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Three Rarely Reported Digeneans Inhabiting Amphibians From Vancouver Island, British Columbia, Canada

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ABSTRACT: Three rarely reported species of digeneans were collected in amphibian hosts from Diversion Reservoir, Sooke, Vancouver Island, British Columbia, Canada. The endangered anuran *Rana aurora* hosted *Glythelmins californiensis*, *Gorgoderina multilobata*, and *Megalodiscus microphagus*. In addition, the anuran *Pseudacris regilla* and the caudatan *Taricha granulosa* hosted *Megalodiscus microphagus*. *Gorgoderina multilobata* has been reported only once since 1936 and has never been reported outside of California. We note several characters not previously reported for *Gorgoderina multilobata* and *G. aurora*.

During the summer of 2002, a single specimen of the red-legged frog, *Rana aurora*; 3 rough-skinned newts, *Taricha granulosa*; and 4 Pacific tree frogs, *Pseudacris regilla*; were collected from Diversion Reservoir, Sooke, Vancouver Island (123°58'18.4"W; 48°30'0.54"N) and examined for parasitic helminths. Digenetic trematodes were collected while alive and fixed by shaking in hot formalin, and then stored in 70% ethanol. Hosts were euthenized with benzocaine. Specimens were stained with Mayer's haematoxylin, dehydrated, and mounted in Canada balsam for identification and deposited at the United States National Parasite Collection in Beltsville, Maryland (USNPC).

The anuran *R. aurora* hosted 2 specimens of *Glythelmins californiensis* (Cort, 1919) Miller, 1930, in the anteriormost part of the small intestine (USNPC 95033). *Glythelmins californiensis* was originally described in this host from a frog farm near San Francisco by Cort (1919). Ingles (1936) later reported it in *Rana boylei* from Butte County, California. Other reports of *G. californiensis* in *R. aurora* include western Washington and Oregon (Lehmann, 1960), Langley and Duncan, British Columbia (O'Grady, 1987); and Sooke and Nanaimo, British Columbia (Moravec, 1984). This species has also been reported from Mexico in *Rana pipiens* (Caballero y Caballero, 1942) (actually an undetermined

member of the "*Rana pipiens* group" because *Rana pipiens* sensu stricto does not occur in Mexico); *Rana dunni* (Pulido-Flores, 1994; Razo-Mendivil et al., 1999; Perez-Ponce de León et al., 2000); *R. megapoda* (Razo-Mendivil et al., 1999); *Rana montezumae* (Caballero y Caballero, 1942; Leon-Regagnon, 1992; Razo-Mendivil et al., 1999; Perez-Ponce de León et al., 2000), and *R. neovolcanica* (Razo-Mendivil et al., 1999).

All 3 amphibian species from Diversion Reservoir hosted *Megalodiscus microphagus* Ingles, 1936. The *R. aurora* specimen hosted 8 *M. microphagus* in the intestine (USNPC 95034), and 1 of the 4 *P. regilla* was infected with 2 intestinal *M. microphagus* (USNPC 95035). Two of the 3 rough-skinned newts, *T. granulosa*, were infected with *M. microphagus*, 1 with a single specimen in the esophagus and the other with 7 in the esophagus (USNPC 95037) and 2 in the intestine (USNPC 95036). Ingles (1936) described *M. microphagus* in *Bufo boreas* from Shasta County, California, noting that immature specimens occurred in the intestine, rectum, and bladder with mature specimens, having lost eyespots, found only in the bladder. We, on the other hand, found immature specimens in the esophagus of *T. granulosa* and mature specimens only in the intestine of all 3 host species. This is an unusual find, because all other species of *Megalodiscus* occur in the rectum.

Previous reports of *M. microphagus* in *Rana aurora* include Oregon (Macy, 1960), Nanaimo, Sooke, and Marion Lake, British Columbia (Efford and Tsumura, 1969; Moravec, 1984); and Endert's Beach, Crescent City, Del Norte County, California and Redwood National Park, Humboldt County, California (Goldberg et al., 2004). *Megalodiscus microphagus* has also been reported previously in *Taricha granulosa* and *Pseudacris regilla* from Oregon (Macy, 1960) and from Marion Lake, British Columbia (Efford and Tsumura, 1969), and in *T. granulosa* from Nanaimo and Sooke, British Columbia (Moravec, 1984). Other reported hosts include the brown and long-toed salamanders, *Ambystoma gracile*

and *A. macrodactylum*, from Marion Lake, British Columbia (Efford and Tsumura, 1969); the Pacific giant salamander, *Dicamptodon ensatus*, from Oregon (McCauley and Pratt, 1959); the boreal toad, *Bufo boreas*, from Marion Lake, British Columbia (Efford and Tsumura, 1969); the foothill yellow-legged frog, *Rana boylei*, from Humboldt, California (Walker, 1965); and the western spotted frog, *Rana pretiosa*, from Bear Spring, Oregon (Panitz and Briggs, 1968).

Rosen and Manis (1976) reported *M. microphagus* in 1 of 70 bullfrogs, *R. catesbeiana*, from Arkansas. However, there is no record of these authors having deposited voucher specimens in any museum collection, so we are unable to confirm this identification. We believe this report might have been a misidentification. *Rana catesbeiana* has previously been reported as hosting *M. temperatus* (Stafford, 1905) Harwood, 1932 (e.g., Slagle, 1966; Brooks, 1977; Muzzall, 1991; Andrews et al., 1992; McAlpine and Burt, 1998) and *M. intermedius* (Hunter, 1930), Harwood, 1932 (e.g., Brandt, 1936), which might be a synonym of *M. temperatus*.

Two specimens of *Gorgoderina multilobata* Ingles and Langston, 1933 were found to infect the urinary bladder of the *R. aurora* specimen (USNPC 95032). *Gorgoderina multilobata* was originally reported in *R. boylei* from the Santa Cruz Mountains and streams near Chico, California and in *R. aurora* from a small pond near Berkeley, California. Ingles (1936) later described *G. aurora* from *R. aurora*, stating that specimens of *G. aurora* had been considered to be young ovigerous adults of *G. multilobata*, on the basis of smaller body size (5–8 mm vs. 7.1–11.7 mm), and greater body width to body length ratio (1:8.4–12 vs. 1:3.8–8.5). Ingles (1936), however, noted that the small (*G. aurora*) and large (*G. multilobata*) ovigerous forms could be differentiated consistently on the basis of the ratio of oral sucker length to ventral sucker length (1:2.0–2.4 vs. 1:1.69, respectively) and in the morphology of the vitellarium (irregularly lobed vitellarium vs. 2 clusters of 7–9 lobes, respectively). Ingles (1936) also reported *G. multilobata* in *R. pretiosa* (= *R. cascadae*, Turner, 1958), extending the range from the Santa Cruz Mountains north to the Oregon-California border. However, since the Ingles (1936) description, there has been no other report of *G. aurora* and there has been only one other report of *G. multilobata*, also from California (Walker, 1965).

In attempting to identify the material collected from Vancouver Island, we examined the holotype and 2 paratypes each of *G. multilobata* (USNPC 8665) and *G. aurora* (USNPC 8932). Ingles and Langston (1933:244) described the vitellarium of *G. multilobata* as “two grape-like clusters, each possessing from seven to nine lobes,” and the vitellarium of *G. aurora* as “not having regularly lobed vitellaria” (Ingles, 1936:77). Examination of deposited material revealed that *G. multilobata* has 2 clusters of 8 lobes, whereas *G. aurora* specimens have 6 lobes on the sinistral cluster and 7 lobes on the dextral cluster. The specimens we collected from Sooke, Vancouver Island have 7 lobes in each cluster.

Ingles and Langston (1933) described a bipartite internal seminal vesicle (but did not mention or illustrate a pars prostatica) and esophagus surrounded by glands for *G. multilobata*. We confirm herein, that these structures are also present in *G. aurora* and the specimens from Sooke, Vancouver Island.

Ingles and Langston (1933) described *G. multilobata* as having ceca of unequal length, whereas Ingles (1936) reported that *G. aurora* has ceca of equal length. Our examination of type material indicates that the sinistral cecum is longer than the dextral in *G. aurora*, whereas in *G. multilobata* the dextral cecum is slightly longer than the sinistral. The specimens from Sooke, Vancouver Island resemble *G. multilobata* in having a shorter sinistral cecum and longer dextral cecum. Ingles and Langston (1933) described the ovary of *G. multilobata* as being on either side of the body. Our examination of type material indicates that the ovary is located on the sinistral side of the body in *G. aurora* and on the dextral side in *G. multilobata*. The specimens from Vancouver Island resemble *G. aurora* by having ovaries on the sinistral side of the body. Finally, our examination of the type material indicates that the uterus does not overlap the testes in *G. multilobata* but does so in *G. aurora*. In this regard, the specimens from Vancouver Island resemble *G. multilobata*. Finally, specimens collected in this report have an oral : ventral sucker width ratio less than 2 (1:1.79), resembling *G. multilobata* more than *G. aurora*.

There appear to be 2 possible interpretations of the morphological diversity encountered in the present study. One possibility is that both

G. multilobata and *G. aurora* are valid species, and the material from Vancouver Island represents a third species. The other possibility is that all material pertains to a single highly variable species, *G. multilobata*. Resolving this conundrum will require additional collections, both to provide substantial numbers of specimens for morphological analysis, and to provide material for molecular analysis to strengthen inferences about the significance of the morphological diversity encountered.

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