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Report of the chromosome numbers of some Costa Rican anurans

by

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(Received for publication September 14, 1968)

Cytogenetic studies have proved useful in establishing phylogenetic relationships among anurans. Evolution in this group seems to have proceeded with a general reduction of the chromosome numbers, so that the more advanced families have lower numbers than the more primitive ones. The mechanism of this reduction probably involves centric fusion of two acrocentric chromosomes with the formation of a metacentric one, and the subsequent loss of the heterochromatic fragment. The fact that in anurans acrocentrics have been reported only among the more primitive families (5) seems to support this view. DUELLMAN (2) has pointed out some variations in this general trend that have not yet been explained.

The object of this report is to present the data so far collected on the chromosome numbers of 21 previously unrecorded Costa Rican species, representing eleven genera and seven families of the Anura.

MATERIAL AND METHODS

Chromosomes were studied on temporary slide-mounts of testicular material from 21 species of Costa Rican anurans. Voucher specimens and photographs of chromosome spreads of each species are deposited in the Museo de Zoología (UCR) of the Universidad de Costa Rica.

Most counts were determined from cells in the stage of diakinesis. Diplotene, metaphase I, metaphase II and the mitotic metaphase were also examined. Gonadal material was stained and mounted according to the technique outlined by DUELLMAN and COLE (3), except that the testicular fragments were immersed in 18% HCl for three minutes before staining with propiono-orcein. All

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specimens studied were injected intra-peritoneally with approximately 0.02 ml of 0.05 % colchicine per gram of body weight five hours before killing. Since the foregoing procedure was done directly in the laboratory, the fixative recommended by these authors was considered unnecessary.

RESULTS

Unless otherwise indicated, the following chromosome counts have not been previously reported:

RHINOPHRYNIDAE: *Rhinophrynus dorsalis* Duméril & Bibron, $N=11$.

DENDROBATIDAE: *Dendrobates auratus* (Girard), $N=9$. A population of *D. auratus* showing considerable pattern variation was studied. The haploid number of 9, which is also found in *Eleutherodactylus podiciferus*, is to my knowledge the lowest reported among the anurans. Counts were made on two populations of *Dendrobates pumilio* from Siquirres, Provincia de Limón; the results ($N=10$; $2N=20$), being in accordance with the figure obtained by DUELLMAN (2) from eight Panamanian populations.

LEPTODACTYLIDAE: *Eleutherodactylus podiciferus* (Cope), $N=9$; *E. diastema* (Cope), $N=10$; *E. fitzingeri* (Schmidt), *Engystomops pustulosus* (Cope), *Leptodactylus labialis* (Cope), and *L. bolivianus* Boulenger, all with haploid numbers of 11.

HYLIDAE: *Hyla angustilineata* Taylor, *H. pseudopuma* Günther, *H. rivularis* Taylor, *H. rosenbergi* Boulenger, *H. tica* Starrett, *H. uranochroa* Cope, all with $N=12$; *Agalychnis annae* (Duellman), and *Phyllomedusa lemur* Boulenger, with $N=13$.

MICROHYLIDAE: *Hypopachus variolosus* (Cope), $N=11$; *Glossostoma aterrimum* Günther, $N=13$.

BUFONIDAE: *Bufo coccifer* Cope, $N=11$.

RANIDAE: *Rana vibicaria* (Cope), and *R. warschewitschi* (Schmidt), both with haploid numbers of 13.

DISCUSSION

The application of cytogenetic evidence to indicate phylogenetic relationships in anurans will be of limited value until a substantial number of the species in each family has been studied. The counts presented here support some of the already established trends and point out possible future lines of research.

The families Bufonidae and Ranidae display consistent chromosome counts of 11 and 13 respectively. The Hylidae have so far presented the greatest diversity in chromosome numbers. They are reported to include species with haploid numbers ranging from 11 through 15 (2, 3). All the species studied of *Agalychnis* and *Phyllomedusa* have a haploid number of 13. The *Hyla* species presented here all have haploid numbers of 12. Other members of this genus are known to possess haploid numbers of 13 and 15 (3).

The counts presented for the two microhylids include the first reports on American members of this family. The higher chromosome count found in *Glossostoma* as compared to *Hypopachus* supports the long held view concerning the primitiveness of the former (6), and is in agreement with the conclusion of CARVALHO (1) regarding the origin of *Glossostoma* as being "...from some form ancestral to *Hypopachus*..." Additional chromosome studies in this family are highly desirable. Comments on the family Dendrobatidae are postponed until more data are available.

The leptodactylids *Eleutherodactylus podiciferus*, *E. diastema*, and *E. fitzingeri* were found to possess haploid numbers of 9, 10 and 11 respectively. *Engystomops* and two species of *Leptodactylus* presented here have haploid numbers of 11. Further studies in the genus *Eleutherodactylus* might prove helpful in establishing natural groupings.

The chromosome number obtained from *Rhinophrynus dorsalis* sheds no light at this time on the possible origin of this monotypic family.

Incidental counts were done on species previously reported in the literature. Published accounts (2, 3, 4) of the chromosome numbers of the following species were confirmed during the present study: *Centrolenella (Centrolene) prosoblepon*, *C. (Cochranella) fleishmanni*, *Hyla microcephala*, *Agalychnis callidryas* and *Smilisca baudini*.

SPECIMENS EXAMINED

RHINOPHRYNIDAE: *Rhinophrynus dorsalis* (UCR-2421). DENDROBATIDAE: *Dendrobates auratus* (UCR-2238). LEPTODACTYLIDAE: *Eleutherodactylus diastema* (UCR-2396); *E. fitzingeri* (UCR-3137); *E. podiciferus* (UCR-2371); *Engystomops pustulosus* (UCR-2408); *Leptodactylus bolivianus* (UCR-2404); *L. labialis* (UCR-2407). HYLIDAE: *Hyla angustilineata* (UCR-2220); *H. pseudopuma* (UCR-2416); *H. rivularis* (UCR-2222); *H. rosenbergi* (UCR-2406); *H. tica* (UCR-2357); *H. uvanochroa* (UCR-2229); *Agalychnis annae* (UCR-2342); *Phyllomedusa lemur* (UCR-2387). MICROHYLIDAE: *Hypopachus variolosus* (UCR-2343); *Glossostoma aterrimum* (UCR-2206). BUFONIDAE: *Bufo coccifer* (UCR-2340). RANIDAE: *Rana vibicaria* (UCR-2417); *R. warschewitschi* (UCR-2393).

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SUMMARY

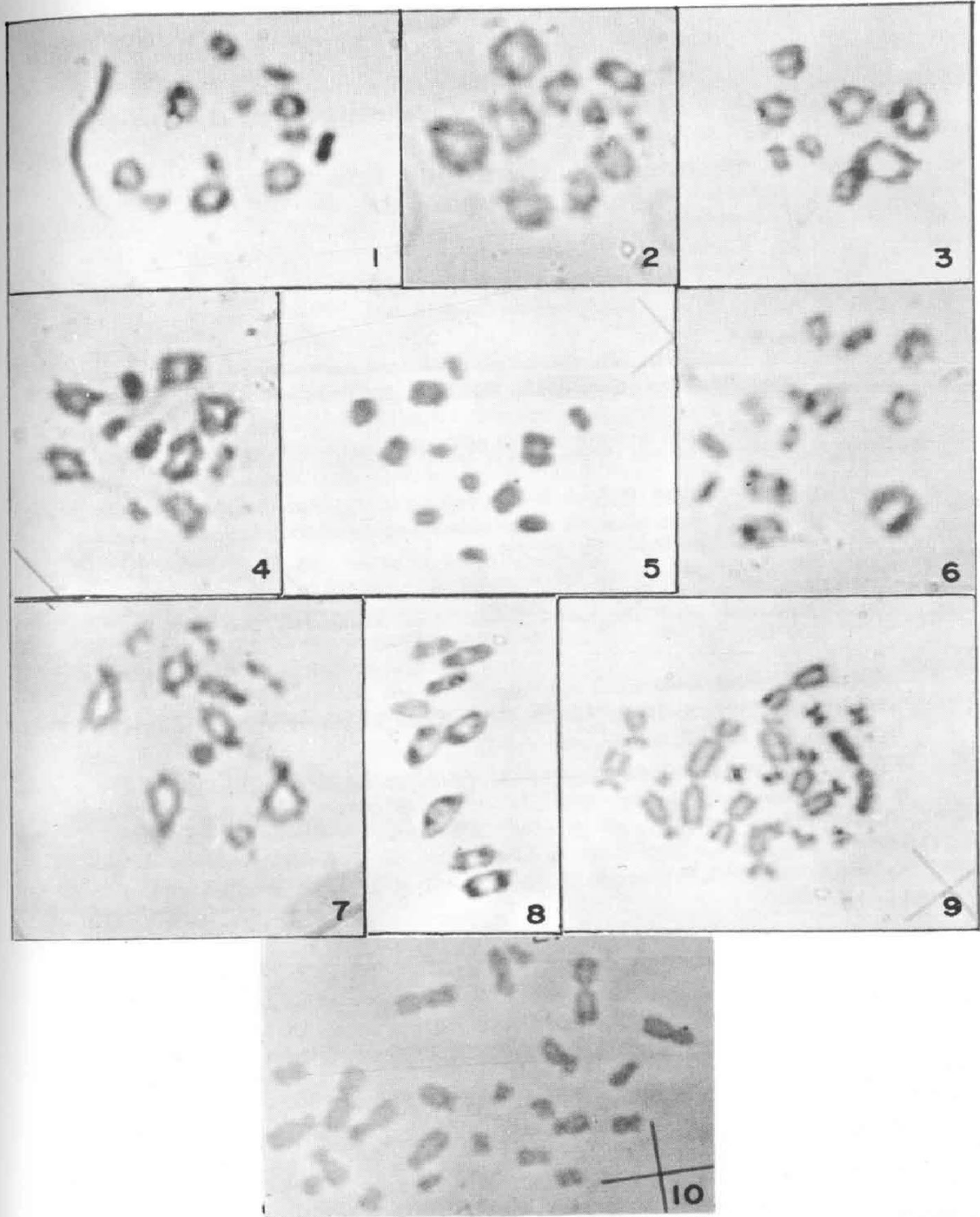
The chromosome numbers of 21 Costa Rican anurans, representing eleven genera and seven families were determined from testicular meiotic cells stained

with propiono-orcein. In this group higher numbers are generally present in the more primitive taxa. *Dendrobates auratus* and *Eleutherodactylus podiciferus* present a haploid number of 9, the lowest reported among anurans. The highest haploid number, 13, was found in members of the Hylidae, Microhylidae and Ranidae.

RESUMEN

Se determinó el número de cromosomas de 21 anuros de Costa Rica representando 11 géneros y 7 familias, en células meióticas testiculares teñidas con propiono-orceína. Entre los anuros, los números más altos se presentan generalmente en las unidades taxonómicas más primitivas. Los dos microhílidos estudiados, *Glossostoma aterrimum* e *Hypopachus variolosus*, presentan números haploides de 13 y 11 respectivamente. *Rhinophrynus dorsalis* posee el número haploide de 11. *Dendrobates auratus* y *Eleutherodactylus podiciferus* presentan números haploides de 9, el más bajo conocido entre los anuros. Otras especies del género *Eleutherodactylus* presentan números haploides más altos. *Engystomops pustulosus* y las especies del género *Leptodactylus* que fueron examinadas, presentan números haploides de 11. Los miembros estudiados de las familias Bufonidae y Ranidae poseen números haploides que están de acuerdo con determinaciones previamente reportadas, a saber, 11 y 13 respectivamente. Los hylidos estudiados del género *Hyla* dieron números haploides de 12, mientras que los de los géneros *Phyllomedusa* y *Agalychnis* presentan número haploide de 13.

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- Fig. 1. *Hyla pseudopuma* N=12.
 Fig. 2. *Eleutherodactylus diastema* N=10.
 Fig. 3. *Dendrobates auratus* N=9.
 Fig. 4. *Rhinophrynus dorsalis* N=11.
 Fig. 5. *Hyla angustilineata* N=12.
 Fig. 6. *Glossostoma aterrimum* N=13.
 Fig. 7. *Hypopachus variolosus* N=11.
 Fig. 8. *Eleutherodactylus podiciferus* N=9.
 Fig. 9. *Dendrobates pumilio* 2N=20.
 Fig. 10. *Hyla rivularis* 2N=24.



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