The Chromosomes of *Biomphalaria tenagophila* (Gastropoda—Basommatophora—Planorbidae)

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The subfamilies Bulininae and Biomphalariinae are of medical importance being potential intermediate hosts of human and herbivore schistosomiasis. South América is inhabited by the subfamilie Biomphalariinae.

Biomphalaria glabrata (= *Australorbis glabratus*) is the principal host of *Schistosoma mansoni* in Brazil. In Argentina there are no cases of schistosomiasis as yet although *Biomphalaria tenagophila*, of extensive distribution in this country, has proved to be also host (Rey 1956).

Previously eight papers were devoted to the cytogenetics of the subfamily Biomphalariinae (Rangel 1951, Fraga and Goncalves 1956, Burch 1960 a, b, 1965, Natarajan *et al.* 1965, Narang 1974, Raghunathan 1976) but, excepting in the papers of Narang (1974) and Raghunathan (1976), karyotypes have not been shown.

The aim of this research is to report the study of the meiotic and mitotic chromosomes of three populations of *Biomphalaria tenagophila*.

Materials and methods

Twenty nine specimens of *Biomphalaria tenagophila* (Spix 1827) were analyzed. The animals studied were colleted from the Botanical Garden of Buenos Aires and San Miguel de Tucumán in Argentina, and from Carrasco in Uruguay. Identification was made on the basis of the shell characteristics according to the description and figures presented by Paranse and Deslandes (1955).

Chromosome preparations were made from ovotestis of adult animals and total body of young animals. The method applied was as follows: the material was placed in distilled water for hypotonic treatment for 20-30 min. Then, it was immediately fixed in three changes of fresh Carnoy (3:1) of 45' each. The cells were spread on Carnoy moisted slides and they were air dried at room temperature. 4% Giemsa Gurr R66 in phoshate buffer at pH 6.8 during 10 min was used as stain.

Results

The chromosomes of the fresh water snail *Biomphalaria tenagophila* were studied in three populations. The somatic complement is $2n=36$ (Fig. 1 a, b). The fundamental number of the species is $FN=68$. The karyotypes of this

snail in Argentina do not differ consistently by different populations. The karyogram shown in Fig. 1a is representative of the populations from Buenos Aires and San Miguel de Tucumán, and it consists of 16 pairs of meta- and submetacentric elements and 2 pairs (3, 16) of acrocentric chromosomes. Eight pairs (1, 5, 8, 10-12, 15, 17) are metacentric and eight pairs (2, 4, 6, 7, 9, 13, 14, 18) are submetacentric. In the karyotype of the population from Carrasco, Uruguay, a distal secondary constriction can be observed in the first pair (Fig. 1 b).

Table 1. The chromosome numbers of the subfamily biomphaliariinae (Mollusca-Gastropoda-Euthyneura-Basommatophora-Planorbidae-Biomphaliariinae)

Species	2n	n	Source	References
<i>Biomphalaria alexandrina</i>		18	Egypt	Natarajan <i>et al.</i> 1965
<i>Biomphalaria choanomphala</i>		18	Tanzania	Burch 1965
<i>Biomphalaria glabrata</i>	36	18	Brasil	Rangal 1951, Fraga and Goncalves 1956, Burch 1960, b
			Puerto Rico	Narang 1974. Raghunathan 1976.
<i>Biomphalaria olivaceus</i>	18		?	Burch 1960 a
<i>Biomphalaria pfeifferi</i>	18		Tanganyika	Natarajan <i>et al.</i> 1965
			Rhodesia	
<i>Biomphalaria pfeifferi gaudi</i>	18		Liberia	Natarajan <i>et al.</i> 1965
			Ghana	
<i>Bimopharia pfeifferi</i>	18		Madagascar	Natarajan <i>et al.</i> 1965
<i>madagascariensis</i>				
<i>Biomphalaria sudanica sudanica</i>	18		Sudan	Burch 1960 b
<i>Biomphalaria sudanica</i>	18		Tanganyika	Natarajan <i>et al.</i> 1965
<i>tanganyicensis</i>				
<i>Biomphalaria tenagophila</i>	36	18	Argentina	Giacomozzi, Riva and Vidal
			Uruguay	(This paper)

The sequential stages of meiosis were studied in all adult animals observed. A pachytene nucleus as illustrated in Fig. 2a showed 18 bivalents. Initial to medial diplotene stages were not present. Final diplotene features were scarce. The paired chromosomes during diakinesis were held together by one or more chiasmata (Fig. 2b). The bivalents appear at metaphase I well condensed (Fig. 2 c). Eighteen typical dyads were seen at metaphase II. At this stage all chromosomes corresponding to the somatic chromosomes could be identified (Fig. 2 d). Through the meiotic and mitotic stages, there was no morphological differentiation of chromosomes that could be interpreted as the sex chromosomes.

Figs. 1 a-b. a, karyogram of *Biomphalaria tenagophila* from Buenos Aires, Argentina. b, karyogram of *B. tenagophila* from Carrasco, Uruguay, the pair N° 1 exhibits satellites. (Scale, 5 micra). Figs. 2 a-d. Meiotic features of the chromosomes of *B. tenagophila*. a, pachytene. b, diakinesis, persistent nucleoli associated with bivalents and separated from them as indicated by arrows. c, early metaphase I. d, metaphase II. (Scale, 5 micra).

Discussion

Cytology of seven African and two American species and subspecies of *Biomphalaria* have been studied previously (Table 1). As in other *Biomphalarinae* snails the haploid number of *Biomphalaria tenagophila* is $n=18$. Karyotypes have been shown only for *B. glabrata* (Narang 1974, Raghunathan 1976). In snails from Brazil, Narang (1974) found that the chromosome complement of *B. glabrata* is composed of 11 pairs of metacentric, 6 pairs of submetacentric and one pair of acrocentric chromosomes. Nevertheless, this author showed in the figure 1 of his paper one pair of elements (pair 11) included in the 'metacentrics' group that, in our view has characteristics of acrocentric chromosome, being probably similar to our small pair of acrocentrics. If this is accepted, the karyotypes of *B. glabrata* from Brazil and *B. tenagophila* are karyotypically similar.

Raghunathan (1976) working on *B. glabrata* from Puerto Rico, described that the karyotype consisted of 10 pairs of metacentric, 4 pairs of submetacentric, 2 pairs of telocentric and 2 pairs of acrocentric chromosomes. This author also pointed out that the pair 8 had a secondary constriction and pair 9 carried satellites ("the nucleolus organizer"). The text and figures of this work need several comments: 1) the pairs 14 and 15 (see figure 3 of Raghunathan's paper) are considered as acrocentric, while in our view they are submetacentric chromosomes, 2) the pairs 8 and 9 designated as telocentric, are probably acrocentrics because it is difficult to accept the chromosomes with satellites as telocentrics, and 3) the secondary constriction of pair 8 is not apparent in the pointed figure.

In the papers by Narang (1974) and Raghunathan (1976), the information on meiosis was not available at all.

In our samples, no satellites were observable in the pair of small acrocentrics, neither in somatic metaphase nor in metaphase II. The presence of the secondary constriction in pair 1 of the sample from Carrasco could be explained as a population or racial variation of the chromosomal phenotype.

It would be necessary to investigate the possible existence of polyploids species in the genus *Biomphalaria*, in the light of Burch's hypothesis (Burch 1960 b) that there may exist a possible connection between polyploidy and susceptibility to infection with schistosomes.

Summary

The study of mitotic and meiotic chromosomes of the fresh water snail *Biomphalaria tenagophila* is reported. Three populations, two from Argentina (Buenos Aires and San Miguel de Tucumán) and one from Uruguay (Carrasco), were investigated. The haploid number of the species is $n=18$ and the diploid number is $2n=36$. The fundamental number is $FN=68$.

The karyotype is composed of 8 pairs of metacentric, 8 pairs of submetacentric and 2 pairs of acrocentric chromosomes. Secondary constrictions were apparent in the first pair of the Carrasco's population.

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