



New record of the invasive snail *Melanoides tuberculata* (Gastropoda, Thiaridae) - Ceará State, Brazil

M. R. F. Barros^{a*} , R. A. Chagas^a , M. Herrmann^a and A. M. Bezerra^b

^aPrograma de Pós-graduação em Aquicultura e Recursos Aquáticos Tropicais, Instituto Socioambiental e dos Recursos Hídricos – ISARH, Universidade Federal Rural da Amazônia – UFRA, Av. Presidente Tancredo Neves, 2501, Montese, CP 917, CEP 66077-530, Belém, PA, Brasil

^bPrograma de Pós-graduação em Saúde e Produção Animal na Amazônia, Universidade Federal Rural da Amazônia – UFRA, Av. Presidente Tancredo Neves, 2501, Montese, CP 917, CEP 66077-530, Belém, PA, Brasil

*e-mail: eng.p.marabarro@gmail.com

Received: August 3, 2018 – Accepted: January 11, 2019 – Distributed: May 31, 2020
(With 3 figures)

Abstract

Melanoides tuberculata is a freshwater gastropod native to Africa and Asia, and currently presents distributed worldwide. The first record of *Melanoides tuberculata* in South America occurred in Brazil, in the city of Santos, state of São Paulo, in 1967. Therefore, the objective of the present study is to report the occurrence of *Melanoides tuberculata* for Banana Lagoon and to characterize the current population situation. The gastropods were collected through direct visualization of shells, morphometric measurements of *Melanoides tuberculata* shells were determined using a digital caliper. After identifying the gastropod and its first distribution in the area, a preliminary descriptive analysis of the population was carried out. The gastropods were classified by grouping the individuals into four classes according to the shell width (mm). This is the first record of the occurrence of *Melanoides tuberculata*, in Banana Lagoon, Caucaia municipality, Ceará state, Northeast Brazil, being the largest specimen cited in the literature (33.77 mm) and 92.65% of the gastropods present in sizes above the first reproduction stages.

Keywords: molluscs, gastropod, invasive species, distribution, freshwater.

Novo registro do gastrópode invasor *Melanoides tuberculata* (Gastropoda, Thiaridae) – Estado do Ceará, Brasil

Resumo

Melanoides tuberculata é um gastrópode de água doce nativo da África e da Ásia e atualmente apresenta-se distribuído mundialmente. O primeiro registro de *Melanoides tuberculata* na América do Sul ocorreu no Brasil, na cidade de Santos, estado de São Paulo, em 1967. Portanto, o objetivo do presente estudo é relatar a ocorrência de *Melanoides tuberculata* para Lagoa do Banana, caracterizando a situação atual da população no local. Os gastrópodes foram coletados através da visualização direta de conchas, e as medidas morfométricas das conchas de *Melanoides tuberculata* foram determinadas por meio de um paquímetro digital. Após identificar o gastrópode e constando sua primeira distribuição na área, realizou-se uma análise preliminar descritiva da população. Os gastrópodes foram classificados agrupando os indivíduos em quatro classes de acordo com a largura da concha (mm). Este é o primeiro registro da ocorrência de *Melanoides tuberculata*, na Lagoa da Banana, município de Caucaia, estado do Ceará, Nordeste do Brasil, sendo o maior exemplar citado na literatura (33,77 mm) já registrado e 92,65% dos gastrópodes apresentam-se em tamanhos acima do primeiro estágios de reprodução.

Palavras-chave: molusco, gastrópode, espécie invasora, distribuição, água doce.

1. Introduction

Melanoides tuberculata (OF Müller, 1774) (Gastropoda: Thiaridae) (WoRMS, 2018), is a freshwater gastropod, popularly known as “caramujo trombeta” and “snail” (Santos et al., 2016). It is native to Northeastern Africa and Southeast Asia (Vaz et al., 1986), and is currently distributed worldwide (Facon et al., 2003) favored mainly by its high

adaptation to new environments. The species is adapted to high salinity variation (Santos and Eskinazi-Sant’Anna, 2010; Silva and Barros, 2015), and high temperatures (Gerald and Spezzano Junior, 2005; Elkarmi and Ismail, 2007). *M. tuberculata* is generally associated with habitats with high vegetation (Medeiros and Henry-Silva, 2017) and

anthropogenic alterations, which allow shaded areas for their protection (Rocha-Miranda and Martins-Silva, 2006). Its diet consists preferably of fine debris and periphytic biofilms (Raw et al., 2016). It reaches maximum shell length of 31 mm (Gonçalves, 2015), initiating its sexual maturity above 7 mm of shell height (Heller and Farstay, 1989). Dioecious and ovoviviparous animals, they can be found in aggregations up to 842 individuals from early stage to juveniles in a single litter and adults breed all year long (Gonçalves, 2015).

The first record of *Melanoides tuberculata* in South America occurred in Brazil, in the city of Santos, state of São Paulo, in 1967 (Vaz et al., 1986). Its recent distribution in the Brazilian territory is wide, with records in 19 states and in the Federal District (Santos et al., 2016). The main routes of introduction/dispersal of *M. tuberculata* occur through trade in plants and animals (Vaz et al., 1986). However, recently, in a brief review, Coelho et al. (2018) cite other causes of dispersal: fish stocks (Coelho et al., 2017), aquaculture (Duggan, 2010; Assis et al., 2014), water diversions where gastropod is present (Azevêdo et al., 2014), or of ship ballast and transport of sand and other porous wet materials may contribute to the dispersion of this species.

A high abundance of *M. tuberculata* directly affects the diversity and balance of the native benthic community, mainly the populations of *Biomphalaria* spp. because of competition for food and density (m^2) (Pointier, 1993; Guimarães et al., 2001; Nascimento Filho et al., 2014). The gastropod can also serve as an occasional host for *Schistosoma mansoni* (endemic in some areas in Brazil) and

several others Platyhelminthes (Vaz et al., 1986; Mitchell et al., 2005; Pinto and Melo, 2010; Paula-Andrade et al., 2012; Ximenes et al., 2017). According to Bezerra (2007) and Lima et al. (2013), the dispersal of *Melanoides tuberculata* in the Brazilian Northeast was facilitated mainly by the courses of artificial waters between rivers and basins, jeopardize the native fauna. With this, the author points out, the importance of monitoring the spatial distribution of this invasive snail. Therefore, the objective of the present study is to report the occurrence of *Melanoides tuberculata* for Banana Lagoon and to characterize the current population situation.

2. Material and Methods

2.1. Study site

The gastropods were collected near the margin of Banana lagoon ($3^{\circ}37'31.44''S$ and $38^{\circ}45'36.18''W$), municipality of Caucaia, the State of Ceará, Northeast Brazil (Figure 1). Surrounding this freshwater lagoon, a riparian vegetation predominates, however, being one of the main tourist attractions of the state, several residences and leisure and entertainment establishments coexist.

2.2. Methods

After the visualization of the gastropods in the pond, which occurred in February 2016, the specimens were collected manually, fixing them in 70% ethanol and deposited at the Zoology Museum "Professor Carlos Alberto Moreira de Melo" under deposit number MZUFRA Moll 081 from the Universidade Federal Rural da Amazônia.

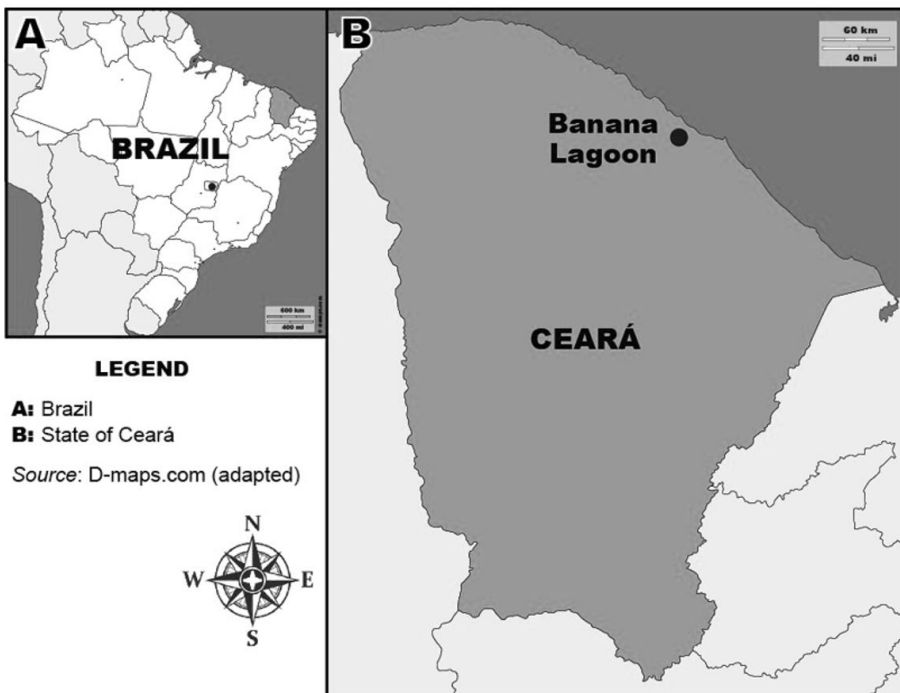


Figure 1. Localization of the Banana lagoon at Ceará state, Northeastern Brazil.

2.3. Morphometry

Morphometric measurements of *M. tuberculata* shells were determined using a digital caliper (Tesa - DATAdirect, with accuracy of 0.01 mm). To perform a preliminary descriptive analysis of the population, the gastropods were classified according to Miyahira (2010), grouping the individuals into four classes according to shell width (mm): it is considered: class 1: non-reproductive (0.01 to 2.99 mm); class 2: beginning of the reproduction stage (3 to 5.99 mm); class 3: full reproduction stage (6 to 8.99 mm) and class 4: reproduced more than once (> 9 mm). A total of 381 individuals of *M. tuberculata* (Figure 2) were collected along the banks of Banana Lagoon, a touristic region with presence of riparian, vegetation providing, that for providing shading and foliar material that can favor the establishment of *M. tuberculata* (Souto et al., 2011).



Figure 2. *Melanoides tuberculata* collected at Banana Lagoon, Ceará State, Brazil, on February, 2016. Scale: 5 mm.

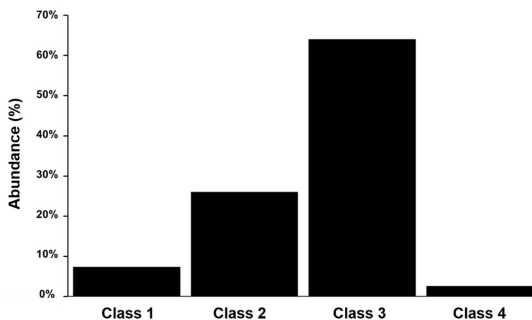


Figure 3. Classes of length of *Melanoides tuberculata* according to their reproductive stage: class 1: non-reproductive (0.01 to 2.99 mm); class 2: beginning of the reproduction stage (3 to 5.99 mm); class 3: full reproduction stage (6 to 8.99 mm); and class 4: reproduced more than once (> 9 mm).

3. Results

The raw morphometric data of *M. tuberculata* are available on the digital platform *Data Publisher for Earth & Environmental Science* - PANGAEA (Chagas et al., 2018). The total length of the shell varied from 1.15 to 33.77 mm, with an average of 19.82 ± 6.25 mm ($\pm$ SD), width between 0.65 and 9.35 mm (6.17 ± 1.80 mm) and height between 0.73 and 8.88 mm (5.76 ± 1.66 mm). We also registered, in the sampled area, the largest length for *M. tuberculata* (33.77 mm) already observed, which registered a length of 33.00 mm in Rio de Janeiro – RJ (Bogéa et al., 2005).

Due to the fragmentation of the protoconch, a common fact in adults due to its life habit (Miyahira, 2010), the description of the population of the gastropods found relied on the width measurement to estimate the reproductive stages of the molluscs (Miyahira, 2010). We verified that 64.05% of the gastropods were in full reproductive stage (class 3), 25.98% started their reproductive cycle (class 2), 7.35% still did not reach the reproductive stage (class 1), and 2.62% were in their second reproductive stage (class 4). Through the description of the reproductive stages of the morphometry of *M. tuberculata* shell (Figure 3), we can consider that the species is established, since 90% of the individuals present at Banana Lagoon were able to reproduce, or already reproduced at least once.

4. Discussion

In the Banana lagoon, the presence of *Biomphalaria* sp. was observed in a reduced number. Several authors mention that the introduction of *M. tuberculata* into Brazilian territory occurred since the species was used at the biological control of *Biomphalaria* (Leão et al., 2011) and through fishkeeping due to the transport of aquatic plants (Vaz et al., 1986). Thus, it is inferred that the presence of *M. tuberculata* decimated, almost in total, the population of the native gastropod.

Little is known about the route of introduction of *M. tuberculata* in the state of Ceará. Coelho et al. (2018), in its brief review, presents the current records of *M. tuberculata* in the state, including for the municipality of Caucaia, however, in a region different from that mentioned in this study, located at the border with the capital Fortaleza. The dispersion of the gastropod to Banana Lake (Caucaia) that may have occurred through the construction of the Integration Channel downstream of the Açude Castanhão dam, which began in 2004 (Bezerra, 2007).

The introduction of exotic species generates large negative impacts on the environment, since it becomes the species with the highest number of records, when established (Souto et al., 2011; Paula et al., 2017). However, its great density can be controlled by aquatic birds, not being necessary to introduce new species for the control, which would avoid greater environmental problems (Silva and Gomes, 2014).

Through the obtained results, it is considered that *Melanoides tuberculata* is established at Banana Lagoon, mainly because it presents individuals in all the reproductive

stages, which guarantees that the species presents complete life cycle in the region. However, there are still no studies on the impacts caused by the species on the native community, but it is possible to infer that local environmental conditions are favoring its development, which may explain the largest individual ever recorded in scientific literature. Several authors cite control measures of *M. tuberculata* dispersion. among the mitigating measures are: care when cleaning and discarding plants and organisms from the aquarium, mainly through the use of molluscicides. However, several studies cite the impossibility of controlling the molluscs, after consolidation in the environment, and advise the monitoring of dispersion as the best methodology for population control of the species.

In the foregoing, it is pointed out that the rapid dissemination and ease of establishment of the species in the environments are strong indications for such studies to be carried out, especially on issues related to the monitoring of species dispersal.

References

- ASSIS, D.A.S., CAVALCANTE, S.S. and BRITO, M.F.G., 2014. Aquarium trade as a potential disseminator of non-native invertebrates in Northeastern Brazil. *Neotropical Biology and Conservation*, vol. 9, no. 2, pp. 115-119.
- AZEVEDO, E.D.L., BARBOSA, J.E.D.L., VIDIGAL, T.H., CALLISTO, M. and MOLOZZI, J., 2014. First record of *Corbicula largillierii* (Philippi, 1844) in the Paraíba River Basin and potential implications from water diversion of the São Francisco River. *Biota Neotropica*, vol. 14, no. 4, pp. e20140036. <http://dx.doi.org/10.1590/1676-0603003614>.
- BEZERRA, F.S.M., 2007. Alterações na fauna malacológica límica do Estado do Ceará. In: *Anais do XX Encontro Brasileiro de Malacologia*, 2007, Rio de Janeiro. Rio de Janeiro: Instituto de Biologia Roberto Alcântara Gomes, pp. 404.
- BOGÉA, T., CORDEIRO, F.M. and GOUVEIA, J.S., 2005. *Melanoides tuberculatus* (Gastropoda: Thiaridae) as intermediate host of Heterophyidae (Trematoda: Digenea) in Rio de Janeiro metropolitan area, Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*, vol. 47, no. 2, pp. 87-90. <http://dx.doi.org/10.1590/S0036-46652005000200005>. PMID:15880219.
- CHAGAS, R.A., BARROS, M.R.F. and BEZERRA, A.M., 2018. Morphometry of the bioinvader gastropod *Melanoides tuberculata* (Müller, 1774), Banana Lagoon, Ceará, Northeastern Brazil. Bremerhaven: PANGAEA. <https://doi.org/10.1594/PANGAEA.884524>.
- COELHO, P.N., CESAR, D.A.S. and RUOCCO, A.M., 2017. Aquicultura como potencial dispersor de espécies não nativas em microbacias. In: V.A. RODRIGUES, H.E. SIQUEIRA, P.J.D. OLIVEIRA, L.Z. PINHEIRO and L.A. BUCCI, eds. *Biomias brasileiros: conservação da biodiversidade, solo, floresta e água*. Botucatu: FEPAP, pp. 607-609.
- COELHO, P.N., FERNANDEZ, M.A., CESAR, D.A.S., RUOCCO, A.M.C. and HENRY, R., 2018. Updated distribution and range expansion of the gastropod invader *Melanoides tuberculata* (Müller, 1774) in Brazilian waters. *BioInvasions Records*, vol. 7, no. 4, pp. 405-409. <http://dx.doi.org/10.3391/bir.2018.7.4.08>.
- DUGGAN, I.C., 2010. The freshwater aquarium trade as a vector for incidental invertebrate fauna. *Biological Invasions*, vol. 12, no. 11, pp. 3757-3770. <http://dx.doi.org/10.1007/s10530-010-9768-x>.
- ELKARMI, A.Z. and ISMAIL, N.S., 2007. Growth models and shell morphometrics of two populations of *Melanoides tuberculata* (Thiaridae) living in hot springs and freshwater pools. *Journal of Limnology*, vol. 66, no. 2, pp. 90-96. <http://dx.doi.org/10.4081/jlimnol.2007.90>.
- FACON, B., POINTIER, J.P., GLAUBRECHT, M., POUX, C., JARNE, P. and DAVID, P., 2003. A molecular phylogeography approach to biological invasions of the New World by parthenogenetic Thiarid snails. *Molecular Ecology*, vol. 12, no. 11, pp. 3027-3039. <http://dx.doi.org/10.1046/j.1365-294X.2003.01972.x>. PMID:14629383.
- GERALD, G.W. and SPEZZANO JUNIOR, L.C., 2005. The influence of chemical cues and conspecific density on the temperature selection of a freshwater snail (*Melanoides tuberculata*). *Journal of Thermal Biology*, vol. 30, no. 3, pp. 237-245. <http://dx.doi.org/10.1016/j.jtherbio.2004.12.002>.
- GONÇALVES, C.T.P., 2015. *Aspectos populacionais e reprodutivos do gastrópode invasor Melanoides tuberculata* (Mollusca: Thiaridae) no Lago Paranoá, Brasília, Brasil. Brasília: Universidade de Brasília, 108 p. Dissertação de Mestrado em Zoologia.
- GUIMARÃES, C.T., SOUZA, C.P. and SOARES, D.M., 2001. Possible competitive displacement of planorbids by *Melanoides tuberculata* in Minas Gerais, Brazil. *Memórias do Instituto Oswaldo Cruz*, vol. 96, suppl., pp. 173-176. <http://dx.doi.org/10.1590/S0074-02762001000900027>. PMID:11586446.
- HELLER, J. and FARSTAY, V., 1989. A field method to separate males and females of the freshwater snail *Melanoides tuberculata*. *The Journal of Molluscan Studies*, vol. 55, no. 3, pp. 427-429. <http://dx.doi.org/10.1093/mollus/55.3.427>.
- LEÃO, T.C.C., ALMEIDA, W.R., DECHOUM, M.S. and ZILLER, S.R., 2011. *Espécies exóticas invasoras no nordeste do Brasil*. Recife: Cepan, 99 p.
- LIMA, L.F.O., BRASIL, B.I.A.L. and MARTINS-SILVA, M.J., 2013. *Melanoides tuberculata* (Müller, 1774): northeastern dispersal in the São Francisco basin, Brazil. *Check List*, vol. 9, no. 1, pp. 162-164. <http://dx.doi.org/10.15560/9.1.162>.
- MEDEIROS, E.L. and HENRY-SILVA, G.G., 2017. Evaluation of the feeding preference between the aquatic macrophytes *Egeria densa* and *Chara indica* by the invasive mollusk *Melanoides tuberculata*. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 77, no. 2, pp. 234-239. <http://dx.doi.org/10.1590/1519-6984.08415>. PMID:28300942.
- MITCHELL, A.J., OVERSTREET, R.M., GOODWIN, A.E. and BRANDT, T.M., 2005. Spread of an exotic fish-gill trematode. *Fisheries*, vol. 30, no. 8, pp. 11-16. [http://dx.doi.org/10.1577/1548-8446\(2005\)30\[11:SOAEFT\]2.0.CO;2](http://dx.doi.org/10.1577/1548-8446(2005)30[11:SOAEFT]2.0.CO;2).
- MIYAHIRA, I.C., 2010. *Dinâmica populacional de Melanoides tuberculatus* (Müller, 1774) em um riacho impactado da Vila do Abraão, Ilha Grande, Angra dos Reis, RJ, Brasil. Rio de Janeiro: Universidade do Estado do Rio de Janeiro, 111 p. Dissertação de Mestrado em Ecologia e Evolução.
- NASCIMENTO FILHO, S.L., VIANA, G.F.S. and GOMES, R.L.M., 2014. Inventário da malacofauna límica de três grandes reservatórios do sertão de Pernambuco, Brasil. *Scientia Plena*, vol. 10, no. 11, pp. 1-7.

- PAULA, C.M., VAZ, A.A., VAZ, A.A., PELIZARI, G.P., ROBAYO, H.M.S., GARCIA, T.D., AVELINO, D., ZACARIN, G.G. and SMITH, W.S., 2017. Ocorrência de um molusco invasor (*Melanoides tuberculata*, Müller, 1774), em diferentes sistemas aquáticos da bacia hidrográfica do Rio Sorocaba, SP, Brasil. *Revista Ambiente & Água*, vol. 12, no. 5, pp. 829-841. <http://dx.doi.org/10.4136/ambi-agua.1971>.
- PAULA-ANDRADE, C., PINTO, H.A., COSCARELLI, D., VIDIGAL, T.H. and MELO, A.L., 2012. The natural infection of *Melanoides tuberculata* (Müller, 1774) (Mollusca: Gastropoda) by *Entrocestus formosanus* (Nishigori, 1924) (Platyhelminthes: Trematoda) in Paranoá lake, Brasília, Brazil. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 72, no. 2, pp. 419-420. <http://dx.doi.org/10.1590/S1519-69842012000200026>. PMID:22735153.
- PINTO, H.A. and MELO, A.L., 2010. *Melanoides tuberculata* as intermediate host of *Philophthalmus gralli* in Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*, vol. 52, no. 6, pp. 323-327. <http://dx.doi.org/10.1590/S0036-46652010000600007>. PMID:21225216.
- POINTIER, J.P., 1993. The introduction of *Melanoides tuberculata* (Mollusca: Thiariidae) to the island of Saint Lucia (West Indies) and its role in the decline of *Biomphalaria glabrata*, the snail intermediate host of *Schistosoma mansoni*. *Acta Tropica*, vol. 54, no. 1, pp. 13-18. [http://dx.doi.org/10.1016/0001-706X\(93\)90064-1](http://dx.doi.org/10.1016/0001-706X(93)90064-1). PMID:8103624.
- RAW, J.L., PERISSINOTTO, R., MIRANDA, N.A.F. and PEER, N., 2016. Feeding dynamics of *Melanoides tuberculata* (Müller, 1774). *The Journal of Molluscan Studies*, vol. 82, no. 2, pp. 328-335. <http://dx.doi.org/10.1093/mollus/eyv070>.
- ROCHA-MIRANDA, F. and MARTINS-SILVA, M.J., 2006. First record of the invasive snail *Melanoides tuberculatus* (Gastropoda: Prosobranchia: Thiariidae) in the Paranã river basin, Go, Brazil. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 66, no. 4, pp. 1109-1115. <http://dx.doi.org/10.1590/S1519-69842006000600018>. PMID:17299947.
- SANTOS, C. and ESKINAZI-SANT'ANNA, E., 2010. The introduced snail *Melanoides tuberculatus* (Müller, 1774) (Mollusca: Thiariidae) in aquatic ecosystems of the Brazilian Semiarid Northeast (Piranhas-Assu River basin, State of Rio Grande do Norte). *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 70, no. 1, pp. 1-7. <http://dx.doi.org/10.1590/S1519-69842010000100003>. PMID:20231954.
- SANTOS, S.B., THIENGO, S.C., FERNANDEZ, M.A., MIYAHIRA, I.C., SILVA, E.F., LOPES, B.G., GONÇALVES, I.C.B., XIMENES, R.F. and LACERDA, L.E.M., 2016. Moluscos límnicos: gastrópodes. In: A.O. LATINI, D.C. RESENDE, V.B. POMBO and L. CORADIN, eds. *Espécies exóticas invasoras de águas continentais no Brasil*. Brasília: MMA, pp. 123-250.
- SILVA, E.C. and BARROS, F., 2015. Sensibility of the invasive snail *Melanoides tuberculatu* (Müller, 1774) to salinity variations. *Malacologia*, vol. 58, no. 1-2, pp. 365-369. <http://dx.doi.org/10.4002/040.058.0215>.
- SILVA, E.C. and GOMES, L.E.O., 2014. *Melanoides tuberculatus* (Müller, 1774): occurrence extension of the invasive gastropod in Bahia, Brazil. *Pan-American Journal of Aquatic Sciences*, vol. 9, no. 2, pp. 145-149.
- SOUTO, L.S., BRITO, M.F.G. and ROSA, L.C., 2011. *Melanoides tuberculatus* (Müller, 1774): a new threat to the conservation of native aquatic species in Sergipe, Brazil. *Scientia Plena.*, vol. 7, no. 4, pp. 1-6.
- VAZ, J.F., TELES, H.M.S., CORREA, M.A. and LEITE, S.P.D.S., 1986. Ocorrência no Brasil de *Thiara* (*Melanoides*) *tuberculata* (O.F. Müller, 1774) (Gastropoda, Prosobranchia), primeiro hospedeiro intermediário de *Clonorchis sinensis* (Cobbold, 1875) (Trematoda, Platyhelminthes). *Revista de Saúde Pública*, vol. 20, no. 4, pp. 318-322. <http://dx.doi.org/10.1590/S0034-89101986000400008>. PMID:3554478.
- WORLD REGISTER OF MARINE SPECIES – WoRMS, 2018 [viewed 8 August 2018]. *Melanoides tuberculata* (OF Müller, 1774) [online]. Available from: <http://www.marinespecies.org/aphia.php?p=taxdetails&id=225694>
- XIMENES, R.F., GONÇALVES, I.C.B., MIYAHIRA, I.C., PINTO, H.A., MELO, A.L. and SANTOS, S.B., 2017. *Centrocestus formosanus* (Trematoda: Heterophyidae) in *Melanoides tuberculata* (Gastropoda: Thiariidae) from Vila do Abraão, Ilha Grande, Rio de Janeiro, Brazil. *Brazilian Journal of Biology = Revista Brasileira de Biologia*, vol. 77, no. 2, pp. 318-322. <http://dx.doi.org/10.1590/1519-6984.13615>. PMID:27599102.