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**Ecologia populacional e social do boto-cinza (*Sotalia guianensis*) na Baía Babitonga,
Santa Catarina, Brasil**

Florianópolis

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Santa Catarina, Brasil**

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Orientadora: Prof. Dra. Marta Jussara Cremer

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Babitonga, Santa Catarina, Brasil**

O presente trabalho em nível de doutorado foi avaliado e aprovado por banca
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Certificamos que esta é a **versão original e final** do trabalho de conclusão que foi
julgado adequado para obtenção do título de doutor em Ecologia pelo Programa de Pós-
Graduação em Ecologia.

Coordenação do Programa de Pós-Graduação

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Orientadora

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À minha pequena Teresa,
que ainda não conheceu as loucuras desse mundo
mas que já sabe me fazer tão forte.

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RESUMO

A ecologia tem como um dos objetivos centrais descrever os padrões de ocorrência e interações de uma espécie com o meio adjacente. Neste contexto, avaliar parâmetros que descrevem a dinâmica populacional de forma integrada e a interpretação de padrões sociais de uma espécie/população, é chave para não só descrever os processos ecológicos, mas também para a proposição de estratégias de conservação ou manejo. Parâmetros populacionais nos permitem descrever tendências populacionais e sua dinâmica no tempo. Já a descrição de padrões sociais pode nos dizer muito sobre a ecologia, genética e biologia de uma população. Assim, este trabalho utilizou um banco de dados de 9 anos de fotoidentificação em um intervalo de 12 anos, da população de botos-cinza (*Sotalia guianensis*) da Baía Babitonga, sul do Brasil, com o objetivo de estimar parâmetros e tendência populacional e descrever padrões sociais. No primeiro capítulo, foram aplicados modelos de marcação e recaptura, utilizando o Desenho Robusto de Pollock para estimar a abundância, sobrevivência aparente e probabilidade de emigração temporária, e os modelos de Pradel combinados ao Desenho Robusto para estimar a taxa de crescimento e detectar a tendência da população. A sobrevivência aparente de indivíduos adultos foi considerada alta e constante ao longo do período de estudo ($\phi=0,98$; 95% IC: 0,97-0,99). As probabilidades de captura e recaptura variaram entre as ocasiões de captura de 0.03 (SE: 0.03 and 95% CI: 0.005 to 0.19) to 0.83 (SE: 0.06 and 95% CI: 0.66-0.91). A abundância total (N_t) estimada variou entre anos, de 101 (95% CI: 93-110) em 2007, a 127 (95% CI: 117-137) indivíduos em 2011. Foi detectado um padrão Markoviano de emigração temporária e a taxa anual de crescimento populacional (λ) foi de 1,00 para um período de 12 anos, indicando que a o número de indivíduos adultos da população permaneceu estável no período de estudo. No segundo capítulo, descreveu-se a estrutura social desta mesma população e avaliou-se se a mesma é determinada pelo sexo e área de vida dos indivíduos, usando modelos de *Multiple Regression Quadratic Assignment Procedures* (MRQAP) e teste de Mantel. Amostras de pele de 22 indivíduos foto-identificados foram coletadas para a análise do sexo. A média de sobreposição das áreas de vida entre dois indivíduos foi de 40%. Os botos-cinza apresentam baixos índices de associação social (HWI médio = 0,09) e os modelos MRQAP e teste de Mantel sugerem que apenas a sobreposição de área de vida está correlacionada à associação social. Em nível de população, este estudo levantou importantes informações para a conservação do boto-cinza, como estimativas de abundância, e sobrevivência e, a primeira estimativa de taxa de crescimento populacional para a população da Baía Babitonga. Em nível de espécie, este é o primeiro estudo combinando dados de foto-identificação, associação social, sexo e área de vida de botos-cinza, trazendo informações inéditas para a espécie, importantes para monitoramentos futuros que possam identificar eventuais tendências negativas no tamanho da população.

Palavras-chave: Parâmetros populacionais, índices de associação, padrões sociais, *Sotalia guianensis*.

ABSTRACT

One of the main objectives of ecology is to describe the patterns of occurrence and interactions of a species with its adjacent environment. In this context, evaluating parameters that describe population dynamics in an integrated way and the interpretation of social patterns of a species/population is key to not only describe ecological processes, but also to propose conservation or management strategies. Population parameters allow us to describe population trends and their dynamics over time. The description of social patterns can tell us a lot about the ecology, genetics and biology of a population. Thus, this work used a 9-year database of photoidentification, over a 12-year interval, of the dolphin population (*Sotalia guianensis*) from Babitonga Bay, southern Brazil, in order to estimate parameters and population trends and describe social standards. In the first chapter, mark-recapture models were applied, using Pollock's Robust Design to estimate abundance, apparent survival and probability of temporary emigration, and Pradel's models combined with Robust Design to estimate growth rate and detect a trend of the population. The apparent survival of adult subjects was considered high and constant throughout the study period ($\phi=0.98$; 95% CI: 0.97-0.99). Capture and recapture probabilities ranged between capture occasions from 0.03 (SE: 0.03 and 95% CI: 0.005 to 0.19) to 0.83 (SE: 0.06 and 95% CI: 0.66-0.91). The estimated total abundance (N_t) ranged between years, from 101 (95% CI: 93-110) in 2007 to 127 (95% CI: 117-137) individuals in 2011. A Markovian pattern of temporary emigration and an annual population growth rate (λ) was 1.00 for a 12-year period, indicating that the number of adult individuals in the population remained stable over the study period. In the second chapter, the social structure of this population was described and it was evaluated whether it is determined by the sex and home range overlap of the individuals, using Multiple Regression Quadratic Assignment Procedures (MRQAP) models and the Mantel test. Skin samples from 22 photo-identified individuals were collected for sex analysis. The average overlap of home ranges between two individuals was 40%. Guiana dolphins have low social association indices (mean HWI = 0.09) and the MRQAP models and Mantel test suggest that only home range overlap is correlated with social association. At the population level, this study raised important information for the conservation of the dolphin, such as abundance and survival estimates, and the first estimate of population growth rate for the Babitonga Bay population. At the species level, this is the first study combining data from photo-identification, social association, sex and home range of Guiana dolphins, providing new information for the species, important for future monitoring that can identify any negative trends in the size of the dolphin population.

Keywords: population parameters, association index, social patterns, *Sotalia guianensis*.

LISTA DE FIGURAS

CAPÍTULO 1:

Figure 1. Study area: Babitonga Bay, south Brazil. The hatched area indicates the area covered by the routes taken during data collection.....28

Figure 2. Rarefaction curves with 95% confidence intervals of cumulative photo-identified Guiana dolphins in Babitonga Bay, in the 3 different routes conducted for data collection....35

Figure 3. Total abundance (Nt) estimate for the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, between 2001 and 2017. The vertical bars indicate a 95% confidence interval. * Total abundance estimate using the transect method with distance estimate. Source: Cremer et al. (2011).....37

CAPÍTULO 2:

Figure 1. Study area: Babitonga Bay, on the northern coast of Santa Catarina, southern Brazil.....55

Figure 2. Standardized Lagged Association Rate (SLAR) (orange line) for all individuals, suggesting rapid dissociations over time. The blue line represents the null association rates (SNAR), should individuals associate randomly. The yellow line represents the model that best fitted the data (Table 3). Vertical bars indicate jackknifed standard errors.....64

LISTA DE TABELAS

CAPÍTULO 1:

Table 1. Variation in sampling effort for photo-identification of the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, south Brazil, from April 2006 to December 2017.....24

Table 2. Sampling effort for photo-identification of the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, south Brazil, from April 2006 to December 2017.....28

Table 3. Robust Design model selection by Quasi-Akaike Information Criteria of population parameters (survival rate (ϕ), temporary emigration (γ), capture probability (p) and recapture probability (c)) estimated for the Guiana dolphin (*Sotalia guianensis*) in the Babitonga Bay between the years 2006-2017. Model notation: constant parameter (.); time dependence between years (t); time dependence between months (s).....30

Table 4. Annual abundance estimates of marked individuals (N -hat) in the population of the Guiana dolphin (*Sotalia guianensis*) from the Babitonga Bay with their respective total values (N_t) corrected by the ratio of marked and unmarked individuals (θ). Standart error (SE); coefficient of variation (CV); confidence interval (CI).....31

Table 5. Pradel model selection by Quasi-Akaike Information Criteria of population parameters (survival rates (ϕ), population growth rate (λ), capture probability (p) and recapture probabilitly (c)) estimated for the Guiana dolphin (*Sotalia guianensis*) in the Babitonga Bay between the years 2006-2017. Model notation: constant parameter (.); time dependence between years (t); time dependence between months (s).....32

CAPÍTULO 2:

Table 1. Overlapping matrix considering the home range overlap between individuals (HRO) of photo-identified Guiana dolphin females in Babitonga Bay with at least 10 records. The

highlighted values indicate the maximum and minimum overlap between two females. Mean = 0.40.....62

Table 3. Standardized Lagged Association Rate (SLAR), $g'(t)$, models ranked by the lowest quasi-Akaike Information Criteria (QAIC). Δ QAIC, QAIC weights and Likelihood indicate the relative support for each model.....63

Table 4. Multiple Regression Quadratic Assignment Procedure (MRQAP) and Matel test. HWI = Half Weight Index; HRO = Home Range Overlap; SEX = Sex of Individuals.....65

SUMÁRIO

RESUMO.....	6
ABSTRACT.....	7
1.INTRODUÇÃO GERAL.....	13
1.1. O estudo da ecologia populacional e social a partir de dados de marcação-recptura.....	14
1.2. O boto-cinza.....	14
REFERÊNCIAS.....	17
2.OBJETIVOS.....	22
2.1. Objetivo Geral.....	22
2.2. Objetivos Específicos.....	22
CAPÍTULO 1: POPULATION PARAMETERS AND TRENDS OF GUIANA DOLPHINS (SOTALIA GUIANENSIS) IN BABITONGA BAY, SOUTHERN BRAZIL.....	23
ABSTRACT.....	24
1. INTRODUCTION.....	25
2. METHODS.....	27
2.2. Data collection.....	28
2.3. Photo-identification.....	29
2.4. Estimation of population parameters and trends.....	31
3. RESULTS.....	33
4. DISCUSSION.....	38
ACKNOWLEDGEMENTS.....	41
REFERENCES.....	41
CAPÍTULO 2: SOCIAL PATTERNS IN GUIANA DOLPHINS (SOTALIA GUIANENSIS) FROM SOUTHERN BRAZIL.....	51
ABSTRACT:.....	52

INTRODUCTION.....	53
METHODS.....	54
Study area.....	55
Photo-identification and data collection.....	56
Biopsy sampling.....	56
Sexing.....	57
Home range overlap.....	57
Social patterns.....	58
Correlations of association index with home range and sex.....	60
RESULTS.....	61
Home range overlap.....	61
Association between individuals.....	62
HWI, HRO and sex correlation.....	64
DISCUSSION.....	65
ACKNOWLEDGMENTS.....	66
REFERENCES.....	67
3.CONCLUSÃO GERAL.....	75

1. INTRODUÇÃO GERAL

Grande parte da ciência da ecologia está focada em tentar entender o que determina a abundância e a organização das populações nas áreas de ocorrência de uma espécie (BEAGON et al. 2006).

A partir de experimentos de marcação-recaptura, é possível levantar informações sobre a ecologia de uma população por longos períodos de tempo. Dentre as diversas aplicações deste método, podemos destacar a estimativa de parâmetros populacionais como abundância, sobrevivência, recrutamento, migração e emigração, taxas de crescimento populacional e padrões sociais de uma população. A marcação-recaptura pode ser utilizada em qualquer grupo animal, desde que os animais possam ser, de alguma forma, identificados individualmente (LETTIK & ARMSTRONG 2003).

A fotoidentificação é uma das técnicas de reconhecimento individual amplamente utilizada no estudo de populações de mamíferos, incluindo os cetáceos (WÜRSIG & JEFFERSON 1990). Esta técnica foi desenvolvida para a identificação individual de animais terrestres (CALSTROM & EDELSTAM 1946) e por volta dos anos 1970 foi adaptada com sucesso para várias espécies de cetáceos (WÜRSIG & WÜRSIG 1977; WÜRSIG 1978; FORD et al. 1994). Hoje em dia é a principal forma de identificar individualmente os cetáceos, baseada principalmente em marcas naturais, como arranhões, cicatrizes e cortes que estão presentes na nadadeira e região dorsal dos animais (WÜRSIG & WÜRSIG 1977; WÜRSIG & JEFFERSON 1990). De maneira geral, as marcas naturais são decorrentes das atividades diárias dos animais, como arranhões causados por abrasão do corpo com o fundo do mar, durante a perseguição a presas ou mordidas decorrentes do contato intra- ou interespecífico (WÜRSIG & WÜRSIG 1977).

1.1 O ESTUDO DA ECOLOGIA POPULACIONAL E SOCIAL A PARTIR DE DADOS DE MARCAÇÃO-RECPTURA

Aliando os dados de fotoidentificação com o método da marcação-recaptura, a estimativa de parâmetros populacionais depende do número de animais marcados e sua proporção em amostragens subsequentes (SEBER 1982). É construído um histórico de capturas onde são registradas as avistagens de cada indivíduo fotoidentificado e aplicam-se modelos de populações fechadas e abertas, onde, por tanto, são ou não consideradas a entrada

e saída de indivíduos por meio de nascimentos, mortes, imigrações e emigrações durante o período amostral (AMSTRUP et al. 2005). Ou ainda, é possível combinar os modelos de população aberta e fechada com a abordagem de Desenho Robusto, sendo possível assim, além da abundância, estimar outros parâmetros demográficos como sobrevivência aparente e emigração temporária (POLLOCK 1982). Pode-se também aplicar os modelos de Pradel, que observando o histórico de capturas de trás para frente, avalia a entrada de indivíduos na população e estima a taxa de recrutamento e taxa de crescimento da população (PRADEL 1996).

Dados utilizados para construir o histórico de capturas são parte importante no cálculo do índice de associação entre os indivíduos, que é a base para os estudos sociais de uma população indicando a qualidade, o conteúdo e o padrão dos relacionamentos sociais entre indivíduos (HINDE 1976). A estrutura social é uma chave determinante da biologia populacional, influenciando a forma, fluxo genético, padrões espaciais e escala de movimentos. A força e a estabilidade de uma associação estão ligadas aos benefícios sócio-ecológicos, como defesa contra predação e captura de alimento (WILSON et al. 1975).

O conhecimento detalhado da dinâmica social da maioria das espécies de mamíferos marinhos continua limitado, em grande parte devido às dificuldades logísticas que tornam seu estudo em ambiente natural desafiador, custoso e lento (TAYLOR & GERRODETE 1993).

1.2 . O BOTO-CINZA

O boto-cinza (*Sotalia guianensis*) ainda permanece como lacuna no conhecimento das dinâmicas populacional e social de cetáceos, pois os estudos abordando essas temáticas ainda são pontuais. Endêmico da costa ocidental do Oceano Atlântico, o boto-cinza distribui-se desde Honduras (FLORES & DA SILVA 2009) até a Baía Norte em Florianópolis (SIMÕES-LOPES 1988). E, por habitar a região costeira e estar fortemente associada a manguezais, esta espécie é altamente afetada pelas atividades antrópicas, como a captura acidental em redes de pesca, a poluição química e sonora do ambiente marinho e a perda de hábitat.

Na Baía Babitonga uma população de botos-cinza é estudada desde 1996 (CREMER 2000). Esta população é considerada residente (HARDT et al. 2010) e vive em simpatria com outra população de pequeno cetáceo, a toninha (*Pontoporia blainvillei*) (CREMER et al. 2017).

Os grupos de botos-cinza variam na sua composição e tamanho, com cerca de dois a 29 indivíduos em geral (EDWARDS & SCHNELL 2001; DAURA-JORGE et al. 2005; NASCIMENTO et al. 2008). Na Baía Babitonga o tamanho médio dos grupos é de cerca de seis indivíduos (CREMER 2000), mas grandes agrupamentos são frequentemente avistados em comportamento de pesca e interagindo com aves marinhas, como trinta-réis, biguás e atobás (CREMER et al. 2004).

Os botos-cinza distribuem-se de forma heterogênea na Baía Babitonga, com o uso do hábitat variando diariamente com o ciclo da maré e acompanhando o deslocamento das presas (PAITACH et al. 2017). Variações na área de vida foram observadas ao longo dos anos, sendo que as áreas preferenciais estão localizadas sempre próximas às ilhas (CREMER et al. 2004; CREMER et al. 2009).

Estimativas pontuais de abundância foram realizadas na Babitonga em 2001, 2002, 2003, e 2011. De 2001 a 2003, foram estimados 245 (95% IC: 142-422), 186 (95% IC: 93-374) e 179 (95% IC: 93-144) indivíduos respectivamente, com o método de transecção linear (CREMER et al. 2011). Já em 2011, 209 (95% IC: 174-252) indivíduos foram estimados pelo método da marcação-recaptura, com dados de fotoidentificação (SCHULZE 2012). Outros parâmetros populacionais como sobrevivência, migração e emigração também foram estimados, mas ainda faltam dados a respeito da dinâmica e tendência populacional para esta população.

A organização social de populações de botos-cinza já foi estudada em quatro localidades, todas na costa brasileira: em Cananéia-SP (SANTOS & ROSSO 2008), em Sepetiba-RJ (BEIRÃO-CAMPOS et al. 2015), em Caravelas-BA (CANTOR ET al. 2012) e na Baía de Pipa-RN (LUNARDI & FERREIRA 2014). A ecologia social em delfínídeos é variada, pois os indivíduos podem estabelecer longas associações relacionadas à consanguinidade, ou estabelecer elos de curta duração com vários indivíduos (RENDELL & WHITEHEAD 2001; LUSSEAU et al. 2003). Em uma população, os grupos podem se dividir em subgrupos, aumentando a eficiência do forrageamento, minimizando os riscos da predação e/ou para reprodução, e em seguida se reagruparem (WÜRSIG 1978). Desta forma, os indivíduos associam-se em uma dinâmica social do tipo fissão-fusão (AURELI et al. 2008). Para o boto-cinza, estudos revelaram que há um padrão de agrupamentos fluidos indicando um sistema social com uma alta dinâmica tipo fissão-fusão em uma rede social modular (CANTOR et al. 2012). Uma grande população que habita a mesma área pode dividir-se em grupos menores com número variado de indivíduos onde todos interagem dentro da

população, mas em taxas diferentes (BEIRÃO-CAMPOS et al. 2015). Associações estáveis parecem não ser características (SANTOS & ROSSO 2008), com grupos coesos no espaço, mas instáveis no tempo, com alterações na composição e no número de indivíduos (LUNARDI & FERREIRA 2014).

Porém na Baía Babitonga este tema ainda não foi abordado e o levantamento de informações sobre parâmetros populacionais em conjunto com padrões sociais são fatores chave na identificação do potencial efeito das atividades de origem antrópica sobre uma população de vida livre. A construção de um banco de dados com informações base definem a condição de uma população e servem como comparativo na investigação de mudanças na abundância, comportamento e surgimento de doenças, por exemplo, para que se chegue a conclusões a respeito do surgimento e/ou magnitude de uma atividade antrópica com possível impacto sobre a população estudada.

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2. OBJETIVOS

2.1. OBJETIVO GERAL

Este trabalho tem como objetivo estimar parâmetros populacionais, assim como descrever os padrões sociais da população de botos-cinza (*Sotalia guianensis*) da Baía Babitonga, usando um banco de dados de 9 anos de fotoidentificação.

2.2. OBJETIVOS ESPECÍFICOS

Estimar abundância, sobrevivência, probabilidade de migração temporária e taxa de crescimento populacional para avaliar a dinâmica populacional.

Descrever os padrões sociais da população, e avaliar se as interações entre os indivíduos são determinadas pelo sexo ou compartilhamento da mesma área de vida.

CAPÍTULO 1:

POPULATION PARAMETERS AND TRENDS OF GUIANA DOLPHINS (*SOTALIA GUIANENSIS*) IN BABITONGA BAY, SOUTHERN BRAZIL

Abstract

1. Populations of Guiana dolphins (*Sotalia guianensis*) are discontinuously distributed in the coastal waters of the western Atlantic Ocean and many populations have been affected by different impacts. This study used a 9-year photo-identification database to estimate trends and population parameters of a small population unit of Guiana dolphins in Babitonga Bay, southern Brazil, that is exposed to multiple anthropic pressures.
2. Data collection was conducted from April 2006 to December 2017. Individual identification was made through the recognition of permanent or long-term natural marks. We applied Pollock Robust Design to estimate abundance (N), apparent survival (ϕ) and temporary emigration probabilities (γ), and Pradel models were combined to the Robust Design to estimate the growth rate of the population (λ).
3. The apparent survival of adults was high and constant ($\phi=0.98$; SE: 0.004 and 95% CI: 0.97-0.99). The capture and recapture probabilities varied among capture events from 0.03 (SE: 0.03 and 95% CI: 0.005 to 0.19) to 0.83 (SE: 0.06 and 95% CI: 0.66-0.91). The total abundance (N_t) estimated ranged from 101 (95% CI: 93-110) individuals in 2007 to 127 (95% CI: 117-137) individuals in 2011. Markovian temporary emigration pattern ($\gamma^* \gamma^{**}$) was detected, indicating that individuals move in and out of the study area in a non-random manner. The annual population growth rate (λ) was equal to 1.00 for a period of 12 years, and indicates that this population remained stable.
4. This study raised important issues for the conservation of Guiana dolphins like abundance and survival rates, providing the first estimates of population growth rate in southern Brazil, the austral limit of the species' distribution. The results indicate stability in population size, which is key information for the species in an area that is exposed to a considerable level of anthropogenic pressure.

Key words: population trends, abundance, survival, population growth rate, mark-recapture, photo-identification.

1. Introduction

Information about population parameters enables the detection of population trends, which are important for the identification of impacts and threats that can generate population decline, and is critical for management of threats to wildlife (Hooker et al., 2011). Population trend is the major component in the conservation of wildlife species which may indicate growth, stability, or reduction in the growth rate of a population (Wade, 2002), and is among the criteria established by the International Union for Conservation of Nature and Natural Resources (IUCN) to determine whether a species is at risk of extinction (Mace et al., 2008).

Populations of Guiana dolphins (*Sotalia guianensis*) are discontinuously distributed in the coastal waters of the western Atlantic Ocean (15°N to 27°S, Silva & Best, 1996). These populations inhabit coastal areas, estuaries, and bays, and their abundance may vary from less than 40 to more than 800 individuals (e.g. Edwards & Schnell, 2001; Flach et al., 2008; Cantor et al., 2012; Azevedo et al., 2017; Cremer et al., 2017). Species with small populations and discontinuous distribution are more vulnerable to environmental and demographic fluctuations (Thompson et al., 2000). Although the abundance is influenced by the availability of resources and habitat carrying capacity (Hawley, 1982), many Guiana dolphin populations have been affected by different pressures related to human activities, including vessel traffic (e.g. Araújo et al., 2008), marine pollution (e. g. Yogui et al., 2003) and accidental capture in fishing gears (e. g. Di Benedetto, 2003; Crespo et al., 2010;). Multiple environmental impacts were associated with the decline of Guiana dolphin in Guanabara Bay (Azevedo et al., 2017), while a morbillivirus outbreak was responsible for a mass mortality in the population of Sepetiba Bay in 2017 (Groch et al., 2018).

Although some populations are well studied, including information about population parameters (Flach et al., 2008; Cantor et al., 2012; Azevedo et al., 2017), very little is known regarding population parameters for the vast majority of Guiana dolphin populations. For

many years, the International Union for the Conservation of Nature – IUCN considered that the data available about abundance, trends, and mortality levels of the Guiana dolphin was insufficient for an evaluation of the status, and the species was listed as “Data Deficient” (DD). Recently, the species was listed as “Near Threatened” (NT) (Secchi et al., 2018) considering that it is affected by several threat factors including bycatch in fisheries, deliberate capture for bait, pollution, and habitat deterioration (see Crespo et al., 2010). In Brazil the species is considered endangered (Ministério do Meio Ambiente, 2014).

A resident population of Guiana dolphins in Babitonga Bay, southern Brazil, has been studied since 1996. Information about home range, habitat use, distribution, abundance, acoustic behavior, diet and trophic ecology are available (Cremer et al., 2009; Hardt et al., 2010; Cremer et al., 2011; Cremer et al., 2012; Cremer et al., 2017), but there is no information on population trends for this population. The increasing pressures observed in this area over the last 20 years include overfishing (Serafini et al., 2014), chemical and organic contamination (Barros et al., 2010; Martins et al., 2014), and the increase of harbour activities (Cremer et al., 2017; Herbst et al., 2020) have raised concerns about the status of the Guiana dolphin population. Evidence regarding the threats present in Babitonga Bay are already being documented, as is the case of contamination by PCBs and pyrethroids (Dorneles et al., 2013; Vidal et al., 2020) and the accidental capture in fishing nets (Pinheiro & Cremer, 2004).

In the period from 2000 to 2017, 71 Guiana dolphins’ carcasses were recovered in Babitonga Bay and adjacent areas, being that, 25 carcasses were found only in 2016 (unpublished data), suggesting that this population could be in decline considering that the population was estimated to be around 203 individuals between 2001 to 2003 (Cremer et al., 2011). In this context, the present study used a 9-year photo identification database in a interval of 12 years (from 2006 to 2017) to estimate trends and population parameters of

Guiana dolphins in the Babitonga Bay. Knowledge about the dynamics of this population is very important for its conservation, making it possible to propose management and conservation measures for the species.

2. Methods

2.1. Study Area

Babitonga is a 153.7 km² bay located in southern Brazil (26°02'-26°28'S / 48°28'-48°50'W; Figure 1). It is surrounded by six cities, including the largest industrial park in the State, totalling more than 600,000 inhabitants. The region's economy is dominated by tourism, agriculture, ports, and industries (mainly textile, metallurgical, and foundry industries) (Niederle & Mattei, 2012). There are also artisanal fisheries, with more than 10 types of common practices in the region (Serafini et al., 2014). The dumping of untreated domestic and industrial effluents is the main contributor to the chemical and organic pollution of the Babitonga river system (Barros et al., 2008), which affects the habitat quality for marine fauna, including small cetaceans (Cremer, 2006).

2.2. Data collection

Data collection was conducted from April 2006 to December 2017. A 5.5 m aluminum boat powered by a 60 hp outboard engine, and a 6.2 m inflatable boat with an 200 hp outboard engine were used in three pre-established routes over the years (Figure 1), that covered the entire bay to search for Guiana dolphins (only in sea conditions from 0 to 2 on the Beaufort scale). From 2006 to 2009 (Period 1 - Route 1), data collection was performed in routes directed to the species' core areas. From 2010 to 2012 (Period 2 - Route 2), data collection was undertaken over 2 consecutive days or with two boats simultaneously, to maximize the area covered. From 2013 to 2017 (Period 3 - Route 3), data collection was performed following a single continuous route, that covered the entire bay. The extent of the routes was limited by the existence of navigable areas, since there are extensive submerged sand banks

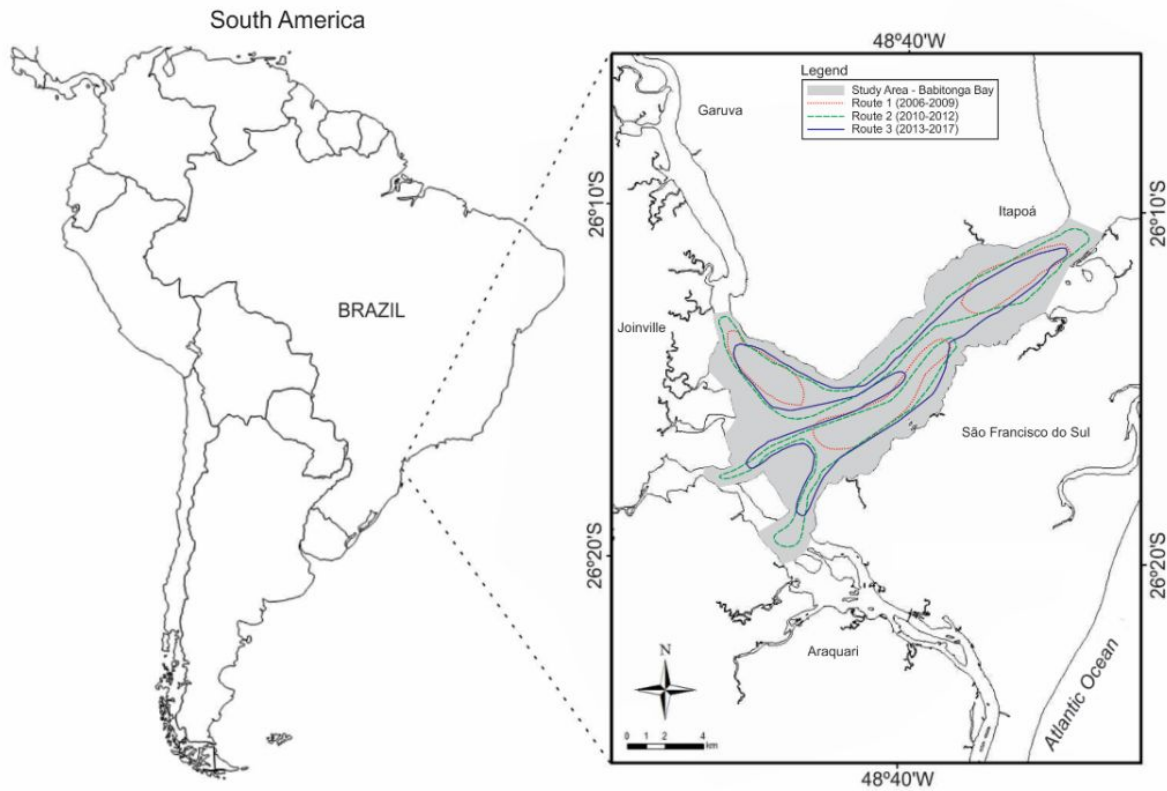


Figure 1. Study area: Babitonga Bay, south Brazil. The hatched area indicates the area covered by the routes taken during data collection.

that do not have sufficient depth for the passage of the research boats (Figure 1). Intervals between data collection were not equal during the study period (Table 1), therefore, the sightings were grouped by month forming a capture occasion (see data analysis below). Whenever a group of Guiana dolphins was sighted, it was followed at a distance of at least 20 meters and for no more than 30 minutes to obtain digital photographs of the dorsal fins (cameras CANON 20D and Canon 7D with Zoom Lens 100-300mm and 100-400mm).

2.3. Photo-identification

Individual dolphins were identified through the recognition of permanent or long-term natural marks, such as nicks, scars, and mutilations present on the dorsal fin (Würsig & Jefferson, 1990). Calves are born without marks, but dolphins acquire marks throughout their

lives, so older animals tend to have more marks; thus, it was assumed that the identified individuals were adults (Würsig et al., 2017).

Table 1. Variation in sampling effort for photo-identification of the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, south Brazil, from April 2006 to December 2017.

	JA	FE	M	A	M	JU	J	A	S		N	D
	N	B	AR	PR	AY	N	L	G	P	CT	V	EC
2006				X		X	X	X		X		
2007			X	X	X	X	X					
2008		X	X	X	X	X	X					
2009				X	X	X	X					
2010				X	X	X	X	X	X	X	X	
2011		X	X	X	X	X		X	X	X		
2012		X	X	X	X		X	X	X	X	X	
2013		X	X	X	X	X	X	X	X	X	X	
2014				X				X				
2017				X	X	X		X	X	X	X	X

Only high-quality photographs (good focus, clarity and contrast, proximity and perpendicular angle) were used in the analyses and whenever possible the animals were photographed on both sides. The identification of individuals at each sampling occasion (month) comprised a presence/absence capture history.

Two possible types of identification errors are described in the literature: false positives (when the same individual is catalogued more than once) and false negatives (when different individuals are catalogued as being the same individual) (Stevick et al., 2011). These errors were minimized by cataloguing only individuals with distinctive marks, selecting only good quality photographs, and maintaining the constant monitoring of individuals through photo-identification in order to detect changes in the marks used for identification during the study period (Gunnlaugsson & Sigurjónsson, 1990).

The following assumptions were made: (1) identification marks are not lost over time, (2) individuals are properly recognized on recapture occasions, (3) all individuals in the

population have equal probability of capture and recapture, (4) all individuals have the same probability of survival, (5) the study area did not vary geographically, (6) the population remained closed (births, deaths, immigration, and emigration did not occur) within primary periods, and (7) the capture of an individual did not affect its subsequent recapture probability during the secondary period (Lindberg & Rexstad, 2002; Williams et al., 2002; Pollock & Alpizar-Jara, 2005). Violation of assumptions may cause bias in the results, such as decreased precision and accuracy of the estimated parameters (Lindberg & Rexstad, 2002).

To minimize the violation of assumptions (1) and (2), only permanent and long-term marks were used to identify individuals; only individuals with distinctive marks were incorporated into the study; and only good quality photographs were used for identification (Urian et al., 2014). Assumptions (3) and (4) are likely to be already violated due to heterogeneity in individual behaviour, changes in staff, the acquisition of higher quality cameras and lenses, and variations in fieldwork logistics, which may cause differences in the probability of capture and recapture and hence the probability of survival. Thus, violation of conditions (3) and (4) were analyzed by tests 2 and 3 in the Release program (see below).

Discovery curves were generated using the method for sample-based rarefaction curves (Gotelli & Colwell, 2001) to visualize differences in sampling effort periods that were conducted over three different routes (Figure 1). These curves provide a valid comparison of the number of marked dolphins under varying sampling effort. Resampling was done setting 1,000 interactions by Monte Carlo methods in the software EstimateS (Colwell RK, <http://purl.oclc.org/estimates>).

2.4. Estimation of population parameters and trends

Robust Design was applied to estimate abundance (N), apparent survival (ϕ) and temporary emigration probabilities (γ) (Pollock, 1982), and Pradel models were combined to the Robust Design to estimate the growth rate of the population (λ , where $\lambda < 1$ indicates

decline, $\lambda = 1$ indicates stability, $\lambda > 1$ indicates growth). The Robust Design approach combines characteristics of both open and closed models in an integrated framework, and accommodates multiple temporary movements in or out of the study population (Pollock, 1982; Kendall et al., 1997). The Robust Design (RD) organizes data into two hierarchical sampling periods, primary and secondary periods, where the population can be assumed as constant over the secondary sampling period (that are brief enough that one can assume no births, deaths, emigration, or immigration) (Pollock, 1982). On the other hand, the Pradel approach reverses the encounter history going backwards in time; thereby, it is possible to estimate parameters that allowed the probability of entering a population (Pradel, 1996). All population parameters were estimated using program MARK (Cooch & White, 2014).

Each sampled year was considered a primary period (10 periods - 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, and 2017), used to estimate the apparent survival, temporary emigration, capture probability and population growth rate. It should be highlighted that these parameters were estimated mainly for the adults of the population, which were considered to be the marked individuals in the population (Würsig et al., 2017). Each month was considered a secondary period, used to estimate the abundance (N) of each primary period. The population was assumed to be closed within a year, considering the high residence rates in the area (Hardt et al., 2010) and the low number of marked individuals found dead per year.

The models were built considering the time effect within the primary periods (t) and the time effect between primary periods (s). Robust Design with Huggins' and Robust Design with Huggins' Heterogeneity parameterization were used. The models were built following the ecology and biology of the species and the logic of the most complex to the simplest, maintaining the configuration that best fits the data following the AIC method. To test the possible violation of assumptions and overdispersion, the "goodness-of-fit" test was applied for collapsed data in primary occasions adjusted to Cormack-Jolly-Seber (CJS) models, using

the RELEASE program (Cooch & White, 2014), and the tests 2 and 3 were applied to evaluate the homogeneity and equal probability of survival among individuals, respectively (assumptions 3 and 4). These procedures provide an assessment of the data settings to the models (Burnham et al., 1987; Lebreton et al., 1992). The variance inflation factor (\hat{c}) is a measure that quantifies over- and underdispersion and was calculated by summing the chi-square of tests 2 and 3, divided by the sum of the respective degrees of freedom. The \hat{c} is included as a dispersion parameter in the calculation of the parameter error. If $\hat{c} > 1$, the selection of the model was performed by QAIC (Quasi-Akaike Information Criteria, Burnham & Anderson, 2002), which fixes the AIC considering overdispersion. Models were selected using Quasi-Akaike Information Criteria (AIC; Anderson et al., 1994).

The proportion of marked individuals in the population (θ) was estimated as the number of marked individuals divided by the total number of individuals observed in each group, averaged over all groups, to estimate the total abundance (N_t), that include animals without natural marks (juveniles and calves) (Silva et al., 2009). Only groups with a maximum of 12 individuals were considered to ensure accuracy in the total number of individuals counted, representing 54% of all sightings. The total abundance was estimated as

$N_t = \frac{N - h}{\theta}$, where N is the number of individuals estimated by the Robust Design model, divided by θ , which is the average proportion of the number of marked individuals. The variance of the total abundance was estimated according to Delta method (see Williams et al., 2002). N and $var(N)$ were estimated by the model selected by QAIC. The confidence interval (95%) was estimated by N_t/r (lower) and $N_t \cdot r$ (upper), where

$$r = \exp \left\{ 1.96 \sqrt{\ln \left(1 + \left(\frac{CV(N)}{N} \right)^2 \right)} \right\}.$$

For comparison purposes, the abundances estimated by Cremer et al. (2011) in 2001, 2002 and 2003 were included in the results graph.

3. RESULTS

Guiana dolphin groups were sampled from April 2006 to December 2017 during all 77 capture occasions, totaling 447:50h of effort. During this period, 113 individuals were photo-identified in 348 groups, varying between 28 and 58 individuals per year. From that, 96 individuals have been seen at least twice. The number of sampled months per year (secondary periods) ranged from four to 10 (Table 2). The size of the groups varied from one to more than 80 individuals (mean = 16; median = 12; mode = 8). Calves were seen in 70% of the groups and in every sampled months of the sampled years.

Table 2. Sampling effort for photo-identification of the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, south Brazil, from April 2006 to December 2017.

Year	Secondary occasions	N_{ID}	N_c	N_R	Effort (hours)	Sighted groups
2006	5	37	37	-	25:53	12
2007	5	46	18	28	60:42	24
2008	6	47	8	39	19:16	23
2009	4	28	3	25	14:29	7
2010	8	56	11	45	27:09	32
2011	8	58	7	51	69:28	74
2012	9	51	2	49	78:02	48
2013	10	56	7	49	124:48	88
2017	8	53	20	33	53:56	40
Total	63	-	113	-	447:50	348

N_{ID} = number of photo-identified individuals, N_c = number of new photo-identified individuals N_R = number of recaptured individuals.

The rarefaction curves showed that regardless of the route used to collect data, in the three sampling periods the sampling was efficient and enough for the analyzes performed (Figure 2).

Tests 2 ($p = 0.013$; $df = 6$) and 3 ($p = 0.15$; $df = 13$) from the Release program indicated violation of homogeneity on capture probability. C-hat was changed from 1.00 to 1.80 to offset overdispersion and the selection of the model was performed by QAIC.

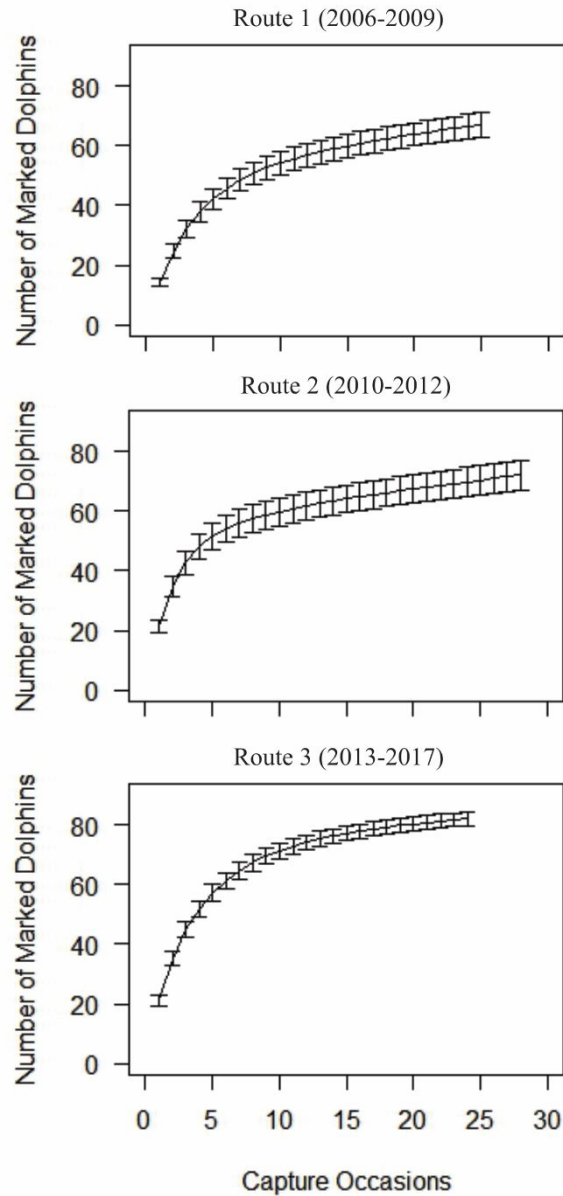


Figure 2. Rarefaction curves with 95% confidence intervals of cumulative photo-identified Guiana dolphins in Babitonga Bay, in the 3 different routes conducted for data collection.

Best-fitting Robust Design model (Table 3) indicated that the apparent survival of adults was high and constant ($\phi=0.98$; SE: 0.004 and 95% CI: 0.97-0.99). The capture probability was equal to the recapture probability and varied among capture occasions (primary and secondary periods) from 0.03 (SE: 0.03 and 95% CI: 0.005 to 0.19) to 0.83 (SE: 0.06 and 95% CI: 0.66-0.91).

Table 3. Robust Design model selection by Quasi-Akaike Information Criteria of population parameters (survival rate (ϕ), temporary emigration (y), capture probability (p) and recapture probability (c)) estimated for the Guiana dolphin (*Sotalia guianensis*) in the Babitonga Bay between the years 2006-2017. Model notation: constant parameter (.); time dependence between years (t); time dependence between months (s).

Model	QAICc	Delta QAICc	AICc Weights	No. Parameters	QDeviance
{ $\phi(.) y''(.) y'(.) p=c(t*s)$ }	2378.59	0	0.95	66	3073.67
{ $\phi(t) y''(.) y'(.) p=c(t*s)$ }	2385.31	6.72	0.03	73	3064.73
{ $\phi(t) y''= y'(.) p=c(t*s)$ }	2386.17	7.58	0.02	72	3067.84
{ $\phi(t) y''(t) y'(t) p=c(t*s)$ }	2400.02	21.43	0.00	82	3059.04
{ $\phi(t) y''= y'=0 p=c(t*s)$ }	2408.08	29.49	0.00	71	3091.99
{ $\phi(t) y''(t) y'(t) p=c(t)$ }	2639.06	260.47	0.00	30	3411.92

The total abundance (N_t) estimate ranged from 101 (95% CI: 93-110) individuals in 2007 to 127 (95% CI: 117-137) individuals in 2011 (Table 4 and Figure 3). Markovian emigration pattern ($y''(x) y'(x)$), constant in time, was detected, indicating that individuals move in and out of the study area in a non-random manner. The probability that an individual available for capture on previous occasions temporarily emigrated from the study area (y'') was 0.09 (SE: 0.03 and 95% CI: 0.04-0.18), and the probability that an individual that was outside the study area on a previous occasion remained outside it (y') was 0.78 (SE: 0.19 and 95% CI: 0.29-0.96). Thus, the probability of dolphins remaining in the study area between capture occasions ($1-y''$) was 0.91 and the return rate of temporary emigrants to the study area ($1-y'$) was 0.22, confirming the residency and fidelity of the population to the area.

The Pradel model that best fitted the data (Table 5) indicates that this population has remained stable during the period of 12 years, with an annual population growth rate (λ) equal to 1.00.

Table 4. Annual abundance estimates of marked individuals (N-hat) in the population of the Guiana dolphin (*Sotalia guianensis*) from the Babitonga Bay with their respective total values (Nt) corrected by the ratio of marked and unmarked individuals (θ). Standart error (SE); coefficient of variation (CV); confidence interval (CI).

Year	Estimated abundance (Robust Design)		Proportion of marked individuals		Total abundance		
	N-hat	SE	(θ)	CV(θ)	N _t	CV(N _t)	95% CI
2006	52.02	7.44	0.46	0.04	113	0.15	85-151
2007	46.47	0.73	0.46	0.04	101	0.04	93-110
2008	52.02	2.80	0.46	0.04	113	0.07	99-129
2009	48.88	8.16	0.46	0.04	106	0.17	76-148
2010	56.12	0.35	0.46	0.04	122	0.04	113-132
2011	58.33	0.59	0.46	0.04	127	0.04	117-137
2012	51.47	0.72	0.46	0.04	112	0.04	103-121
2013	56.09	0.31	0.46	0.04	122	0.04	113-131
2017	54.21	1.87	0.46	0.04	118	0.05	107-130

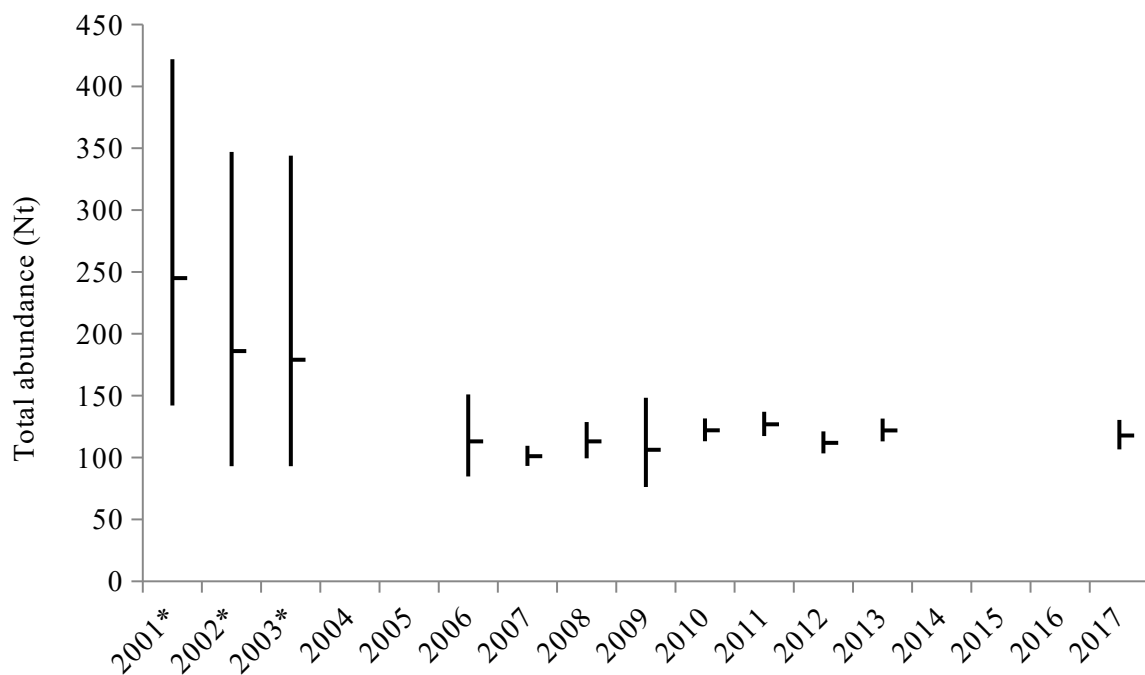


Figure 3. Total abundance (Nt) estimate for the Guiana dolphin (*Sotalia guianensis*) population in the Babitonga Bay, between 2001 and 2017. The vertical bars indicate a 95% confidence interval. * Total abundance estimate using the transect method with distance estimate. Source: Cremer et al. (2011).

Table 5. Pradel model selection by Quasi-Akaike Information Criteria of population parameters (survival rates (ϕ), population growth rate (λ), capture probability (p) and recapture probability (c)) estimated for the Guiana dolphin (*Sotalia guianensis*) in the Babitonga Bay between the years 2006-2017. Model notation: constant parameter (.); time dependence between years (t); time dependence between months (s).

Model	QAICc	Delta QAICc	AICc Weights	No. Parameters	QDeviance
{ $\phi(t) \lambda(.) p=c(t*s)$ }	2651.9796	0	0.9425	72	1917.1202
{ $\phi(.) \lambda(.) p=c(t*s)$ }	2657.6742	5.6946	0.05467	65	1938.4496
{ $\phi(t)\lambda(t) p=c(t*s)$ }	2663.5966	11.617	0.00283	79	1912.9209
{ $\phi(t) \lambda(t) p=c(t)$ }	2890.9	238.9204	0	25	2257.6812

4. DISCUSSION

This study provides the first estimates of several population parameters of the Guiana dolphin in southern Brazil. The population stability is an indicator of population health, reflecting the conditions of the population to maintain reproduction, high residency patterns and low mortality rates (Williams et al., 2002), having been used to assess the threat level of some populations of Guiana dolphins throughout the species' distribution (Cantor et al., 2012; Azevedo et al., 2017). An apparent stability in the Guiana dolphin population in Babitonga Bay was found.

This result indicates the importance of long-term studies to detect a trend, since analyses made over shorter periods could lead to misinterpretations about some population parameters. It should be noted that the stability in abundance found in this study refers mainly to the adults, which are the mainly marked individuals. Cantor et al. (2012), in a long-term study, also found an apparently stable population of Guiana dolphins in the Caravelas River Estuary, in north-east Brazil. But a decline in a population was recently found in Guanabara Bay, in southeast Brazil (Azevedo et al., 2017).

In the Babitonga Bay, 17 individuals sighted in 2006 were also sighted in 2017, which represents a resident period of 12 years in the area; 55 individuals were sighted for at least five years. A low emigration probability was estimated, so as a high return rate from the emigrants, confirming the high degree of residence already reported through photo-

identification for the Babitonga Bay population (Hardt et al., 2010). High residence levels were also reported in other areas throughout Guiana dolphin's distribution (Flores, 1999; Azevedo et al., 2004; Rossi-Santos et al., 2007; Batista et al., 2014). Movements in and out of Babitonga Bay seem to be lower than that of other populations, like that in Caravelas estuary (Cantor et al., 2012), although it is evident that the species occupy a larger home range, considering the representative number of Guiana dolphin carcasses recorded on the beaches outside Babitonga Bay (Cremer et al., 2018). These data underscore the importance of Babitonga Bay for the survival of this population, which supports two marine mammal populations, considered top predators in this system. Babitonga Bay is the only area where a Guiana dolphin population lives in direct sympatry with another species of small cetacean – the franciscana dolphin (*Pontoporia blainvillei*) (Cremer et al., 2017). This condition, even if it does not involve agonistic interactions and direct competition for food resources, implies at least some level of niche overlap between the two species.

A previous study estimated higher values for total abundance of Guiana dolphins in Babitonga Bay in the period of 2001 to 2003 (Cremer et al., 2011), although comparisons are not recommended since they used different methods to estimate abundance. These results may be overestimated because the mark-recapture method tends to be more accurate than the linear distance transection sampling method (Evans & Hammond 2004, Daura-Jorge & Simões-Lopes 2016). Despite that, most individuals found dead in the area from 2000 to 2016 were juveniles (Cremer et al., 2017) and, just as recorded for franciscana dolphins, juveniles tend to make up the largest proportion of individuals that are accidentally caught in gillnets (Botta et al., 2010). The same pattern appears to be found in some populations of Guiana dolphins, with a higher frequency of stranding for immature individuals (Ramos et al., 2000; Rosas & Monteiro-Filho 2002; Ramos et al., 2008). Although adult survival is the primary factor influencing population growth rates, low survivorship of calves inevitably reduces the

population recruitment rate and may be a cause for concern (Wade, 2009). Thus, recruitment may have been affected and may be influencing the abundance estimates.

Because it was not possible to distinguish mortality from permanent emigration, the apparent survival rate ($\phi = 0.98$) was estimated for the adults in the population. The apparent survival was higher than in other regions (0.88 in Caravelas, Brazil - Cantor et al., 2012; 0.86 in Cananéia, Brazil – Mello et al., 2019; 0.85 in Paraty, Brazil – Souza, 2013) and corresponds to that expected for adults in long-lived species (Wells, 1991). The apparent survival rate equal to 0.98 means a 2% loss of the adult population from one year to the next.

Although the apparent survival rate is high, a slow growth rate exposes the population to risks associated with small populations, such as fragmentation of social bonds, decrease of reproductive rates, and loss of genetic variability (Belovsky, 1987; Nunney & Campbell, 1993). Anthropogenic impacts on coastal ecosystems, especially bays and estuaries, have increased in recent years throughout the Guiana dolphins' distribution area. In the Guanabara Bay, in south-eastern Brazil, the population is in decline, with only 39 individuals estimated in 2015 compared to the 62 individuals estimated in 2000 (Azevedo et al., 2017). In Caravelas, north-eastern Brazil, the population is also small but apparently stable, ranging from 57 to 124 dolphins (Cantor et al., 2012). In Babitonga Bay, the main threats to dolphin populations are accidental capture in fishing nets, noise disturbance, boat traffic and contaminants of the marine environment (Cremer et al., 2017). Considering that the accidental capture in fishing nets is the main pressure on this population, affecting mainly the juveniles, management measures are needed, involving the fishing activity to help with the dolphins conservation. Management measures should mainly include reducing effort with gillnets, the main fishing device responsible for accidental catches in the area (Pinheiro & Cremer, 2004); the use of pingers in the area should be considered with great caution, as they can cause habitat loss (Dawson et al., 2013; Kyhn et al., 2015). Although accidental captures represent

the main direct threat, changes in habitat caused by port activities must also be assessed with restriction due to the great impact they can cause on dolphin populations in the region (Cremer et al., 2017), with long-term chronic effects that can be difficult to detect in time.

In Babitonga Bay the apparent population stability should be interpreted with caution. It is important to be aware of the loss of young and calves, considering the late age of sexual maturity of the species and the small size of the population. An important advance in understanding the real situation of this population would be the increase of the number of identifies individuals and the inclusion of the juveniles in the analyzes, which could be done using also the short-term marks, such as scratches, in samples concentrated in time, as proposed by Maieski et al (2020). Considering that the proportion of marked individuals in the population directly affects the abundance estimates, it is important that the field team is cohesive and constantly trained in order to improve the counting of the group size and that the photographs are taken by experienced people and with quality photographic equipment. It is also important that data collection is maintained in a systematic manner, in order to avoid gaps in population monitoring and improve the accuracy of population parameters estimates.

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CAPÍTULO 2:

SOCIAL PATTERNS IN GUIANA DOLPHINS (*SOTALIA GUIANENSIS*) FROM SOUTHERN BRAZIL

Abstract: We described the social structure of Guiana dolphins in southern Brazil, by combining sex, home range and social association among individuals. We analyzed photo-identification data from April 2006 to December 2017. Skin samples were collected from 22 individuals (6 males 16 and females). Overlap average of home range between two individuals was 40%. The dolphins showed low association indices (average HWI = 0.09) and the MRQAP model and Mantel test suggested that only the home range overlap was correlated with the social associations. Our results reinforce that this population is organized in a fission-fusion dynamics, whose dyadic associations are weak and short-lived, and related to the degree of similarity in space use.

KEYWORDS: Biopsy, fission-fusion, genetics, home range overlap, photo-identification, social patterns, *Sotalia guianensis*.

INTRODUCTION

Social animals live together and interact, forming complex social relationships and structures (Wey, Blumstein, Shen, & Jordán, 2008). For most social species, the composition of groups is dynamic and individuals move between groups, leading to the formation of a highly interconnected social network (Foster et al., 2012). Investigation into the sociobiology may therefore not only provide opportunities to study responses to anthropogenic disturbance, but can also inform conservation and management initiatives (Lusseau 2007). The social structure can be correlated to ecological and biological parameters, such as genetics, survival, foraging strategies and mating system of the population (Whitehead & Van Parijs, 2010).

Most dolphin species live in societies with fission-fusion dynamics, where the size, composition and cohesion of groups undergo frequent variations (Aureli, Schaffner & Boesch, 2008; Connor, Mann, Tyack & Whitehead, 2000). However, long-standing associations can be found in some species, either related to consanguinity, such as the formation of matrilineal groups of *Orcinus orca* (Bigg, Olesiuk, Ellis, Ford & Balcomb, 1990) and *Physeter macrocephalus* (Whitehead, Waters & Lyrholm, 1991); or to the sex of individuals, such as alliances between males in *Tursiops truncatus* (Parsons et al., 2003) and in *Tursiops aduncus* (Connor & Krützen, 2015).

The Guiana dolphin (*Sotalia guianensis*) is a small delphinid endemic to the western coast of the South Atlantic Ocean. Studies on Guiana dolphins throughout its distribution range have revealed some key features of the social system of this species. For example, genetic analyses of multiple populations from mitochondrial and nuclear markers suggested that females are the philopatric sex, while males disperse (Cunha 2007; Cunha, Da Silva & Solé-Cava, 2010) although there is no evidence of these displacements. Moreover, most

populations seem to experience high fission-fusion dynamics, in which social groups form and disband at multiple spatial-temporal scales (Beirão-Campos, Cantor, Flach & Simões-Lopes, 2015; Cantor et al., 2012; Lunardi & Ferreira, 2014; Santos & Rosso, 2008) while the populations themselves also change in composition and size (Cantor, Wedekin, Daura-Jorge, Rossi-Santos & Simões-Lopes, 2011; Cantor et al. 2012). Home range studies have described the specie range pattern and habitat use throughout its distribution (Rossi-Santos et al., 2006; Azevedo et al., 2007; Wedekin et al., 2007; Paitach et al., 2017), providing significant insights on social organization and interaction, but there is no information on the relationship of home range and the characteristics of the individuals. No study in *Sotalia* to date has analyzed the social dynamics of the species by combining these data streams linking association rates among photo-identified and genetically-sexed individuals over time with their home range. This integrative approach enables one investigates the contribution of sex and home range overlap in forming dyadic social associations and shaping the emergent social structure (e.g. Diaz-Aguirre, Parra, Passadore, & Möller, 2019; Hunt, Allen, Bejder & Parra 2019; Machado et al. 2019), which remains to be done for Guiana dolphins.

We describe the social structure of a resident Guiana dolphin population from the Babitonga Bay, southern Brazil. Combining individual photo-identification, spatial and genetic data, we seek to understand social associations among individual dolphins in relation to sex and home range overlap. We test the hypothesis that these dolphins follow the fission-fusion dynamics pattern due to the small size of the population, and that they are correlated with same-sex individuals with whom they share their home range.

METHODS

Study area

Babitonga Bay is located on the northern coast of Santa Catarina, southern Brazil ($26^{\circ}02' - 26^{\circ}28'S / 48^{\circ}28' - 48^{\circ}50'W$) (Figure 1). The region has one of the largest and most important mangrove formations in southern Brazil (IBAMA 1998). With about 24 islands in its interior, in addition to slabs and tidal flats, it has a water depth of 153.7 km². Its banks are formed predominantly of mangroves, but also sandy beaches and rocky shores. Babitonga Bay is characterized by the dominance of the flood tide (Truccolo & Schettini, 1999) and is sheltered from the waves, with calm and warm waters. The bay is surrounded by six cities, including the largest industrial park in the state and totaling more than 600,000 inhabitants, which represent a significant anthropic pressure on the system.

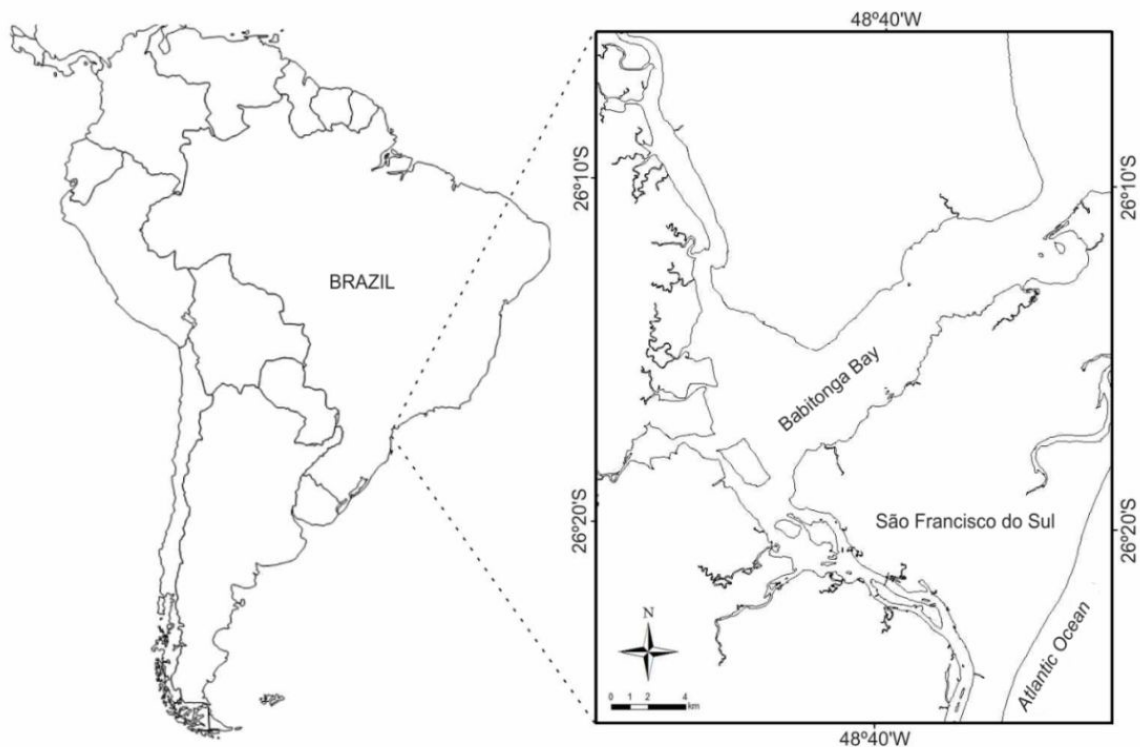


Figure 1. Study area: Babitonga Bay, on the northern coast of Santa Catarina, southern Brazil.

Photo-identification and data collection

Digital photographs of Guiana dolphins' dorsal fins were obtained from 2006 to 2017. The photo-identification technique allows recognizing each individual by comparing natural marks present in the dolphins' dorsal region (Würsig & Jefferson, 1990). Two vessels were used to obtain the records: a 5.5 meter long aluminum boat with a 60 Hp outboard motor and a 6.2 meter long inflatable boat and a 200 Hp outboard motor. Predetermined routes were followed and only in good navigable conditions, favorable to the observation of groups of dolphins (sea condition between 0 and 2 on the Beaufort scale). A group was considered to be all individuals spotted within the same area and performing the same activity. In each group found, we tried to photograph all individuals, without preferences between marked and unmarked individuals, within a maximum time of 30 minutes. During image acquisition the geographical location (GPS - Global Positioning System) and the number of individuals in each group were also recorded.

The photographs were screened for selection (sharpness, light, proximity and angle of the dorsal fin). The individuals were identified visually, without the aid of identification programs, only by permanent or long-lasting natural marks, such as nicks, scars and mutilations (Würsig & Jefferson, 1990). Only photo-identified individuals with at least two long-lasting marks with sufficient size to be easily identified were included in the catalog, thus reducing identification errors such as false positives and false negatives (Gunnlaugsson & Sigurjónsson, 1990).

Biopsy sampling

Skin samples were collected from photo-identified individuals in the population from 2016 to 2018. These samples were collected through biopsies performed using a 120-pound crossbow

and CETA-Dart darts with tips specifically developed for use in small cetaceans (8x25mm, stainless steel). The focal group methodology was used (Lehner, 1996), which consists of visually monitoring a group for a certain period of time; in this case, until it was possible to detect photo-identified individuals and they assumed a stable behavior that would allow a closer approach to the vessel. The dart was fired at a distance between 5 to 10 meters from the animal, perpendicular to the dolphin, at the same time that a second researcher took a photograph of the dorsal fin of the individual being sampled. After collection, the samples were kept in absolute alcohol until analysis in the laboratory.

Sexing

To identify the sex of the individuals, sex-specific nuclear markers ZFX/ZFY were amplified by PCR using the primers described by Berube and Palsbøll (1996), as validated and optimized for the species (Cunha & Solé-Cava, 2007). The reactions were performed in 15 μ L, containing 1U of Taq polymerase, 0.2 mM dNTPs, 2.5 mM MgCl₂, 1 μ g / μ L BSA and 0.5 μ M of each primer and 1 μ L of DNA (5-50ng). The amplifications were performed in an automatic thermocycler (Veriti - AppliedBiosystems) following the cycling profiles of Cunha and Solé-Cava (2007). Positive and negative controls were included in all amplifications. The fragments were separated on 2% agarose gel in 0.5X TBE buffer, later stained with ethidium bromide. The bands were visualized through UV-light and the gel was photographed.

Home range overlap

The Kernel density estimator is considered an efficient method to determine home range in cetacean studies (Seaman & Powell, 1996). It is a method that takes into account the intensity of samples per unit area (Laver & Kelly, 2008). The home range of the individuals was

calculated by the Kernel 95% method and the radius of influence was selected by the Least Square Cross-Validation method (Seaman & Powell, 1996) in the ArcGis 9.3 program. The home range was calculated for all photo-identified individuals with at least 10 sighting records on different dates (in the case of re-sightings on the same day, only the first record was considered). The percentage of home range overlap between individuals (HRO) was

calculated as: $HRO_{ij} = \left(\frac{hr_{ij}}{hr_i} \right) \cdot \left(\frac{hr_{ij}}{hr_j} \right)$, where hr_i is the home range of individual i , hr_j is the home range of individual j and hr_{ij} is the area of overlap of the two areas (Silva et al. 2008). Land home range areas were excluded.

Social patterns

To describe social patterns among identified Guiana dolphins, we calculated association indices based on group-membership data, that is, assuming that co-occurrence in a group implies that individuals can interact with each other (the gambit of group; Whitehead & Dufault, 1999). The association between pairs of individuals was estimated using the Half-Weight Index (HWI) (Cairns & Schwager, 1987). This index represents the proportion of time that a pair of individuals is observed in the same group, in relation to the time they were

observed in different groups, defined as: $HWI = x / \left(x + yab + \left(\frac{ya + yb}{2} \right) 2 \right)$ where x is the number of times individuals a and b were sighted in the same group; yab is the number of times that individuals a and b were observed separately; ya and yb are the number of times that only the individual a or b , respectively, were observed. Only individuals with at least five photographic records were included in order to avoid spurious associations (Whitehead 2008).

The large-scale structure of a network was described using density (i.e. the proportion of the realized links), clustering coefficient (i.e. the degree to which the individuals tend to cluster together), and modularity (i.e. the tendency of the network to be structured into cohesive subgroups of individuals that are more connected to each other than with the rest of the network (see Whitehead 2009)).

We performed Monte Carlo permutations to test the null hypothesis that individuals associate randomly against the alternative that there are preferred or avoided associations between sample periods (Bejder, Fletcher & Bräger, 1998.). The group membership of individuals seen more than five times was randomized (20,000 interactions) using a swapping algorithm that kept key features of the original data constant: group size, individual frequency of observation, and total number of observations (Bejder et al., 1998). The permutations were restricted within secondary capture occasions (one month) to avoid bias due to demographic changes across occasions (Whitehead, 1999) and due to differences in the sampling effort over the entire study. For each of the 20,000 permuted group-membership matrices, a new HWI association matrix was generated, from which the coefficient of variation (CV) was calculated. The CV of the observed HWI association matrix was then compared to this theoretical distribution of CV of the permuted data. The null hypothesis was rejected when the CV of the observed HWI was significantly higher than the CV of the permuted matrices, suggesting that the observed associations were more variable than expected by chance. Unexpectedly high HWI values suggest preferred associations, while unexpectedly low values suggest avoided associations between sample periods (Whitehead, 2008).

Temporal patterns in associations were evaluated using Standardized Lagged Association Rates (SLAR), which estimates the probability, $g'(t)$, that two individuals seen together in the same group at a given time t will remain associated at a subsequent time, $t + 1$

(Whitehead, 1995). We fitted four exponential decay models to the association data to describe the population-level variation, if any, of association probability over time (Whitehead, 1995; Whitehead, 2008). In the SLAR1 model, the average association rate between individuals remains constant over time, representing permanent associations. The SLAR2 model describes rapid dissociations, where the association rate drops to zero after a given time lag and does not recur. The SLAR3 describes an association rate that drops during a short time lag then levels off, suggesting the combination of rapid dissociations with some stronger associations. Finally, the SLAR4 model describes two levels of dissociation, one after a shorter and another after a longer time lag. As a benchmark, we also fitted the Standardized Null Association Rate (SNAR) representing the expected rate if the associations were random (Whitehead, 2007). The models were ranked by quasi-Akaike-Information-Criteria (QAIC), which compensates for data overdispersion (Burnham & Anderson, 2002). The standard errors of the SLAR and SNAR were calculated via jackknife resampling, omitting 30-day periods at each iteration. Social analyses were conducted in SOCPROG 2.8 program (Whitehead, 2009).

Correlations of association index with home range and sex

To evaluate the relationship, if any, between the social associations and the other pairwise traits, we built Multiple Regression Quadratic Assignment Procedure (MRQAP) models using the double-semi-partial method and 20,000 iterations (Dekker, Krackhardt & Snijders, 2007). We described the association among pairs of individuals (HWI) in function of their home range overlap and sex similarity. The construction of the models was done in two steps, aiming to use the largest number of samples as possible. First, we used the most complex model, that included all the variables (HWI, Sex and HRO). Then, we evaluate the

significance of each variable in the model, and those that not showed significance were excluded in the next model. To test the correlation between pairs of variables, the Mantel test was performed with 20,000 mutations o test the significance of the result. This test calculates the correlation between two matrices by making numerous permutations on the values of these matrices and calculates the value of R (correlation) in each permutation. Thus, it is possible to test whether the real R is different from chance (Whitehead 2009; Smouse et al 1986). To evaluate the effect size, we used Pearson's correlation (Smouse, Long & Sokal, 1986; Whitehead, 2009). All correlation analyses were performed using the SOCPROG 2.8 program.

RESULTS

We analyzed data from 384 groups of Guiana dolphins containing at least two individuals. From April 2006 to December 2017, 115 individuals were sighted. photo-identified and included in the catalog.

Twenty-two dolphins were biopsied. Skin samples were collected from six males and 16 females photo-identified Guiana dolphins.

Home range overlap

Home range area (Kernel 95%) was calculated for 55 photo-identified individuals, which were sighted on at least 10 capture occasions on different days. The average individual home range size was $35.4 \text{ km}^2 \pm 11.7 \text{ SD}$, ranging from a minimum of 15.5 km^2 to a maximum of 84.3 km^2 . Considering the 22 dolphins that were genetically sexed, the average home range of the eight photo-identified females with at least 10 records was $32.1 \text{ km}^2 \pm 5.01 \text{ SD}$. The

average male home range could not be calculated, as only one biopsied male obtained at least 10 records (# 054, area = 49.9 km²).

The average overlap of home range area between two individuals was 40%, with a maximum of 91% between individuals # 019 and # 044 and a minimum of 9% between individuals #048 and #112. Considering the females, the average overlap of home range area with other individuals was 39%, and with other females was 40% (Table 2).

Table 1. Overlapping matrix considering the home range overlap between individuals (HRO) of photo-identified Guiana dolphin females in Babitonga Bay with at least 10 records. The highlighted values indicate the maximum and minimum overlap between two females. Mean = 0.40.

Females	#032	#045	#047	#057	#058	#059	#112
#026	0.31	0.27	0.64	0.64	0.51	0.30	0.52
#032	-	0.63	0.17	0.24	0.30	0.53	0.15
#045		-	0.21	0.31	0.38	0.60	0.16
#047			-	0.69	0.46	0.23	0.58
#057				-	0.50	0.36	0.53
#058					-	0.36	0.37
#059						-	0.13

Association between individuals

Of the 115 photo-identified individuals, 76 were photographed at least five times between 2006 and 2017 and their associations were analyzed. About 37% of all possible pairs among these individuals (n=2,850) were never seen in the same group. Overall, the Guiana dolphins from the Babitonga Bay showed low association indices (average HWI = 0.09, \pm 0.1 SD, median = 0.06, mode =0.00, range = 0.00- 0.81).

The permutation test indicated that the coefficient of variation of the observed association data was greater than the null expectation (observed HWI CV = 1.065; random HWI = 0.823; p = 0.001). This greater variation among the observed associations suggested

the existence of preferred and avoided associations during the sampling period—that is, that some pairs were observed in the same group more and less often, respectively, than expected by chance.

The SLAR reinforced that most associations were ephemeral, with about five ($a_1 = 5.1 \pm 13.46$ SE) dissociations per day (Table 3). The probability of two individuals remaining associated decreased over time, with a significant low chance of individuals remaining associated after about three years, when the association rates were similar to the null expectation (Figure 3).

The social network was densely connected (63% of realized links), and with 0,18 clustering coefficient ($SD = 0.02$). The low modularity, $Q = 0.186$ did not suggest that any partition into subsets of individuals are more connected to each other than the rest of the population.

Table 2. Standardized Lagged Association Rate (SLAR), $g'(t)$, models ranked by the lowest quasi-Akaike Information Criteria (QAIC). Δ QAIC, QAIC weights and Likelihood indicate the relative support for each model.

	Model	QAIC	ΔQAIC	QAIC weights	Likelihood
SLAR4	$g'(t) = 1.36 * e^{(-5.11 * t)} + 0.03 * e^{(-0.0003 * t)}$	139380.06	0	0.51	1.00
SLAR2	$g'(t) = 0.03 * e^{(-0.0003 * t)}$	139380.16	0.1	0.49	0.95
SLAR3	$g'(t) = 0.02 + 0.05 * e^{(-1.23 * t)}$	139941.22	561.16	0.00	0.00
SLAR1	$g'(t) = 0.02$	139953.58	573.52	0.00	0.00

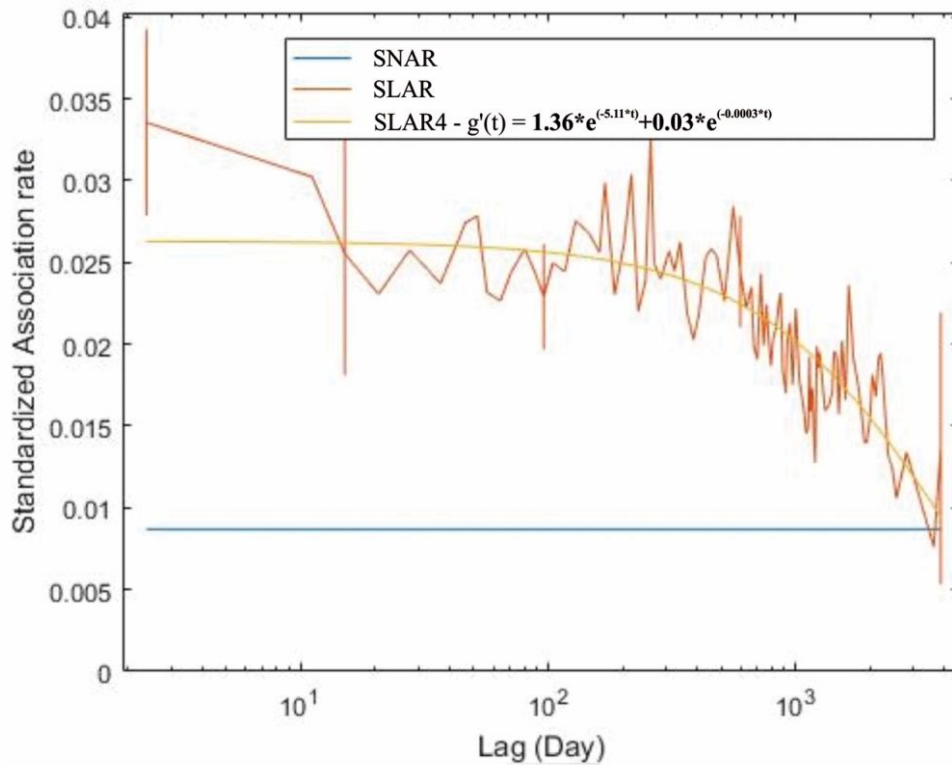


Figure 2. Standardized Lagged Association Rate (SLAR) (orange line) for all individuals, suggesting rapid dissociations over time. The blue line represents the null association rates (SNAR), should individuals associate randomly. The yellow line represents the model that best fitted the data (Table 3). Vertical bars indicate jackknifed standard errors.

HWI, HRO and sex correlation

The MRQAP model and Mantel test (table 4) suggested that only the home range overlap was correlated with the social associations.

Tabela 3. Multiple Regression Quadratic Assignment Procedure (MRQAP) and Mantel test. HWI = Half Weight Index; HRO = Home Range Overlap; SEX = Sex of Individuals.

Test	Dependent variable	Predictors	Regression Coefficient	P	Sample Size
MRQAP	HWI	HRO	0.7670	0.000	9
		SEX	0.4266	0.1158	9
Mantel	HWI	HRO	0.4556	0.000	55

DISCUSSION

We described the social patterns of Guiana dolphins in Babitonga Bay, southern Brazil, by combining genetics, home range and social association among individuals. Our results reinforce that social structure of Guiana dolphins is typically influenced by high fusion-fission dynamics. Associations were overall weak, short-lived and more influenced by similar ranging behavior than sex similarity.

All studies on social patterns of Guiana dolphin populations indicate low or moderately low association rates among individuals. The average association index among the individuals in Babitonga Bay, 0.09, was lower than that of other populations (Caravelas: average HWI = 0.167; Sepetiba = 0.21; Cananéia = 0.15) (Beirão-Campos et al., 2015; Cantor et al., 2012; Santos & Rosso, 2008). In addition, we found that most of these associations tend to be short-lived, with a few preferred pairs of individuals seen more often than expected (see also Cantor et al., 2012). Babitonga Bay has a small population of Guiana dolphins, what may be influencing the HWI through demographic process. The interplay between demographic processes and social behavioral (how the loss and gain of individuals affects social interactions, and how social interactions affect survival, reproduction, or movement) may have a profound impact on social networks (Shizuka & Johnson, 2019).

A small home range spatially approximates individuals, increasing the opportunities for all individuals of the population to interact. In species with high fidelity to the area, the social context is highly related to the use of the habitat (Best et al., 2013; Vezuela-Sánchez, Harding, Cunningham, Chirgwin & Soto-Azat, 2014). Individuals with overlapping home range may have more opportunities to interact and thus associate with each other more often than with those who do not have home range overlap (Best et al., 2014). In Babitonga Bay,

the home range overlap is the variable that best explains the association between individuals, with individuals making intense use of the central region of the bay, the core area of the population, known to be an important feeding area for the population (Cremer et al., 2017). The most likely explanation is that the association between individuals is a consequence of sharing the same area, mainly for foraging activities, since large aggregations, with more than 40 individuals, are frequently observed in fishing activity. The majority of the population tends to remain aggregated in this manner and concentrated within the same region, at least for part of the time (Cremer et al., 2017).

The densely connected social network found in this study, did not have any partition into subsets of individuals who could suggest that are more connected individuals to each other than the rest of the population. The fission-fusion dynamic is the norm, but preferential associations were found that are not explained by gender between individuals, as expected.

In conclusion, this study reveals that the Guiana dolphin population of Babitonga Bay is organized in a population with fission-fusion dynamics, whose dyadic associations are weak and short-lived, and related to the degree of similarity in space use. Fission-fusion dynamics has profound implications for the transmission of disease and socially transferable information. A small population with weak and short-lived associations makes this population particularly vulnerable to anthropogenic pressures, and thus, increases the need to protect the habitat where it is inserted.

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3. CONCLUSÃO GERAL

Em nível de população, este estudo levantou importantes informações para a conservação do boto-cinza, como dados genéticos, estimativas de abundância e sobrevivência e, a primeira estimativa de taxa de crescimento populacional da Baía Babitonga. Para essa espécie, este é o primeiro estudo combinando dados de foto-identificação, associação social, genética e área de vida de botos-cinza.

A população de botos-cinza da Baía Babitonga está em aparente estabilidade, mas é importante monitorar a mortalidade de juvenis e filhotes, considerando o longo período de maturidade sexual da espécie, o pequeno tamanho da população e as ameaças de origem antrópica presentes na região.

Assim como já detectado em outras localidades, os botos-cinza da Baía Babitonga estão organizados em uma dinâmica do tipo fissão-fusão, onde as associações são breves e fracas. Mas foi possível correlacionar a associação social entre os indivíduos com a similaridade no uso do espaço.

Considerando o atual cenário da região, com a presença da pesca artesanal, aumento da atividade portuária e, baixa abrangência do tratamento de esgoto das cidades ao redor da baía que juntas somam mais de 600.000 habitantes, é importante a continuidade do monitoramento desta população, a fim de detectar uma possível tendência de declínio populacional a tempo de se tomar medidas de manejo adequadas para a conservação do boto-cinza que se encontra na Lista de Espécies Ameaçadas de Extinção de Santa Catarina.