

BIOMPHALARIA TENAGOPHILA GUAIBENSIS SSP.N. FROM SOUTHERN BRAZIL AND URUGUAY (PULMONATA: PLANORBIDAE). I. MORPHOLOGY

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A new subspecies of planorbid snail, Biomphalaria tenagophila guaibensis, is described. It has been found along the coastal belt of the Brazilian state of Rio Grande do Sul and the middle part of Uruguay, from Porto Alegre to Mercedes. It differs from the nominate subspecies, Biomphalaria tenagophila tenagophila, in the appearance of the penial complex (prepuce longer and proportionally slenderer in B. t. guaibensis, shorter and proportionally stouter in B. t. tenagophila), in the ratio between the lengths of the penial sheath and the prepuce, in the ratio between the lengths of the uterine complex and the penial complex, and in a coefficient of difference of 2.44 for the ratio between the penis sheath and prepuce and of 2.02 for the ratio between the uterine complex and penial complex. The shell and the other organs of the genital system are similar in both subspecies.

B. t. guaibensis is very similar to Biomphalaria occidentalis Paraense, 1981, but is readily separated from it by the presence of a vaginal pouch, which is lacking in the latter, besides showing highly significant difference in the penis sheath: prepuce and uterine complex: penial complex ratios.

Crossbreeding experiments which lend additional support to the recognition of B. t. guaibensis as a subspecies will be reported elsewhere.

*Biomphalaria tenagophila, described by Orbigny in 1835, is a planorbid snail which occupies a wide range in South America east of the Andes. It was originally referred to the Argentine province of Corrientes (Orbigny, 1835:26), and subsequently restricted to the "canton de las Ensenadas" (Orbigny, 1837:347; see Paraense, 1981:201). It was collected again by Orbigny (1835:26) in Rio de Janeiro, Brazil, and treated as a different species under the name of *Planorbis ferrugineus* Spix, 1827. As shown by Paraense (1961a, b), the specimens from Rio de Janeiro belong to *B. tenagophila*, and *P. ferrugineus*, described by Spix from Bahia, Brazil, is a synonym of *Biomphalaria glabrata*.*

*A description of the shell and anatomy of B. tenagophila, under the synonymic name of Australorbis nigricans, based on the study of 50 specimens from Itajubá, state of Minas Gerais, was published by Paraense & Deslandes (1955). The ratio between the lengths of the penial sheath and prepuce was about 1:1 that sample (mean 0.84 ± 0.12). Two additional samples of 40 and 50 specimens from Santos, state of São Paulo, were also examined (Paraense & Deslandes, 1956), again with a ratio of about 1:1, ranging from 0.6 to 1.2 (mean 0.87 ± 0.13) in the former sample and from 0.7 to 1.3 (mean 1.01 ± 0.10) in the latter. During subsequent years many other samples from 113 localities over the whole range of *B. tenagophila* were examined, with a penial sheath to prepuce ratio varying from 0.5 to 1.5.*

*In November 1957 I collected from a drainage ditch at Guaíba, state of Rio Grande do Sul, 8 specimens of a planorbid similar in shell and anatomic characters to *B. tenagophila*, except for the appearance of the penial complex. Four of them showed a proportionally very long and slender prepuce from 2.0 to 2.4 times as long as the penial sheath (ratio between the lengths of penial sheath and prepuce from 0.4 to 0.5); in two others an anomaly was observed, consisting in the intromission of the distal portion of the vas deferens and proximal portion of the penial complex more or less deeply into the renal vein (Paraense, to be published). The remaining two specimens were aphyllic. My incipient experience at that time led me to consider that sample as a small collection of anomalous *B. tenagophila*. Thenceforth, however, specimens from other localities resembling those from Guaíba have been observed, and investigations to elucidate their taxonomic status led me to conclude that they constitute a new subspecies, which is described below under the name of *Biomphalaria tenagophila guaibensis*.*

MATERIAL AND METHODS

Fifty specimens measuring 12 ± 1 mm in shell diameter, collected on April 27, 1983 from a drainage ditch at the district of Sans-Souci, Guaíba municipality, were preserved in Railliet-Henry fixative for anatomical study, as described by Paraense (1981), with the only difference that they were previously relaxed in a 0.05% (not 0.1%) solution of nembutal. Additional specimens of different sizes from the same breeding-place were also collected and preserved.

Another sample of 50 specimens from Viamão, 30km eastward of Guaíba, also measuring 12 ± 1 mm in shell diameter and collected on April 27, 1983, was treated in the same way for comparison with the Guaíba sample. Previous observations had shown that the Viamão population was morphologically

indistinguishable from the nominate subspecies, *Biomphalaria tenagophila tenagophila*, since they agree in shell and anatomic characters with topotypic specimens of *B. tenagophila* (see Paraense, 1981, Fig. 4).

Smaller samples from the following localities were also dealt with in this study: *B. tenagophila tenagophila* from Tramandaí, state of Rio Grande do Sul, and from Arroyo Juan Santos, in Paysandú, Uruguay; and *B. tenagophila guaibensis* from Porto Alegre, Tapes, Pelotas, Capão do Leão, Rio Grande, Arroio Grande and Jaguarão, in the state of Rio Grande do Sul; Arroyo Salsipuedes Grande, between the Departments of Río Negro and Tacuarembó, and Cañada del Sauce, left affluent of Arroyo Dacá, near Mercedes, Department of Soriano, Uruguay, both sent for identification by Dr. José Olazarri. Each of these samples consisted of 5 to 10 dissected specimens (Table I).

All the mentioned localities are shown in Fig. 5.

TABLE I

Ratios between penial sheath and prepuce, and between uterine complex and penial complex, in *Biomphalaria tenagophila tenagophila* and *B. tenagophila guaibensis*.

Subspecies	Locality	No. specimens	Penial sheath : prepuce		Uterine complex : penial complex	
			Range	Mean \pm S.D.	Range	Mean \pm S.D.
<i>B. t. tenagophila</i>	Viamão	50	0.68-1.35	0.92 0.11	1.31-2.16	1.65 0.14
	Tramandaí	10	0.80-1.20	1.01 0.14	1.29-2.14	1.66 0.28
	Paysandú	10	0.65-1.07	0.82 0.13	1.23-1.81	1.42 0.23
<i>B. t. guaibensis</i>	Guaíba	50	0.37-0.72	0.51 0.06	0.84-1.55	1.06 0.15
	Porto Alegre	10	0.43-0.67	0.53 0.07	0.85-1.65	1.21 0.24
	Tapes	5	0.46-0.60	0.52 0.06	0.97-1.61	1.32 0.28
	Pelotas	10	0.42-0.54	0.46 0.04	1.09-1.29	1.20 0.07
	Capão do Leão	7	0.43-0.51	0.46 0.04	1.15-1.56	1.34 0.16
	Rio Grande	1	0.48	-	1.22	-
	Arroio Grande	10	0.40-0.51	0.46 0.04	1.15-1.36	1.27 0.08
	Jaguarão	5	0.43-0.53	0.49 0.04	1.09-1.25	1.16 0.06
	Arroyo Salsipuedes Grande	5	0.61-0.74	0.65 0.05	0.92-1.02	0.98 0.04
	Mercedes	10	0.36-0.65	0.43 0.08	0.98-1.18	1.07 0.06

For comparison, 50 specimens of *Biomphalaria occidentalis* from Campo Grande, state of Mato Grosso do Sul, measuring from 8 to 14 mm in shell diameter (12 mm in 23 specimens), were also observed.

Mensurations of the organs concerned were made on camera lucida drawings. The prostatic diverticula were counted by crosscutting with a needle the prostatic duct between contiguous stalks; this procedure, which destroys the prostatic gland, is the surest one to single out the interwoven diverticula with very close stalks.

The statistics were subjected to the *t* test, and subspecific distinctness was determined by the coefficient of difference - CD (Mayr, Linsley & Usinger, 1953).

RESULTS

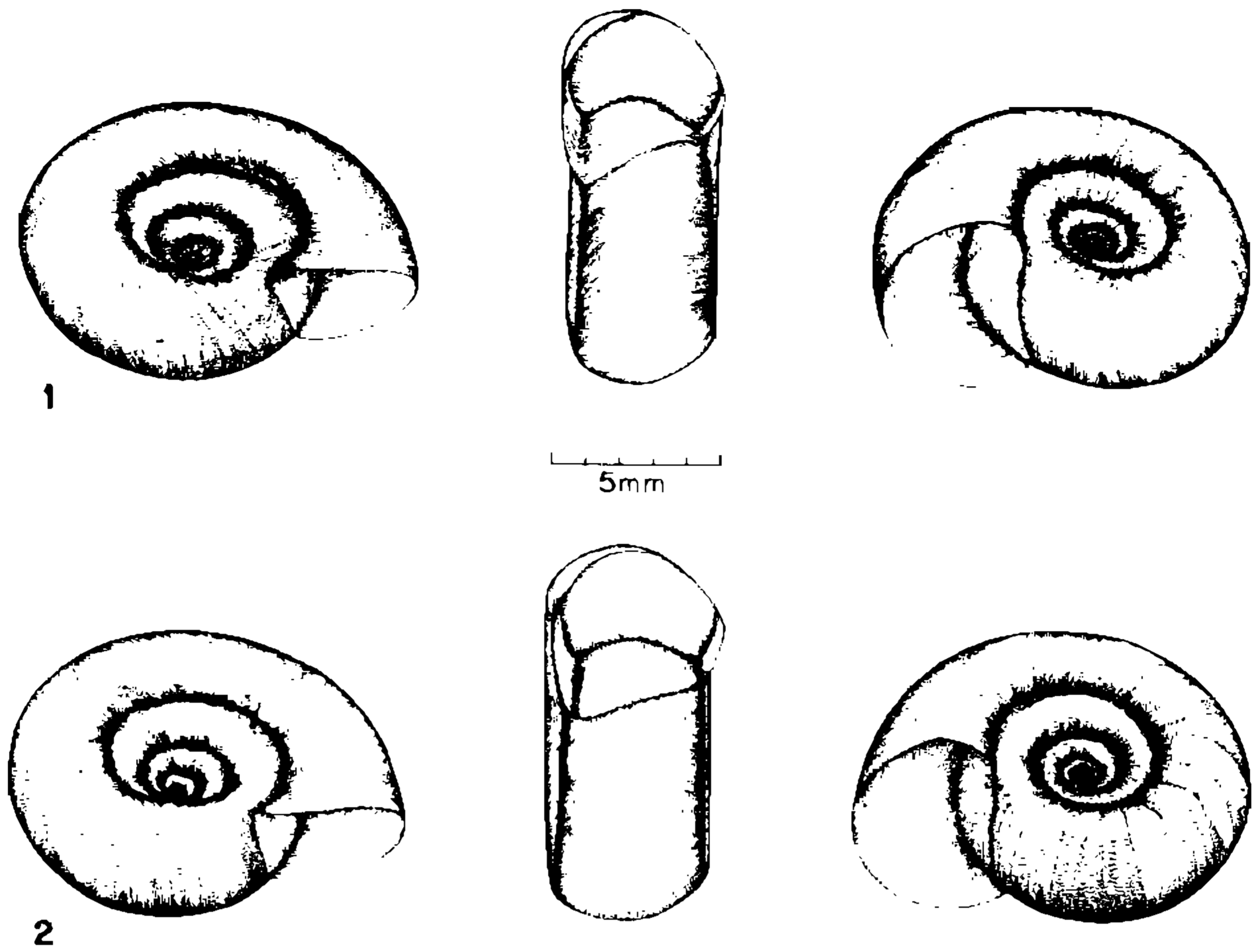
The largest shell collected at Guaíba measures 18.5 mm in diameter by 7.5 mm in width and has 6 whorls. The shell of the Guaíba specimens (Fig. 2) is indistinguishable from that of *B. tenagophila* as described by Paraense & Deslandes (1955). As to the Viamão specimens (Fig. 1), the largest of which measures 21 mm in diameter by 6.5 mm in width and has 6.5 whorls, they do not differ from the Guaíba ones.

With regard to the anatomic features, no differences were found in the organs of the hermaphroditic tract (ovotestis, ovispermiduct and seminal vesicle) and in the female organs (oviduct, nidamental gland, uterus, spermatheca and vagina).

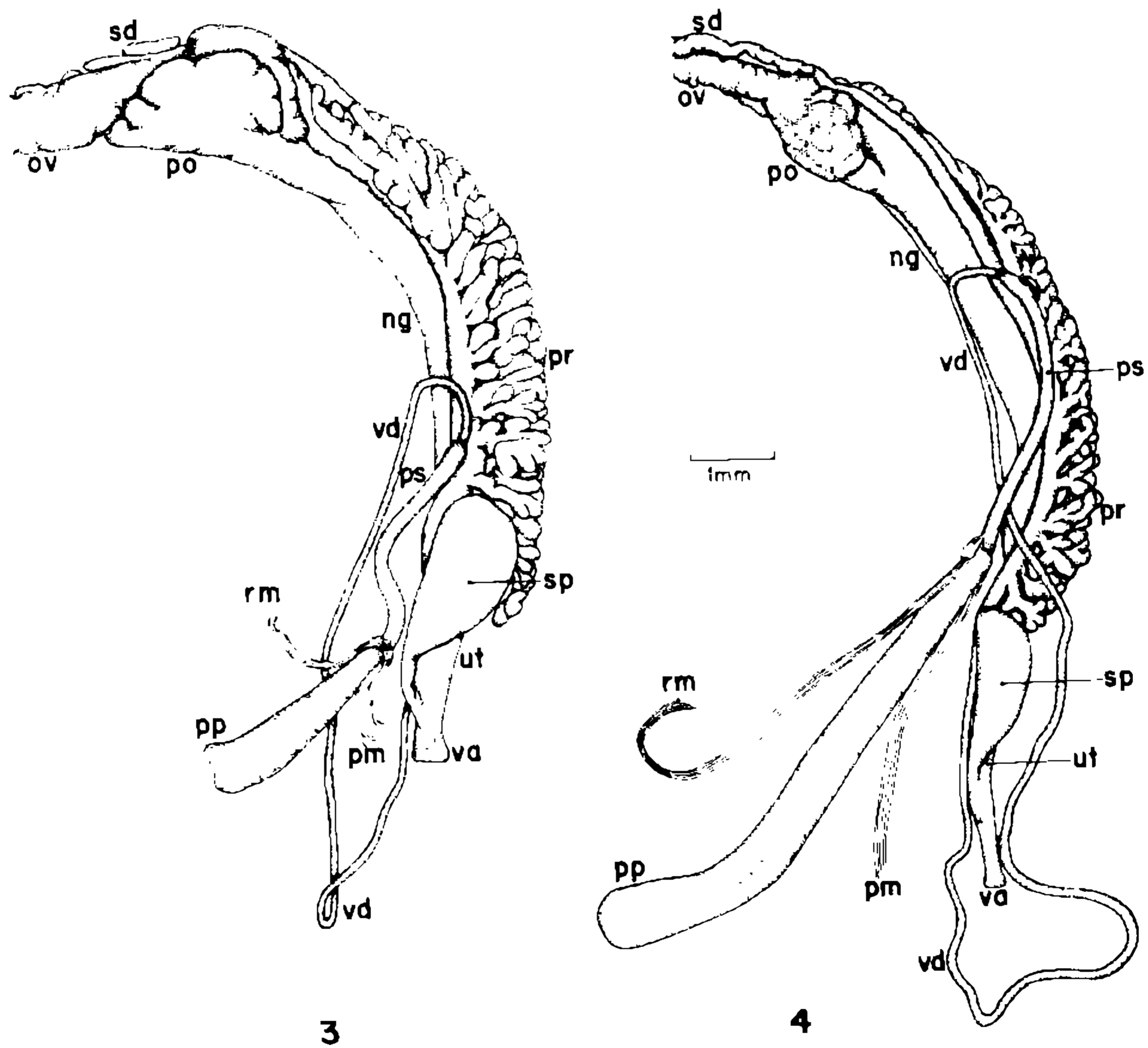
B. tenagophila guaibensis (Fig. 4) is characterized by its penial sheath being shorter than the prepuce, the ratio between the lengths of the two organs (ps: pp) ranging from 0.37 to 0.72 (mean 0.51 ± 0.06) in the 50 dissected specimens from Guaíba. In *B. tenagophila tenagophila* from Viamão (Fig. 3) that ratio ranged from 0.68 to 1.35 (mean 0.92 ± 0.11). The lowest ratio (0.68) was observed in only one specimen, the next lower having been 0.79 (1) and 0.80 (1). So the difference between the two means is highly significant ($p < 0.001$).

Comparing Figs. 3 and 5, it can be seen that, for specimens of the same size, the penial complexes look quite different, with shorter and proportionally stouter prepuce in *B. t. tenagophila*, longer and proportionally slenderer in *B. t. guaibensis*, their respective retractor and protractor muscles partaking of such features.

Another difference is in the ratio between the lengths of the uterine complex (uc, meaning the portion of the female duct from the cephalic border of the pouch of the oviduct to the vaginal opening) and of the penial complex (pc, penial sheath plus prepuce). That ratio (uc: pc) ranged from 0.84 to 1.55 (mean 1.06 ± 0.15) in *B. t. guaibensis*, and from 1.31 to 2.16 (mean 1.65 ± 0.14) in *B. t. tenagophila*. Again in this case the difference between the two means is highly significant ($p < 0.001$). Figs. 3 and 4 show that



Biomphalaria tenagophila – Fig. 1: shell of *Biomphalaria tenagophila tenagophila* from Viamão, 12 mm in diameter. Fig. 2: shell of *Biomphalaria tenagophila guaibensis* ssp. n. from Guaíba, 12 mm in diameter.



Biomphalaria tenagophila – Fig. 3: genital organs of *Biomphalaria tenagophila tenagophila* from Viamão, 12 mm in shell diameter. Fig. 4: genital organs of *Biomphalaria tenagophila guaibensis* ssp. n. from Guaíba, 12 mm in shell diameter. ng = nidamental gland, ov = oviduct, pm = protractor muscle of penial complex, po = pouch of oviduct, pp = prepuce, pr = prostate, ps = penial sheath, rm = retractor muscle of penial complex, sd = spermiduct, sp = spermatheca, ut = uterus, va = vagina, vd = vas deferens.

the penial complex tends to be as long as the uterine complex in *B. t. guaibensis*, and shorter than the latter in *B. t. tenagophila*. Actually, in the 50 examined specimens of each sample, the penial complex was as long as or longer than the uterine complex in 34 *B. t. guaibensis*, and shorter in all *B. t. tenagophila*.

In *B. occidentalis* the ps: pp ratio ranged from 0.52 to 0.75 (mean 0.64 ± 0.06), and uc: pc from 1.11 to 2.52 (mean 1.72 ± 0.36). It showed highly significant difference ($p < 0.001$) in both ratios as compared with *B. t. guaibensis*, and no significant difference ($p > 0.1$) as compared with *B. t. tenagophila*.

The numbers of prostatic diverticula varied from 9 to 24 (mean 15.48 ± 3.13) in *B. t. guaibensis*, and from 11 to 21 (mean 16.54 ± 2.46) in *B. t. tenagophila*, showing no significant difference ($p > 0.05$).

The CD values for *B. t. guaibensis* and *B. t. tenagophila* were 2.44 for ps: pp and 2.02 for uc: pc.

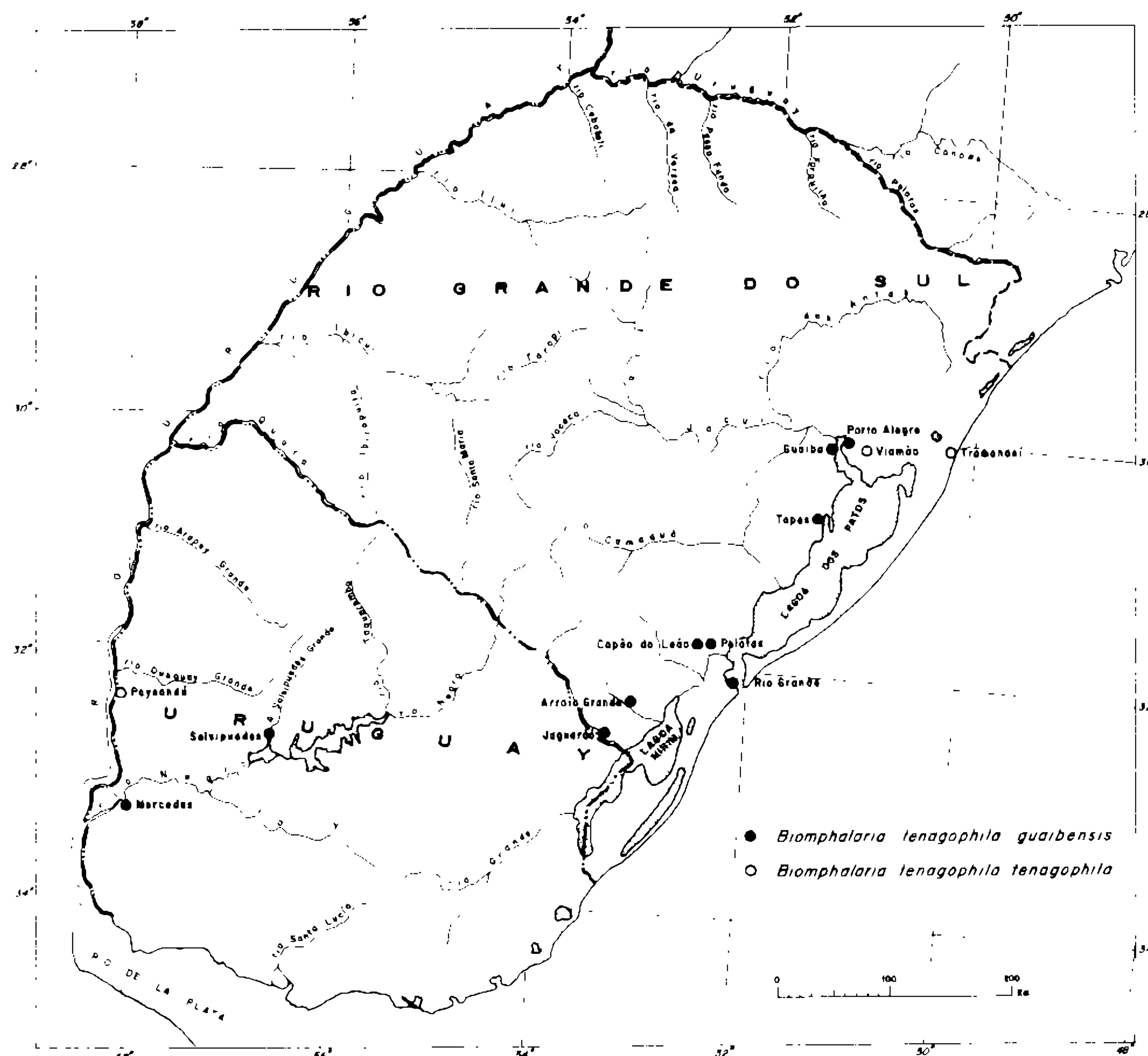


Fig. 5: distribution of populations of *Biomphalaria tenagophila guaibensis* and *B. tenagophila tenagophila* examined in this study.

DISCUSSION

In the area represented in Fig. 5, searched several times by the author, *B. t. tenagophila* was found so far just at Viamão, Tramandaí and Paysandú, seeming to be less frequent than *B. t. guaibensis*. The dominant species in that area is *Biomphalaria peregrina* (Orbigny, 1835), the distribution of which was studied by Paraense (1966), and *B. t. guaibensis* seems not to extend far beyond the represented belt. Such spatial limitation satisfies the definition of a subspecies as a geographically defined aggregate of local populations which differs taxonomically from other such subdivisions of a species (Mayr, Linsley & Usinger, 1953). Besides being statistically different, as mentioned above, the two subspecies are also taxonomically different, as they show coefficients of difference much higher than 1.5, considered sufficient by most exigent authors to justify subspecies recognition. The CDs in our case indicate that about 98% of the individuals of Guaíba are expected to be different from about 98% of the individuals of Viamão.

A species that shows a high degree of similarity to *B. tenagophila* is *B. occidentalis*. Their shells are indistinguishable and they only can be separated by the characteristics of some genital organs, besides being reproductively isolated and occupying separate ranges (Paraense, 1981). *B. occidentalis* is highly significantly different statistically from *B. t. guaibensis* as regards the ps: pp and uc: pc ratios, and shows no such difference with respect to *B. t. tenagophila*. However, it differs from either *B. tenagophila* subspecies in the absence of a qualitative character, the vaginal pouch, which is present in the latter.

So far three subspecies of *B. tenagophila* were proposed, all of them on the basis of shell characters: *Planorbis tenagophilus orbignyana* Beck, 1837, from Argentina; *P. tenagophilus chemnitziana* Beck, 1837, from Bolivia; and *Australorbis bahiensis megas* Pilsbry, 1951, from Petrópolis, state of Rio de Janeiro, Brazil.

P. tenagophilus orbignyana and *P. t. chemnitziana* are catalog names given to shells deposited in the Museum of Copenhagen, not accompanied by figures or descriptions. The description and figures of *A. bahiensis megas* leave no doubt about its identity with *B. tenagophila*, the dominant species in the area. In those three instances the old-style proposal of new subspecies followed a purely typological concept.

For a clear distinction between *B. t. guaibensis* and *B. t. tenagophila* an essential condition is that the animal body is preserved for dissection in a well-relaxed condition.

The characterization of *B. t. guaibensis* and *B. t. tenagophila* as subspecies was also based on cross-breeding experiments, which will be reported in a subsequent paper.

RESUMO

É descrita uma nova subespécie de molusco planorbídeo, *Biomphalaria tenagophila guaibensis*, que tem sido encontrada ao longo da faixa costeira do Estado do Rio Grande do Sul e da parte média do Uruguai, de Porto Alegre a Mercedes. Distingue-se da espécie nominativa, *Biomphalaria tenagophila tenagophila*, pelo aspecto do complexo peniano (prepúcio mais longo e proporcionalmente mais delgado em *B. t. guaibensis*, mais curto e proporcionalmente mais grosso em *B. t. tenagophila*), pela razão entre os comprimentos da bainha do pênis e do prepúcio, pela razão entre os comprimentos do complexo uterino e do complexo peniano, e por um coeficiente de diferença de 2.44 para a razão entre a bainha do pênis e o prepúcio e de 2.02 para a razão entre o complexo uterino e o complexo peniano. A concha e os outros órgãos do sistema genital são semelhantes nas duas subespécies.

A *B. t. guaibensis* tem grande semelhança com a *Biomphalaria occidentalis* Paraense, 1981, mas dela se distingue facilmente pela presença de uma bolsa vaginal, que falta na *B. occidentalis*, além de apresentar diferença altamente significativa nas razões bainha do pênis : prepúcio e complexo uterino : complexo peniano.

Experiências de cruzamento que trazem apoio adicional ao reconhecimento da *B. t. guaibensis* como subespécie serão relatadas em outra publicação.

ACKNOWLEDGEMENTS

To Dr. Antonio A. Pereira da Silva, Regional Director of SUCAM (Superintendência de Campanhas de Saúde Pública, Ministry of Health), Rio Grande do Sul, for samples of snails from Tapes, Capão do Leão, Rio Grande, Arroio Grande and Jaguarão; to Lic. José Olazarri, for the samples of *B. t. guaibensis* from Uruguay; and to Mr. José Eduardo Prado, artist of this Department, for the illustrations.

REFERENCES

- BECK, H., 1837. Index molluscorum praesentis aevi Musei Principis Augustissimi Christiani Frederici. Fasciculus secundus. Mollusca Gastraeopoda Pulmonata. Hafniae.
- MAYR, E.; LINSLEY, E.G. & USINGER, R.L., 1953. Methods and principles of systematic zoology. McGraw Hill, New York. ix + 328p.
- ORBIGNY, A., 1835. Synopsis terrestrium et fluviatilium molluscorum, in suo per Americam meridionalem itinere collectorum. *Mag. Zool.*, 5, Classe 5 (61-62) :1-44.
- ORBIGNY, A., 1837. Voyage dans l'Amérique méridionale, vol. 5, 3^e partie: Mollusques. P. Bertrand, Paris.
- PARAENSE, W.L., 1961a. The nomenclature of Brazilian planorbids. I. *Australorbis glabratus* (Say, 1818). *Rev. Brasil. Biol.*, 21 (3) :287-296.
- PARAENSE, W.L., 1961b. The nomenclature of Brazilian planorbids. II. *Australorbis tenagophilus* (Orbigny, 1835). *Rev. Brasil. Biol.*, 21 (4) :343-349.
- PARAENSE, W.L., 1966. The synonymy and distribution of *Biomphalaria peregrina* in the Neotropical region. *Rev. Brasil. Biol.*, 26 (3) :269-296.
- PARAENSE, W.L., 1981. *Biomphalaria occidentalis* sp.n. from South America (Mollusca Basommatophora Pulmonata). *Mem. Inst. Oswaldo Cruz*, 76 (2) :199-211.
- PARAENSE, W.L. & DESLANDES, N., 1955. Observations on the morphology of *Australorbis nigricans*. *Mem. Inst. Oswaldo Cruz*, 53 (1) :121-134.
- PARAENSE, W.L. & DESLANDES, N., 1956. *Australorbis nigricans* as the transmitter of schistosomiasis in Santos, State of São Paulo. *Rev. Brasil. Malariol. Doen. Trop.*, 8 (1) :235-245.
- PILSBRY, H.A., 1951. Notes on some Brazilian Planorbidae. *Nautilus*, 65 (1) :3-6. Figures in 64 (4) Pl. 9.