Studies on Puerto Rican Physidae

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MEMBERS of the family Physidae are among the most common and widespread fresh-water mollusks in Puerto Rico and are frequently associated with Australorbis glabratus, intermediate snail host of Schistosoma mansoni. In several recent papers on the ecology of A. glabratus in Puerto Rico (1-4) and in a key to the fresh-water mollusks of Puerto Rico by Van der Schalie (5), the only physid referred to is Physa cubensis Pfeiffer. Ferguson and Richards (6) reviewed listings of physid mollusks from Puerto Rico by earlier collectors (including several species of Physa and Aplexa), confirming P. cubensis and adding Aplexa marmorata (Guilding) from recent collections. As a potential intermediate host of Fasciola hepatica, P. cubensis (7) must be differentiated from Aplexa if molluscacidal control of sheep liver fluke disease is attempted locally.

Methods

Our studies of fresh-water mollusks, including these two genera, consisted of extensive field studies and collections in Puerto Rico and the U.S. Virgin Islands; laboratory culture and experimental exposure to *S. mansoni* miracidia; and examination of internal structure, both in specimens dissected alive and stained with Janus green and in permanently mounted preparations which had been treated with merthiolate iodine formalin (MIF) stain preservative.

Results

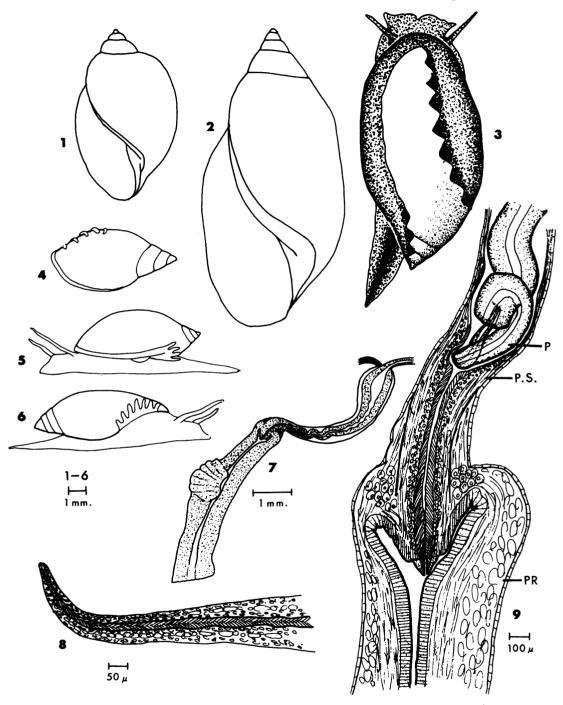
Shell. Although some shells were difficult to identify with certainty, the typical shells of common Physa and Aplexa of Puerto Rico and the Virgin Islands were distinctive. In the

Physa (fig. 1), considered to be *P. cubensis*, the body whorl was broadly expanded, the sutures deep, and the surface usually dull and rough with growth lines. In *A. marmorata* (fig. 2) the body whorl was narrow with parallel sides, the sutures very shallow, and the surface typically smooth and shiny.

Morphology. In P. cubensis the black body pigment was diffused, while in A. marmorata it was concentrated to form a medial black stripe down the posterior part of the foot as well as spots on each projection of the mantle margin (fig. 3). The body surface of A. marmorata contained numerous small yellow-green vacuoles, which gave the body a characteristic tint and released their pigment when the body was crushed or dissected. This feature was absent in P. cubensis. In the field when identification was not apparent from the morphology of the shell or body, snails crushed in a paper towel were distinguished by the yellowgreen pigment released by A. marmorata. This feature was consistent in collections throughout Puerto Rico and the Virgin Islands.

The mantle margin of P. cubensis had prominent fingerlike projections, while that of A. marmorata was typically plain or serrated as in figure 3. In several localities a small A plexa was collected with a mantle margin that had a few fingerlike projections (figs. 4-6). Whether this form is a variant of the larger, more typical A. marmorata or whether more than one species is involved was not determined.

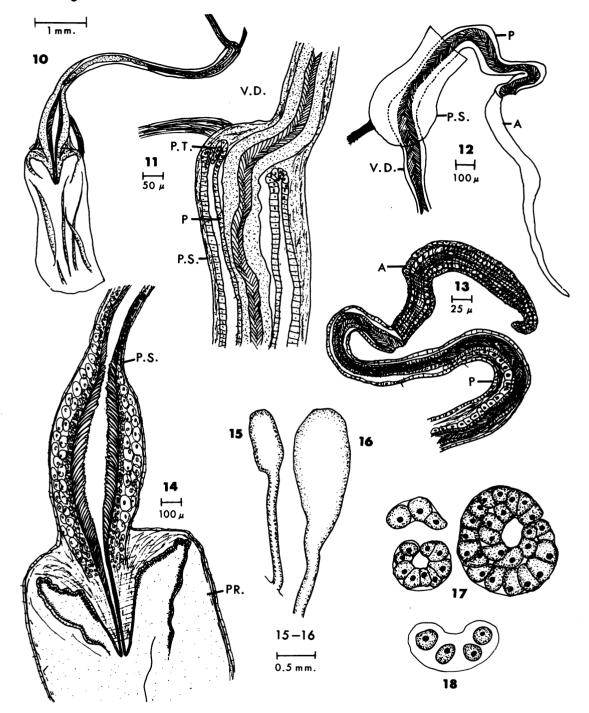
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A----appendage; P----penis; PR----preputium; P.S.---penis sac; P.T.---proximal tubules; V.D.---vas deferens

 Physa cubensis, shell.
Aplexa marmorata, shell.
A. marmorata, dorsal view, alive.
Aplexa, small variety, three views, alive.
P. cubensis, preputium with lateral accessory organ, penis sac, and penis, prepared slide (MIF). 8. P. cubensis, end of penis (MIF). 9. P. cubensis, junction of penis sac and preputium (MIF).

Figures 10-18.



10. A. marmorata, preputium, penis sac, and penis (MIF). 11. A. marmorata, proximal end of penis sac showing muscle attachment, entry of vas deferens, proximal tubules (MIF). 12. A. marmorata, proximal portion of penis sac and penis showing lateral opening of penal duct and appendage (MIF). 13. A. marmorata, distal portion of penis showing lateral penal duct opening and appendage, fresh preparation with Janus green. 14. A. marmorata, junction of penis sac and preputium (MIF). 15. A. marmorata, seminal receptacle (MIF). 16. P. cubensis, seminal receptacle (MIF). 17. A. marmorata, three egg clutches. 18. Aplexa, small variety, egg clutch. This form also has the medial black stripe on the posterior part of the foot and the yellowgreen pigment vacuoles.

The mantle of P. cubensis was frequently reflected over the edge of the shell. However, the mantle of the Puerto Rican A. marmorata was reflected over the shell even more (fig. 3), sometimes almost completely hiding the shell.

Our studies of internal morphology were limited primarily to the reproductive system. particularly the penis, penis sac, and preputium. In P. cubensis (figs. 7-9) the penis was swollen proximally, then tapered evenly to the end, the vas deferens joined the proximal end of the penis sac centrally, and the penal duct opened out at the end of the penis; the wall of the distal end of the penis sac was thickened but not bulbous; the walls of the preputium were thick and muscular and included a well-developed lateral accessory organ. In A. marmorata (figs. 10-14) the penis was not swollen proximally; the vas deferens joined the proximal end of the penis sac laterally so that the duct turned at a right angle; the penal duct opened out laterally, with a long swollen and muscular appendage extending beyond the orifice of the penal duct; the cavity of the penis sac had small, poorly developed "proximal tubules;" the distal wall of the penis sac was swollen with the elongate, fingerlike inner epithelial cells; the preputium was thin walled, with poorly developed pilasters, was ciliated proximally, and lacked an accessory organ. The lateral opening of the penal duct was consistent in both forms of Aplexa; emission of sperm was observed in fresh preparations.

In the larger A plexa the seminal receptacle (fig. 15) was elongate, narrowing abruptly to a long stalk; the seminal vesicles were all white. In the smaller A plexa the seminal receptacle was oval with a short stalk; the seminal vesicles were orange proximally and white distally. In *Physa* (fig. 16) the seminal receptacle was broadly rounded distally, tapering gradually to the stalk.

Eggs of the larger A plexa (fig. 17) were laid in clutches with a small gelatinous envelope; those of the smaller A plexa (fig. 18), with a relatively larger envelope.

Parasites. In a series of collections from St. Croix and the Virgin Islands and in a few Puerto Rican collections most of the *P. cubensis* were heavily infected with *Trichodina chlorophora* (8). Collections of *A. marmorata* from nearby locations were all negative for this parasite. As mentioned, *P. cubensis* is an intermediate host for *F. hepatica* in Puerto Rico. Although larvae were not observed to develop to the infective stage, miracidia of *S. mansoni* penetrated *Aplexa*, but not *Physa*, in several experimental exposures (9). *Aplexa* was the only nonplanorbid in the experimental exposures penetrated by *S. mansoni* miracidia.

Distribution and ecology. In a study based on 750 collections from Puerto Rico and the U.S. Virgin Islands, Richards and Ferguson (10) collected Physa 116 times and Aplexa 112; these were among the 5 most commonly occurring species. Physa was most common in ponds, streams, drainage ditches, and lakes; Aplexa, in marshes, limesink ponds, streams, and drains. Physa was the common species along the south coast of Puerto Rico; Aplexa, in the higher areas, western Puerto Rico, limestone regions, and the Vieques and St. Kitts Islands. Both were abundant in the Virgin Although both were common and Islands. widely distributed, they were found in association in the same habitats only about 10 percent of the times collected. Aplexa was associated with A. glabratus in 72 percent of its collections; Physa, in 32 percent.

Discussion

Among criteria for separation of the genus Physa from Aplexa, Aguayo (7), Pennak (11), and others include a description of reflection of the mantle over the shell in Physa but not in Aplexa. As noted in the Puerto Rican Aplexa, the extreme reflection of the mantle sometimes almost completely hides the shell. If the Caribbean form identified as A. marmorata is congeneric with species lacking the reflexed mantle, then this characteristic varies within the genus and should not be used in distinguishing Aplexa from Physa.

In addition to differentiating between Puerto Rican *Physa* and *Aplexa*, these two should be distinguished from the elongate, sinistral planorbid *Plesiophysa*. This snail was found to be the most widespread mollusk in a series of 22 ponds sampled in the limestone area of north central Puerto Rico (10, 12). Plesiophysa was found associated with Aplexa in 44 percent of its collection localities; with Physa, in 8 percent. The shell of Plesiophysa has a prominent, dull periostracum, a blunt apex, deep sutures, and shouldered whorls. Crushing of Plesiophysa releases hemoglobin, in contrast to the release of yellow-green pigment by Aplexa and the lack of apparent pigment release in Physa; internal morphology includes multiple flagella at the proximal end and a circle of glands at the distal end of the verge sac.

Pimentel and White (4) listed P. cubensis as the most common molluscan associate of A. glabratus found in a study in Puerto Rico. Their table shows that 45.5 percent of the habitats of A. glabratus include also P. cubensis, but it does not list Aplexa. They concluded that the snails Physa cubensis, Lymnaea spp., and Drepanotrema anatinum indicate the presence of A. glabratus at odds greater than 1:1. Richards and Ferguson (10) found D. anatinum associated with A. marmorata about five times as often as with P. cubensis and A. glabratus associated with A. marmorata about twice as often as with P. cubensis.

Summary

Two genera of snails in Puerto Rico, *Physa* and *Aplexa*, need to be differentiated if molluscacidal control of sheep liver fluke disease is to be tested locally. The appearance of typical shells and the snails' release or nonrelease of characteristic pigment upon crushing are aids in differentiation. Studies of the internal morphology of their reproductive systems also reveal distinctive differences. The species *Physa* cubensis and *Aplexa marmorata* are among the dominant fresh-water mollusks of the area; both are about equally common and widespread but rarely occur together. *Australorbis gla*- bratus, the intermediate snail host of Schistosoma mansoni, was found associated with A. marmorata about twice as often as with P. cubensis.

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