

INTESTINAL HELMINTHS OF FRANCISCANA (*Pontoporia blainvilliei*) AND GUIANA DOLPHINS (*Sotalia guianensis*) FROM THE NORTH COAST OF SANTA CATARINA STATE, BRAZIL

Ana Kassia de Moraes Alves^{1*}, Elizabeth Annie Lourdes Souza¹,
Juliana Marigo² & Marta Jussara Cremer¹

¹ Universidade da Região de Joinville (UNIVILLE), Laboratório de Nectologia, Departamento de Ciências Biológicas, Rodovia Duque de Caxias, nº6.365, Campus São Francisco, São Francisco do Sul, SC, Brasil. CEP: 89240-000

² Universidade de São Paulo (USP), Laboratório de Patologia Comparada de Animais Selvagens (LAPCOM), Departamento de Patologia, Faculdade de Medicina Veterinária e Zootecnia, Avenida Professor Doutor Orlando Marques de Paiva, nº 87, Cidade Universitária, São Paulo, SP, Brasil. CEP: 05508-270

E-mails: kassia_moraes@hotmail.com; mjc2209@yahoo.com.br; ethannie@gmail.com; jumarigo@hotmail.com.

ABSTRACT

The study of helminth parasites of cetaceans from distinct areas helps to identify ecological differences among groups. This study reports the helminth fauna present in the intestinal tract of *Pontoporia blainvilliei* (n=28) and *Sotalia guianensis* (n=21) found dead on beaches along the north coast of Santa Catarina and the differences in intensity levels and infection prevalence among individuals from the inner region of Babitonga Bay in comparison with those collected in the adjacent coastal region. We analyzed 49 intestines removed from carcasses recovered from 2000 to 2014. The trematode *Synthesium pontoporiae* was the only parasite identified in the intestines of *P. blainvilliei*. Individuals from Babitonga Bay had lower intensity and prevalence values than those collected in the adjacent coastal region. These results corroborate those of previous studies that indicated some level of insulation for the population living in Babitonga Bay. The trematode species *Synthesium tursionis* and the acanthocephalan genus *Bolbosoma* were found in *Sotalia guianensis*. There was no difference in intensity and prevalence indices between genders for both cetacean species.

Keywords: *Bolbosoma* spp.; intensity; prevalence; *Synthesium tursionis*; *Synthesium pontoporiae*.

The franciscana dolphin, *Pontoporia blainvilliei* (Gervais & D'Orbigny 1844), is a small cetacean endemic to the eastern coast of South America, with the northern limit in Espírito Santo State, Brazil (18°25'S) (Moreira & Siciliano 1991), and southern limit in Golfo Nuevo, Argentina (42°35'S) (Crespo *et al.* 1998). Restricted to shallow coastal waters, *P. blainvilliei* is highly vulnerable to anthropogenic impacts, mainly accidental capture in fishing nets (Secchi *et al.* 2002, Siciliano *et al.* 2006). It is considered "critically endangered" in the Brazilian National List of Endangered Species (MMA 2014) and "vulnerable" in the Santa Catarina State List of Endangered Species (Consema 2011).

The Guiana dolphin, *Sotalia guianensis* (Van Bénéden 1864), is continuously distributed along the tropical and subtropical coasts of Central and South America, from Nicaragua (13°40'N) (Carr & Bonde 2000) to North Bay, Florianópolis, Santa Catarina State (27°35'S) (Simões-Lopes 1988). The species is mainly

associated with estuarine regions (Borobia *et al.* 1991). The southern limit of its occurrence coincides with the southern limit of the mangrove ecosystem distribution (Wedekin *et al.* 2007). This species is considered as "data deficient" in the IUCN Red List of Threatened Species (Secchi 2012).

Comparing parasite infection levels of one cetacean species from different geographical areas can demonstrate differences in ecological relationships among groups (Balbuena *et al.* 1995). In addition, some parasites are used as biological markers for differentiation of population stocks, dispersion routes and feeding habits (Marigo *et al.* 2002, Secchi *et al.* 2002, Marigo 2003, Marigo & Andrade 2005).

Sotalia guianensis populations, with high residence levels, have already been confirmed to occur in North Bay, Florianópolis (Flores 1999, Wedekin *et al.* 2007) and in the Babitonga Bay (Hardt *et al.* 2010, Schulze 2012) in Santa Catarina State. However, there are no records of the species along

the adjacent coastline of Santa Catarina outside of these bays. The Babitonga Bay, located in the north of Santa Catarina State, is surrounded by an extensive mangrove forest, considered the southernmost ecosystem of this kind in the Southern Hemisphere. This large estuarine area hosts resident populations of *P. blainvillici* and of *S. guianensis* (Hardt *et al.* 2010, Sartori 2014). Both cetacean species also occur in the adjacent coast (ACR), but for this region, the only data available are the stranding records of dead animals. In this sense, the number of *P. blainvillici* strandings is higher than that for *S. guianensis* (Paitach 2015). The aim of this study was to analyze the intestinal helminths of *P. blainvillici* and *S. guianensis* and evaluate differences in the parasitism between the populations that live inside the Babitonga Bay and in the adjacent coastal region.

The study area was located in Santa Catarina State, between 26°07'S and 26°37'S, and between 48° 36'W and 48°40'W (including Itapoá, São Francisco do Sul, Araquari, Barra do Sul and Barra Velha municipalities), and the Babitonga Bay estuary, located between 26°02' and 26°28'S, and between 48°28' and 48°50'W. The *P. blainvillici* and *S. guianensis* intestines were collected from carcasses in different stages of decomposition recovered in the period of 2000 to 2014. All the individuals are registered in the Iperoba Biological collection at the University of the Joinville Region (UNIVILLE).

Small and large intestines of *P. blainvillici* specimens were recovered during necropsy according to Yamasaki *et al.* (1975). Only complete intestines were analyzed based on the method used by Andrade (1996). The small intestine was divided into five equal-sized sections and only the first third of each section was screened. The entire large intestine was examined. For *S. guianensis*, the small and large intestines may not be distinguishable. Thus, the intestine was divided in three equal parts and each one was analyzed completely (Dierauf 1990, Marigo 2003).

The intestinal contents were washed in a sieve (mesh=150 µm) and examined under a stereoscopic microscope (10x) to collect the parasites (Marigo *et al.* 2002). All parasites were fixed in 70% ethanol according to Dailey (1978). Species identification

were based mainly on Price (1932), Raga *et al.* (1994), Fernandez *et al.* (1995), Yamaguti (1963) and Marigo *et al.* (2008). For the identification of the parasites in the samples, two slides were mounted and used as reference material. The parasites were stained with Gomori trichrome, Delafield hematoxylin, or alcoholic chloridic carmine, cleared in beechwood creosote, and mounted with Canada balsam.

The prevalence and intensity of infection were analyzed according to Bush *et al.* (1997) for each cetacean species. The intensity and prevalence were calculated for each dolphin species and for each studied area, considering each parasite taxa. The intensity was calculated as the number of parasites divided by the number of infected hosts. The prevalence was calculated as the proportion of hosts with a certain parasite divided by the total number of hosts. Parasite intensities were compared between individuals collected inside the Babitonga Bay (BB) and those collected in the adjacent coastal region (ACR), and between males and females using the Mann-Whitney test and the Student t-test ($p<0.05$), considering each species of parasite separately. In some cases, it was not possible to identify the gender due to the advanced degree of decomposition of the carcasses, so those individuals were excluded from this analysis.

A total of 49 intestines, 28 from *P. blainvillici* (12 males, 14 females and 2 unidentified) and 21 from *S. guianensis* (13 males, 6 females and 2 unidentified) were examined. Intestines of *P. blainvillici* presented average length of 23.75 m (9.7-39.80 m) and only one species of trematode parasite was found, *Synthesium pontoporiae* (Raga *et al.* 1994) (Digenea, Brachycladiidae), which was registered in both areas (Table 1).

The *S. guianensis* intestines had an average length of 26.77 m (6.75-12.75 m) and contained the trematode species *Synthesium tursionis* (Marchi 1873) Stunkard and Alvey, 1930 (Digenea, Brachycladiidae) and the acanthocephalans from the genus *Bolbosoma* (Porta, 1908) (Polimorphida, Polimorphidae), which was not identified to the species level. The individuals of the Babitonga Bay showed lower intensity for *S. tursionis* and *Bolbosoma* spp. than the ones from the ACR. (Table 1).

Table 1. Prevalence and intensity of intestinal helminths of *Pontoporia blainvilliei* and *Sotalia guianensis* in the State of Santa Catarina, southern Brazil, in the period of 2000 to 2014.

Cetacean species	Region	Total number of individuals	Parasite species	Intensity	Prevalence (%)	ALI (m)
<i>P. blainvilliei</i>	(ACR)	11	<i>S. pontoporiae</i>	96.8	81.8	23.57
	(BB)	17	<i>S. pontoporiae</i>	32.7	41.1	26.77
<i>S. guianensis</i>	(ACR)	3	<i>Bolbosoma</i> spp.	80	33.3	10.7
			<i>S. tursionis</i>	486	66.6	
	(BB)	18	<i>Bolbosoma</i> spp.	1.5	11.1	8.26
			<i>S. tursionis</i>	8.0	66.6	

(ACR) Adjacent coastal region; (BB) Babitonga Bay; (ALI) Average length of the intestines.

However, the number of individuals recovered in the ACR was too low ($n=3$) to allow statistical comparisons. Differences in parasites intensities were not detected between gender for both host species (*P. blainvilliei*: $U=68.5$, $p=0.425$) (*S. guianensis*: $t=0.64$, $df=17$, $p=0.530$). For this reason, individuals with unidentified gender were also included in the analysis of intensity and prevalence. For *P. blainvilliei*, the individuals of *S. pontoporiae* recovered inside the Babitonga Bay had lower intensity values when compared to the ones from the ACR ($U=51.5$, $p=0.048$), and the prevalence in the ACR was twice the Babitonga Bay ones (Table 1).

The parasite *S. pontoporiae*, recorded for *P. blainvilliei* in this study, has been previously reported for this species along the Brazilian coast (Marigo 2003) and was also recorded in *S. guianensis* (Marigo *et al.* 2002, 2008, 2010, Luque *et al.* 2010). This parasite was suggested as a biological tag for *P. blainvilliei* populations or stocks identification (Aznar *et al.* 1994, Andrade *et al.* 1997, Secchi *et al.* 2002, Marigo *et al.* 2008). *Synthesium tursionis* and *Bolbosoma* spp., found in this study in *S. guianensis*, has been recorded mainly in the intestines of the striped dolphin *Stenella coeruleoalba* and of the false killer whale *Pseudorca crassidens*, and occasionally in the stomachs of *S. guianensis* and *P. blainvilliei* (Andrade 1996, Andrade *et al.* 2001, Di Beneditto & Ramos 2001, Rosas 2002).

Based on the ecological differences observed in the helminth parasitism on the franciscana dolphins, we suggest the hypothesis that the *P. blainvilliei* population inside the Babitonga Bay has some degree of isolation in relation to that in the coastal region. This hypothesis has been previously discussed considering cranial morphology (Alves 2013), movement patterns and satellite transmitter data, which showed that individuals living inside the bay do not leave it (Cremer *et al.* 2012). In addition, recent DNA studies have also indicated that individuals inside the bay show small haplotype and nucleotide diversity, which indicates phylopatry and genetic isolation (Dias *et al.* 2013). In Uruguay, Costa *et al.* (2008) suggested that *P. blainvilliei* populations, living inside the La Plata River estuary, should be considered as a distinct management stock from those living off the estuary. Considering this, the lower intensity of the parasites found in hosts inside the Babitonga Bay could indicate an environment less suitable for the helminths life cycles.

ACKNOWLEDGMENTS

We would like to thank all who helped in the collection of the dead animals and in sorting the samples, and Ana Luisa Valente and Mariana Ebert for their help in the parasites identification. Research support was funded by UNIVILLE (FAP), PETROBRAS - Petróleo Brasileiro.

REFERENCES

- Alves, A. K. M. 2013. Caracterização morfométrica sincrâniana de *Pontoporia blainvilliei* (Pontoporiidae) na Baía da Babitonga norte de Santa Catarina, Brasil. Departamento de Ciências biológicas da Universidade da Região de Joinville. p. 42.
- Andrade, A. L. V. 1996. Comunidade componente de helmintos gastrintestinais de franciscana no Rio Grande do Sul, Brasil, e sua utilização como marcador biológico na identificação de estoques. Universidade Federal do Rio Grande. p. 82.
- Andrade, A., Pinedo, M. C. & Pereira J. r. 1997. The gastrointestinal helminths of Franciscana, *Pontoporia blainvilliei*, in Southern Brazil. Report International Whaling Commission, 47, 669-73.
- Andrade, A. L. V., Pinedo, M. C., & Barreto, A. S. 2001. Gastrointestinal parasites and prey items from a mass stranding of false killer whales, *Pseudorca crassidens*, in Rio Grande do Sul, southern Brazil. Revista Brasileira de Biologia, 61(1), 55-61. DOI: 10.1590/S0034-71082001000100008
- Aznar, F. J., Raga, J. A., Corcuera, J., & Fonzón, F. 1994. Parásitos como marcadores biológicos de las poblaciones de Franciscana (*Pontoporia blainvilliei*) en aguas argentinas y uruguayas. In: 6^a Reunião de Trabalho de Especialistas em Mamíferos Aquáticos da América do Sul. Universidade Federal de Santa Catarina, Florianópolis, Brazil, 142, p.117.
- Balbuena, J.A., Aznar, F.J., Fernandez, M. & Raga, J.A. 1995. Parasites as indicators of social structure and stock identity of marine mammals. In: A. S. Blix, L. Walloe, and O. Ultang (Eds.), Proceedings of the international Symposium on the Biology of Marine Mammals in the east Atlantic. Whales, seals, fish and man: developments in marine biology. pp. 720. Amsterdam: Elsevier Science publisher Ltd.
- Borobia, M., Siciliano, S., Lodi, L. & Hoek, W. 1991. Distribution of the South American dolphin *Sotalia fluviatilis*. Canadian Journal of Zoology, 69(4), 1025-1039. DOI: 10.1139/z91-148
- Bush A. O., Lafferty K. D., Lotz J. M. & Shostak, A.W. 1997. Parasitology meets ecology on its own terms: Margolis *et al.* revisited. The Journal of Parasitology, 83(4), 575-583.
- Carr, T. & Bonde, R. K. 2000. Tucuxi (*Sotalia fluviatilis*) occurs in Nicaragua, 800 km north of its previously known range. Marine Mammals Science, 16(2), 447-452.
- Cremer, M. J., Holz, A. C., Schulze, B., Sartori, C. M., Rocha-Campos, C. C., Simões-Lopes, P. C., Bordino, P. & Wells, R. S. 2012. Satellite-linked telemetry and visual monitoring of tagged franciscanas in south Brazil. In: 15^a Reunião de Trabajo de Expertos en Mamíferos Acuáticos de América del Sur. Puerto Madryn, Argentina.
- Crespo, E. A., Harris, G. & González, R. 1998. Group size and distributional range of the franciscana, *Pontoporia blainvilliei*. Marine Mammal Science, 14(4), 845-849. DOI: 10.1111/j.1748-7692.1998.tb00768.x
- Consema. 2011. Resolução nº002, de 6 de dezembro de 2011. Conselho Estadual do Meio Ambiente. Disponível em: http://www.fatma.sc.gov.br/upload/Fauna/resolucao_fauna_002_11_fauna.pdf
- Costa, P., Lessa, E. P. & Secchi, E. R. 2008. Estructura de grupos del delfín franciscana, *Pontoporia blainvilliei*. In: XII Reunión de Trabajo de Especialistas en Mamíferos Acuáticos de América del Sur y 7ºCongresso SOLAMAC; Montevideo, Uruguay. pp. 81.
- Dailey, M. D. & Stroud, R. 1978. Parasites and associated pathology observed in cetaceans stranded along the Oregon coast. Journal of Wildlife Diseases, 14, 503-511.
- Dias, C. P., Cremer, M. J., Azevedo, A. F. & Cunha, H. A. 2013. Baixa diversidade mitocondrial sugere filopatria das toninhas *Pontoporia blainvilliei* (Gervais & D'Orbigny 1844) da Baía da Babitonga, SC. In: II Encontro de Mastozoólogos do Estado do Rio de Janeiro. Rio de Janeiro, Brazil. pp. 17.
- Dierauf, L. A. 1990. Marine mammal parasitology. In: L. A Dierauf. (Ed.), Handbook of marine mammal medicine: health, diseases and rehabilitation. pp. 89- 96. Florida: CRC Press.
- Di Beneditto, A. P. M. & Ramos, R. M. A. 2001. Biologia e conservação de pequenos cetáceos no norte do estado do Rio de Janeiro. Universidade Estadual do Norte Fluminense, p. 94.
- Fernández, M., Balbuena, J. A., Pertusa, J. F. & Raga, J. A. 1995. Biometric variability of *Hadwenius tursionis* (Marchi, 1873) (Digenea, Campulidae) from the intestine of the bottlenose dolphin *Tursiops truncatus* (Montagu, 1821). Systematic Parasitology, 30, 67-76.
- Flores, P. A. C. 1999. Preliminary results of a photoidentification study of the marine tucuxi, *Sotalia fluviatilis*, in Southern Brazil. Marine Mammal Science, 15(3), 840-847.
- Hardt, F., Cremer M. J., Tonello, A. J. & Simões-Lopes, P. C. 2010. Residence patterns of the Guiana-dolphin *Sotalia guianensis* in Babitonga Bay, south coast of Brazil. Latin American Journal of Aquatic Mammals, 8(1-2), 117-121. DOI: 10.5597/lajam00160
- Luque, J. L., Muniz-Pereira, L. C., Siciliano, S., Siqueira, L. R., Oliveira, M. S. & Vieira, F. M. 2010. Checklist of helminth parasites of cetaceans from Brazil. Zootaxa, 2548, 57-68.
- Marigo, J., Rosas, F. C. W., Andrade, A. L. V., Oliveira, M. R., Dias, R. A. & Catão-Dias, J. L. 2002. Parasites of *Pontoporia blainvilliei* from São Paulo and Paraná States, Brazil. Latin American Journal of Aquatic Mammals, 1(1), 115-122.
- Marigo, J. 2003. Patología Comparada das Principais Enfermidades Parasitárias de Mamíferos Marinhos encontrados na Costa Sudeste e Sul do Brasil. Departamento de Patología da Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo, p.131.
- Marigo, J. & Andrade, A. L. V. 2005. Protocolo de conduta para encalhes de Mamíferos aquáticos: Parasitología. REMANE. Recife, p. 298.
- Marigo, J., Vicente, A. C. P., Valente, A. L. S., Measures, L. & Santos C. P. 2008. Redescription of *Synthesium pontoporiae* n. comb. with notes on *S. tursionis* and *S. seymouri* n. comb. (Digenea: Brachycladiidae Odhner, 1905). The Journal of Parasitology, 94, 505-514.
- Marigo, J., Ruoppolo, V., Rosas, F. C. W., Valente, A. L., Oliveira, M. R., Dias, R. & Catão-Dias, J. 2010. Helminths of *Sotalia guianensis* (Cetacea, Delphinidae) from south and southeastern coasts of Brazil. Journal of Wildlife Diseases, 46(2), 599-602.
- Ministério do Meio Ambiente 2014. Portarias nº 444/2014, de 17 de dezembro de 2014. Lista de espécies brasileiras ameaçadas

- de extinção. Brasília, DF. Disponível em: http://www.icmbio.gov.br/portal/images/stories/biodiversidade/fauna-brasileira/avaliacao-do-risco/PORTARIA_N%C2%BA_444_DE_17_DE_DEZEMBRO_DE_2014.pdf
- Moreira, L. M. & Siciliano, S. 1991. Northward extension range for *Pontoporia blainvilliei*. In: Biennial conference on the biology of marine mammal. Chicago, Illinois, USA, pp.9.
- Paitach, R. L. 2015. Ecologia alimentar e espacial da toninha (*Pontoporia blainvilliei*) e do Boto-cinza (*Sotalia guianensis*) na baía da Babitonga, sul do Brasil. Departamento de Ecologia e Zoologia do Centro de Ciências Biológicas da Universidade Federal de Santa Catarina. p. 1-142.
- Price, E. 1932. The trematode parasite of marine mammals. Proceedings of United States National Museum, 81(13), 68.
- Raga, J. A., Aznar, J.; Balbuena, J. A. & Dailey, M. D. 1994. *Hadwenius pontoporiae* sp. n. (Digenea: Campulidae) from the intestine of franciscana (Cetacea: Pontoporiidae) in Argentinian Waters. Journal of Helminthology, 61(1), 45-49.
- Rosas, F. C. W., Monteiro-Filho, E. L. A. 2002. Reproductive parameters of *Pontoporia blainvilliei* (cetacea: Pontoporiidae), on the coast of São Paulo and Paraná States, Brazil. Mammalia, 66(2), 231-245.
- Sartori, C. M. 2014. Fotoidentificação, abundância e área de vida de toninhas, *Pontoporia blainvilliei*, na Baía da Babitonga, Santa Catarina, Brasil. Departamento de Ciências Biológicas da Universidade da Região de Joinville. p. 96.
- Secchi, E. R., Ott, P. H. & Danilewicz, D. 2002. Report of the Fourth Workshop for Coordinated Research and Conservation of the Franciscana Dolphin (*Pontoporia blainvilliei*) in the Western South Atlantic. Latin American Journal of Aquatic Mammals, 1(1), 11-20.
- Secchi, E. 2012. *Sotalia guianensis*. In: IUCN 2012. IUCN Red List of Threatened Species. Version 2012.2. Disponível em: <http://www.iucnredlist.org/details/181359/0>
- Schulze, B. 2012. Estimativa populacional e área de vida do boto-cinza, *Sotalia guianensis*, (Cetácea, Delphinidae) na Baía da Babitonga, Santa Catarina, Brasil. Departamento de Ciências Biológicas da Universidade Federal de Santa Catarina. p. 1-121.
- Simões-Lopes, P. C. 1988. Ocorrência de uma população de *Sotalia fluviatilis* (Gervais 1853) (Cetacea, Dephinidae) no limite sul de sua distribuição, Santa Catarina, Brasil. Biotemas, 1(1), 57-62.
- Siciliano, S., Moreno, I. B., Silva, E. D. & Alves, V. C. 2006. Baleias, botos e golfinhos na Bacia de Campos. Série Guias de Campo: Fauna Marinha da Bacia de Campos. São Miguel, Caxias do Sul. p. 1-100.
- Wedekin, L. L., Daura-Jorge, F. G., Piacentini, V. Q. & Simões-Lopes, P. C. 2007. Seasonal variations in spatial usage by 84 estuarine dolphin, *Sotalia guianensis* (van Bénédén, 1864) (Cetacea; Delphinidae) at its southern limit of distribution. Brazilian Journal of Biology, 67(1), 1-8.
- Yamazaki, F. Takahashi, K. & Kamiya, T. 1975. Digestive tract of la Plata dolphin, *Pontoporia blainvilliei*. II. Small and large intestines. Okajimas Folia Anatomica Japonica, 52, 1-26.
- Yamaguti, S. 1963. Acanthocephala. In: Systema Helminthum, Vol. 5. Interscience Publications, John Wiley and Sons edit., New York - London, p. 423. DOI:19642901689

Submitted: April 13, 2016.

Accepted: November 3, 2016.