

Research Note

Parasites of the African Clawed Frog, *Xenopus laevis*, in Southern California, U.S.A.

BORIS I. KUPERMAN,¹† VICTORIA E. MATEY,^{1,4} ROBERT N. FISHER,² EDWARD L. ERVIN,²
MANNA L. WARBURTON,² LUDMILA BAKHIREVA,³ AND CYNTHIA A. LEHMAN¹

¹ Center for Inland Waters and Department of Biology, San Diego State University,
5500 Campanile Drive, San Diego, California 92182, U.S.A.,

² United States Geological Survey–Biological Resources Division, 5745 Kearny Villa Road,
San Diego, California 92123, U.S.A., and

³ Department of Epidemiology and Biostatistics, School of Public Health, San Diego State University,
San Diego, California 92182, U.S.A.

ABSTRACT: A total of 230 feral African clawed frogs, *Xenopus laevis*, from 3 localities in southern California were examined for parasites. The following species were found: 3 species of Protozoa, *Nyctotherus* sp., *Balantidium xenopodis*, *Protoopalina xenopodis*; 2 species of Monogenea, *Protopolystoma xenopodis*, *Gyrdicotylus gallieni*; 1 species of Digenea, *Clinostomum* sp. (as metacercariae); 1 species of Cestoda, *Cephalochlamys namaquensis*; 2 species of Nematoda, *Contracaecum* sp. (as larvae), *Eustrongylides* sp. (as larvae); and 1 species of Acanthocephala, *Acanthocephalus* sp. (as cystacanth). Of these, the protozoans *P. xenopodis* and *B. xenopodis*, both monogeneans, and the cestode have an African origin. *Contracaecum* sp., *Eustrongylides* sp., and *Acanthocephalus* sp. have not been previously reported from *X. laevis*.

KEY WORDS: Protozoa, Monogenea, Digenea, Cestoda, Nematoda, Acanthocephala, survey, African clawed frog, *Xenopus laevis*, southern California.

The African clawed frog, *Xenopus laevis* (Daudin, 1802), is a pipid anuran native to sub-Saharan Africa (Tinsley et al., 1996). *Xenopus laevis* was used for pregnancy assays in humans, and beginning in the 1930s, thousands of these frogs were exported from Africa to other continents, mainly Europe and the Americas. Specimens of feral *X. laevis* were recorded in the early 1960s in U.K., Germany, and the United States (Tinsley and McCoid, 1996) and in the early 1970s in southern California, U.S.A., in Los Angeles, Orange, Riverside, San Diego, and Ventura counties (Marhdt and Knefler, 1972; St. Amant et al., 1973; McCoid and Fritts, 1980). Now the African clawed frog can be found in most ponds, rivers, and streams in southwestern California (Tinsley and McCoid, 1996).

In South Africa, *X. laevis* harbors a diverse parasite fauna, with most species unique to this host. Twenty-five genera from 7 taxonomic groups (Protozoa, Monogenea, Digenea, Cestoda, Nematoda, Hirudinea, Acari) have been reported from *X. laevis* over some 60 yr of study (see Tinsley, 1996). In contrast, little is known about the parasites of *X. laevis* outside Africa. The monogenean *Protopolystoma xenopodis* (Price, 1943) has been reported from populations of *X. laevis* in Wales, U.K., and southern California, U.S.A., and the pseudophyllidean cestode *Cephalochlamys namaquensis* (Cohn, 1906) from populations on the Isle of Wight, U.K., and southern California (Lafferty and Page, 1997; Tinsley and Jackson, 1998; Jackson and Tinsley, 2001a, b). This note presents parasites of *X. laevis* collected in southern California and compares them with those found in African populations of *X. laevis*.

A total of 230 *X. laevis* (mean snout–vent length, 61 ± 25 mm SD, range 16–90 mm) were collected in 1999–2001 from 3 localities in southern California, U.S.A.: 132 from ponds of the Rancho Jamul System (32°40′03″N; 116°51′48″W), San Diego County; 68 from ponds of the Dulzura Creek System (32°37′30″N; 116°46′34″W), San Diego County; 30 from a backwater of the Santa Ana River (33°58′00″N; 117°38′43″W), Riverside County. The frogs were trapped using Gee[®] minnow traps or seines, transported to San Diego State University, and killed with an overdose of MS-222. After snout–vent measurement and external examination, each frog was examined internally. The body was opened by an incision from vent to throat and the gastrointestinal tract, kidney, urinary bladder, lungs, liver, heart, gonads, and body cavity were examined separately using a dissection microscope. Helminths were collected, counted, and selected specimens fixed for light microscopy (LM) or scanning electron micros-

⁴ Corresponding author.

† Deceased August 10, 2002.

Table 1. Prevalence, range, and infection sites of parasites of *Xenopus laevis* from 3 localities in southern California, U.S.A., 1999–2001.

| Parasite | Prevalence (%) | | | Range | | | Infection Site* | Present in Africa |
|-----------------------------------|----------------|-----|-----|-------|------|------|-----------------|-------------------|
| | RJS | DCS | SAR | RJS† | DCS† | SAR† | | |
| Protozoa | | | | | | | | |
| <i>Nyctotherus</i> sp. | 100 | 100 | 100 | | | | R | Uncertain‡ |
| <i>Balantidium xenopodis</i> | 81 | 79 | 77 | | | | R | Yes |
| <i>Protoopalina xenopodus</i> | 45 | 44 | 40 | | | | R | Yes |
| Monogenea | | | | | | | | |
| <i>Protopolystoma xenopodis</i> | 48 | 46 | 47 | 1–24 | 1–31 | 1–12 | UB | Yes |
| <i>Gyrdicotylus gallieni</i> | 10 | 18 | 10 | 1–13 | 1–20 | 1–9 | M, E, S | Yes |
| Digenea | | | | | | | | |
| <i>Clinostomum</i> sp. | 0 | 72 | 0 | | 1–4 | | LC, P, BC | Uncertain‡ |
| Cestoda | | | | | | | | |
| <i>Cephalochlamys namaquensis</i> | 51 | 46 | 30 | 1–55 | 1–37 | 1–18 | SI | Yes |
| Nematoda | | | | | | | | |
| <i>Contraecum</i> sp. | 3 | 4 | 0 | 1–3 | 1–2 | | BC | No |
| <i>Eustrongylides</i> sp. | 1 | 0 | 0 | 1–2 | | | ST | No |
| Acanthocephala | | | | | | | | |
| <i>Acanthocephalus</i> sp. | 1 | 0 | 0 | 1 | | | L | No |

* BC, body cavity; E, esophagus; L, liver; LC, lymph cavities; M, mouth cavity; P, pericardium; R, rectum; SI, small intestine, ST, subcutaneous tissue; UB, urinary bladder.

† RJS, Rancho Jamul System ($n = 132$); DCS, Dulzura Creek System ($n = 68$); SAR, Santa Ana River ($n = 30$).

‡ Neither African nor California specimens identified beyond genus.

copy (SEM) for precise identification. Helminths for LM were fixed in 70% ethanol, AFA (alcohol, formalin, acetic acid), or 5% formalin, stained with hematoxylin, and examined using a compound microscope. Protozoans for LM were collected by pipetting from rectal contents and examined alive or fecal smears were fixed in Schaudinn's fixative and stained with iron hematoxylin. Both helminths and protozoans selected for SEM were fixed in Karnovsky's solution, processed by standard methods, and examined with a Hitachi S-2700 scanning electron microscope. Voucher frogs were deposited in the herpetology collection of the California Academy of Sciences (CAS), San Francisco, California, U.S.A. (CAS 220089-220108).

Ten species of parasites were found: 3 species of Protozoa, *Nyctotherus* sp., *Balantidium xenopodis* De Puytorac and Grain, 1965, *Protoopalina xenopodus* Metcalf, 1923; 2 species of Monogenea, *P. xenopodis*, *Gyrdicotylus gallieni* Vercammen-Grandjean, 1960; 1 species of Digenea, *Clinostomum* sp. (as metacercariae); 1 species of Cestoda, *C. namaquensis*; 2 species of Nematoda (as larvae), *Contraecum* sp. and *Eustrongylides* sp.; and 1 species of Acanthocephala, *Acanthocephalus* sp. (as cystacanth). Selected specimens were deposited in the

Harold W. Manter Laboratory of Parasitology (HWML), University of Nebraska State Museum, Lincoln, Nebraska, U.S.A.: *Nyctotherus* sp., HWML 16618; *B. xenopodis*, HWML 16619; *P. xenopodus*, HWML 16620; *P. xenopodis*, HWML 16130; *G. gallieni*, HWML 16623; *Clinostomum* sp., HWML 16622, *C. namaquensis*, HWML 16132; *Contraecum* sp., HWML 16733; *Eustrongylides* sp., HWML 16734; and *Acanthocephalus* sp., HWML 16732. Prevalence and infection sites for each parasite and ranges of infection for helminths are given in Table 1.

The protozoans *B. xenopodis* and *P. xenopodus* were previously recorded for *X. laevis* in Africa (Thurston, 1970). The morphology of *Nyctotherus* sp. from California is similar to that reported for the African specimens only identified to generic level by Thurston (1970). Further study is required to identify the species of this parasite and to determine whether our material represents the same species as the African material. Both species of monogeneans have been reported from Africa (Tinsley, 1996). In addition, *P. xenopodis* has been recorded in *Xenopus* from U.K. and the United States (Tinsley and Jackson, 1998; Jackson and Tinsley, 2001a). Metacercariae of *Clinostomum* sp. have previously been

reported from *X. laevis* in Africa (Macnae et al., 1973) as well as ranid and hyloid frogs in the United States (Ingles, 1936; Goldberg et al., 1998; Goldberg and Bursey, 2001). However, it is unknown whether the same species of *Clinostomum* infects *X. laevis* in Africa and in California. *Cephalochlamys namaquensis* is the only cestode known to infect *X. laevis*. It has been reported from Africa, U.K., and United States (Thurston, 1967; Ferguson and Appleton, 1988; Tinsley, 1996; Lafferty and Page, 1997; Jackson and Tinsley, 2001b). In Africa, *Xenopus* harbors 2 camallanid, 1 capillarid, and 1 filariid species of nematode (Thurston, 1970; Wade, 1982; Jackson and Tinsley, 1995). We did not find any of these species but did find juvenile stages of 2 other species, *Contracaecum* sp. and *Eustrongylides* sp., which have not been reported in Africa. Acanthocephala is the only parasite phylum that has not been reported from *X. laevis* in Africa (see Tinsley, 1996). Our specimen was located in the liver and was assigned to *Acanthocephalus* sp. based on the structure of its trunk and proboscis.

In California, *X. laevis* harbors parasite species of African origin that apparently were carried by the frog to its new environment as well as species acquired after introduction. Introduced African species include the protozoans *B. xenopodis* and *P. xenopodus*, the monogeneans *P. xenopodis* and *G. gallieni*, and the cestode *C. namaquensis*, all species unique to *X. laevis* (see Tinsley, 1996). Neither did we find them in other frogs, i.e., *Hyla regilla*, *Hyla cadaverina*, *Rana catesbeiana*, *Bufo boreas*, or *Spea hammondi*, that we collected in the same localities as *X. laevis* (Kuperman, unpublished data) nor have they been reported in frogs of other areas of North America (Ingles, 1936; Baker, 1987; Goldberg et al., 1995; Goldberg, Bursey, Gergus et al., 1996; Goldberg, Bursey, Sullivan, et al., 1996; Goldberg et al., 1998).

African species with direct life cycles (protozoans and monogeneans) dominate the list of parasites carried to new environments. Of 13 African parasites with indirect life cycles unique to *Xenopus* (see Tinsley, 1996), only the cestode *C. namaquensis* seems to have found a suitable intermediate host, a cyclopoid copepod, that allowed its survival in California. Newly acquired parasites of *X. laevis* in California are predominantly bird parasites that use fish as an intermediate host, i.e., the nematodes *Contracaecum* sp. and *Eustrongylides* sp. and the acanthocephalan *Acanthocephalus* sp. (Yamaguti, 1961, 1963). Because *X. laevis* has a fully aquatic life history, it is perhaps more similar to fish than to

semiterrestrial frogs and may serve as a paratenic host. Species of *Clinostomum* are known to use both fish and amphibians as intermediate hosts (Yamaguti, 1961; Levine, 1980).

Populations of *X. laevis* in California harbor fewer species of parasites than African populations. The number of protozoan species is reduced from 9 to 3, digeneans from 10 to 1, nematodes from 5 to 2, and parasites of Hirudinea and Acari are missing. These data are in accord with a major principle of ecological parasitology (Dogiel, 1938; Kennedy and Bush, 1994): a host species with a particular parasite fauna in its native range will lose a number of parasite species as a result of introduction to a new environment and will acquire additional non-host-specific parasites in the new habitat.

We thank Terri Stewart, California Department of Fish and Game, Joe Funk, Bureau of Land Management, and Keith Greer, City of San Diego, for providing access to collection sites; Adam Backlin, Carmon Burton, Rob Lovich, Drew Stokes, Camm Swift, and Peter Tang for their assistance in the collection of frogs. We are deeply indebted to Charles R. Bursey, John M. Kinsella, and Bahram S. Dezfuli for their valuable help in the identification of trematodes, nematodes, and acanthocephalans. We express our gratitude to Stuart Hurlbert and Charles Bursey for their critical reading of the manuscript and to Richard C. Tinsley for useful discussion. This research was partially funded by the U.S. Geological Survey, Amphibian Research Monitoring Initiative, and NAQWA programs.

LITERATURE CITED

- Baker, M. R.** 1987. Synopsis of the Nematoda parasitic in amphibian and reptiles. Memorial University of Newfoundland, Occasional Papers in Biology 11:1–325.
- Dogiel, V. A.** 1938. Some achievements in the field of parasitology. Zoologicheskii Zhurnal 17:889–904. (In Russian.)
- Ferguson, R. R., and C. C. Appleton.** 1988. Some aspects of the morphology, population structure and larval biology of *Cephalochlamys namaquensis* (Cestoda: Diphyllidea), a parasite of the clawed toad, *Xenopus laevis*. South African Journal of Zoology 23:117–123.
- Goldberg, S. R., and C. R. Bursey.** 2001. Persistence of the nematode, *Oswaldo cruzia pipiens* (Molineidae), in the Pacific treefrog, *Hyla regilla* (Hylidae), from California. Bulletin of the Southern California Academy of Sciences 100:44–50.
- Goldberg, S. R., C. R. Bursey, and H. Cheam.** 1998. Helminths of two native frog species (*Rana chiricahuensis*, *Rana yavapaiensis*) and one introduced frog species (*Rana catesbeiana*) from Arizona. Journal of Parasitology 84:175–177.

- Goldberg, S. R., C. R. Bursey, E. W. A. Gergus, B. K. Sullivan, and Q. A. Truong.** 1996. Helminths from three treefrogs *Hyla arenicolor*, *Hyla wrightorum*, and *Pseudacris triseriata* (Hyllidae) from Arizona. *Journal of Parasitology* 82:833–835.
- Goldberg, S. R., C. R. Bursey, and I. Ramos.** 1995. The component parasite community of three sympatric toad species, *Bufo cognatus*, *Bufo debilis* (Bufonidae), and *Spea multiplicata* (Pelobatidae) from New Mexico. *Journal of the Helminthological Society of Washington* 62:57–61.
- Goldberg, S. R., C. R. Bursey, B. K. Sullivan, and Q. A. Truong.** 1996. Helminths of the Sonoran green toad, *Bufo retiformis* (Bufonidae), from southern Arizona. *Journal of the Helminthological Society of Washington* 63:120–122.
- Ingles, L. G.** 1936. Worm parasites of California amphibia. *Transactions of the American Microscopical Society* 55:73–92.
- Jackson, J. A., and R. C. Tinsley.** 1995. Evolutionary relationships, host range and geographical distribution of *Camallanus* Railliet & Henry, 1915 (Nematoda: Camallanidae) from clawed toads of genus *Xenopus* (Anura: Pipidae). *Systematic Parasitology* 32:1–21.
- Jackson, J. A., and R. C. Tinsley.** 2001a. *Protopolystoma xenopodis* (Monogenea) primary and secondary infections in *Xenopus laevis*. *Parasitology* 123:455–463.
- Jackson, J. A., and R. C. Tinsley.** 2001b. Host-specificity and distribution of cephalochlamydid cestodes: correlation with allopolyploid evolution of pipid anuran hosts. *Journal of Zoology* 254:405–419.
- Kennedy, C. R., and A. O. Bush.** 1994. The relationship between pattern and scale in parasite community: a stranger in a strange land. *Parasitology* 109:187–196.
- Lafferty, K. D., and C. J. Page.** 1997. Predation on the endangered tidewater goby, *Eucyclogobius newberryi*, by the introduced African clawed frog, *Xenopus laevis*, with notes on the frog's parasites. *Copeia* 1997: 589–592.
- Levine, N. D.** 1980. *Nematode Parasites of Domestic Animals and of Man*, 2nd ed. Burgess Publishing, Minneapolis, Minnesota. 477 pp.
- Macnae, W., L. Rock, and M. Makowski.** 1973. Platyhelminths from the South African clawed toad, or platanna (*Xenopus laevis*). *Journal of Helminthology* 47:199–235.
- Marhdt, C. R., and F. T. Knefler.** 1972. Pet or pest? *Environmental Southwest* 466:2–5.
- McCoid, M. J., and T. H. Fritts.** 1980. Observations of feral populations of *Xenopus laevis* (Pipidae) in southern California. *Bulletin of the Southern California Academy of Sciences* 79:82–86.
- St. Amant, J. A., F. G. Hoover, and G. R. Stewart.** 1973. African clawed frog, *Xenopus laevis* (Daudin) established in California. *California Fish and Game* 59: 151–153.
- Thurston, J. P.** 1967. The morphology and life-cycle of *Cephalochlamys namaquensis* (Cohn, 1906) (Cestoda: Pseudophyllidae) from *Xenopus muelleri* and *X. laevis*. *Parasitology* 57:187–200.
- Thurston, J. P.** 1970. Studies of some Protozoa and helminth parasites of *Xenopus*, the African clawed toad. *Revue de Zoologie et de Botanique Africaines* 82:349–369.
- Tinsley, R. C.** 1996. Parasites of *Xenopus*. Pages 233–261 in R. C. Tinsley and H. R. Kobel, eds. *The Biology of Xenopus*. Oxford University Press, Oxford, U.K.
- Tinsley, R. C., and J. A. Jackson.** 1998. Speciation of *Protopolystoma* Bychovsky, 1957 (Monogenea: Polystomatidae) in hosts of the genus *Xenopus* (Anura: Pipidae). *Systematic Parasitology* 40:93–141.
- Tinsley, R. C., C. Loumont, and H. R. Kobel.** 1996. Geographical distribution and ecology. Pages 35–39 in R. C. Tinsley and H. R. Kobel, eds. *The Biology of Xenopus*. Oxford University Press, Oxford, U.K.
- Tinsley, R. C., and M. J. McCoid.** 1996. Feral populations of *Xenopus* outside Africa. Pages 81–94 in R. C. Tinsley and H. R. Kobel, eds. *The Biology of Xenopus*. Oxford University Press, Oxford, U.K.
- Wade, S. E.** 1982. *Capillaria xenopodis* sp. n. (Nematoda: Trichuroidea) from the epidermis of the South African clawed frog (*Xenopus laevis* Daudin). *Proceedings of the Helminthological Society of Washington* 49:86–92.
- Yamaguti, S.** 1961. *Systema Helminthum*. Vol. 3, Part I: The Nematodes of Vertebrates. Interscience Publishers Inc., New York. 679 pp.
- Yamaguti, S.** 1963. *Systema Helminthum*. Vol. 5: Acanthocephala. Interscience Publishers Inc., New York. 423 pp.