest. However, little is known about the cytogenetics of this group because the very small size and high number of chromosomes has made the analysis of karyotypes extremely difficult. It would appear that centromere positions and length variation of chromosomes within complements have not been clearly established for any lampreys. ZANANDREA and CAPANNA¹, in the only study which deals in some detail with lamprey chromosomes, have given counts for 3 species within the genus *Lampetra*. Other references to lamprey diploid numbers²⁻⁴ also refer to northern-hemisphere species.

The exclusively southern-hemisphere genus *Mordacia* has been regarded by most authors as sufficiently different from northern genera to merit being placed in at least a separate sub-family⁵. The current study was carried out on *M. mordax* (Richardson), to provide morphological details of the chromosomes of a member of the Petromyzonidae and to determine the diploid number of 1 of the 4 species of southern-hemisphere lampreys for comparison with those already recorded for northern species.

Material and methods. *M. mordax* occurs in rivers in south-eastern Australia. The difficulty of obtaining adults precluded the use of gonadal material for the study of mitotic and meiotic configurations. Ammocoete larvae were therefore employed for counts, the source of material being the epithelial cells lining the gut. Tissue was re-

moved from animals, after they had been placed in 0.01% colchicine solution for 5 h, and macerated in saline. The resulting cell suspension was treated hypotonically and fixed in 3:1 methanol-glacial acetic acid. Counts were made from photographs of air dried preparations stained with either Giemsa or 2% aceto-orcin. Bright field and phase microscopy helped to distinguish individual chromosomes which could not be resolved from photographic prints.

Results and discussion. Many preparations, made from the guts of numerous ammocoetes, were examined in order to find spreads clear enough for accurate counting. The distribution of counts obtained from the 23 satisfactory spreads is shown in the Table.

Despite the small size, large number and occasional overlapping of the chromosomes, a range in counts of 75-79, and the occurrence of 76 chromosomes in more than half the total number of counts, indicated that

¹ G. ZANANDREA and E. CAPANNA, Boll. Zool. 31, 669 (1964).

² L. A. CHUBAREVA, Vest. lenigr. gos. Univ. 9, 83 (1957).

³ W. J. R. LANZING, *Studies on the River Lamprey, Lampetra fluvialis, during its Anadromous Migration* (Uitgeversmaatschappij Neerlandia, Utrecht 1959).

⁴ S. NOGUSA, Mem. Hyogo Univ. Agric. 3, 1 (1960).

⁵ I. C. POTTER and R. STRAHAN, Proc. Linn. Soc. Lond. 179, in press.