UFRGS
UNIVERSIDADE FEDERAL
DO RIO GRANDE DO SUL

PPGBAN
PROGRAMA DE PÓS-GRADUAÇÃO EM BIOLOGIA ANIMAL

INSTITUTO DE BIOCIÊNCIAS
PROGRAMA DE PÓS-GRADUAÇÃO EM BIOLOGIA ANIMAL

KARINA BOHRER DO AMARAL

INFLUÊNCIA DO AMBIENTE MARINHO NO PADRÃO DE DISTRIBUIÇÃO E NA ESTRUTURA GENÉTICA DE MAMÍFEROS MARINHOS PREDADORES DE TOPO DE CADEIA

PORTO ALEGRE
2018

KARINA BOHRER DO AMARAL

INFLUÊNCIA DO AMBIENTE MARINHO NO PADRÃO DE DISTRIBUIÇÃO E NA ESTRUTURA GENÉTICA DE MAMÍFEROS MARINHOS PREDADORES DE TOPO DE CADEIA

Tese apresentada ao Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências da Universidade Federal do Rio Grande do Sul, como requisito parcial à obtenção do título de Mestre/Doutor em Biologia Animal.

Área de concentração: Biologia Comparada

Orientador(a): Prof. Dr. Ignacio Benites Moreno

Co-orientador(a): Dr. Ana Rita Amaral
Prof. Dr. Nelson Fagundes

PORTO ALEGRE
2018

KARINA BOHRER DO AMARAL

INFLUÊNCIA DO AMBIENTE MARINHO NO PADRÃO DE DISTRIBUIÇÃO E NA ESTRUTURA GENÉTICA DE MAMÍFEROS MARINHOS PREDADORES DE TOPO DE CADEIA

Aprovada em _____ de _____ de _____.

BANCA EXAMINADORA

_____

Dr/a. Maria João Ramos Pereira

_____

Dr/a. Renan Maestri

_____

Dr/a. Juan Pablo Torres Florez

Dedico este trabalho aos meus pais, Marta e Carlos Alberto,
e a minha irmã, Caroline,
que me ensinaram a ser persistentes, lutar,
e, principalmente, acreditar que tudo seria possível
mesmo quando tudo parecia impossível
em vários momentos das nossas vidas.

[...]Longe se vai sonhando demais
Mas onde se chega assim
Vou descobrir o que me faz sentir
Eu, caçador de mim

Nada a temer
Senão o correr da luta
Nada a fazer
Senão esquecer o medo
Abrir o peito à força
Numa procura
Fugir às armadilhas da mata escura

Vou descobrir o que me faz sentir
Eu, caçador de mim

Sergio Magrão e Luiz Carlos Sá

AGRADECIMENTOS

Ao se concluir esta etapa, faz-se necessário agradecer:

- à minha família, em especial a minha mana querida (Caroline) por ser meu melhor e maior exemplo. Aos meus pais Marta e Carlos Alberto pelo amor, carinho, segurança e apoio. E, principalmente, por me ensinarem a acreditar em mim. Um agradecimento aos meus avós queridos, todos tios(as) e primos(as) que fizeram parte da minha construção como pessoa;

- à Pança, essa cachorrinha especial que enriqueceu a minha vida de todas as formas. Ela fez eu me reencontrar no pior momento da minha vida e me trouxe equilíbrio todas as vezes que eu precisei. Para mim, ela é o amor mais puro e verdadeiro.

- ao Murilo, que felizmente teve a oportunidade de me conhecer antes do doutorado (rsrsrs). E, ao longo desses anos, foi um parceiro incrível e essencial para a realização do Doutorado. Esteve sempre me dando apoio e fazendo eu acreditar que eu era capaz; me ensinou Inglês e, me ajudou a entender o mundo de outra forma. Realizou um dos meus maiores sonhos, aprender a surfar, e junto comigo cuida e compartilha Pança. Obrigada Namorzinho!

- à família do Murilo que, hoje, é como uma segunda família para mim. Um porto seguro;

- aos amigos mais queridos Bibiane Godoy, Valentina Zaffaroni e Filipe Michels Bianchi. A Bibi por ser uma irmã querida que a vida me presentou em 2006. A Valen por ser a minha parceira de luta e o Filipe por ser o meu exemplo de vida acadêmica, um pesquisador justo, correto e, mais importante que tudo: muito engraçado! Valen e Filipe obrigada por todos momentos incríveis que partilhamos nesses últimos anos, que só nós mesmos somos capazes de nos proporcionar (rsrsrs). Obrigada Filipe por nossos saltos de

para-quedas sem medo e nossos grãozinhos de areia. Valen, obrigada por estar comigo na luta pelos nossos sonhos e ser essa amiga impecável na academia e no coração.

- à família Labsmar que desde 2010 compartilhamos aprendizagens e experiências profissionais e de vida;

- aos amigos que conheci pelo mundo nestes quatro anos por causa dos golfinhos, em especial a Giovanna Figueiredo, Dalia Barregán-Barrera , Nohelia Farias, Raissa Miranda, Maura Sousa: obrigada pelos incríveis e maravilhosos momentos em São Francisco, Halifax, São Paulo, na minha casa e em Cambará;

- aos amigos queridos que fiz na Universidade de Michigan durante o doutorado sanduíche e que me ensinaram demais (sem eles essa tese não existiria!): Andrea Thomaz, Fernanda Zaidan, Jeronymo Dallapicola, Luciana Resende, Mariah Kenney, Joyce Prado, Renata Magalhães e todos demais membros do laboratório da Dr. Lacey Knowles;

- aos professores do PPGBAN/UFRGS, em especial a Prof. Dr. Maria João Ramos Pereira pelas significativas contribuições que foram fundamentais para meu crescimento ao longo desses quatro anos;

- aos "amigos orientadores" que foram parte fundamental da minha formação desde a Iniciação Científica : Juliano Boldo, Charley & Lívia e Larissa Heinzelmann;

- aos colaboradores pelo mundo, em especial Marcos César de Oliveira Santos e Paula Mendéz-Fernandez;

- em especial a Dr. Ana Rita Amaral, a melhor co-orientadora que existe no Universo! Sem ela, também NADA disso seria possível que literalmente me orientou com o Oceano Atlântico de distância!

- em especial também a Dr. Lacey Knowles que me deu a oportunidade de estudar quatro meses em uma universidade de primeiro mundo, em que o pesquisador tem, realmente, as condições necessárias para pesquisar!!!

- em especial, ao Prof. Ignacio, pela orientação, confiança, dedicação, paciência, conselhos e amizade ao longo desses 8 anos de parceria. E, principalmente, por tudo que me ensinou, as gentis e grandes oportunidades que me deu, como a bolsa de IC para fazer a tal "modelagem de nicho ecológico" em 2010...nunca se sabe até onde uma porta aberta pode levar/transformar alguém...esta porta me levou até aqui e eu só tenho a te agradecer por toda essa trajetória!

SUMÁRIO

RESUMO

Duas espécies de cetáceos apresentam padrões de distribuição peculiares ao longo da costa brasileira, muito provavelmente em resposta às condições hidrográficas e topográficas que ocorrem entre 20 e 33°S. A primeira espécie, a franciscana ou toninha (*Pontoporia blainvillei)*, é um golfinho de distribuição restrita do Brasil até a Argentina, que ocorre primariamente na plataforma continental interna, raramente ultrapassando os 50 m de profundidade. Já a segunda espécie, o golfinho-pintado-do-Atlântico (*Stenella frontalis)*, é um golfinho de distribuição restrita ao Oceano Atlântico, que ocupa principalmente a plataforma continental. Estas duas espécies apresentam hiatos ao longo da sua distribuição no Brasil que tem consequências na morfologia e estrutura genética das espécies. Através da aplicação de diferentes métodos, o principal objetivo deste estudo foi investigar a influência do ambiente marinho no padrão de distribuição e na estrutura genética destas duas espécies com ênfase na costa brasileira. No primeiro capítulo, investigou-se a relação do ambiente marinho com o padrão de distribuição da franciscana. Para tanto, uma revisão e atualização da distribuição das áreas de manejo da franciscana (FMA), e dos limites dos hiatos, ao longo do Brasil foram realizadas. Análises de nicho ecológico sugerem que os hiatos fazem parte do nicho fundamental da franciscana que seriam, portanto, relativamente adequados para a espécie. No entanto, o estreitamento da plataforma continental parece ser o principal fator que explica a ausência da espécie nos hiatos e, inclusive poderia explicar a diferenciação genética entre algumas FMAs. No segundo e terceiro capítulos, a relação entre similaridade genética e distâncias geográficas e ambientais foram investigadas para o golfinho-pintado-do-Atlântico em duas escalas: ao longo de praticamente toda distribuição e em uma escala

mais restrita com ênfase no Brasil. Populações geneticamente distintas ao longo de toda distribuição da espécie foram identificadas com base em um marcador mitocondrial, que podem ser resultado Isolamento por Distância e Isolamento por Resistência, relacionados tanto com condições ambientais contemporâneas quanto do passado (Último Glacial Máximo). As análises de estrutura populacional do golfinho-pintado-do-Atlântico no Brasil, investigada mais profundamente com marcadores genômicos, indicam ao menos a existência de três populações (Brasil, Colômbia e Oceânica) suportanto, portanto, a hipótese de uma população isolada no sudeste do Brasil. De forma geral, conclui-se que o ambiente marinho e, principalmente, fatores como extensão da plataforma continental, batimetria e temperatura tem um papel fundamental para explicar o padrão de distribuição destas espécies no Brasil. Além disso, outros processos podem estar envolvidos na estruturação genética do golfinho-pintado-do-Atlântico e também da franciscana como, por exemplo, estrutura social, filopatria e a história evolutiva destas espécies. O maior desafio para conservação da franciscana é seu status de Criticamente Ameaçada no Brasil e, em relação ao golfinho-pintado-do-Atlântico é a deficiência de dados. Uma vez que ambas espécies ocorrem na porção mais desenvolvida do país, os resultados aqui obtidos têm impacto direto na conservação destas espécies, porque trazem informações que podem ser utilizadas em planos futuros de conservação e manejo.

PALAVRAS-CHAVE: modelagem de nicho ecológico, genômica, golfinhos, franciscana, golfinho-pintado-do-Atlântico

ABSTRACT

Along Brazilian coastal waters, either franciscana and Atlantic spotted dolphins exhibited distributional gaps, which is most likely resulting from changes in the environmental features between 20 and 33°S. The former species, franciscana (*Pontoporia blainvillei)*, is a river dolphin with restricted distribution from Brazil to Argentina, recorded mainly up to 50 m deep over the inner shelf. The second species, Atlantic spotted dolphin (*Stenella frontalis*), is a delphinine dolphin distributed across the Atlantic Ocean, being mainly recorded over the continental shelf. The distribution patterns that these species showed in Brazil have a direct influence on the morphology/ecology and genetic struture of both species. Different approaches were applied to address the main goal of this study, which was investigating the influence of marine environment in shaping the distribution pattern, as well as genetic strcuture of franciscana and Atlantic spotted dolphin with emphasis in the Brazilian coastal waters. In the first chapter, I investigated the franciscana distribution in Brazil using an ecological niche modeling approach. In order to do that, I performed a review of records of the species along Brazial and, updated the limits of franciscana management areas (FMAs) and distributional gaps. The results suggested that gaps are within franciscana fundamental niche and, therefore, both gaps would be suitable for franciscana. However, the narrow of continental shelf seems to be the main factor inhibiting the presence of franciscana in these areas. Furthermore, the narrowing of continental shelf play a role to explain the genetic differentiation among FMAs. In the second and third chapters, the relationship between genetic distances and geographic and environmental distances were investigated both in a restrict and a broad scale. I found genetically distinct populations

across Atlantic spotted dolphin distribution based on mtDNA, that are probably resulting of Isolation-by-Distance and Isolation-by-Resistance related both with contemporary and past conditions (e.g. Last Glacial Maximum). Furthermore, I investigated population struture using genomic markers (Single Nucleotide Polymorphisms, SNPs) across Western South Atlantic, Caribbean and Eastern Atlantic. The results suggested at least three different populations, and therefore, confirmed previous hypothesis of an isolated population in the southeastern Brazil. Overall, I concluded that marine envinronment, especially the extension of continental shelf, bathymetry and sea surface temperature, are the main factors that explaning the distribution pattern of franciscana and Atlantic spotted dolphin in Brazil. Besides that, other process such as, social structure and phylopatry, as well as biogeographical process might be investigated in further studies. Franciscana is considered "Critically endangered" in Brazil, and Atlantic spotted dolphin has not enough data to determine its conservation status. Since both species are recorded in the most developed region of the country with high anthropic pressure, my results could help in future management and conservation plans for both species in a regional scale.

KEY-WORDS: ecological niche modeling, genomic, dolphins, franciscana, Altantic spotted dolphin

INTRODUÇÃO GERAL

A distribuição geográfica de uma espécie é a manifestação de complexas interações entre as características intrínsecas de um organismo (principalmente, suas tolerâncias ambientais, os recursos que necessita, história de vida, parâmetros demográficos e, capacidade de dispersão) com as características do ambiente (principalmente, aquelas que variam no espaço e tempo e que tem influência na limitação da distribuição de uma espécie)(BROWN; STEVENS; KAUFMAN, 1996). As consequências dessas interações influenciam todas as características da distribuição geográfica de uma espécie: tamanho, forma, limites, e estrutura interna (BROWN; STEVENS; KAUFMAN, 1996). Dessa forma, o tamanho da distribuição geográfica de uma espécie e como ela varia ao longo do tempo é uma das principais características ecológicas e evolutivas de uma espécie (BROWN; STEVENS; KAUFMAN, 1996). Além disso, o tamanho da distribuição geográfica de uma espécie é um forte preditor do risco de extinção de uma espécie, uma vez que espécies com distribuição restrita são mais vulneráveis que espécies com ampla distribuição (GASTON; FULLER, 2009a).

Descrever precisamente e entender os processos que determinam a distribuição dos organismos é um problema fundamental em ecologia, com importantes aplicações em conservação e manejo (GASTON; FULLER, 2009b; PEARSON, 2007). Uma vez que as espécies estão intimamente relacionadas com o ambiente que elas ocupam, compreender os padrões de distribuição de uma espécie e, se possível, delimitar sua distribuição são etapas fundamentais para seu completo conhecimento. Estudos biogeográficos possibilitam o entendimento de padrões que influenciam na divergência populacional e

especiação, além de auxiliar na identificação de processos que estruturam a diversidade de organismos em uma variedade de escalas geográficas e taxonômicas.

Os cetáceos são predadores marinhos de topo de cadeia com grande capacidade de dispersão. Cetáceos fazem parte da ordem Cetartiodactyla (AGNARSSON; MAY-COLLADO, 2008), que se caracterizam por uma variedade de formas e adaptações aos mais diversos ambientes aquáticos. Atualmente dentro de Cetacea são reconhecidas aproximadamente 90 espécies (Committee on Taxonomy 2014), que se distribuem amplamente no ambiente marinho, desde regiões tropicais a polares, com algumas espécies ocorrendo em bacias hidrográficas (MCGOWEN; SPAULDING; GATESY, 2009; STEEMAN et al., 2009). Cetacea está dividido em dois grupos monofiléticos: Mysticeti e Odontoceti (MCGOWEN; SPAULDING; GATESY, 2009). Mysticeti é representado pelas baleias com cerdas bucais, ao passo que Odontoceti é representado pelas "baleias com dentes" (*e.g.* orca, baleia-piloto), botos e golfinhos.

Através da perspectiva da história natural, os cetáceos são um grupo completamente adaptado ao meio aquático e, consequentemente tridimensionalmente conectados. Dessa forma, explicar a distribuição de animais marinhos é frequentemente desafiador nesse "mundo 3D". Este desafio é a particularmente real para os cetáceos, já que estes são animais geralmente grandes, altamente móveis, endotérmicos, homeotérmicos, e presumivelmente euritérmicos, podendo ser encontrados virtualmente em qualquer ambiente aquático. No entanto, mesmo com tantas adaptações muitos cetáceos exibem padrões claramente delimitados de distribuição, sendo poucos considerados "cosmopolitas" (DO AMARAL et al., 2018).

São muitos os fatores que afetam a distribuição dos cetáceos, entre eles: fatores demográficos (abundância, idade e estrutura sexual das populações, status reprodutivo e ciclo de vida dos indivíduos), fatores evolutivos (morfologia, fisiologia, adaptações comportamentais), fatores ecológicos (produção biológica, uso e distribuição de presas, predadores e competidores), fatores ambientais (temperatura da água, salinidade, densidade, profundidade da termoclina, tipo de substrato e batimetria) e fatores antropogênicos (poluição, capturas acidentais ou diretas, efeitos sonoros) (PALACIOS et al., 2013; REDFERN et al., 2006).

Para os grupos atuais de cetáceos está bem estabelecido que a distribuição das espécies relaciona-se intimamente tanto com características hidrográficas, quanto com características fisiográficas dos oceanos. Estes dados oceanográficos são os principais delimitadores das espécies de presas e consequentemente da distribuição dos cetáceos, uma vez que o habitat é primariamente delimitado pela disponibilidade de alimento (BAUMGARTNER; MULLIN; MAY, 2001). Portanto, a estrutura, comportamento e distribuição global de muitos cetáceos viventes está fortemente ligada a disponibilidade de alimento e, por sua vez, massas de água e padrões climáticos globais.

Relações significantes entre variáveis topográficas, como batimetria e gradientes de profundidade e a distribuição de populações de cetáceos foram observadas para muitas espécies de cetáceos (e.g. BAUMGARTNER; MULLIN; MAY, 2001; DANILEWICZ et al., 2010). A influência primária do meio físico sobre a distribuição dos cetáceos é devido, provavelmente, a agregação de presas. A distribuição de espécies de presas bentônicas ou demersais é diretamente limitada pela fisiografia através da profundidade e/ou seu gradiente e o tipo de substrato. Já para espécies de presas, como peixes pelágicos

ou cefalópodes, a fisiografia atua indiretamente, através de mecanismos que são induzidos pela topografia, como a ressurgência de nutrientes. Tais mecanismos levam ao aumento da produção primária e agregação de zooplâncton, levando ao aumento da produção secundária (PALACIOS et al., 2013; REDFERN et al., 2006).

As características hidrográficas, como temperatura da água, salinidade, concentração de clorofila *a*, entre outras, são importantes características que estão correlacionadas com a distribuição dos cetáceos, uma vez que podem secundariamente afetar a disponibilidade de presas. A temperatura e salinidade da água são consideradas importantes variáveis oceanográficas, uma vez que influenciam diretamente a vida marinha. E, a clorofila *a* representa o status trófico da superfície das águas (e.g., BALLANCE; PITMAN; FIEDLER, 2006; DAVIES, 1963; SÃO PEDRO et al., 2015; XU et al., 2013).

Nos últimos anos, o tradicional conceito de grandes e homogêneas populações de organismos marinhos tem sido transformado pela genética/genômica molecular (KELLEY et al., 2016). A ausência de barreiras físicas óbvias nos oceanos e o alto fluxo gênico entre populações desafiam os modelos de especiação alopátrica. No entanto, extensiva estruturação genética tem sido recuperada, sugerindo uma dinâmica complexa no recrutamento de espécies marinhas. Por esses motivos, a especiação marinha é considerada um paradoxo (BIERNE; BONHOMME; DAVID, 2003; HAUSER; CARVALHO, 2008).

A identificação de barreiras geográficas no ambiente marinho é um desafio, já que elas não são tão óbvias quanto no ambiente terrestre (DO AMARAL et al., 2018). Uma vez que muitas espécies marinhas apresentam ampla e rápida dispersão e assume-se

que as correntes e passagens oceânicas permitem uma constante mistura de *pools* gênicos, inibindo mudanças evolutivas. Assim, o requerimento de isolamento durante especiação alopátrica parece ser mais difícil de ser satisfeito nos oceanos (NORRIS, 2000; STEEMAN et al., 2009), embora a especiação também possa ocorrer em simpatria. No entanto, especializações intraespecíficas durante o forrageio parecem ser importantes delimitadores do fluxo gênico entre populações simpátricas e parapátricas (HOELZEL, 1998). Diversos estudos têm revelado estruturação populacional em fina escala como resultado de divergência ecológica em espécies consideradas cosmopolitas como ocorre, por exemplo, com os golfinhos-nariz-de-garrafa (*Tursiops truncatus*) e as orcas (*Orcinus orca*) (MORIN et al., 2015; SEGURA-GARCÍA et al., 2018; TEZANOS-PINTO et al., 2008).

Em relação aos cetáceos, espécies associadas a um habitat específico, forageio, especializações e interações sociais e culturais, em combinação com processos demográficos como efeito fundador e contrações/expansões populacionais podem acarretar em relações descontínuas entre distâncias genética e geográficas ((FOOTE et al., 2016; MORIN et al., 2015; SEGURA-GARCÍA et al., 2018; VACHON; WHITEHEAD; FRASIER, 2018; WHITEHEAD, 2017). Por exemplo, diferenças e mudanças em correntes, ambientais, na distribuição de presas e comportamento foram já identificadas como fatores que influenciam a estrutura genética de algumas espécies de golfinhos da família Delphinidae (FOOTE et al., 2016; MÖLLER et al., 2011).

No hemisfério sul, a maior diversidade de cetáceos ocorre no Oceano Atlântico, onde pelo menos 57 espécies têm registros confirmados (DO AMARAL et al., 2018). Na costa brasileira, são registradas 46 espécies de cetáceos, sendo que nas regiões sul e

sudeste concentram-se o maior número de registros (n=44). Esta região de alta diversidade de cetáceos, localizada aproximadamente entre as latitudes de 20° e 33°S, engloba dois grandes sistemas oceanográficos a Zona de Convergência Subtropical e o "*Brazilian Bight*" (Figura 1).
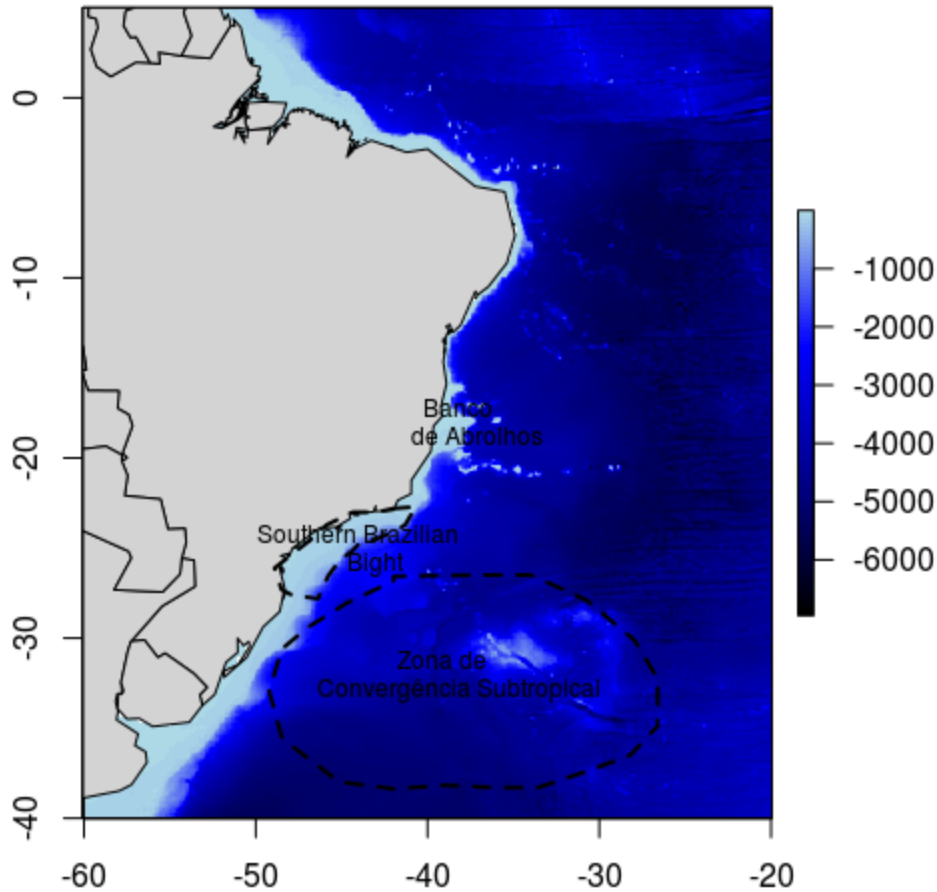

Figura 1. Mapa batimétrico do Atlântic Sul Ocidental com indicação do Banco de Abrolhos, *Southeast Brazilian Bight* e Zona de Convergência Subtropical.

A zona de convergência é resultado do encontro de duas correntes importantes: a Corrente do Brasil e a Corrente das Malvinas. A Corrente do Brasil tem origem na divisão da Corrente Sul Equatorial, que também forma a Corrente do Norte do Brasil, próximo dos 10ºS (SILVEIRA et al., 2000). A Corrente do Brasil carrega a Água Tropical,

oligotrófica, ao longo da plataforma continental em direção ao Sul (EMÍLSSON, 1961; SILVEIRA et al., 2000). À medida que se dirige para o sul, a Água Tropical perde calor para atmosfera e se mistura com águas de baixa salinidade e temperatura, resultando na Água Subtropical, que se caracteriza pela salinidade entre 35 e 36 ppm e temperatura variando entre 10 e 20ºC (EMÍLSSON, 1961). A Água Subtropical faz parte da Água Central do Atlântico Sul (EMÍLSSON, 1961), que é uma massa de água que flui para o norte em camadas profundas e pode alcançar a margem continental. Esta massa de água tem temperaturas maiores que 6ºC e menores que 20ºC, e salinidade entre 34,6 e 36 ppm (SILVEIRA et al., 2000).

A Corrente do Brasil encontra com a Corrente das Malvinas entre 33 e 40ºS. A Corrente das Malvinas é uma corrente de contorno oeste subpolar, cuja origem está na Corrente Circumpolar Antártica (MATANO; SCHLAX; CHELTON, 1993). É caracterizada por carregar Água Subantártica (temperatura entre 4 e 15ºC; salinidade de 33 ppm; rica em nutrientes) para o norte (SEELINGER; ODEBRECHT; CASTELLO, 1998). Ao se cruzarem, as duas correntes são forçadas em direção leste e originam a zona de Convergência Subtropical do Oceano Atlântico Sul Ocidental, uma das regiões oceânicas mais energéticas do mundo (MATANO; SCHLAX; CHELTON, 1993).

A zona de convergência entre a Corrente do Brasil e a Corrente das Malvinas apresenta uma variação sazonal, ocorrendo mais ao norte durante o inverno do que no verão austral. Este fenômeno tem consequências importantes tanto para o clima local quanto para as populações marinhas, porque marca o limite entre as águas quentes e as águas frias oriundas da Corrente Circumpolar Antártica (MATANO; SCHLAX; CHELTON, 1993).

Um exemplo da influência da Convergência Subtropical está em uma grande porção da plataforma continental e do talude entre o Cabo de Santa Marta (28º40'S) e Uruguai (34º40'S). Esta é uma zona de transição biogeográfica entre a Patagônia e o Brasil tropical. A dominância sazonal de diferentes massas de água sobre a plataforma e o talude continental condicionam a composição e abundância das espécies, a distribuição das comunidades e suas relações tróficas, além da produção biológica (SEELINGER; ODEBRECHT; CASTELLO, 1998).

O Oceano Atlântico Sul Ocidental apresenta outra áreas importantes para a vida marinha: o "*Southeast Brazilian Bight*" (SBB), entre Cabo Frio (23ºS) e o Cabo de Santa Marta (28ºS) (Emílsson, 1961). O SBB é também uma área de alta produtividade e presença de cetáceos, devido a ressurgência de águas ricas em nutrientes (CAMPOS; VELHOTE; DA SILVEIRA, 2000; CASTRO; MIRANDA, 1998).

Em relação ao relevo, a costa leste da América do Sul entre aproximadamente 20ºS e 34ºS apresenta variações na plataforma continental (CASTRO; MIRANDA, 1998; MAHIQUES; SOUSA; FURTADO, 2010) (Figura 1). Entre 5 e 16ºS, a plataforma continental é bastante estreita, tendo de 20 a 50 km de extensão. Já entre 16 e 20ºS, ocorre uma expansão da plataforma continental, que corresponde ao Banco de Abrolhos. A partir do Banco de Abrolhos até Cabo Frio (23ºS), a plataforma continental fica estreita novamente. Além disso, a linha costa muda de orientação abruptamente from NE-SW para E-W. A partir deste ponto, a plataforma amplia novamente e correspondendo ao SBB (20 a 28ºS). Na região do Cabo de Santa Marta (29ºS), a quebra da plataforma continental ocorre próximo da costa, mas a partir desta localização a plataforma continental torna-se bastante ampla até o sul do continente.

Duas espécies de cetáceos apresentam padrões de distribuição peculiares ao longo da costa brasileira, muito provavelmente em resposta às condições hidrogŕaficas e topográficas descritas acima. E, por isso, se tornaram o alvo deste estudo: a franciscana *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) e o golfinho-pintado-do-Atlântico *Stenella frontalis* (Cuvier, 1829). A primeira espécie é um golfinho de distribuição restrita do Brasil até a Argentina, que a ocorre primariamente na plataforma continental interna, raramente ultrapassando os 50 m de profundidade (CRESPO; HARRIS; GONZÁLEZ, 1998; DANILEWICZ et al., 2010). Já a segunda espécie é um golfinho de distribuição restrita ao Oceano Atlântico, ocupando principalmente a platafora continental com registros esporádicos até os 1.000 m de profundidae (MORENO et al., 2005; PERRIN, 2009). Interessantemente, as duas espécies apresentam hiatos ao longo da sua distribuição no Brasil (BORDINO et al., 2002; DO AMARAL et al., 2015; MORENO et al., 2005). A seguir apresenta-se uma breve descrição das duas espécies.

Franciscana (*Pontoporia blainvillei*)

Pontoporia é uma das linhagens de golfinhos de rio que habitaram e se diversificaram em um complexo sistema fluvial-marinho da América do Sul que existiu durante períodos de nível do mar elevado, registrados no Mioceno (HAMILTON et al., 2001). No entanto, esta foi a única linhagem que se dispersou para o ambiente marinho a partir do recuo da Bacia do Paraná, colonizando a zona de plataforma interna ao norte e ao sul do estuário do Rio de La Plata na América do Sul (HAMILTON et al., 2001).

Desta linhagem, a única espécie existente é a franciscana ou toninha (*Pontoporia blainvillei*) que é encontrada a partir de Itaúnas, Espírito Santo, Brasil, até Golfo Nuevo, Península Valdés, Argentina) (CRESPO; HARRIS; GONZÁLEZ, 1998; SICILIANO; DI

BENEDITTO; RAMOS, 2002). A distribuição da espécie não é contínua e dois hiatos são registrados no sudeste no Brasil. Até o momento, considera-se que o hiato norte é de Regência, Espírito Santo (19º40'S) até Barra do Itabapoana, Rio de Janeiro (21º18'S); e o hiato sul é de Macaé, Rio de Janeiro, (22º25'S) até Ilha Grande, Rio de Janeiro (23º09'S) (Figura 2).
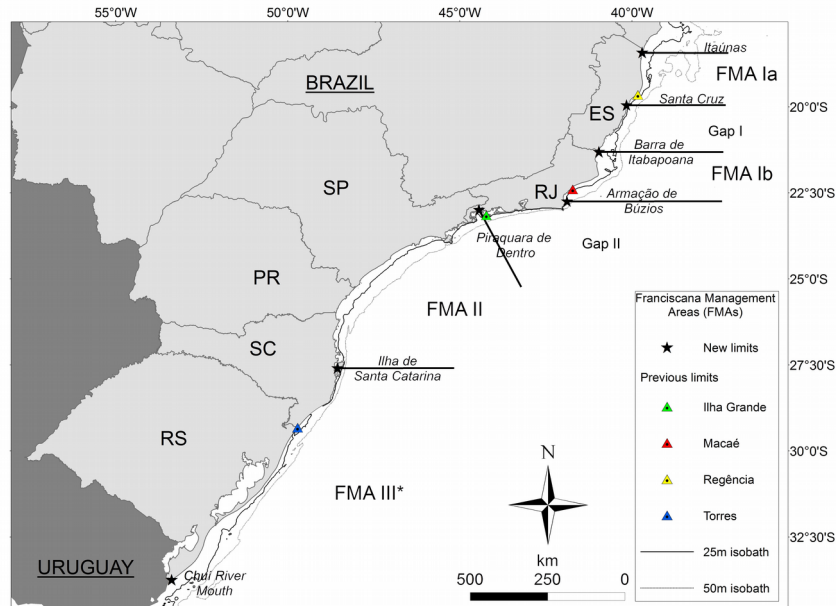

Figura 2. Mapa de distribuição da franciscana com os limites considerados até o momento de cada área de manejo (Franciscana Management Areas, FMAs) e hiatos, bem como os novos limites propostos no capítulo 1.

Atualmente, a franciscana é considerada o golfinho mais ameaçado da América do Sul. Seu status de conservação é "Vulnerável" na Lista Vermelha de espécies ameaçadas da União Internacional para Conservação da Natureza (*The IUCN Red List*) (ZERBINI et al., 2017); regionalmente, a espécie é listada oficialmente como "Criticamente Ameaçada"(MMA 2014), devido principalmente a capturas acidentais em rede de pesca (FRAINER; HUGGENBERGER; MORENO, 2015; PRADO; SECCHI; KINAS, 2013). Para promover a conservação da espécie, Secchi et al. (2003) propuseram quatro

23

unidades de manejo (*Franciscana Management Areas,* FMAs) ao longo da distribuição da franciscana: FMA I e FMA II localizadas exclusivamente no sudeste e sul do Brasil, FMA III entre sul do do Brasil e Uruguai, e FMA IV ao longo da Argentina. A divisão da espécie nestas áreas de manejo foi posteriormente suportada por diferentes tipos de dados (poluentes, dieta, morfologia externa, parasitas) (ALONSO et al., 2012; BARBATO et al., 2012; COSTA-URRUTIA et al., 2012; CUNHA et al., 2014; TORRE; ALONSO; MARTÍNEZ, 2012). Recentemente, novos estudos sugerem a reformulação destas FMAs, no sentido de incluir subdivisões(e.g. MENDEZ et al., 2010). Por exemplo, a FMA I já é formalmente reconhecida em duas unidades de manejo distintas separadas pelo hiato norte (CUNHA et al., 2014). Também foi proposto que a FMA I constitui uma Unidade Evolutiva Significante (*Evolutionary Significant Unit, ESU*) distinta de todas as demais FMAs (CUNHA et al., 2014).

Golfinho-pintado-do-Atlântico (*Stenella frontalis*)

Delphinidae é a mais diversa família de cetáceos existentes e apresenta uma variedade de formas de crânio, dentes e adaptações corporais que refletem suas dietas variadas e métodos de locomoção sendo, portanto, animais ecologicamente versáteis (BARNES, 1990; STEEMAN et al., 2009). Esta é a família de golfinhos que apresenta o maior número de espécies de mamíferos marinhos, atualmente existindo 38 espécies reconhecidas/válidas (CABALLERO et al., 2007; GEISLER et al., 2011; MCGOWEN, 2011; WICKERT et al., 2016). Os delfinídeos são altamente diversos em águas tropicais e em latitudes quentes a temperadas, onde são encontrados diversos gêneros: *Delphinus, Sotalia, Sousa, Stenella, Steno, Tursiops* (MCGOWEN, 2011).

O golfinho-pintado-do-Atlântico (*Stenella frontalis)* é um golfinho endêmico das águas tropicais, subtropicais e temperadas do Oceano Atlântico (PERRIN, 2009; PERRIN et al., 1987) e, diferentemente das demais espécies do gênero, sua distribuição parece ser mais restrita à águas relativamente rasas (PERRIN, 2009). A espécie é altamente variável geograficamente, levando a confusões taxonômicas e má identificação dos espécimes, já que também apresenta o padrão de pintas pelo corpo e muitas vezes é confundido com o golfinho-pintado-pantropical (*S. attenuata*). Na Lista Vermelha da IUCN, a espécie apresenta dados insuficientes e a maior ameaça à espécie é captura acidental ao longo de sua distribuição (HAMMOND et al., 2012).

A espécie é encontrada em águas quentes do Oceano Atlântico, principalmente sobre a plataforma continental, mas também pode ser encontrada esporadicamente em águas profundas. Ocorre também nas ilhas oceânicas dos Açores e Canárias. Existem formas menores e menos pintadas que habitam águas oceânicas pelágicas e, estes golfinhos juntamente com aqueles que ocorrem em ilhas, tem suas preferências ambientais menos conhecidas. No geral, a espécie tem uma distribuição geográfica complexa, na qual ao menos seis distintos morfótipos foram identificados (PERRIN et al., 1987). Estudos genéticos baseados em marcadores mitocondriais e nucleares identificaram dois *clusters* correspondentes aos que foram previamente descritos como os morfótipos costeiros e oceânicos ao longo do oeste do Atlântico Norte e Golfo do México. Além disso, o *cluster* costeiro parece estar sub-estruturado em três grupos que parecem estar correlacionados com distintos requerimentos ambientais (VIRICEL; ROSEL, 2014).

MORENO et al. (2005) observou que os registros de *Stenella frontalis* no Oceano Atlântico Sul Ocidental ocorrem ao norte de 6ºS e entre 21 e 33ºS. Existindo, portanto, uma grande área no Nordeste do Brasil (aproximadamente 1.500 km), entre 6 e 18ºS, sem a ocorrência da espécie (DANILEWICZ et al., 2013).

Análises de modelo de nicho ecológico indicam a ausência de condições ambientais ótimas para os golfinhos-pintados-do-Atlântico a partir do Banco de Abrolhos, e inclusive ao norte de 6ºS (DO AMARAL et al., 2015) (Figura 3). Provavelmente o estreita da plataforma continental brasileira desde o Banco de Abrolhos (~18ºS) até 6ºS explica o hiato da distribuição da espécie na costa brasileira. No Golfo do México, o estreitamento da plataforma continental foi considerado uma barreira física para a espécie, causando uma diminuição no habitat requerido preferencialmente pelo morfótipo considerado costeiro (VIRICEL; ROSEL, 2014).

Segundo MORENO et al., (2005), a distribuição descontínua desta espécie na costa do Brasil poderia indicar a existência de uma população no sul/sudeste do Brasil distinta e isolada daqueles animais que ocorrem no nordeste brasileiro e de outras regiões do Atlântico (e.g. Caribe e Atlântico Norte). Análises morfológicas, genéticas e evidências ecológicas suportam a hipótese de que os golfinhos-pintados-do-Atlântico que ocupam a porção sul/sudeste da costa brasileira podem, de fato, constituir uma linhagem evolutiva distinta (CABALLERO et al., 2013; DO AMARAL et al., 2015; MORENO, 2002). Esta população estaria, portanto, distribuída entre as regiões sul e sudeste da Costa Brasileira onde o ambiente marinho preferencial da espécie é altamente impactado por ações antrópicas das mais variadas (e.g. poluição, capturas acidentais em redes de pesca, exploração de óleo e gás, etc., portos, tráfego de embarcações), uma vez que esta região é

a economicamente mais desenvolvida do país (MÉNDEZ-FERNANDEZ et al., 2018; SANTOS et al., 2018).
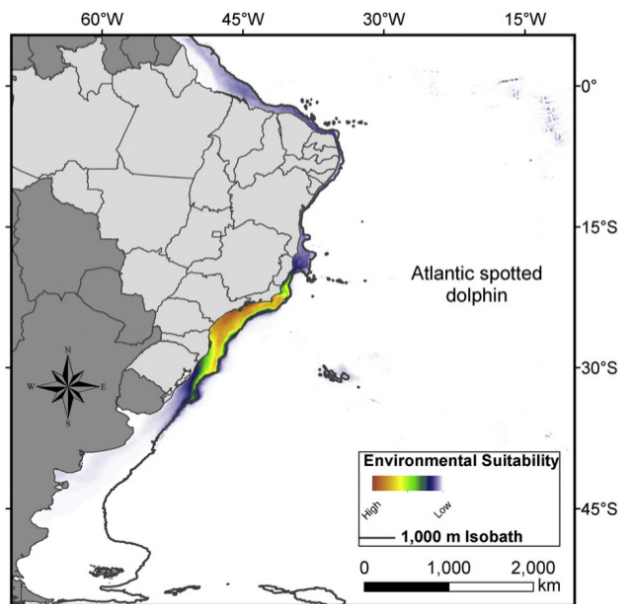

Figura 3. Mapa da distribuição potencial do golfinho-pintado-do-Atlântico no Atlântico Sul Ocidental. Retirado de DO AMARAL et al., 2015.

OBJETIVOS

Objetivo geral

Investigar a influência do ambiente marinho no padrão de distribuição e na estrutura genética de duas espécies de golfinhos: a franciscana e o golfinho-pintado-do-Atlântico com ênfase na costa brasileira.

Objetivos específicos

1) Revisar a distribuição da franciscana no Brasil, incluindo uma atualização dos limites das FMAs I, II e III e, também dos hiatos;

2) Investigar os fatores que potencialmente explicam a ausência da franciscana nos hiatos através da modelagem de nicho ecológico;

3) Investigar a estrutura genética do golfinho-pintado-do-Atlântico ao longo de toda sua distribuição, usando a região controle do DNA mitocondrial (mtDNA);

4) Investigar a influência do ambiente marinho na diferenciação genética do golfinho-pintado-do-Atlântico através de metodologias relacionadas a *"Seascape genetics",* uma adaptação da genética de paisagem para o ambiente marinho;

5) Investigar a estrutura genética do golfinho-pintado-do-Atlântico com ênfase nos indivíduos que ocorrem ao longo da costa brasileira através da análise de Nucleotídeos de Polimorfismo Único (*Singe Nucleotide Polymorsphism*, SNPs) obtidos através do sequenciamento de nova geração.

ESTRUTURA DA TESE

A tese é composta por três capítulos que serão apresentados na forma de manuscrito eforam escritos na língua inglesa. Espera-se que os manuscritos sejam submetidos o mais brevemente possível, com exceção do capítulo 1 que já está publicado. Os periódicos a qual cada manuscrito foi formatado é indicado no início de cada capítulo. Os três capítulos que compõem a tese são referentes aos seguintes manuscritos:

Capítulo I

"*Reassessment of the franciscana* Pontoporia blainvillei *(Gervais & d'Orbigny, 1844) distribution and niche characteristics in Brazil*"

Artigo publicado no *Journal of Experimental Marine Biology and Ecology*: https://doi.org/10.1016/j.jembe.2018.07.010.

Neste capítulo, atingiram-se os objetivos específicos 1 e 2 que visavam uma revisão e atualização da distribuição das áreas de manejo da franciscana (FMA) ao longo do Brasil, e também dos limites dos hiatos. Além disso, procurou-se através da aplicação de diferentes métodos esclarecer a ausência da franciscana nestas áreas com base nas diferenças das caractecterísticas ambientais destas áreas em relação às áreas onde a espécie é registrada.

No total, 788 registros de franciscana foram compilados ao longo de sua distribuição no Brasil. Estes dados foram utilizados para confirmar os limites das FMAs propostos por especialistas e, também novos limites para os hiatos de distribuição foram determinados: o hiato norte estende-se da desembocadura do Piraquê-Açu (19º57'S), Santa Cruz,

Espírito Santo até Barra de Itabapoana (21º18'S), Rio de Janeiro; o hiato sul estende-se de Armação dos Búzios (22º44'S) até Piraquara de Dentro (22º59'S), Rio de Janeiro. Análises de nicho ecológico sugerem que os hiatos fazem parte do nicho fundamental da franciscana sendo, portanto, relativamente adequados para a espécie em termos de salinidade, temperatura, turbidez e profundidade. No entanto, o estreitamento da plataforma continental parece ser o principal fator que explica a ausência da espécie nos hiatos e, inclusive poderia explicar a diferenciação genética entre algumas FMAs. Aparentemente, uma plataforma continental estreita poderia intensificar as interações bióticas levando, por exemplo, a maior competição por alimento e/ou causando uma limitação geográfica para manutenção de tamanho populacional mínimo viável em períodos de tempo presente ou passado.

Capítulo II

"*Seascape genetics of the Atlantic spotted dolphin* (Stenella frontalis) *based on mtDNA control region*"

Neste capítulo, cumpriram-se os objetivos 3 e 4 citados anteriormente. Uma vez que foram encontradas populações distintas geneticamente ao longo de toda distribuição do golfinho-pintado-do-Atlântico baseado nas análises de mtDNA, investigou-se a relação das distâncias genéticas com diferentes tipos de distâncias geográficas, de resistência e ambientais.

Através da análise de 545 sequências da região controle do mtDNA(incluindo sequências já publicadas e dados inéditos), foram indentificadas diferentes populações no Atlântico Norte Ocidental e Golfo do México, além de uma população oceânica que inclui

indivíduos que habitam os arquipélagos do Atlântico Oriental e, também as águas oceânicas do Atlântico Norte Ocidental. Indivíduos que ocorrem no sul/sudeste do Brasil parecem formar uma população distinta, exibindo baixos níveis de diversidade genética de acordo com os resultados obtidos. Análises de paisagem genética para o ambiente marinho ("*Seascape genetics*") indicam certo grau de Isolamento por Distância e Isolamento por Resistência, relacionados tanto com condições ambientais contemporâneas quanto do passado (Último Glacial Máximo). Além disso, sugere-se que outros processos podem estar envolvidos na diferenciação das populações como, por exemplo, estrutura social e filopatria.

Capítulo III

"*Population genomics of the Atlantic spotted dolphins* (Stenella frontalis) *across the Atlantic Ocean*"

Neste capítulo, cumpriu-se o objetivo 5 citado anteriormente. Considerando-se o impacto das técnicas genômicas que aumentam a qualidade, resolução e confiabilidade das análises genéticas, neste capítulo utilizou-se de uma destas técnicas disponíveis para investigar a estrutura populacional do golfinho-pintado-do-Atlântico ao longo do Caribe, Ilhas Canárias Brasil e Uruguai. Além disso, a relação da estruturação genética com o ambiente marinho foi investigada.

Dados genômicos foram gerados para indivíduos coletados no Atlântico Sul Ocidental (Brasil e Uruguai), Caribe (Colômbia e Ilha Guadaloupe) e Atlântico Oriental (Ilhas Canárias). Estes dados foram utilizados para testar padrões de diferenciação genética entre estas regiões e, principalmente, avaliar a distinção genética dos indivíduos que

ocorrem no Atlântico Sul Ocidental. As análises indicam ao menos a existência de três populações: uma no sudeste do Brasil, uma na Colômbia e uma oceânica. No entanto, os níveis diferenciação recuperados foram baixos, sendo necessários mais amostras e, principalmente, amostragem em outros locais para aumentar o poder estatístico das análises e melhor delineamento dos limites populacionais regionais. Os resultados também indicam que a similaridade genética está correlacionada com a geografia; mas o ambiente marinho também parece ter alguma influência na estrutura genética, principalmente a profundidade e a temperatura. Finalmente, nossos dados suportam a hipótese de uma população isolada no sudeste do Brasil.

REFERÊNCIAS*

* Conforme Normas da ABNT (Autoria Abreviada) disponibilizadas pelo SABIUFRGS.

AGNARSSON, I.; MAY-COLLADO, L. J. The phylogeny of Cetartiodactyla: The importance of dense taxon sampling, missing data, and the remarkable promise of cytochrome b to provide reliable species-level phylogenies. **Molecular Phylogenetics and Evolution**, [s. l.], v. 48, n. 3, p. 964–985, 2008. Disponível em: <https://www-sciencedirect-com.proxy.lib.umich.edu/science/article/pii/S1055790308002996>. Acesso em: 12 jun. 2018.

ALONSO, M. B. et al. Natural and anthropogenically-produced brominated compounds in endemic dolphins from Western South Atlantic: Another risk to a vulnerable species. **Environmental Pollution**, [s. l.], v. 170, p. 152–160, 2012.

BALLANCE, L. T.; PITMAN, R. L.; FIEDLER, P. C. Oceanographic influences on seabirds and cetaceans of the eastern tropical Pacific: a review. **Progress in Oceanography**, [s. l.], v. 69, n. 2, p. 360–390, 2006.

BARBATO, B. H. A. et al. Geographical variation in franciscana (*Pontoporia blainvillei*) external morphology. **Journal of the Marine Biological Association of the United Kingdom**, [s. l.], v. 92, n. 08, p. 1645–1656, 2012.

BARNES, L. G. **The fossil record and evolutionary relationships of the genus** *Tursiops*. San Diego: Academic Press, 1990.

BAUMGARTNER, M.; MULLIN, K.; MAY, L. Cetacean habitats in the northern Gulf of Mexico. **FISHERY BULLETIN**, [s. l.], 2001. Disponível em: <http://www.biomedsearch.com/article/Cetacean-habitats-in-northern-Gulf/75434038.html>. Acesso em: 7 ago. 2016.

BIERNE, N.; BONHOMME, F.; DAVID, P. Habitat preference and the marine-speciation paradox. **Proceedings of the Royal Society B: Biological Sciences**, [s. l.], v. 270, n. 1522, p. 1399–1406, 2003. Disponível em: <http://rspb.royalsocietypublishing.org/cgi/doi/10.1098/rspb.2003.2404>

BORDINO, P. et al. Report of the Working Group on Distribution and Behavior. **Latin American Journal of Aquatic Mammals**, [s. l.], v. 1, n. 1, p. 21–23, 2002.

BROWN, J.; STEVENS, G.; KAUFMAN, D. The geographic range: size, shape, boundaries, and internal structure. **Annual review of ecology and**, [s. l.], 1996. Disponível em: <http://www.jstor.org/stable/2097247>. Acesso em: 7 ago. 2016.

CABALLERO, S. et al. Taxonomic status of the genus *Sotalia*: Species level ranking for "tucuxi" (*Sotalia fluviatilis*) and "costero" (*Sotalia guianensis*) dolphins. **Marine Mammal Science**, [s. l.], v. 23, n. 2, p. 358–386, 2007. Disponível em: <http://doi.wiley.com/10.1111/j.1748-7692.2007.00110.x>. Acesso em: 4 jun. 2018.

CABALLERO, S. et al. Initial description of the phylogeography, population structure and genetic diversity of Atlantic spotted dolphins from Brazil and the Caribbean, inferred from analyses of mitochondrial and nuclear DNA. **Biochemical Systematics and Ecology**, [s. l.], v. 48, p. 263–270, 2013.

CAMPOS, E. J. D.; VELHOTE, D.; DA SILVEIRA, I. C. A. Shelf break upwelling driven by Brazil current cyclonic meanders. **Geophysical Research Letters**, [s. l.], v. 27, n. 6, p. 751–754, 2000.

CASTRO, B. M.; MIRANDA, L. B. Physical oceanography of the western Atlantic continental shelf located between 4º N and 34º S. In: ROBINSON, A.; BRINK, K. H. (Eds.). **The sea**. [s.l.] : John Wiley & Sons, Inc., 1998. p. 209–251.

COSTA-URRUTIA, P. et al. Population genetic structure and social kin associations of franciscana dolphin, *Pontoporia blainvillei*. **The Journal of heredity**, [s. l.], v. 103, n. 1, p. 92–102, 2012. Disponível em: <http://www.ncbi.nlm.nih.gov/pubmed/22013080>. Acesso em: 7 ago. 2016.

CRESPO, E. A.; HARRIS, G.; GONZÁLEZ, R. Group size and distributional range of the franciscana, *Pontoporia blainvillei*. **Marine Mammal Science**, [s. l.], v. 14, n. 4, p. 845–849, 1998.

CUNHA, H. A. et al. Population structure of the endangered franciscana dolphin (*Pontoporia blainvillei)*: reassessing management units. **PloS one**, [s. l.], v. 9, n. 1, p. e85633, 2014.

DANILEWICZ, D. et al. Abundance estimate for a threatened population of franciscana dolphins in southern coastal Brazil: uncertainties and management implications. **Journal of the Marine Biological Association of the United Kingdom**, [s. l.], v. 90, n. 08, p. 1649–1657, 2010. Disponível em: <http://www.journals.cambridge.org/abstract_S0025315409991482>. Acesso em: 10 ago. 2016.

DANILEWICZ, D. et al. Occurrence of the Atlantic spotted dolphin, *Stenella frontalis*, in southern Abrolhos Bank, Brazil. **Marine Biodiversity Records**, [s. l.], v. 6, 2013.

DAVIES, J. L. The antitropical factor in cetacean speciation. **Evolution**, [s. l.], v. 17, p. 107–116, 1963.

DO AMARAL, K. B. et al. Ecological niche modeling of *Stenella* dolphins (Cetartiodactyla: Delphinidae) in the southwestern Atlantic Ocean. **Journal of Experimental Marine Biology and Ecology**, [s. l.], v. 472, p. 166–179, 2015.

DO AMARAL, K. B. et al. Historical Biogeography of Delphininae Dolphins and Related Taxa (Artiodactyla: Delphinidae). **Journal of Mammalian Evolution**, [s. l.], v. 25, n. 2, p. 241–259, 2018. Disponível em: <http://link.springer.com/10.1007/s10914-016-9376-3>. Acesso em: 4 jun. 2018.

EMÍLSSON, I. The shelf and coastal waters off southern Brazil. **Boletim do Instituto Oceanográfico**, [s. l.], v. 11, n. 2, p. 101–112, 1961. Disponível em: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0373-55241961000100004&lng=en&tlng=en>. Acesso em: 12 jun. 2018.

FOOTE, A. D. et al. Genome-culture coevolution promotes rapid divergence of killer whale ecotypes. **Nature Communications**, [s. l.], v. 7, 2016.

FRAINER, G.; HUGGENBERGER, S.; MORENO, I. B. Postnatal development of franciscana's (*Pontoporia blainvillei*) biosonar relevant structures with potential implications for function, life history, and bycatch. **Marine Mammal Science**, [s. l.], v. 31, n. 3, p. 1193–1212, 2015. Disponível em: <http://doi.wiley.com/10.1111/mms.12211>. Acesso em: 12 jun. 2018.

GASTON, K. J.; FULLER, R. A. The sizes of species' geographic ranges. **Journal of Applied Ecology**, [s. l.], v. 46, n. 1, p. 1–9, 2009. a. Disponível em: <http://doi.wiley.com/10.1111/j.1365-2664.2008.01596.x>. Acesso em: 7 ago. 2016.

GEISLER, J. H. et al. A supermatrix analysis of genomic, morphological, and paleontological data from crown Cetacea. **BMC Evolutionary Biology**, [s. l.], v. 11, n. 1, p. 112, 2011. Disponível em: <http://bmcevolbiol.biomedcentral.com/articles/10.1186/1471-2148-11-112>. Acesso em: 11 ago. 2016.

HAMILTON, H. et al. Evolution of river dolphins. **Proceedings of the Royal Society of London B: Biological Sciences**, [s. l.], v. 268, n. 1466, p. 549–556, 2001.

HAMMOND, P. S. et al. *Stenella frontalis.* 2012. Disponível em: <http://www.iucnredlist.org/details/20732/0>. Acesso em: 9 jun. 2018.

HAUSER, L.; CARVALHO, G. R. Paradigm shifts in marine fisheries genetics: Ugly hypotheses slain by beautiful facts. **Fish and Fisheries**, [s. l.], v. 9, n. 4, p. 333–362, 2008.

HOELZEL, A. R. Genetic structure of cetacean populations in sympatry, parapatry, and mixed assemblages: implications for conservation policy. **Journal of Heredity**, [s. l.], v.

89, n. 5, p. 451–458, 1998. Disponível em: <https://academic.oup.com/jhered/article-lookup/doi/10.1093/jhered/89.5.451>. Acesso em: 12 jun. 2018.

KELLEY, J. L. et al. The life aquatic: advances in marine vertebrate genomics. **Nature Reviews Genetics**, [s. l.], v. 17, n. 9, p. 523–534, 2016. Disponível em: <http://www.nature.com/articles/nrg.2016.66>. Acesso em: 4 jun. 2018.

MAHIQUES, M.; SOUSA, S.; FURTADO, V. The Southern Brazilian shelf: general characteristics, quaternary evolution and sediment distribution. **Brazilian Journal of**, [s. l.], 2010. Disponível em: <http://www.scielo.br/scielo.php?pid=S1679-87592010000600004&script=sci_arttext>. Acesso em: 7 ago. 2016.

MATANO, R. P.; SCHLAX, M. G.; CHELTON, D. B. Seasonal variability in the southwestern Atlantic. **Journal of Geophysical Research**, [s. l.], v. 98, n. C10, p. 18027, 1993. Disponível em: <http://doi.wiley.com/10.1029/93JC01602>. Acesso em: 12 jun. 2018.

MCGOWEN, M. R. Toward the resolution of an explosive radiation—A multilocus phylogeny of oceanic dolphins (Delphinidae). **Molecular Phylogenetics and Evolution**, [s. l.], v. 60, n. 3, p. 345–357, 2011. Disponível em: <https://www-sciencedirect-com.proxy.lib.umich.edu/science/article/pii/S1055790311002235>. Acesso em: 12 jun. 2018.

MCGOWEN, M. R.; SPAULDING, M.; GATESY, J. Divergence date estimation and a comprehensive molecular tree of extant cetaceans. **Molecular Phylogenetics and Evolution**, [s. l.], v. 53, n. 3, p. 891–906, 2009.

MÉNDEZ-FERNANDEZ, P. et al. Contamination status by persistent organic pollutants of the Atlantic spotted dolphin (*Stenella frontalis*) at the metapopulation level. **Environmental Pollution**, [s. l.], v. 236, p. 785–794, 2018. Disponível em: <https://www-sciencedirect-com.proxy.lib.umich.edu/science/article/pii/S0269749117312769>. Acesso em: 4 jun. 2018.

MENDEZ, M. et al. Isolation by environmental distance in mobile marine species: molecular ecology of franciscana dolphins at their southern range. **Molecular Ecology**, [s. l.], v. 19, n. 11, p. 2212–2228, 2010. Disponível em: <http://doi.wiley.com/10.1111/j.1365-294X.2010.04647.x>. Acesso em: 7 ago. 2016.

MMA, 2014. Lista Nacional Oficial de Espécies da Fauna Ameaçadas de Extinção – Mamíferos, Aves, Répteis, Anfíbios e Invertebrados Terrestres. Portaria MMA nº. 444, de 17 de dezembro de 2014, Brazil.

MÖLLER, L. et al. Fine-scale genetic structure in short-beaked common dolphins (*Delphinus delphis*) along the East Australian Current. **Marine Biology**, [s. l.], v. 158, n. 1, p. 113–126, 2011.

MORENO, I. B. **Padrão de distribuição dos golfinhos do gênero** *Stenella* **(Delphinidae: Cetacea) no oceano Atlântico sul-ocidental e morfometria craniana dos golfinhos- pintados (***Stenella frontalis* **e** *S. attenuata***)**. 2002. Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Brazil, [s. l.], 2002.

MORENO, I. B. et al. Distribution and habitat characteristics of dolphins of the genus *Stenella* (Cetacea: Delphinidae) in the southwest Atlantic Ocean. **Marine Ecology Progress Series**, [s. l.], v. 300, p. 229–240, 2005.

MORIN, P. A. et al. Geographic and temporal dynamics of a global radiation and diversification in the killer whale. **Molecular Ecology**, [s. l.], v. 24, n. 15, p. 3964–3979, 2015.

NORRIS, R. D. Pelagic species diversity, biogeography, and evolution. **Paleobiology**, [s. l.], v. 26, p. 236–258, 2000. Disponível em: <http://www.bioone.org.proxy.lib.umich.edu/doi/abs/10.1666/0094-8373(2000)26%5B236:PSDBAE%5D2.0.CO%3B2>. Acesso em: 4 jun. 2018.

PALACIOS, D. et al. Beyond correlation: integrating environmentally and behaviourally mediated processes in models of marine mammal distributions. **Endangered Species Research**, [s. l.], v. 22, n. 3, p. 191–203, 2013. Disponível em: <http://www.int-res.com/abstracts/esr/v22/n3/p191-203/>. Acesso em: 7 ago. 2016.

PEARSON, R. G. Species' Distribution Modeling for Conservation Educators and Practitioners. **Lessons in Conservation**, [s. l.], v. 3, p. 54–89, 2007. Disponível em: <https://pdfs.semanticscholar.org/66db/947ee1a6ab91c408f489d17cfb6e068931a6.pdf>. Acesso em: 12 jun. 2018.

PERRIN, W. F. et al. Revision of the Spotted dolphins, *Stenella spp*. **Marine Mammal Science**, [s. l.], v. 3, n. 2, p. 99–170, 1987.

PERRIN, W. F. Atlantic spotted dolphin: *Stenella frontalis*. In: **Encyclopedia of Marine Mammals**. [s.l: s.n.]. p. 54–56.

PRADO, J. H. F.; SECCHI, E. R.; KINAS, P. G. Mark-recapture of the endangered franciscana dolphin (*Pontoporia blainvillei*) killed in gillnet fisheries to estimate past bycatch from time series of stranded carcasses in southern Brazil. **Ecological Indicators**, [s. l.], v. 32, p. 35–41, 2013. Disponível em: <https://www-sciencedirect-com.proxy.lib.umich.edu/science/article/pii/S1470160X13001118>. Acesso em: 12 jun. 2018.

REDFERN, J. V et al. Techniques for cetacean–habitat modeling. **Marine Ecology Progess Series**, [s. l.], v. 310, p. 271–295, 2006.

SANTOS, M. C. de O. et al. Cetaceans using the marine protected area of "Parque Estadual Marinho da Laje de Santos", Southeastern Brazil. **Brazilian Journal of Oceanography**, [s. l.], v. 65, n. 4, p. 605–613, 2018. Disponível em: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S1679-87592017000400605&lng=en&tlng=en>. Acesso em: 4 jun. 2018.

SÃO PEDRO, S. L. et al. Evidence of Positive Selection of Aquaporins Genes from *Pontoporia blainvillei* during the Evolutionary Process of Cetaceans. **PLOS ONE**, [s. l.], v. 10, n. 7, p. e0134516, 2015. Disponível em: <http://dx.plos.org/10.1371/journal.pone.0134516>. Acesso em: 12 ago. 2016.

SEELINGER, U.; ODEBRECHT, C.; CASTELLO, J. P. Subtropical convergence environments: the coast and sea in the southwestern Atlantic. **Oceanographic Literature Review**, [s. l.], v. 8, n. 45, p. 1366, 1998.

SEGURA-GARCÍA, I. et al. Eco-Evolutionary Processes Generating Diversity Among Bottlenose Dolphin, *Tursiops truncatus*, Populations off Baja California, Mexico. **Evolutionary Biology**, [s. l.], v. 45, n. 2, p. 223–236, 2018. Disponível em: <http://link.springer.com/10.1007/s11692-018-9445-z>. Acesso em: 12 jun. 2018.

SICILIANO, S.; DI BENEDITTO, A. P. M.; RAMOS, R. M. A toninha *Pontoporia blainvillei* nos Estados do Rio de Janeiro e Espírito Santo, costa sudeste do Brasil: caracteriazação dos hábitats e fatores de isolamento das populações. **Boletim do Museu Nacional**, [s. l.], v. 476, p. 1–15, 2002.

SILVEIRA, I. C. A. Da et al. A corrente do Brasil ao largo da costa leste brasileira. **Revista Brasileira de Oceanografia**, [s. l.], v. 48, n. 2, p. 171–183, 2000. Disponível em: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S1413-77392000000200008&lng=pt&tlng=pt>. Acesso em: 12 jun. 2018.

STEEMAN, M. E. et al. Radiation of Extant Cetaceans Driven by Restructuring of the Oceans. **Systematic Biology**, [s. l.], v. 58, n. 6, p. 573–585, 2009. Disponível em: <http://sysbio.oxfordjournals.org/cgi/doi/10.1093/sysbio/syp060>. Acesso em: 11 ago. 2016.

TEZANOS-PINTO, G. et al. A Worldwide Perspective on the Population Structure and Genetic Diversity of Bottlenose Dolphins (*Tursiops truncatus*) in New Zealand. **Journal of Heredity**, [s. l.], v. 100, n. 1, p. 11–24, 2008. Disponível em: <http://jhered.oxfordjournals.org/cgi/doi/10.1093/jhered/esn039>

TORRE, A. D. La; ALONSO, M.; MARTÍNEZ, M. Dechlorane-related compounds in franciscana dolphin (*Pontoporia blainvillei*) from Southeastern and Southern Coast of Brazil. **science & technology**, [s. l.], 2012. Disponível em: <http://pubs.acs.org/doi/abs/10.1021/es302934p>. Acesso em: 10 ago. 2016.

VACHON, F.; WHITEHEAD, H.; FRASIER, T. R. What factors shape genetic diversity in cetaceans? **Ecology and Evolution**, [s. l.], v. 8, n. 3, p. 1554–1572, 2018. Disponível em: <http://doi.wiley.com/10.1002/ece3.3727>. Acesso em: 12 jun. 2018.

VIRICEL, A.; ROSEL, P. E. Hierarchical population structure and habitat differences in a highly mobile marine species: The Atlantic spotted dolphin. **Molecular Ecology**, [s. l.], v. 23, n. 20, p. 5018–5035, 2014.

WHITEHEAD, H. Gene-culture coevolution in whales and dolphins. **Proceedings of the National Academy of Sciences of the United States of America**, [s. l.], v. 114, n. 30, p. 7814–7821, 2017. Disponível em: <http://www.ncbi.nlm.nih.gov/pubmed/28739936>. Acesso em: 12 jun. 2018.

WICKERT, J. C. et al. Revalidation of *Tursiops gephyreus* Lahille, 1908 (Cetartiodactyla: Delphinidae) from the southwestern Atlantic Ocean. **Journal of Mammalogy**, [s. l.], v. 97, n. 6, p. 1728–1737, 2016. Disponível em: <https://academic.oup.com/jmammal/article-lookup/doi/10.1093/jmammal/gyw139>. Acesso em: 4 jun. 2018.

XU, S. et al. Adaptive evolution of the osmoregulation-related genes in cetaceans during secondary aquatic adaptation. **BMC Evolutionary Biology**, [s. l.], v. 13, n. 1, p. 189, 2013. Disponível em: <http://bmcevolbiol.biomedcentral.com/articles/10.1186/1471-2148-13-189>. Acesso em: 12 ago. 2016.

ZERBINI, A. et al. *Pontoporia blainvillei*. 2017. Disponível em: <http://www.iucnredlist.org/details/17978/0>. Acesso em: 12 jun. 2018.

CAPÍTULO I

Reassessment of the franciscana *Pontoporia blainvillei* **(Gervais & d'Orbigny, 1844) distribution and niche characteristics in Brazil**

Karina Bohrer do Amaral [*, a, b], Daniel Danilewicz [c, d,e], Alexandre Zerbini [f,g], Ana Paula Di Beneditto [h], Artur Andriolo [d, i], Diego Janisch Alvares [b, j], Eduardo Secchi [k], Emanuel Ferreira [l], Federico Sucunza [c, d,i], Márcio Borges-Martins [b, c, j], Marcos César de Oliveira Santos [m], Marta Cremer [n], Pablo Denuncio [o], Paulo Henrique Ott [c, p], Ignacio Benites Moreno [a, b, q]

* Corresponding author: karinabohrerdoamaral@gmail.com

[a] Laboratório de Sistemática e Ecologia de Aves e Mamíferos Marinhos (LABSMAR), Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, Sala 206, 91501-70, Porto Alegre, RS, Brazil. Telephone: +55 51 3308-9541.

[b] Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, 91501-70, Porto Alegre, RS, Brazil.

[c] Grupo de Estudos de Mamíferos Aquáticos do Rio Grande do Sul (GEMARS), Rua Bento Gonçalves, 165/1002, 95560-000, Torres, RS, Brazil.

[d] Instituto Aqualie, Av. Dr. Paulo Japiassu Coelho, 714, Sala 206, 36033-310, Juiz de Fora, MG, Brazil.

[e] Programa de Pós-Graduação em Zoologia, Universidade Estadual de Santa Cruz, Rodovia Ilhéus/Itabuna, km 16, 45662-900, Ilhéus, BA, Brazil.

.

[f] National Marine Mammal Laboratory, Alaska Fisheries Science Center/NOAA, 7600 Sand Point Way NE, Seattle, WA, 98115-649, USA.

[g] Cascadia Research Collective, 2181/2 W 4th Ave, Olympia, WA, 98501, USA.

[h] Laboratório de Ciências Ambientais (CBB), Universidade Estadual do Norte Fluminense, Avenida Alberto Lamego, 2000, 28013-602, Campos, RJ, Brazil.

[i] Laboratório de Ecologia Comportamental e Bioacústica, Departamento de Zoologia, Instituto de Ciências Biológicas, Universidade Federal de Juiz de Fora, Rua José Lourenço Kelmer, s/n - Campus Universitário, 36036-900, Juiz de Fora, MG, Brazil.

[j] Laboratório de Herpetologia, Departamento de Zoologia, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, sala 102, Porto Alegre, RS 91501-70, Brazil.

[k] Laboratório de Ecologia e Conservação da Megafauna Marinha-EcoMega, Instituto de Oceanografia, Universidade Federal do Rio Grande-FURG, Caixa Postal 474, 96203-900, Rio Grande, RS, Brazil.

[l] Associação R3 Animal, Rodovia João Gualberto Soares – Entrada do Parque Estadual do Rio Vermelho, 88061-500, Florianópolis SC, Brazil.

[m] Laboratório de Biologia da Conservação de Mamíferos Aquáticos (LABCMA), Departamento de Oceanografia Biológica, Instituto Oceanográfico, Universidade de São Paulo, Praça do Oceanográfico, 191, Sala 145-A, 05508-120, São Paulo, SP, Brazil.

[n] Projeto Toninhas, Universidade da Região de Joinville (UNIVILLE), São Francisco do Sul, SC, Brazil. E-mail: mjc2209@yahoo.com.br

[o] Instituto de Investigaciones Marinas y Costeras (IIMYC-CONICET), Departamento de Ciencias Marinas, Universidad Nacional de Mar del Plata (UNMDP), Casilla de Correo 1260, Mar del Plata, Argentina.

[p] Universidade Estadual do Rio Grande do Sul (Uergs), Unidade do Litoral Norte, Rua Machado de Assis, 1456, 95520-000, Osório, RS, Brazil.

[q] Centro de Estudos Costeiros, Limnológicos e Marinhos (CECLIMAR), Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Tramandaí, 976, 95625-000, Imbé, RS, Brazil.

ABSTRACT

The franciscana (*Pontoporia blainvillei*) is the most threatened small cetacean of South America. The species is endemic to coastal waters of the western South Atlantic Ocean, where it is distributed from Itaúnas (Brazil) to Golfo San Matias (Argentina). Its range was divided in four Franciscana Management Areas (FMAs) for conservation purposes. However, the distribution of the franciscana is not continuous along its range, with two hiatuses proposed in southeastern Brazilian coast. The absence of franciscana records in these regions has been confirmed by multiple years of research, however the reasons for this discontinuous distribution is not well understood. In this study, information on the distribution of the franciscana in south and southeastern Brazil is updated and new limits for FMAs are proposed. NicheA 3.0 software was used to investigate the environmental suitability of distributional gaps in relation to four weakly correlated, allegedly relevant descriptors of franciscana's distribution. In total, 788 records from dedicated aerial and boat surveys and bycatch were used to verify and to confirm the new FMAs limits proposed by franciscana's experts previously. The distributional gaps were reshaped and defined as following: Gap I from Piraquê-Açu River Mouth, Santa Cruz (19º57'S) in the state of Espírito Santo to Barra de Itabapoana (21º18'S) in the state of Rio de Janeiro; and Gap II from Armação dos Búzios (22º44'S) to Piraquara de Dentro (22º59'S) in Rio de Janeiro. The ecological niche model indicated that distributional gaps are inside franciscana's fundamental niche, and are relatively suitable in terms of salinity,

temperature, diffuse attenuation and bathymetry. However, the narrow of continental shelf seems to be the main factor explaining the absence of franciscanas in the distributional gaps as well as for the differentiation of some of the FMAs proposed. Narrowness of continental shelf seems to be intensifying the dynamics of biotic interactions promoting food competition for example, and/or causing geographic limitation to maintain minimal viable population size in present or past times periods.

HIGHLIGHTS:

1. The franciscana dolphin is endemic and the most threatened small cetacean of South America

2. We analyzed its distribution in Brazil, mainly in relation to its distributional gaps

3. Narrowness of the continental shelf seems to explain the absence of franciscana within the gaps

4. Loss of shelf habitat could intensify biotic interactions (predation/competition)

5. Historical factors could also play a role to explain this biogeographical pattern

1. INTRODUCTION

The franciscana *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) is the most threatened small cetacean of South America (Secchi et al., 2003a). Mortality in gillnets have been impacting franciscana dolphins throughout their range for at least 50 years (e.g. Ott et al., 2002; Prado et al. 2013, 2016; Secchi et al., 2003a, 2003b), compromising

the viability of its populations (Kinas, 2002; Secchi, 2006). The franciscana faces a high risk of extinction and is listed as "Vulnerable" on a global scale by IUCN (Zerbini et al., 2017), while regionally in Brazil it is officially listed as "Critically Endangered" (MMA 2014).

The franciscana is endemic to coastal waters of Brazil, Uruguay, and Argentina. Currently, the species occurs from Itaúnas (18º25'S), in the state of Espírito Santo, southeastern Brazil (Siciliano et al., 2002) to Golfo San Matias (41º10'S), Rio Negro, Argentina (Crespo et al., 1998). Early studies showed evidence that franciscana is not continuously distributed along its range in Brazil (Siciliano et al., 2002). Many years of bycatch monitoring, beach surveys for stranded animals and aerial surveys confirms the existence of two distributional gaps: (1) from Regência (19º40'S), in Espírito Santo, to Barra do Itabapoana (21º18'S), in the state of Rio de Janeiro, namely northern distributional gap (Gap I); and (2) from Macaé (22º25'S) to Ilha Grande (23º09'S), in Rio de Janeiro, namely southern distributional gap (Gap II) (e.g. Azevedo et al., 2002; Danilewicz et al., 2012; de Moura et al., 2009). Systematic and long-term monitoring has confirmed the absence of franciscanas, mainly in the central portion of these gaps (e.g. de Moura et al., 2009). However, there is no consensus about the exact boundaries of the gaps (e.g. Azevedo et al., 2002; Siciliano et al., 2015) which play an important role in the delineation of management units for the species (Secchi et al., 2003a).

Previous studies revealed the existence of geographical population structure based on external morphology and genetic markers (e.g. Higa et al., 2002; Ott, 2002; Pinedo, 1995; Ramos et al., 2002; Secchi et al., 1998). After applying a multi-methodological approach for identifying stock discreteness, Secchi et al. (2003a) divided the

franciscana's range into four Franciscana Management Areas (FMAs) (please see Fig. 1

of the refereed article). FMA I and FMA II are located exclusively in southeastern and

southern Brazil, FMA III includes southern Brazil and Uruguay, and FMA IV

encompasses the range of the species in Argentina. These management divisions are

supported by recent data on pollutant loads, diet, external morphology and parasites (e.g.

Alonso et al., 2012; Barbato et al., 2012; Costa-Urrutia et al., 2012; de la Torre et al.,

2012; Hoss et al., 2017). New studies have suggested the need of reformulation of the

former FMA's subdivisions (e.g. Gariboldi et al., 2015, 2016; Mendez et al., 2010),

including the separation of FMA I in two distinct management units (FMA Ia and FMA

Ib) separated by the northern distributional gap (Anonymous, 2015; Cunha et al., 2014).

The increased effort from properly designed aerial surveys to estimate

franciscana's abundance (e.g. Danilewicz et al., 2010, 2012; Zerbini et al., 2011) and

long-term projects evaluating franciscana bycatch (see Material and Methods) have

provided many georeferenced at-sea records for the species. These data have been useful

to characterise the distributional ecology of franciscanas' populations in a comprehensive

manner and can be used to perform ecological niche modeling in order to investigate

factors that influence their distribution.

Correlative species distribution models are based on algorithms that estimate

ecological niches and explore potential distributional areas by assessing relationships

between species occurrences and environmental information (Qiao et al., 2016). Niche

modeling approaches dramatically expanded in recent years and currently several

techniques and toolkits are available (Phillips et al., 2006, Qiao et al., 2016). In addition,

these techniques have been widely used in studies on the distribution of cetaceans (e.g. do

Amaral et al., 2015; Palacios et al., 2013; Rossi-Santos and Oliveira, 2016), including

estimates of the potential franciscana distribution (Gomez and Cassini, 2015).

Given the high risk faced by the franciscana, especially the extremely high risk of

extinction observed regionally in Brazil (Rocha-Campo et al., 2010), and the importance

of distributional ecology to either the process of risk assessment and conservation

planning, the aim of this study is (1) to update information on the franciscana distribution

in Brazil, including a review of FMAs I, II and III as well as the distributional gaps

between them, and (2) to investigate the factors that potentially explain the existence of

gaps in the range of the franciscana.

2. MATERIALS AND METHODS

2.1. Study area

The study area includes the Brazilian continental shelf from 18ºS to 34ºS,

including only those waters up to the 50m isobath (Fig. 1A). The area is characterized by

different physical oceanographic processes. Castro and Miranda (1998) therefore

proposed a segmentation of the Brazilian continental shelf into six zones, of which three

zones are encompassed by the study area: Abrolhos – Campos Region (15ºS – 23ºS),

South Brazilian Bight (23ºS – 28º30'S) and Southern Brazilian Shelf (28º30'S – 34ºS)

(Fig. 1B). These areas are characterized by different features in relation to topography,

productivity, sea surface temperatures and salinity due to upwelling, land runoff from

several estuaries and convergence of currents (Figs 1D - F). Conversely to Castro and

Miranda (1998), who proposed a division of the Brazilian continental shelf for practical

47

reasons, Mahiques et al. (2010) suggest a division in terms of geology, bathymetry, declivities and the presence of canyons and channels (Fig. 1C).

2.2. **Franciscana dataset**

Franciscana records used in the present analyses corresponded to observations of live animals *in situ* through dedicated aerial and boat surveys or to specimen entangled in coastal gillnets fisheries in Brazil (for which precise location data were available). Only data from the marine environment were considered, therefore franciscana records previously observed in estuarine areas such as Babitonga Bay (Cremer and Simões-Lopes, 2005, 2008) and Paranaguá Bay (Santos et al., 2009) in southern Brazil were not included. Only sightings data from dedicated surveys and georeferenced data from bycatch were used in the present analysis in order to estimate franciscana's fundamental ecological niche. Sampling effort and potential biases associated with non-uniform sampling effort, especially those related to fishery monitoring, have not been considered in this study.

Data from aerial surveys were obtained through dedicated line transect studies designed to assess franciscana distribution and to estimate abundance (details in Danilewicz et al., 2010, 2012; Zerbini et al., 2010). Bycatch data were obtained directly by some of the authors via onboard surveys or logbook information provided by reliable and well known captains of fishing vessels operating along the Brazilian coast from 1992 to 2004 (Danilewicz, 2007; Danilewicz et al., 2009; Ott, 1998; Secchi et al., 1997, 2004). Additional records were obtained from peer-reviewed literature (Di Beneditto, 2003; Di

Beneditto et al., 2001; Flores, 2009; Moreno et al., 2003; Santos and Netto, 2005; Santos et al., 2002, 2009; Siciliano et al., 2002).

2.3. Environmental dataset

Ten environmental variables that are considered to influence cetaceans distributions (e.g. Baumgartner et al., 2001; Palacios et al., 2013; Redfern et al., 2006) and specifically franciscanas (Gomez and Cassini, 2015; Siciliano et al., 2002) were initially selected to describe the characteristics of the franciscana's habitats and distributional gaps (Table 1).

Environmental information was obtained from Bio-Oracle (Tyberghein et al., 2012) and MARSPEC (Sbrocco and Barber, 2013). These public databases provide a set of user-friendly and high-resolution GIS data layers of the ocean and were designed for species distribution modeling applications (Sbrocco and Barber, 2013; Tyberghein et al., 2012). The layers consist of global coverage satellite-based and *in situ* measured data interpolated and assembled at an annual temporal resolution and at different spatial resolutions (1km and 9km from MARSPEC and Bio-Oracle datasets, respectively). Geophysical layers were derived from the SRTM30_PLUS high resolution bathymetry dataset (Sbrocco and Barber, 2013), and bioclimatic layers were derived from a long term dataset from NOAA's World Ocean Atlas and NASA's MODIS satellite imagery (Sbrocco and Barber, 2013; Tyberghein et al., 2012; for more details about environmental dataset access: http://www.marspec.org/ and http://www.oracle.ugent.be/). All environmental layers were processed in ArcGIS 10.2.2 (ESRI, 2013) in datum WGS 84, using the same spatial extent (18ºS to 34ºS) at a 9km resolution.

In order to assess the shelf habitat available for franciscanas in the study area, distance to shore data was obtained from distance to shore layer, in which we extracted its values at 0.5° latitudinal intervals along the 25m and 50m isobaths.

2.4. Environmental analyses

Non-independence of predictor variables is a well-known problem in ecology (e.g. Dormann et al., 2013), and it is recommended a preliminary selection of layers in order to avoid redundant data layers in ecological niche analysis (e.g. Qiao et al., 2016). Therefore, correlation of environmental layers was assessed, and factorial analyses were used to select variables with low multicollinearity. Collinearity analyses were conducted in R Statistical Software version 3.2.4 (R Development Core Team, 2016) using the corrplot package (Wei and Simko, 2016) on all variables presented in Table 1, with the exception of distance to shore.

Non-parametric tests (Kruskal-Wallis and Dunn tests) were conducted to provide a preliminary assessment of potential differences between occupied and unoccupied areas with respect to environmental variables selected by the factorial analysis. In order to comply with the assumptions of independence and randomization of sampling required by nonparametric tests, sample points randomly distributed throughout the study areas were used. In a first step, polygons were designed representing areas adjacent to the gaps (i.e. FMA Ia, FMA Ib and FMA II) and areas not occupied by franciscana (i.e. gaps). The polygons were constrained longitudinally by the 50m isobath, and latitudinally by the limits for the new FMAs proposed here (see results section). In a second step, a number of random points within each polygon were generated taking into account the proportions of areas (100 points were created within the polygon with the smallest area and so forth).

Data were then grouped as "Area occupied by Franciscana (AOF)", Gap I, and Gap II. AOF corresponded to the region between Itaúnas in Espírito Santo, and the center of Ilha de Santa Catarina in the state of Santa Catarina, without a discrimination of FMAs and the exclusion of distributional gaps. Finally, significant differences were tested among the medians of the variables identified by the factorial analysis using a Kruskal-Wallis test followed by the Dunn test. All statistical tests were performed in software R Statistical Software version 3.2.4 (R Development Core Team, 2016) using the nortest (Gross and Ligges, 2015) and the dunn (Dinno 2017) packages. A significance level of $\alpha=0.05$ was adopted and the p-value for multiple comparisons was adjusted using the Bonferroni method.

2.5. Ecological niche analysis

NicheA 3.0 (Qiao et al., 2016) was used to investigate if the distributional gaps are consistent with franciscana's fundamental ecological niche. NicheA software generates ecological niche models following the Hutchinsonian approach of an n-multidimensional space, and projects these models in geographic space in the form of continuous species suitability models (for details, see Qiao et al., 2016). NicheA assumes that a species' fundamental ecological is convex in shape, and thus can be operationalized as minimum-volume ellipsoids (MVE) (Qiao et al., 2016). Similar to others modeling approaches (e.g. Maxent – Phillips et al., 2006), MVE could be influenced by sampling biases; however, MVE is only influenced by bias in the periphery of the cloud points. If there are sampling biases that affect the concentrations of points in the interior of the cloud, those will have no effect (A. Townsend Peterson 2017, personal communication). This means that MVE is not influenced by the density of the points (Huiji Qiao 2017,

personal communication). Considering the biased nature of the franciscana data set (e.g. uncorrected for effort), NicheA was deemed the most suitable tool to investigate the characteristics of the franciscana's distribution. In order to better represent the franciscana's fundamental niche, all types of records (bycatch, aerial and boats surveys) were pooled.

Finally, MVEs, representing the franciscanas' fundamental ecological niche, were projected to a habitat suitability map. For the MVE, continuous values of suitability were assessed as the Euclidean distance to the niche centroid (Qiao et al., 2016). The most suitable areas are those closest to the niche centroid (with values close to 1), while the most unsuitable are those areas further away from the niche centroid (with values close to 0); areas totally outside of species niche were set to -1 suitability.

3. RESULTS

3.1. Franciscana distribution update

In total, 788 records of franciscanas in Brazil were compiled from Itaúnas (18º25'S) in Espírito Santo to Chuí River Mouth (33º44'S) in the state of Rio Grande do Sul, located on the Brazil-Uruguay border (Fig. 2). Most of the data were collected between 1992 and 2014. Bycatch data represented 78% of these records, sightings from aerial surveys represented 20.9% and sightings from boat surveys accounted for only 1.1% of the overall data (records for each FMA are summarized in Table 2).

Based on the records compiled, a reassessment of the limits of the FMAs and the distributional gaps were proposed (Table 2, Fig. 3). The distributional gaps were defined as following: Gap I is located from Santa Cruz (19º57'S) to Barra de Itabapoana

(21º18'S) in Espírito Santo State; Gap II is located from Armação dos Búzios (22º44'S) to Piraquara de Dentro (22º59'S) in Rio de Janeiro.

3.2. Environmental layers analyses

From nine layers initially considered to have some influence in the franciscana's distribution, four pairs of environmental layers exhibited correlation coefficient higher than 0.7 (Fig. 4). Therefore, the following environmental layers were selected based on the highest value of each factor of factorial analyses (Table 3): Mean Annual Diffuse Attenuation, Annual Range in Sea Surface Temperature, Mean Annual Sea Surface Salinity, and Bathymetry.

Polygons representing areas adjacent to the gaps (i.e. FMA Ia, FMA Ib and FMA II) and areas not occupied by franciscana (i.e. gaps) are presented in Fig. 5. Considering the proportions of areas, the number of random points created for each polygon is presented in Table 4.

Differences between AOF and Gap I were statistically significant for Mean Annual Sea Surface Salinity (Tables 5 and 6, Fig. 6A), and Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B). Differences between AOF and Gap II were statistically significant for Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B) and Mean Annual Diffuse Attenuation (Tables 5 and 6, Fig. 6C). Gap I and Gap II were statistically differentiated in relation to Mean Annual of Sea Surface Salinity (Tables 5 and 6, Fig. 6A), Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B) and Mean Annual of Diffuse Attenuation (Tables 5 and 6, Fig. 6C). Bathymetry was not statistically different among the areas analyzed (Table 5, Fig. 6D). In general, Gap I had the highest median of Mean Annual of Sea Surface Salinity; Gap II had the highest

median of Mean Annual of Diffuse Attenuation; and, AOF had the highest median of Annual Range in Sea Surface Temperature.

3.3. Ecological niche analysis

The franciscana's Minimum-Volume Ellipsoid (MVE, representing the franciscana's fundamental ecological niche) was estimated using 788 occurrence records in a three-dimensional environmental space represented by Mean Annual Diffuse Attenuation, Annual Range in Sea Surface Temperature, Mean Annual Sea Surface Salinity, and Bathymetry in NicheA.

The franciscana's distribution model (i.e. the MVE projected in geographic space) revealed that the waters in the continental shelf up to 25m were closest to niche centroid (values close to 1), therefore these areas corresponded to the most suitable habitat for franciscanas (Fig. 7). On the other hand, water depths between 25m and 50m isobaths exhibited a progressive decrease of environmental suitability and were more distant from franciscana niche centroids (values close to 0). Gap I and Gap II exhibited values of distance to niche centroid lower than 0.75.

3.4. Shelf habitat availability

The 25m isobath was very close to the shore in the areas corresponding to distributional gaps (i.e. very little area with shallow waters), while the areas suitable for franciscanas were characterized by shallow waters (up to the 50m isobath) up to quite some distance from the coast. In the Gap I, 25m isobaths were identified between 5km and 30km of coastal line. In the Gap II, the 25m isobaths were at less than 10 km from shore, as close as just 1km from the coast line at 23ºS (close to Arraial do Cabo in Rio de Janeiro; see Fig. 8).

The location of the 50m isobath was similar to those of 25m, being closest to shore in the areas corresponding to the gaps. In the Gap I, 50m isobaths was more than 30km far from coast line. In the Gap II, 50m isobaths was positioned closest to shore, being less than 1 km far from shore at 23ºS (Fig. 8). In addition to the distributional gaps, a marked narrowing of continental shelf is also observed around the Ilha de Santa Catarina (27º35'S) (Fig. 8).

4. DISCUSSION

The comprehensive review of the franciscana's occurrences along Brazilian coastal waters support the boundaries of FMAs as well as distributional gaps proposed by franciscanas' experts recently (see Anonymous, 2015; Ott et al., 2015). In relation to previous studies (for instance Secchi et al., 2003a; Siciliano et al., 2002), FMA Ia was extended further south from Regência (19º40'S) to Santa Cruz (19º57'S) in Espírito Santo; the southern limit of FMA Ib was relocated southward from Macaé (22º25'S) to Armação de Búzios (22º44'S) in Rio de Janeiro, due to the stranding of a live animal in the locality of Manguinhos, Armação de Búzios, reported by Siciliano et al. (2015). The northern limit of FMA II was established as Piraquara de Dentro (22º59'S) in Rio de Janeiro, while the southern limit was dislocated further northward from Torres (29º20'S) to the center of Ilha de Santa Catarina (27º35'S) in Santa Catarina, based on previous genetic studies (Cunha et al., 2014; Ott, 2002) (see Fig. 3). These changes on FMAs have impact direct on the extension of distributional gaps, which by its turn were reduced in relation to previous studies (Azevedo et al., 2002; Danilewicz et al., 2012; de Moura et al., 2009; Secchi et al., 2003a; Siciliano et al., 2002).

In general, the habitat suitability model presented here confirmed the well-known distribution of franciscanas (Danilewicz et al., 2009), indicating high environmental suitability for the species mainly up to the 25m isobath (Fig. 7). However, this highly suitable environment could extend up to 50m in the southernmost portion of franciscanas' distribution in Brazil as already indicated by Danilewicz et al. (2009). The ecological niche analyses also showed that both distributional gaps seem suitable for franciscanas at some level and they are inside of the fundamental niche of species.

The resulting map of environmental suitability generated here is consistent with that proposed by Gomez and Cassini (2015), where habitat suitability map indicated high suitability for franciscanas in waters up to approximately 30m depth from Brazil to Argentina (Gomez and Cassini, 2015). Even though Gomez and Cassini (2015) did not include bathymetry as a predictor, their resulting map agreed with the IUCN map. On the other hand, the franciscana's IUCN map was proposed by experts based on the 30m isobath to establish the eastern border of franciscanas' distribution. In contrast to Gomez and Cassini (2015), the habitat suitability model proposed here indicated some level of suitability for franciscana in the gaps.

Bathymetry and distance to shore are considered important predictors of franciscanas' distribution (e.g. Danilewicz et al., 2009; Secchi and Ott, 1999), since individuals are rarely recorded beyond 50m isobaths (Danilewicz et al., 2009). However, the present analysis did not indicate that bathymetry differs statistically among the area occupied by franciscanas and the gaps (Fig. 6D). On the other hand, the analysis of shelf habitat availability indeed revealed that the continental platform is extremely narrow in the gaps, reaching just 1km of distance from shore at 23ºS, for instance (Fig. 8). It was

already suggested the narrowing of the continental shelf in the distributional gaps would limit habitat availability for franciscanas (Di Beneditto et al., 2001; Netto and Siciliano, 2007; Siciliano et al., 2002).

A similar example has been demonstrated in the western South Atlantic with the Atlantic spotted dolphin *Stenella frontalis* (G. Cuvier 1829). A gap in the distribution of this dolphin species exists where the Brazilian continental shelf narrows substantially between Abrolhos Bank (~18ºS) and 6ºS (Danilewicz et al., 2013; Moreno et al., 2005). Ecological niche modeling revealed lack of optimal environmental conditions for the species in the region of the coast where the continental shelf narrows (see do Amaral et al., 2015). In relation to franciscanas' distribution it is also interesting to note that a narrowing of continental shelf also exists around Ilha de Santa Catarina (27º35'S), where the limits between the FMA II and FMA III has been proposed (Ott, 2002; Cunha et al., 2014).

Analysis performed here showed that Gap II is located in a very restrict band of continental shelf, where the coastline orientation changes abruptly from NE-SW to E-W (Castro and Miranda, 1998). The continental shelf is almost nonexistent in the Gap II resulting in a drastic reduction of the shelf habitat even if other conditions such as temperature, salinity and productivity could support the existence of species. In relation to Gap I, for example, the narrow shelf associated with higher levels of salinity could play a role to explain the absence of franciscanas in this area.

As suitability was projected in the distributional gaps, the absence of franciscana could be attributed to the reduction in the shelf habitat due to the narrowing of continental shelf. This environmental change could in turn intensify the biotic

interactions such as competition by food and predation with other marine species.

Guiana dolphin *Sotalia guianensis* (Van Bénedén, 1864) is a species with similar habitats

(Da Silva et al., 2010) and could compete with franciscana by food and/or space.

Furthermore, it was already observed a significant overlap in the diet of franciscana and

largehead hairtail *Trichiurus lepturus* Linnaeus, 1758 (see Bittar and Di Beneditto, 2009;

Di Beneditto et al., 2013). Also, the reduction of shelf habitat could enhance the

vulnerability to predation by other cetaceans such as killer whale *Orcinus orca* (Linnaeus

1758) (e.g. Ott and Danilewicz, 1998; Santos and Netto, 2005). Besides the likely

strengthening of these biotic interactions, intrinsic factors, as minimal viable population

size, also can play an important role in this very reduced range of habitat. In fact, a

synergistic effect of biotic and abiotic factors can be determinant for the absence of

franciscanas in the distributional gaps.

As top predators, cetacean distribution is limited by different factors (for example,

productivity, temperature and salinity) that constraint both its prey and predator's

distributions (e.g. Baumgartner et al., 2001; Palacios et al., 2013; Redfern et al., 2006).

Temperature is a well-recognised factor delimiting species distribution (e.g. Jeffree and

Jeffree, 1994) and salinity is also a well-known factor that have influence on cetacean

distribution due to importance of physiological mechanisms to maintain the water and

salt balance in cetaceans (e.g. Xu et al., 2013). Therefore, both salinity and temperature

seem to impose physiological constraints for franciscanas, including stress triggered by

high salt levels (e.g. São Pedro et al., 2015; Xu et al., 2013) and offspring resistance to

cold environment (e.g. Danilewicz 2003).

The presence of franciscana in FMA Ia appears to be associated with the plume of Doce River that probably maintains the levels of salinity more favorable to franciscanas or its prey species (Netto and Siciliano, 2007; Siciliano et al., 2002). Therefore, higher levels of salinity in Gap I could be a potential explanation for the absence of franciscanas in this area, once this gap has the highest median of Sea Surface Salinity. Since the franciscana seems to be associated with areas with great salinity ranges such as estuaries or river mouths (Cremer et al., 2003; Santos et al., 2009; Siciliano et al., 2002), a higher constant salinity could impose some physiological constraint to the franciscana and/or to its prey species. Although franciscana has a fairly opportunist behavior in terms of prey abundance and occurrence (Bassoi, 2005) and shifts in prey composition overtime were already detected in southern Brazil (Secchi et al., 2003b), sciaenid fishes and long-finned squid *Doryteuthis sanpaulensis* (Brakoniecki, 1984) are very representative in the diet of the franciscana along its geographic range (Bassoi, 2005; Danilewicz et al., 2002). Sciaenid species are mainly present in tropical to warm and temperate environments over sandy and muddy bottoms in brackish, estuarine and low-salinity coastal regions (Martins et al., 2016).

Gomez and Cassini (2015) also suggested based on their ecological niche analysis that temperature and salinity could be considered the environmental predictors of franciscana along its entire range as well as the potential distribution of the stripped weakfish *Cynoscion guatucupa* (Cuvier, 1830). They also highlighted the physiological constrains imposed by Sea Surface Temperature and Salinity, and they did not find any support for previous statements that turbidity could be an important ocean determinant of franciscana distribution.

Diffuse attenuation is an indicator of the water turbidity and it is directly related to the presence of scattering particles in the water column. The analysis performed here indicated that the Gap II has the highest median of Diffuse Attenuation. This finding is a little bit controversial in relation to previous studies that suggested a preference for turbid waters by franciscanas (Siciliano et al., 2002). The reader should be aware that these results are influenced by the Guanabara Bay, Rio de Janeiro, and its discharge, which in turn could be increasing the values of Diffuse Attenuation, not reflecting a condition along the entire Gap II.

Finally, it is important to highlight that distributional gaps have an immediate impact on the population structure of franciscanas mainly in relation to franciscanas from FMA I. Genetic evidences based on mtDNA suggests that franciscanas are divided in two evolutionary lineages, franciscanas from FMA I (i.e. those northward Gap II) being a distinct lineage in comparison to all remaining franciscanas (Cunha et al., 2014). Furthermore, FMA I is further sub-structured into FMA Ia and FMA Ib (Cunha et al., 2014). From these findings it is possible to assume that both northern and southern distributional gaps have been acting as a barrier long enough to have an impact on the population structure in the areas adjacent to the gaps. Considering the importance of shelf habitat for franciscanas, it seems to be reasonable to suppose that historical factors such as sea level oscillations and, consequently, fragmentation of coastal platform during glacial and interglacial cycles, play a key role to explain for the absence of franciscana in these areas.

5. CONCLUSIONS

The shelf habitat is very important to franciscana. However, a wide shelf does not necessarily result in an increased presence of franciscana if other conditions such as salinity are not suitable. For example, the Brazilian continental shelf is very large at the north portion of the Espírito Santo, in the region of the Abrolhos Bank, however franciscana range seems to be limited longitudinally, possible due to higher and salt levels recorded there.

The new limits of FMAs and the habitat suitability model presented here could be used as a guide to planning studies and actions that aim the conservation of the franciscana in Brazil. Further studies should investigate franciscanas' prey availability in those areas considered distributional gaps as well the possible relevance of other biotic interactions. Finally, changes in the coastal environment and habitat loss caused by human activities, such as industrial port development, should also be considered in conservation plans for the species.

ACKNOWLEDGEMENTS

REFERENCES

Alonso, M.B., Eljarrat, E., Gorga, M., Secchi, E.R., Bassoi, M., Barbosa, L., Bertozzi, C.P, Marigo, J., Cremer, M., Domit, C., 2012. Natural and anthropogenically-produced brominated compounds in endemic dolphins from Western South Atlantic: Another risk to a vulnerable species. Environ. Pollut. 170, 152–160.

Anonymous, 2015. Report of the VIII Workshop for the Research and Conservation of the Franciscana Dolphin (*Pontoporia blainvillei*) in the Soutwestern Atlantic. UNEP/CMS, São Francisco do Sul, Brazil. http:// www.pontoporia.org

Azevedo, A.F., Fragoso, A.B.L, Lailson-Brito Jr., J., Cunha, H.A., 2002. Records of the franciscana (*Pontoporia blainvillei*) in the southwestern Rio de Janeiro and northernmost São Paulo state coasts - Brazil. LAJAM 1(1), 191 – 192.

Barbato, B.H.A., Secchi, E.R., Di Beneditto, A.P.M., Ramos, R.M.A., Bertozzi, C.,
Marigo, J., Bordino, P., Kinas, P.G., 2012. Geographical variation in franciscana
(*Pontoporia blainvillei*) external morphology. J. Mar. Biol. Assoc. UK 92(8), 1645–
1656.

Bassoi, M., 2005. Feeding ecology of franciscana dolphin, *Pontoporia blainvillei*
(Cetacea: Pontoporiidae), and oceanographic processes on the Southern Brazilian
coast (PhD dissertation) University of Southamptom, Southamptom, England.

Baumgartner, M.F,. Mullin, K.D., May, L.N., Leming, T.D., 2001. Cetacean habitats in
the northern Gulf of Mexico. Fish. Bull. (Wash. DC) 99(2), 219–239.

Bittar, V.T., Di Beneditto, A.P.M., 2009. Diet and potential feeding overlap between
Trichiurus lepturus (Osteichthyes: Perciformes) and *Pontoporia blainvillei*
(Mammalia: Cetacea) in northern Rio de Janeiro, Brazil. Zoologia 26(2), 374-378.

Castro, B.M., Miranda, L.B., 1998. Physical oceanography of the western Atlantic
continental shelf located between 4º N and 34º S - Coastal segment, in: Robinson,
A., Brink, K.H. (Eds) The sea. The Global Coastal Ocean - Regional Studies and
Synthesis. Wiley, New York, pp. 209-251.

Costa-Urrutia, P., Abud, C., Secchi, E.R., Lessa, E.P., 2012. Population genetic structure
and social kin associations of franciscana dolphin, *Pontoporia blainvillei*. J. Hered.
103, 92–102.

Cremer, M.J., Simões-Lopes, P.C., 2005. The occurrence of *Pontoporia blainvillei*
(Gervais & d'Orbigny) (Cetacea, Pontoporiidae) in an estuarine area in southern
Brazil. Rev. Bras. Zool. 22(3), 717 – 723.

Cremer, M.J., Simões-Lopes, P.C., 2008. Distribution, abundance and density estimates

of franciscanas, *Pontoporia blainvillei* (Cetacea: Pontoporiidae), in Babitonga Bay, southern Brazil. Rev. Bras. Zool. 25(3), 397-402.

Crespo, E.A., Harris, G., González, R., 1998. Group size and distributional range of the franciscana, *Pontoporia blainvillei*. Mar. Mamm. Sci. 14(4), 845–849.

Cunha, H.A., Medeiros, B.V., Barbosa, L.A., Cremer, M.J., Marigo, J., Lailson-Brito Jr., J., Azevedo, A., Solé-Cava, A., 2014. Population structure of the endangered franciscana dolphin (*Pontoporia blainvillei*): reassessing management units. PloS One 9(1), e85633.

Da Silva, V.M.F., Fettuccia, D., Rodrigues, E.S., Edwards, H., Moreno, I.B., de Moura, J.F., Wedekin, L., Bazzalo, M., Emin-Lima, R., Carmo, N.A.S., Siciliano, S., Utreras, V., 2010. Report of the working group on distribution, habitat characteristics and preferences, and group size. LAJAM 8 (1-2), 31-38.

Danilewicz, D. 2003. Reproduction of female franciscana (*Pontoporia blainvillei*) in Rio Grande do Sul, southern Brazil. LAJAM 2(2), 67 - 78.

Danilewicz, D., 2007. A toninha, *Pontoporia blainvillei* (Mammalia: Cetacea), no litoral norte do Rio Grande do Sul: mortalidade acidental em redes de pesca, abundância populacional e perspectivas para a conservação da espécie (PhD dissertation) Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil.

Danilewicz, D., Rosas, F., Bastida, R., Marigo, J., Muellbert, M., Rodriguez, D., Lailson-Brito Jr., J., Ruoppolo, V., Ramos, R., Bassoi, M., Ott, P.H., Caon, G., Rocha, A.M., Catão-Dias, J.L., Secchi, E., 2002. Report of the Working Group on Biology and Ecology. LAJAM 1, 25-42.

Danilewicz, D., Secchi, E.R., Ott, P.H., Moreno, I.B., Bassoi, M., Borges-Martins, M.,

    2009. Habitat use patterns of franciscana dolphins (*Pontoporia blainvillei*) off

    southern Brazil in relation to water depth. J. Mar. Biol. Assoc. UK 89, 943–949.

Danilewicz, D., Moreno, I.B., Ott, P.H., Tavares, M., Azevedo, A.F., Secchi, E.R.,

    Andriolo, A., 2010. Abundance estimate for a threatened population of franciscana

    dolphins in southern coastal Brazil: uncertainties and management implications. J.

    Mar. Biol. Assoc. UK 90, 1649–1657.

Danilewicz, D., Zerbini, A., Andriolo, A., Secchi, E.R., Sucunza, F., Ferreira, E.,

    Denuncio, P., Flores, P.A.C., 2012. Abundance and distribution of an isolated

    population of franciscana dolphins (*Pontoporia blainvillei*) in southeastern Brazil:

    red alert for FMA I? Int. Whaling Comm. Working Paper SC/64/SM17.

Danilewicz, D., Ott, P.H., Secchi, E., Andriolo, A., Zerbini, A., 2013. Occurrence of the

    Atlantic spotted dolphin, *Stenella frontalis*, in southern Abrolhos Bank, Brazil. Mar.

    Biodivers. Rec. 6, 1-3.

de la Torre, A., Alonso, M.B., Martínez, M.A., Sanz, P., Shen, L., Reiner, E.J., Lailson-

    Brito Jr., J., Torres, J.P.M., Bertozzi, C., Marigo, J., Barbosa, L., Cremer, M., Secchi,

    E., Malm, O., Eljarrat, E., Barceló, D., 2012. Dechlorane-related compounds in

    franciscana dolphin (*Pontoporia blainvillei*) from Southeastern and Southern Coast

    of Brazil. Environ. Sci. Technol. 46, 12364-12372.

de Moura, J.F., da Silva, É.R., Sholl, T.G.C., Siciliano, S., 2009. Franciscana dolphin

    (*Pontoporia blainvillei*) on the north-east coast of Rio de Janeiro State, Brazil,

    recorded during a long term monitoring programme. Mar. Biodivers. Rec. 2, 1-4.

Di Beneditto, A.P.M., 2003. Interactions between gillnet fisheries and small cetaceans in

northern Rio de Janeiro, Brazil: 2001-2002. LAJAM 2(2), 79-86.

Di Beneditto, A.P.M., Ramos, R.M.A., Lima, N.R.W., 2001. Sightings of *Pontoporia blainvillei* (Gervais & D'Orbigny, 1844) and *Sotalia fluviatilis* (Gervais, 1853) (Cetacea) in South-eastern Brazil. Braz. Arch. of Biol. Techn. 44(3), 291–296.

Di Beneditto, A.P.M., Rezende, C.E., Camargo, P.B., Kehrig, H.A., 2013. Trophic niche comparison between two predators in northern Rio de Janeiro State, Brazil: a stable isotopes approach. Biota Neotrop. 13(3), 29 – 33.

Dinno, A., 2017. dunn.test: Dunn's Test of Multiple Comparisons Using Rank Sums. R package version 1.3.5. https://CRAN.R-project.org/package=dunn.test.

do Amaral, K.B., Alvares, D.J., Heinzelmann, L., Borges-Martins, M., Siciliano, S., Moreno, I.B., 2015. Ecological niche modeling of *Stenella* dolphins (Cetartiodactyla: Delphinidae) in the southwestern Atlantic Ocean. J. Exp. Mar. Biol. Ecol. 472, 166–179.

Dormann, C.F., Elith, J., Bacher, S., Buchmann, C., Carl, G., Carré, G., Marquéz, J.R.G., Gruber, B., Lafourcade, B., Leitão, P.J., Münkemüller, T., McClean, C., Osborne, P.E., Reineking, B., Schröder, B., Skidmore, A.K., Zurell, D., Lautenbach, S., 2013. Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecography 36, 27 – 46.

ESRI, 2013. ArcGIS Desktop: Release 10.2.2. Environmental Systems Research Institute, Redlands, California, USA.

Flores, P.A.C., 2009. Occurrence of franciscana (*Pontoporia blainvillei*) in Baía Norte, Southern Brazil. LAJAM 7(1-2), 93-95.

Gariboldi ,M.C., Túnez, J.I., Dejean, C.B., Failla, M., Vitullo, A.D., Negri, M.F.,

Cappozzo, H.L., 2015. Population genetics of Franciscana Dolphins (*Pontoporia blainvillei*): Introducing a new population from the southern edge of their distribution. PLoS One 10 (7), e0132854.

Gariboldi, M.C., Túnez, J.I., Failla, M., Hevia, M., Panebianco, M.V., Viola, M.N.P., Vitullo, A.D., Cappozzo, H.L., 2016. Patterns of population structure at microsatellite and mitochondrial DNA markers in the franciscana dolphin (*Pontoporia blainvillei*). Ecol. Evol. 1:13 .

Gomez, J.J., Cassini, M.H., 2015. Environmental predictors of habitat suitability and biogeographical range of Franciscana dolphins (*Pontoporia blainvillei*). Global Ecol. Conserv. 3, 90–99.

Gross, J., Ligges, U., 2015. nortest: Tests for Normality. R package version 1.0-4. https://CRAN.R-project.org/package=nortest

Higa, A., Hingst-Zaher, E., De Vivo, M., 2002. Size and shape variability in the skull of *Pontoporia blainvillei* (Cetacea: Pontoporiidae) from Brazillian Coast. LAJAM 1(1), 145–152.

Hoss, D., Valente, A.L., Ott, P.H., 2017. Gastrointestinal helminths of the franciscana *Pontoporia blainvillei* (Mammalia: Cetartiodactyla) in the northern coast of Rio Grande do Sul State, southern Brazil. Oecologia Australis 21(1), 72-82.

Jeffree, E.P., Jeffree, C.E., 1994. Temperature and the biogeographical distributions of species. Funct. Ecol. 8(5), 640-650.

 Kinas, P.G., 2002. The impact of incidental kills by gill nets on the franciscana dolphin (*Pontoporia blainvillei*) in southern Brazil. B. Mar. Sci. 70, 409–421.

Mahiques, M., Sousa, S., Furtado, V., 2010. The Southern Brazilian shelf: general

characteristics, quaternary evolution and sediment distribution. Braz. J. Oceanogr. 58, 25-34.

Martins, A.S., Haimovici, M., 2016. Seasonal mesoscale shifts of demersal nekton assemblages in the subtropical South-western Atlantic. Mar. Biol. Res. 13(1), 88-97.

Mendez, M., Rosenbaum, H.C., Subramaniam, A., Yackulic, C., Bordino, P., 2010. Isolation by environmental distance in mobile marine species: molecular ecology of franciscana dolphins at their southern range. Mol. Ecol. 19, 2212–2228.

MMA, 2014. Lista Nacional Oficial de Espécies da Fauna Ameaçadas de Extinção – Mamíferos, Aves, Répteis, Anfíbios e Invertebrados Terrestres. Portaria MMA n°. 444, de 17 de dezembro de 2014, Brazil. http://www.icmbio.gov.br/cepsul/images/stories/legislacao/Portaria/2014/p_mma_44 4_2014_lista_esp%C3%A9cies_ame%C3%A7adas_extin%C3%A7%C3%A3o.pdf (accessed 15.05.2017).

Moreno, I.B., Martins, C.C.A., Andriolo, A., Engel, M.H., 2003. Sightings of franciscana dolphins (*Pontoporia blainvillei*) off Espírito Santo, Brazil. LAJAM 2(2), 131 – 132.

Moreno, I.B., Zerbini, A.N., Danilewicz, D., Santos, M.C.O., Simões-Lopes, P.C., Laílson-Brito Jr., J., Azevedo, A.F., 2005. Distribution and habitat characteristics of dolphins of the genus *Stenella* (Cetacea: Delphinidae) in the southwest Atlantic Ocean. Mar. Ecol. Prog. Ser. 300, 229–240.

Netto, R.F., Siciliano, S., 2007. Contribuição ao conhecimento da distribuição da toninha *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) no estado do Espírito Santo, sudeste do Brasil. Bol. Mus. Biol. Mello Leitão 21, 35 – 45.

Ott, P.H., 1998. Análise das capturas acidentais da toninha, *Pontoporia blainvillei*, no

litoral norte do Rio Grande do Sul, sul do Brasil (MSc Thesis) Pontifícia

Universidade Católica do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul,

Brazil.

Ott, P.H., 2002. Diversidade genética e estrutura populacional de duas espécies de

cetáceos do Atlântico Sul Ocidental: *Pontoporia blainvillei* e *Eubalaena australis*

(PhD Dissertation) Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio

Grande do Sul, Brazil.

Ott, P.H., Danilewicz, D., 1998. Presence of franciscana dolphins (*Pontoporia blainvillei*)

in the stomach of a killer whale (*Orcinus orca*) stranded in southern Brazil.

Mammalia 62(4), 605-609.

Ott, P.H., Secchi, E.R., Moreno, I.B., Danilewicz, D., Crespo, E.A., Bordino, P., Ramos,

R., Di Beneditto, A.P., Bertozzi, C., Bastida, R., Zanelato, R., Perez, J., Kinas, P.G.,

2002. Report of the working group on fishery interactions. LAJAM 1, 55–64.

Ott, P.H., Domit, C., Siciliano, S., Flores, P.A.C. 2015. Memórias do VII Workshop para

a coordenação de pesquisa e conservação de *Pontoporia blainvillei* (Gervais &

d'Orbigny, 1844). http:// www.pontoporia.org

Palacios, D., Baumgartner, M., Laidre, K., Gregr, E., 2013. Beyond correlation:

integrating environmentally and behaviourally mediated processes in models of

marine mammal distributions. Endanger. Species Res. 22, 191–203.

Pinedo, M.C., 1995. Development and variation in external morphology of the

franciscana, *Pontoporia blainvillei*. Rev. Bras. Biol. 55, 85–96.

Phillips, S.J., Anderson, R.P., Schapire, R.E., 2006. Maximum entropy modeling of

species geographic distributions. Ecol. Model. 190, 231–259.

Prado, J.H.F., Mattos, P.H., Silva, K.G., Secchi, E.R., 2016. Long-term seasonal and interannual patterns of marine mammal strandings in Subtropical Western South Atlantic. PLoS One 11(1), e0146339.

Prado, J.H.F., Secchi, E.R., Kinas, P.G., 2013. Mark-recapture of the endangered franciscana dolphin (*Pontoporia blainvillei*) killed in gillnet fisheries to estimate past bycatch from time series of stranded carcasses in southern Brazil. Ecol. Indic. 32, 35 – 41.

Qiao, H., Peterson, A.T., Campbell, L., Soberón, J., Ji, L., Escobar, L.E., 2016. NicheA: Creating virtual species and ecological niches in multivariate environmental scenarios. Ecography 39, 1-9.

R Development Core Team, 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna (www.r-project.org/. Retrieved 23 July 2016).

Ramos, R.M.A., Di Beneditto, A.P., Siciliano, S., Santos, M.C.O., Zerbini, A., Bertozzi, C., Vicente, A.F.C., Zampirolli, E., Alvarenga, F.S., Lima, N.R.W., 2002. Morphology of the franciscana (*Pontoporia blainvillei*) off southeastern Brazil: sexual dimorphism, growth and geographic variation. LAJAM 1(1), 129 – 144.

Redfern, J.V., Ferguson, M.C., Becker, E.A., Hyrenbach, K.D., Good, C.P., Barlow, J., Kaschner, K., Baumgartner, M., Forney, K., Balance, L., Fauchland, P., Halpin, P., Hamazaki, T., Pershing, A.J., Qian, S.S., Read, A., Reilly, S.B., Torres, L., Werner, F., 2006. Techniques for cetacean–habitat modeling. Mar. Ecol. Prog. Ser. 310, 271-295.

Rocha-Campo, C.C., Danilewicz, D., Siciliano, S., 2010. Plano de ação nacional para a conservação do pequeno cetáceo toninha: *Pontoporia blainvillei.* Série Espécies Ameaçadas n⁰ 10, Instituto Chico Mendes de Conservação da Biodiversidade, Brasília, Brazil.

Rossi-Santos, M.R., Oliveira, G., 2016. Combining cetacean soundscape ecology and niche modeling to contribute in the mapping of the Brazilian Continental Shelf, in: Finkl, C.W., Makowski, C. (Eds.) Seafloor mapping along Continental Shelves. Research and techniques for visualizing benthic environments. Springer International Publishing, Switzerland, pp. 146 – 165.

Santos, M.C.O., Vicente, A.F.C., Zampirolli, E., Alvarenga, F.S., Souza, S., 2002. Records of franciscana (*Pontoporia blainvillei*) from the coastal waters of São Paulo state, southeastern Brazil. LAJAM 1(1), 169-174.

Santos, M.C.O., Netto, D., 2005. Killer whale (*Orcinus orca*) predation on a franciscana dolphin (*Pontoporia blainvillei*) in Brazilian waters. LAJAM 4, 62-72.

Santos, M.C.O., Oshima, J.E.F., da Silva, E., 2009. Sightings of franciscana dolphins (*Pontoporia blainvillei*): the discovery of a population in the Paranaguá Bay estuarine complex, southern Brazil. Braz. J. Oceanogr. 57(1), 57 – 63.

São Pedro, S.L., Alves, J.M.P., Barreto, A.S., Lima, A.O.S., 2015. Evidence of positive selection of aquaporins genes from Pontoporia blainvillei during the evolutionary process of cetaceans. PLoS One 10(7), e0134516.

Sbrocco, E.J., Barber, P.H., 2013. MARSPEC: ocean climate layers for marine spatial ecology. Ecology 94, 979–979.

Secchi, E.R. 2006. Modelling the population dynamics and viability analysis of

franciscana (*Pontoporia blainvillei*) and Hector's dolphins (*Cephalorhynchus hectori*) under the effects of bycatch in fisheries parameter uncertainty and stochasticity (PhD Dissertation) University of Otago, Dunedin, New Zealand.

Secchi, E.R., Zerbini, A.N., Bassoi, M., Dalla-Rosa, L., Möller, L.M., Rocha-Campos, C.C., 1997. Mortality of franciscanas, *Pontoporia blainvillei*, in coastal gillnetting in southern Brazil: 1994-1995. Rep. Int. Whaling Comm. 47, 653-658.

Secchi, E.R., Wang, J.Y., Murray, B.W., Rocha-Campos, C.C., White, B.N., 1998. Population differentiation in the franciscana (*Pontoporia blainvillei*) from two geographic locations in Brazil as determined from mitochondrial DNA control region sequences. Can. J. Zool. 76, 1622–1627.

Secchi, E.R., Ott, P.H., 1999. A profundidade como um fator determinante da distribuioção de toninhas, *Pontoporia blainvillei*, conforme indicado pelos índices CPUE. Report of the Third Workshop for Coordinated Research and Conservation of the Franciscana Dolphin (*Pontoporia blainvillei*) in the Soutwestern Atlantic. http://www.pontoporia.org/.

Secchi, E.R., Danilewicz, D., Ott, P.H., 2003a. Applying the phylogeographic concept to identify franciscana dolphin stocks: implications to meet management objectives. J. Cetac. Res. Manage. 5(1), 61-68.

Secchi, E.R., Ott, P.H., Danilewicz, D., 2003b. Effects of fishing by-catch and conservation status of the franciscana dolphin, *Pontoporia blainvillei*, in: Gales, N., Hindell, M., Kirkwood, K. (Eds.) Marine Mammals: Fisheries, Tourism and Management Issues. CSIRO Publishing, Collinwoop, Australia, pp. 174 – 191.

Secchi, E.R., Kinas, P.G., Muelbert, M., 2004. Incidental catches of franciscana in coastal

gillnets fisheries in the Franciscana Management Area III: period 1999-2000. LAJAM 3(1), 61-68.

Siciliano, S., Di Beneditto, A.P., Ramos, R., 2002. A toninha, *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) (Mammalia, Cetacea, Pontoporidae), nos estados do Rio de Janeiro e Espírito Santo, costa sudeste do Brasil: Caracterização dos hábitats e fatores de isolamento das populações. Boletim do Museu Nacional 476, 1-15.

Siciliano, S., de Moura, J.F., Secco, H.K.C. 2015. Considerações sobre a distribuição da toninha (*Pontoporia blainvillei*, Gervais & d'Orbigny, 1844) na costa centro-norte do estado do Rio de Janeiro, Brasil, in: Ott, P.H., Domit, C., Siciliano, S., Flores, P.A.C. (Eds.) Memórias do VII Workshop para a coordenação de pesquisa e conservação de *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844).

Tyberghein, L., Verbruggen, H., Pauly, K., Troupin, C., Mineur, F., De Clerck, O., 2012. Bio-ORACLE: a global environmental dataset for marine species distribution modelling. Glob. Ecol. Biogeogr. 21, 272–281.

Wei, T., Simko, V., 2016. corrplot: Visualization of a Correlation Matrix. R package version 0.77. http://CRAN.R-project.org/package=corrplot

Xu, S., Yang, Y., Zhou, X., Xu, J., Zhou, K., Yang, G., 2013. Adaptive evolution of the osmoregulation-related genes in cetaceans during secondary aquatic adaptation. BMC Evol. Biol. 13, 189.

Zerbini, A., Secchi, E., Danilewicz, D., Andriolo, A., Laake, J., Azevedo, A., 2011. Abundance and distribution of the franciscana (*Pontoporia blainvillei*) in the Franciscana Management Area II (southeastern and southern Brazil). Int. Whaling Comm. Working Paper SC/62/SM7.

Zerbini, A.N., Secchi, E., Crespo, E., Danilewicz, D., Reeves, R. 2017. *Pontoporia blainvillei*. The IUCN Red List of Threatened Species 2017: e.T17978A123792204. http://www.iucnredlist.org/details/17978/0/ (accessed 15.05.2018).

TABLES

Table 1. List of environmental variables analyzed in this study and its respective source, resolution and unit.

Environmental Variables	Source	Unit	Original Resolution
Bathymetry (Depth of the seafloor)	MARSPEC	meters	1 km
Distance to shore	MARSPEC	kilometres	1 km
Bathymetric Slope	MARSPEC	degrees	1 km
Mean Annual Concentration of Chlorophyll A	Bio-Oracle	mg/m³	9 km
Annual Range in Concentration of Chlorophyll A	Bio-Oracle	mg/m³	9 km
Mean Annual Diffuse Attenuation	Bio-Oracle	m^{-1}	9 km
Mean Annual Sea Surface Salinity	MARSPEC	Psu	1 km
Annual Range in Sea Surface Salinity	MARSPEC	Psu	1 km
Mean Annual Sea Surface Temperature	MARSPEC	degrees °C	1 km
Annual Range in Sea Surface Temperature	MARSPEC	degrees °C	1 km

Table 2. Summary of franciscanas' records by areas and data source and gaps limits. FMAs were established according to Cunha et al., (2014) and limits were updated. Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SC, Santa Catarina; RS, Rio Grande do Sul.

Records Summary Information					
Areas	New limits	Aerial Surveys	Boat Surveys	Bycatch	Total
FMA Ia	Itaúnas, ES (18º25'S) to Santa Cruz, ES (19º57'S)	6	0	0	6
Gap I (north)	Piraquê-Açu River Mouth, Santa Cruz, ES (19º57'S) to Barra de Itabapoana, ES (21º18'S)				
FMA Ib	Barra de Itabapoana, RJ (21º18'S) to Armação de Búzios, RJ (22º44'S)	13	2	11	26
Gap II (south)	Armação dos Búzios, RJ (22º44'S) to Piraquara de Dentro, RJ (22º59'S)				
FMA II	Piraquara de Dentro, RJ (22º59'S) to Ilha de Santa Catarina, SC (27º35'S)	41	7	60	108
FMA III *	Ilha de Santa Catarina, SC (27º35'S) to Chuí River Mouth, RS (33º44'S)	105	0	543	648
TOTAL		165	9	614	788
Percentage (%)		20.9%	1.1%	78.0%	100.0%

*FMA III is partially represented, because it extends into Uruguay.

Tables 3. Factorial Analysis of nine environmental variables used in this study. Abbreviations: bat, Bathymetry; dist, Distance to Shore; slope, Bathymetric Slope; da_mean, Mean Annual Diffuse Attenuation; cl_mean, Mean Annual Concentration of Chlorophyll A; cl_range, Annual Mean in Concentration of Chlorophyll A; sss_mean, Mean Annual Sea Surface Salinity; sss_range, Annual Range in Sea Surface Salinity; sst_mean, Mean Annual Sea Surface Temperature; sst_range, Annual Range in Sea Surface Temperature.

	Factor1	Factor2	Factor3	Factor4	Factor5
bat	0.135			**0.771**	
slope	-0.143	-0.226	0.155		0.126
cl_mean	0.929	0.190		0.289	
cl_range	0.695	0.440	-0.169	-0.190	0.163
da_mean	**0.959**	0.122		0.128	-0.208
sss_mean		-0.121	**0.976**	-0.147	
sss_range	0.118	0.791	-0.271		
sst_mean	-0.123	-0.404	0.837	0.102	
sst_range	0.256	**0.884**	-0.129		
SS loadings	2.402	1.887	1.805	0.786	0.104
Proportion Var	0.267	0.210	0.201	0.087	0.012
Cumulative Var	0.267	0.477	0.677	0.765	0.776

Table 4. Information used to investigate the environmental distinctiveness of areas occupied and not occupied by franciscanas. Abbreaviation: AOF, Area Occupied by Franciscana.

	Areas	Proportion in relation to the smallest polygon (i.e., Gap II)	Number of Random Points Generated
AOF	Area Ia	2.44	244
	Area Ib	2.34	234
	Area II	7.72	772
Gap I		1.48	148
Gap II		1	100

Table 5. Medians comparisons through Kruskal-Wallis. Statistically significant values are in bold.

Environmental Layer	Kruskal-Wallis Test	
Mean Annual Sea Surface Salinity	**χ2 = 106.45**	**p-value < 0.05**
Annual Range in Sea Surface Temperature	**χ2 = 164.01**	**p-value < 0.05**
Mean Annual Diffuse Attenuation	**χ2 = 19.799**	**p-value < 0.05**
Bathymetry	χ2 = 4.0115	p-value =0.13

Table 6. Areas medians comparisons through Dunn tests for each environmental layer, which Kruskall Wallis test was significant. Abbreviation: AOF, Area Occupied by Franciscana. P-value is indicated among parenthesis and statistically significant values are in bold.

	Dunn Test		
	AOF	Gap I	
Mean Annual Sea Surface Salinity	**-10.229 (0.00001)**	-	Gap I
	0.433967 (0.9965)	7.218 (0.00001)	Gap II
Annual Range in Sea Surface Temperature	**12.13 (0.00001)**	-	Gap I
	5.165059 (0.00001)	**-3.999 (0.0001)**	Gap II
Mean Annual Diffuse Attenuation	-0.664 (0.7597)	-	Gap I
	-4.441 (0.00001)	**-3.119 (0.0027)**	Gap II

FIGURE LEGENDS

Figure 1. A) Study area of franciscana dolphin distribution. Brazilian continental shelf zones proposed by B) Castro & Miranda (1998) and C) Mahiques et al. (2010). Representation of annual means of D) Mean Annual Sea Surface Temperature (SST), E) Mean Annual Concentration of Chlorophyll A, and F) Mean Annual Sea Surface Salinity (SSS). Abreviations: ACR, Abrolhos – Campos Region; SBB, South Brazilian Bight; SBS, Southern Brazilian Shelf; SPB, São Paulo Bight; FMB, Florianópolis – Mostardas Bight (FMB); RGC, Rio Grande Cone; TZ, Transitional Zone.

Figure 2. Compiled records of franciscana dolphin along Brazilian coastal waters from Itaúnas (ES) to Chuí River Mouth (RS). Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SP, São Paulo; PR, Paraná; SC, Santa Catarina; RS, Rio Grande do Sul.

Figure 3. New geographic ranges of the Franciscanas Management Areas (FMAs) and distributional gaps. Localities already considered limits are indicated in the map by triangles symbols. Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SP, São Paulo; PR, Paraná; SC, Santa Catarina; RS, Rio Grande do Sul. *FMAIII is partially represented because this management area extends further to the south to include the coast of Uruguay.

Figure 4. Correlation matrix of nine environmental variables evaluated in the study. Abbreviations: bat, Bathymetry; slope, Bathymetric Slope; da_mean, Mean Annual Diffuse Attenuation; cl_mean, Mean Annual Concentration of Chlorophyll A; cl_range, Annual Mean in Concentration of Chlorophyll A; sss_mean, Mean Annual Sea Surface

Salinity; sss_range, Annual Range in Sea Surface Salinity; sst_mean, Mean Annual Sea Surface Temperature; sst_range, Annual Range in Sea Surface Temperature.

Figure 5. Map of polygons used to create random points in order to represent the area occupied by franciscana and those not occupied. Abbreviation: AOF, Area occupied by Franciscana.

Figure 6. Boxplot of environmental values extracted from random points grouped according area occupied by franciscana (AOF) and those not occupied. Boxplot represents median, 25th and 75th percentiles, and 5th and 95th are represented by the errors bars. In A) Mean Annual Sea Surface Salinity; B) Annual Range in Sea Surface Temperature; C) Mean Annual of Diffuse Attenuation; and D) Bathymetry.

Figure 7. A) Habitat suitability model with continuous values of the distance to the niche centroid representing the franciscana's fundamental niche. B) A map zoom is provided to visualize the environmental suitability of northern and southern distributional gaps. Values between 0 and 1 represent the relative distance between the points and the centre of the ellipsoid; -1 represent areas out of the ellipsoid or unsuitable; 0 means areas on the edge of the ellipsoid or with low suitability; and 1 means areas on the center of the ellipsoid or with high environmental suitability (H. Qiao 2017, p.c.).

Figure 8. Distance to shore of the 25m and 50m isobaths in relation to latitude. Area occupied by franciscana are represented in green; distributional gaps are represented in red (left, Gap I; right, Gap II).

slope	0.21	0.26	-0.15	-0.23	-0.23	-0.21	-0.21	-0.21
	sss_mean	0.85	-0.16	-0.36	-0.23	-0.22	-0.19	-0.13
		sst_mean	0.06	-0.55	-0.51	-0.44	-0.24	-0.17
			bat	-0.1	-0.11	-0.08	0.33	0.22
				sss_range	0.77	0.51	0.25	0.22
					sst_range	0.6	0.4	0.33
						cl_rg	0.7	0.67
							cl_mean	0.94
								da_mean
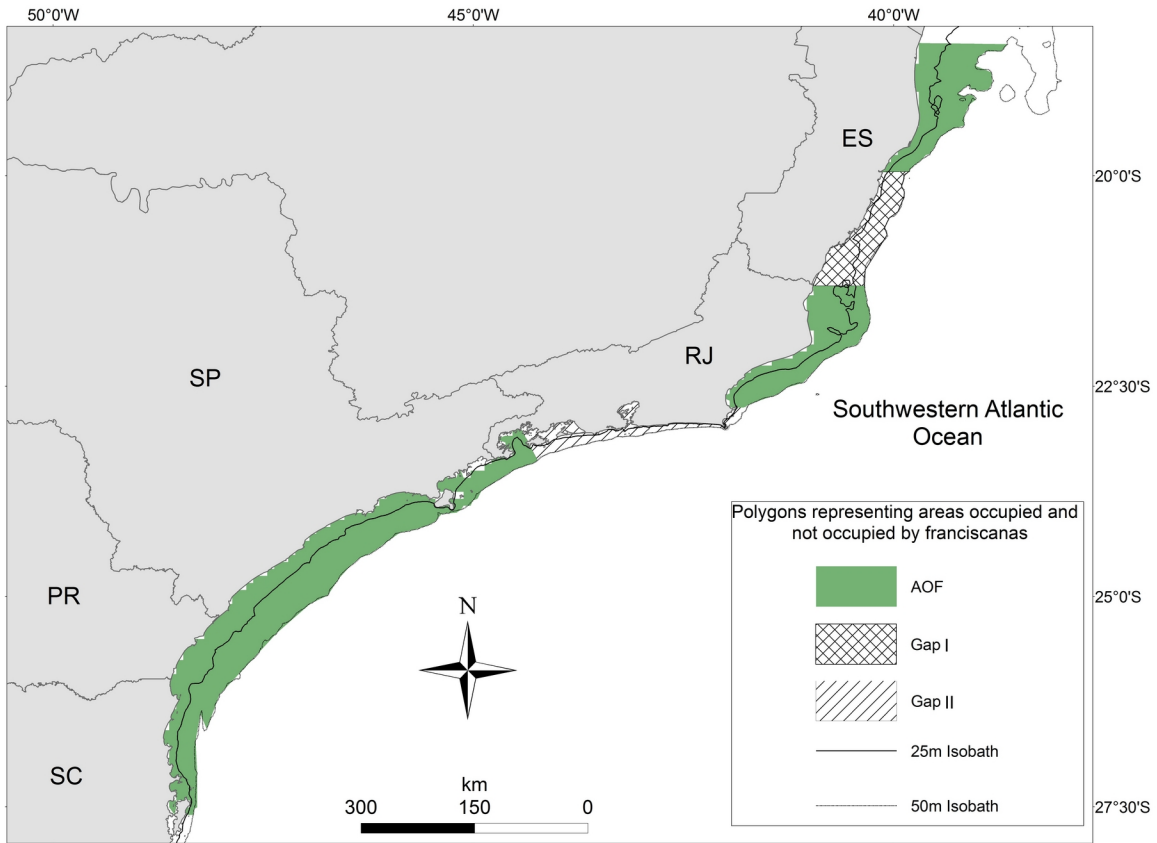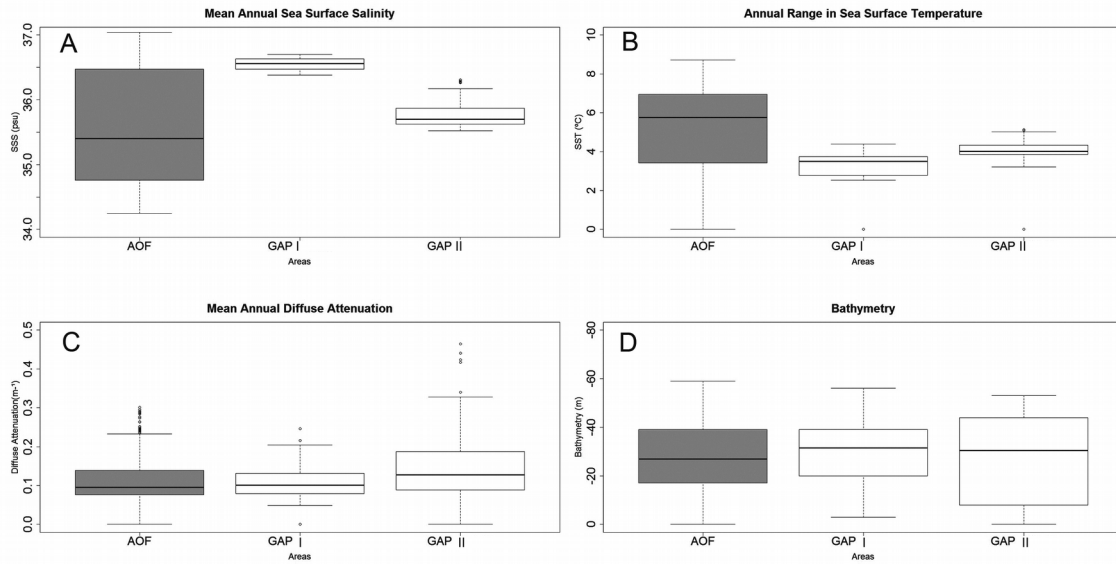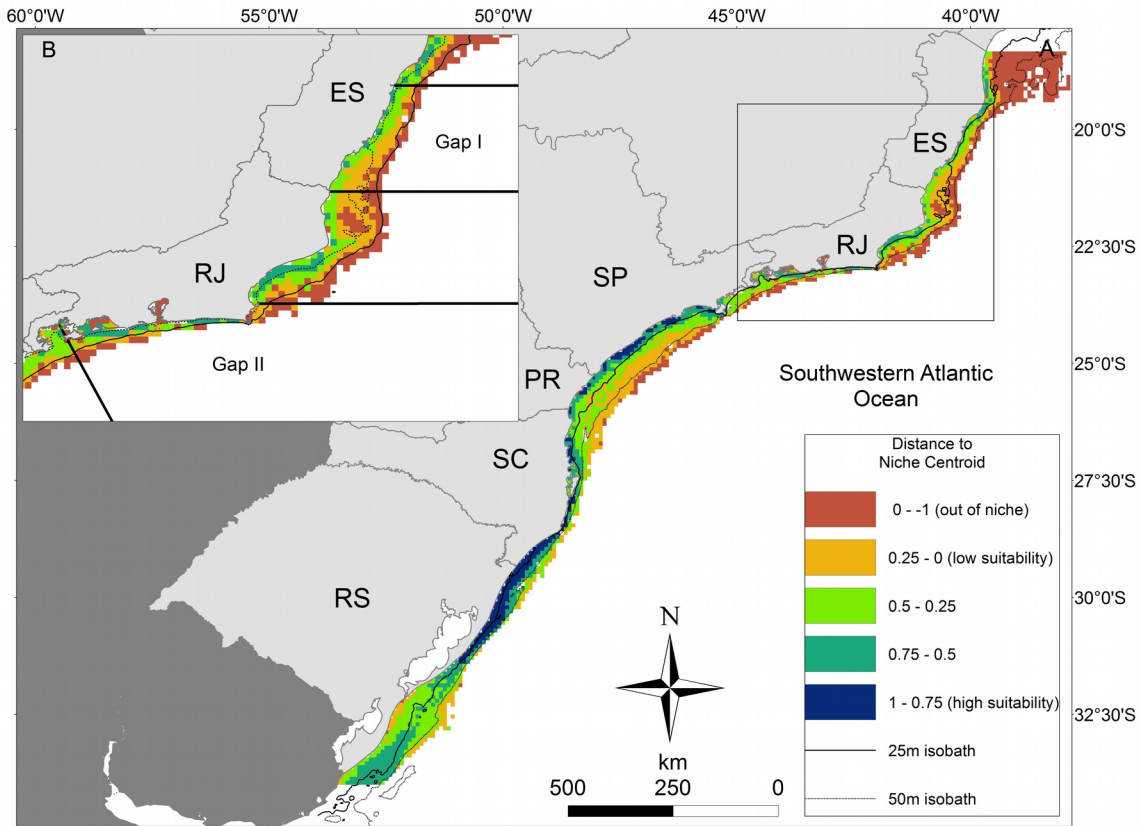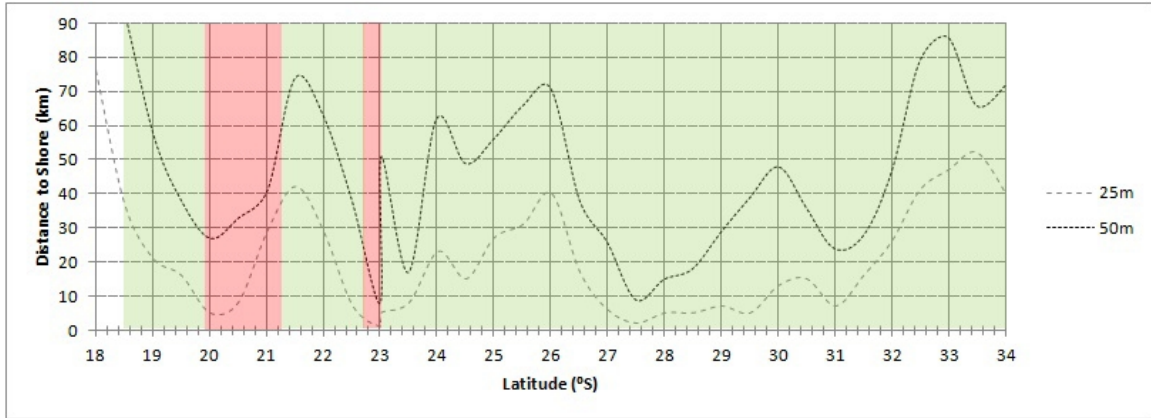
50°0'W   45°0'W   40°0'W

ES

SP   RJ

Southwestern Atlantic Ocean

PR

SC

20°0'S

22°30'S

25°0'S

27°30'S

Polygons representing areas occupied and not occupied by franciscanas

- AOF
- Gap I
- Gap II
- 25m Isobath
- 50m Isobath

N

km
300   150   0

CAPÍTULO II

Seascape genetics of the Atlantic spotted dolphin (*Stenella frontalis*) based on mtDNA control region

Karina Bohrer do Amaral[1,2, *]

Dalia C. Barragán-Barrera[3,4]

Roosevelt A. Mesa-Gutiérrez[5]

Nohelia Farías-Curtidor[3]

Susana Caballero[4]

Paula Mendéz-Fernandez [6]

Marcos C. Oliveira Santos [7]

Salvatore Siciliano [8]

Vidal Martín [9]

Manuel Carrillo[10]

Ana Carolina de Meirelles[11]

Valentina Franco-Trecu[12]

Nelson Fagundes [2, 13]

Ignacio Benites Moreno[1, 2, 14]

Lacey Knowles[15]

 Ana Rita Amaral[16, 17]

[1]Laboratório de Sistemática e Ecologia de Aves e Mamíferos Marinhos (LABSMAR), Universidade Federal do Rio Grande do Sul, RS, Brazil.

[2] Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, 91501-70 Porto Alegre, RS, Brazil.

[3] Fundación Macuáticos Colombia, Colombia, Medellín, Colombia.

[4] Laboratorio de Ecología Molecular de Vertebrados Acuáticos (LEMVA), Department of Biological Sciences, Universidad de los Andes, Carrera 1E No 18A – 12, Bogotá, Colombia.

[5] Nicholas School of the Environment, Duke University, 9 Circuit Dr, Durham, NC 27710, United States of America.

[6] Ingénieur de Recherche, Observatoire Pelagis – UMS 346, Université de La Rochelle/CNRS 5 allés de l'Océan 17000, La Rochelle, France.

[7] Laboratório de Biologia da Conservação de Mamíferos Aquáticos (LABCMA), Departamento de Oceanografia Biológica, Instituto Oceanográfico, Universidade de São Paulo, Praça do Oceanográfico, 191, Sala 145-A, 05508-120 São Paulo, SP, Brazil.

[8] Instituto Oswaldo Cruz/FIOCRUZ, Pavilhão Mourisco – sala 122, Av. Brasil, 4365 - Manguinhos, 21040-360, Rio de Janeiro, RJ, Brazil.

[9] Sociedad para el Estudio de Cetáceos del Archipélago Canario (SECAC), Casa de los Arroyo, Avda. Coll n.6, 35500 Arrecife, Lanzarote, Spain.

[10] Tenerife Conservación, C/Maya No. 8, La Laguna, Tenerife, Canary Islands, Spain.

[11] AQUASIS – Associação de Pesquisa e Preservação de Ecossistemas Aquáticos, Praia de Iparana, s/nº, SESC Iparana, 61600-000, Caucaia, CE, Brazil.

[12] Laboratorio de Ecología Fisiológica y del Comportamiento, Facultad de Ciencias, Universidad de la República, Iguá 4225 Esq. Mataojo, 11400, Montevideo, Uruguay.

[13] Laboratório de Genética Médica e Evolução, Departamento de Genética, Universidade Federal do Rio Grande do Sul. Avenida Bento Gonçalves 9500, Prédio 43312, sala 113, Agronomia, 91501-970 Porto Alegre, RS, Brazil.

[14] Centro de Estudos Costeiros, Limnológicos e Marinhos (CECLIMAR), Campus Litoral Norte, Universidade Federal do Rio Grande do Sul, Avenida Tramandaí, 976, Imbé, Rio Grande do Sul, 95625-000, Brazil.

[15] Museum of Zoology, Department of Ecology and Evolutionary Biology, University of Michigan, room 2010 1105 North University Avenue, Ann Arbor, MI 48109-1085, United States of America.

[16] Centre for Ecology, Evolution and Environmental Changes, Faculdade de Ciências, Universidade de Lisboa, Campo Grande, 1749-016, Lisboa, Portugal.

[17] Sackler Institute for Comparative Genomics, American Museum of Natural History, 79th Street and Central Park West, New York, NY 10024, United States of America.

Corresponding author:

* Karina Bohrer do Amaral

E-mail: karinabohrerdoamaral@gmail.com

Phone: +55 51 3308-9541

Abstract

The Atlantic spotted dolphin (*Stenella frontalis*) is endemic to tropical, subtropical and warm temperate waters of the Atlantic Ocean. The species has a complex geographical distribution, across which population structure has been recovered and it is most likely the result of distinct environmental requirements. Following a seascape genetics approach we investigate population differentiation of Atlantic spotted dolphins along the Atlantic Ocean and its relationship with marine environmental variables. We found different populations in the Western North Atlantic and Gulf of Mexico and individuals inhabiting oceanic islands in the Eastern Atlantic forming one population connected with those from oceanic waters of the Western North Atlantic. We also found that individuals from southeastern Brazil represent one distinct population and exhibit low levels of genetic diversity based on the mtDNA control region marker. We detected some level of Isolation-by-distance and Isolation-by-Resistance, including contemporary and past conditions. We attributed the low levels of correlation between genetic and geography/environment due to such large scale analyzed here and we also hypothesized that different process could play role to explain the genetic patterns recovered such as social structure and some level of phylopatry within populations.

Introduction

Seascape genetics is a derivation of landscape genetics discipline to marine environment (Riginos and Liggins 2013). In general, landscape genetics aims to understand how spatial factors as geographic distance and environmental heterogeneity shape genetic differentiation along species distribution (Manel 2003; Storfer et al. 2007; Holderegger and Wagner 2008; Balkenhol et al. 2009). However, as pointed out by Riginos and Liggins (2013), despite its theoretical similarities, seascape genetics studies should take into account the peculiarities of both the marine environment (e.g., fluidity, three-dimensionality, temporal and spatial scales, currents) and marine organisms (e.g. higher dispersal abilities).

Different patterns of how geography and/or environment shape genetic variation are recognized in landscape genetics. Isolation-by-distance (IBD) (Wright 1943; Manel 2003) is a very common model tested in landscape genetic studies, and postulates that populations separated by greater geographic (i.e. straightline or Euclidean) distances show greater levels of genetic differentiation (Wright 1943; Balkenhol

et al. 2009). More recently, derivations of the "isolation by" generic term have been described, such as isolation-by-environment (IBE) (Wang and Bradburd 2014) and isolation-by-resistance (IBR) (McRae 2006). IBE is defined as a pattern in which genetic differentiation increases with environmental differences, independent of geographic distance (Wang and Bradburd, 2014). On the other hand, IBR model predicts a positive relationship between genetic differentiation and the resistance distance, a graph theoretic distance metric based on circuit theory (McRae 2006). According to the the authors, the resistance distance provides a more appropriate predictor of equilibrium genetic differentiation than Euclidean distance because it accounts for heterogeneity in species' distributions and migration rates, as it incorporates all possible pathways and is better supported by existing analytic theory (McRae 2006).

In recent years, molecular genetic approaches have changed the traditional concept of large and homogeneous marine populations of, for instance, invertebrates and fishes. The absence of obvious physical barriers on the oceans and high gene flow among populations of marine organisms have long challenged the models of allopatric speciation. However, extensive genetic population structure has been discovered in many marine species, suggesting more complex recruitment dynamics in marine species than previously assumed. Thus, marine speciation is considered a paradox (Bierne et al. 2003; Hauser and Carvalho 2008).

Despite of the great dispersal capacity of marine top predators such as cetaceans, several studies have been revealing that these species exhibit also extensive population structure along its distribution. Fine-scale population structure has been extensively recovered and it is most likely the result of ecological divergence in cosmopolitan species like bottlenose dolphins (*Tursiops truncatus*), common dolphis (*Delphinus sp*.) and killer whales (*Orcinus orca*) (e.g., Tezanos-Pinto et al. 2008; Amaral et al. 2012; Foote et al. 2016). In general, habitat association, foraging, specializations and kin interactions, in combination with past bottlenecks and periods of expansion and contraction, can lead to discontinuous relationships between genetic and geographic distance. Habitat discontinuities and changes in oceanographic features, prey distribution and philopatric behaviour have also been identified as influencing the spatial genetic structure of several delphinid species (Möller et al. 2011).

The Atlantic spotted dolphin (*Stenella frontalis*) is a Delphinidae dolphin endemic to tropical, subtropical and warm temperate waters of the Atlantic Ocean (Perrin 2009). The species ranges from 45°N

96

to 35°S in the west Atlantic, and from Azores to at least Gabon in the east Atlantic (Perrin 2009). In general, Atlantic spotted dolphins inhabits continental shelf along its distribution, but also could be recorded in oceanic waters in the western north Atlantic and oceanic islands in the east north Atlantic (Freitas et al. 1989; Jefferson et al. 1997; Baumgartner et al. 2001; Davis et al. 2002; Silva et al. 2003; Moreno et al. 2005; Weir 2010).

In general, *Stenella* dolphins present great dispersal capabilities (e.g., Reilly 1990). Evidences of dispersal across large distances was also demonstrated both direct and indirectly for Atlantic spotted dolphins in North Atlantic (Herzing 1997; Davis et al. 2006; Quérouil et al. 2010). However, the Atlantic spotted dolphin presents a complex geographic distribution, where distinct populations have been identified based on morphology, genetics and pollutants analyses (Perrin et al. 1987; Adams and Rosel 2006; Green et al. 2007; Quérouil et al. 2010; Caballero et al. 2013; Viricel and Rosel 2014; Méndez-Fernandez et al. 2018).

Analysis of mtDNA CR marker is recurrent in all studies conducted so far aiming to understand population structure of Atlantic spotted dolphins among different regions of Atlantic Ocean (see Adams and Rosel 2006; Green et al. 2007; Quérouil et al. 2010; Caballero et al. 2013; Viricel and Rosel 2014). This marker was also considered the most commonly applied molecular marker in genetic studies of cetacean taxonomy (Rosel et al. 2017a, b; Schwartz and Boness 2017). Despite the limitations of mtDNA markers, such as being a matrilineal marker and not providing information on male-biased dispersal (Rosel et al. 2017a), mtDNA is usually considered a useful marker to landscape genetics because it is a neutral and nonrecomninat marker. Holderegger and Wagner (2008) highlighted that in contrast to biparentally inherited marker types such as microsatelites and SNPs, the mtDNA markers are transmitted unchanged from the mother to her offspring, which, in principle, makes them useful for the detection of dispersal events.

In relation to the marine environment, several studies have been conducted based on the framework of seascape genetics (Selkoe et al. 2008; Riginos and Liggins 2013; Wee et al. 2014; Thomas et al. 2015; Benestan et al. 2016) and some of these studies have been conducted in cetaceans species (Mendez et al. 2010, 2011; Amaral et al. 2012). Amaral et al. (2012) revealed that marine productivity and sea surface temperature are correlated with genetic structure in the high mobile and widely distributed

short-beaked common dolphin (*Delphinus delphis*). In a study of humpack dolphin (*Sousa spp.*), Mendez et al. (2011) assessed population structure patterns using mtDNA CR marker and the potential influence of environmental in shaping this patters along western Indian Ocean. The authors showed genetically isolated populations in areas environmentally distinct, and high-lighted the utility of molecular markers in combination with high quality environmental data to address questions related to ecological processes in marine species.

The aim of this study is (1) to assesses population structure of Atlantic spotted dolphin using mtDNA CR along all species distribution and based on seascape genetics framework (2) to investigate how marine environment could be influencing genetic differentiation among populations. This is the first attempt to investigate population structure of Atlantic spotted dolphin as well as its relationship with marine environment along its distribution. In order to do that, we include a comprehensive review of mtDNA CR sequences and its respective geographic coordinates and combine that with high-resolution environmental data. Based on previous findings that related genetics differentiation with environmental conditions (Viricel and Rosel 2014), we expect recover population differentiation across the distribution of the species as resulting from environmental heterogeneity and/or geographic distance.

Material and Methods

Sampling and DNA extraction

Tissue samples of 108 individuals were obtained from remotely-darting biopsies, stranded or incidentally captured Atlantic spotted dolphins from different regions of the Atlantic Ocean, including: Brazil (n=80), Colombia (n=7), Guadaloupe Island (n=1), Uruguay (n=1), and Canary Island (n=19). Samples were preserved in different ways, including ethanol, sodium chloride-saturated 20% dimethyl sulphoxide or lyophilized for long-term preservation.

Total genomic DNA was extracted from tissue samples using the DNeasy Blood and Tissue kit (Qiagen), following the protocol, with the exception of the proteinase K digestion step that was extended overnight (Hancock-Hanser et al. 2013). DNA was eluted in lower volumes than recommended to avoid low concentrations of DNA mainly from samples obtained from stranded animals. DNA quality and concentration was verified using Qubit Fluorometric Quantition (Thermo Fisher Scientific Inc.).

Mitochondrial control region sequencing and GenBank data

We used 1 μl of DNA to amplify a portion of 650 bp of the mitochondrial control region (CR) using the primers t-Pro-whale M13Dlp1.5 (5' TGTAAAACGACAGCCAGTTCACCCAAAGCTGRARTTCTA-3') and Dlp8 (5'-CCATCGWGATGTCTTATTTAAGRGGAA-3') following the protocol by Tezanos-Pinto et al. (2008) for amplification reaction and thermal cycler profile. PCR products were cleaned by adding Shrimp Alkaline Phosphatase and Exonuclease I as recommended by the manufacturer followed by an incubation period at 37°C for 30 min and 80°C for 15 min. Both strands were sequenced on an ABI 3730 automated sequencer (BigDye Terminator Cycle Sequencing; Applied Biosystems).

GenBank data and alignment

In order to include sequences from other regions of the species range, several sequences of mtDNA CR were obtained from GenBank. Accession numbers and original references are available in Table S1. Brazilian samples already analyzed by Caballero et al. (2013) were reanalyzed and resequenced as described above.

All available mtDNA CR sequences from Atlantic spotted dolphins were downloaded, but only those sequences with available geographic information in the refereed publications were considered in the analyses. Geographic coordinates referring to sequences from Bahamas, Azores and Madeira were estimated based on the main local of sampling collection referenced to in the articles. Geographic coordinates from sequences of Western North Atlantic and Gulf of Mexico were not directly available and, thus, were estimated through crossing information of different data banks. We crossed date and region of sampling with dates and geographic coordinates available on GBIF (GBIF, 2018) public database from 1994 to 2001. Only those sequences that exactly matched dates and geographic coordinate with the region of sampling were included in the analyses. Finally, all sequences obtained from GenBank and those obtained by PCR were aligned using the software Sequencher, version 5.4.6 (Genes Codes Corporation).

Genetic diversity and Population differentiation

Populations were defined based on information available from previous studies and geographic localities that better represent the sampling area (Table S1, Fig. 1). Thus, we defined 11 populations:

Azores (AZ), including samples from individuals collected around Azores Archipelago and published by Quérouil et al. (2010); Madeira (MAD), including samples from individuals collected around Madeira Archipelago and also published by Quérouil et al. (2010); Canary (CAN) including sequences collected from animals stranded at Canary Islands and analyzed in this study; MAB (Mid-Atlantic Bight), including haplotypes sequences from individuals collected on coastal and oceanic waters northward Cape Hatteras (~ 35°N) as defined previously by Adams and Rosel (2006) and confirmed by Viricel and Rosel (2014); SAB (South Atlantic Bight), including haplotypes sequences from individuals collected on the continental shelf and southward Cape Hatteras (~ 35°N) as defined previously by Adams and Rosel (2006) and confirmed by Viricel and Rosel (2014); eastern Gulf of Mexico (eGOM), including haplotype sequences from individuals collected at continental shelf and east of the Mobile Bay firstly analyzed by Adams and Rosel (2006), and posteriorly defined by Viricel and Rosel (2014) with analyses of more samples; western Gulf of Mexico (wGOM), including haplotype sequences from individuals collected at continental shelf and westward of Mobile Bay firstly analyzed by Adams and Rosel (2006), and also posteriorly defined by Viricel and Rosel (2014); Bahamas (BAH), including haplotypes sequences from individuals collected on Bahamas and previously analyzed by Green et al. (2007) and Green (2008); Caribbean (CAB), including haplotype sequences collected around Caribbean Sea and previously analyzed by Caballero et al. (2013), but include also sequences from individuals collected in La Guajira (Colombia) and Isla Guadaloupe analyzed in this study by the first time; Northern Brazil (N_Br), included two samples collected from stranded animals in the northern Brazil; finally, Brazil_Uruguay (Br_Uy), included samples collected southward 22°S on the continental shelf, mainly in the Southeast Brazilian Bight(southeastern Brazil), and one sample from an stranded animal at approximately 34°S in Uruguay, all these samples were analyzed in this study (a few of these samples had been previously analyzed by Caballero et al. (2013), but we opted by sequencing these samples again as already explained above).

Haplotypes were defined by DNAsp 6.0 and molecular diversity indexes such as nucleotide and haplotype diversities were estimated in Arlequin 3.5.2 (Excoffier and Lischer 2015). Neutrality tests (Tajima's D and Fu's FS) were also performed to test for population demographic changes in Arlequin 3.5.2 (Excoffier and Lischer 2015). Significance was assesses through 10,000 permutations.

Population differentiation was tested by calculating pairwise F_{ST}, and Φ_{ST} using Tamura-Nei distance in Arlequin 3.5.2 (Excoffier and Lischer 2015). Significance was assessed through 10,000 permutations. Nei's estimate of net divergence (dA) (hereafter, Nei's dA) was also estimated using nucleotideDivergence function of the StrataG package (Archer et al. 2016) with model settled to "TN93". Finally, analysis of molecular variance (AMOVA) was computed in Arlequin 3.5.2 (Excoffier and Lischer 2015) among pairs of populations most closest geographically. Significance was assessed through 1,000 permutations.

Phylogenetic relationships

A median-joining network of haplotypes was constructed in NETWORK 5.0.0.3 (Bandelt et al. 1999). A Bayesian phylogenetic tree was performed in MrBayes v.3.2.6 (Ronquist et al. 2011) including haplotypes. Four simultaneous MCMC chains were run for 2 million generations, with trees sampled at intervals of 100 generations and 20% of trees were discarded as ''burnin''. A sequence of Guiana dolphin (*Sotalia guianensis)* was used as outgroup (GenBank Accession Number KM893401).

Individual level analysis and Procrustes analysis

Since the delimitation of populations *a priori* based on geography is very questionable for cetaceans due to their great dispersal capabilities, an individual level analyses was performed through procrustes analysis approach to find an optimal transformation that maximizes the similarity between Principal Component Analysis (PCA) maps of genetic variation and geographic maps of population locations (Wang et al. 2012).

First, we applied procrustes analysis to compare the individual-level coordinates of the first two components (PC1 and PC2) in the PCA performed on the mtDNA CR data to the geographic coordinates. PCA of mtDNA CR was perfomed using the function dudi.pca of ade4 package (Dray and Dufour 2007). We used procrustes function in order to rotate a configuration to maximum similarity with another configuration, and the function protest to test the significance between two configurations. We used procrustes and protest functions from the vegan package (Oksanen et al. 2017) in R 3.4.2 (R Core Team

2017). Posteriorly, we applied the residual function to estimate the residuals of the first procrustes, and performed a second one using a PCA of environmental rasters (see below).

The consistence of individual level analysis with the determination of population *a priori* by literature and geography was visually checked.

Environmental and Spatial data

Our study area encompasses the complete distribution of Atlantic spotted dolphins along tropical and subtropical Atlantic Ocean (Fig. 1). Currently, the distribution of several species is available in shapefile format by the The IUCN Red List (Hammond et al. 2012). Therefore, we utilized this map as a background for subsequent analyses. However, taking into account that we have access to a sample of an individual (male) collected at southernmost record of species in the Western Atlantic Ocean, we modified the original shapefile in order to consider the environmental information from this area. In the last years, the species have been recorded in Uruguayan waters (Valentina Franco-Trecu personal communication; Paro et al. 2014).

The final range of study area extended longitudinally from 20°E to 100°W, and latitudinally from 45°N to 40°S. This such greater extension of Atlantic Ocean encompasses several different environmental conditions, including coastal and oceanic waters; ranges of Atlantic seafloor such as islands, seamounts and the Mid-Atlantic Ridge; changes in the orientation of coast; oceanic currents and gyres that provide an exceptional environmental heterogeneity along Atlantic spotted dolphin distribution.

Along its distribution, Atlantic spotted dolphins are recorded mainly in those waters above continental shelf up to 1,000 m isobath, being recorded off of continental shelf in specific regions such as Western North Atlantic and in the oceanic islands of the Eastern Atlantic (Perrin et al. 1987; Freitas et al. 1989; Jefferson et al. 1997; Baumgartner et al. 2001a; Silva et al. 2003; Moreno et al. 2005; Fernández et al. 2009; do Amaral et al. 2015). Therefore, the study area includes the following coastal and shelf areas from north to southward in the Western Atlantic Ocean: Southern Atlantic Bight, Gulf of Mexico, Caribbean, north South America, southeastern Brazil and Uruguay. Oceanic waters include the Mid-Atlantic Bight (off United States) and data from Azores, Madeira and Canary Archipelagos in the Eastern Atlantic Ocean. These regions are represented by different oceanic systems and oceanic currents that were

systematically described by Spalding et al. (2007) for coastal and shelf areas and by Spalding et al. (2012) for pelagic areas.

Environmental layers were gathered from MARSPEC (Sbrocco and Barber 2013), an ocean climate layers for marine spatial ecology. This public bank provides both geophysical and bioclimate information in ESRI grid format at ~ 5 km seconds of resolution.

Considering what is known as habitat preferences of cetaceans and specifically Atlantic spotted dolphins (e.g., Baumgartner et al. 2001; do Amaral et al. 2015), we pre-selected the following environmental layers from MARSPEC dataset: bathymetry (m), slope (degrees), distance to shore (km), sea surface salinity (psu, SSS) and sea surface temperature (°C, SST). Three different metrics of SSS and SST were included: mean annual, annual range and annual variance. SSS of the freshest month and SSS of the saltiest month as well as SST from the coldest month and SST from the warmest were also selected. We used a custom R script written by Elizabeth J. Sbrocco to crop MARSPEC ESRI grids to our area of interest as well to convert it to an ASCII file.

Correlation among layers were investigated using the function pairs of raster package (Hijmans et al. 2014). Layers highly correlated were excluded from the following analyses. A PCA was also performed using the function rasterPCA of the package RStoolbox (Leutner and Horning 2016).

Kruskal-Wallis with bonferroni correction were performed to investigate if medians of environmental variables were equal among putative populations (we just considered populations with more than one record, therefore AZ, MAD and BAH were not included). This test was followed by Dunn test using dunn package (Dinno 2017) to test differences between pairwise putative populations. Visualization of medians among different population was provided by boxplot graphics.

"Isolation by" analyses

All populations (except AZ, MAD and BAH) were represented by several individuals with different sampling geographic coordinates. In order to represent each population with one geographic coordinate, we estimated the centroid of each population based on geographic coordinates of individuals collected.

103

One geographic matrix representing geographic distances were estimated using least-cost distances using different functions of marmap package (Pante and Simon-Bouhet 2013). We estimated least-cost distance with no constraint among centroids, and least-cost distances with constrains of a maximum of 300 m, 600 m, 900 m depths.

Three matrices representing environment were generated using the values of bathymetry, Mean Annual SSS and Mean Annual SST extracted for the centroid of each population. These environmental layers are considered important predictors of cetaceans' distribution (Baumgartner et al. 2001; Redfern et al. 2006; Palacios et al. 2013). We also generated a matrix from the principal component 1 (PC1) of the environmental PCA, and another one was generated calculating Euclidean distances of PC1.

Two matrices representing resistance distances (McRae 2006) were calculated using the software Circuitscape v. 3.5.8 (Shah and McRae 2008). Resistance distances were estimated among centroids of populations based on maps of environmental suitability generated for Atlantic spotted dolphins for both contemporary and Last Glacial Maximum (LGM) climatic and geophysical conditions. Maps of environmental suitability were generated using the maxent function of the dismo package (Hijmans et al. 2011) in order to build a MaxEnt ("Maximum Entropy") species distribution model (Phillips et al. 2006). MaxEnt model settings were defined through ENMevaluate function of the package ENMeval package (Muscarella et al. 2014), which provide species-specific tuning of settings to generate models. The model with the lowest value of the Akaike Information Criterion corrected for small samples sizes reflects both model goodness-of-fit and complexity (Muscarella et al. 2014). We used the block as data partitioning method. Environmental layers not correlated were used. We gathered the same set of environmental layers for contemporary and LGM time frames and cropped them to encompass the study area. Occurrence records were the same dataset compiled for sequences, with exception that we remove duplicate records and records representing the same pixel. We used 10,000 points to determine the MaxEnt distribution (background points).

Initially to explore the data, we performed a simple linear regression of data using lm function. We tested three different matrices of genetic differentiation data (linearized F_{ST}, Φ_{ST} and Nei's dA) against the different matrices of geographic distance (without and with restrictions) and the resistance matrices based on suitability models of contemporary and LGM conditions.

Finally, IBD and IBE were tested using Distance-based Redundancy Analysis (dbRDA) (Legendre and Anderson 1999). We also tested environmental distances conditioned on geographic distances. IBE was also tested through Mantel tests and Partial Mantel to test the correlation among genetic matrices and the Euclidean distance of environment, and also controlling for geographic distances. In order to test IBR, Mantel testes were performed between contemporary and LGM resistance matrices and against genetic matrices.

Canonical analysis is the simultaneous analysis of two, or several data tables combining the concepts of ordination and regression. Among canonical analysis, Redundance analysis (RDA) is related to multiple regression analysis and it is used when the X variables display linear relationships with the Y variables. In distance-based redundancy analysis (dbRDA), a resemblance matrix is computed among the sites using a similarity measure appropriate to species data. Principal coordinate analysis (PCoA) is applied to this matrix to obtain new Euclidean axes (matrix Y) fully representing the relationships among the sites; a correction for negative values may be required. The experimental factors and their interactions are coded as orthogonal dummy variables. RDA is applied to the new matrix Y to test the significance of the factor (or interaction) coded into matrix X, with all the other factors (and interactions) coded into a matrix of covariables (Legendre and Legendre 1998). Here dbRDA was performed using the funcion capscale in the vegan package (Oksanen et al. 2017) that is an alternative implementation of dbRDA, in which the dissimilarity data are first ordinated using metric scaling, and the ordination results are analyzed with redundance analysis. We assessed the significance of test using anova function.

Mantel statistic formula is a linear model that brings out the linear component of the relationship between the values in two distance matrices. Strong nonlinearity may prevent relationships from being identified in the Mantel test. Mantel and Partial Mantel testes were performed using mantel and mantel. partial functions in the vegan package (Oksanen et al. 2017) using pearson method. Significance was assessed with 999 permutations.

All R analyses were performed on R 3.4.2 (R Core Team 2017).

Results

mtDNA sequences and genetic diversity

Our final dataset of mtDNA CR was composed of 545 sequences considering both previously analyzed sequences and those generated in this study (Table S1, Fig. 1). From 108 tissue samples analyzed here, we are able to successfully generated sequences from 80 samples. GenBank data included thousands of individual sequences or haplotypes sequences already published or unpublished and we performed a rigorous selection of GenBank sequences. Therefore, we included 465 sequences in our dataset for which we were able to obtain geographic coordinates with a high degree of reliability. When haplotype sequences were available we just selected those for which we could determine the number of individuals included in that haplotype. Although we have been extremely cautious with the selection of GenBank information, we are aware of possible error associated with analyses performed here, mainly those conducted with F_{ST} values due to its dependency on the frequency of haplotypes in the populations. Thus, we highlighted the importance of researchers to be willing to provide key information on their publications in order to allow for the replication of their data in different studies. Some samples obtained from stranded animals have its geographic coordinates slightly modified to capture environmental information from the region (Table S1).

Fig. 1 Representation of bathymetry of study area and 545 geographic coordinates of Atlantic spotted dolphins analyzed. Circles symbol represent sequences from individuals and/or haplotypes already analyzed in other studies; diamond symbol represents samples from individuals analyzed by the first time. Colors represent data grouping in different putative population. Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico

Our mtDNA CR dataset included 344 bp with a total of 103 haplotypes recovered among populations. Molecular diversity indexes are presented on Table 1. Nucleotide diversity was higher than 0.01 for AZ, MAD, CAN, MAB, wGOM, CAB and N_Br. These same populations and also eGOM had haplotype diversities higher than 0.8. Despite their relatively higher sample size, Bahamas and Br_Uy had the lowest values of both indexes.

Table 1. Molecular diversity indexes, and neutrality tests for Atlantic spotted dolphin putative populations. Abbreviations: N, number of samples; Nh, number of haplotypes; π, nucleotide diversity; AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico. Statistical significant values are represented in bold

	AZ	MAD	CAN	MAB	SAB	eGOM	wGOM	BAH	CAB	N_Br	Br_Uy
N	145	46	12	17	82	59	15	93	14	2	60
Nh	54	31	11	11	11	17	6	6	11	2	8
Exclusive haplotypes	31	12	2	2	5	10	1	0	5	0	3
π	0.0210	0.0202	0.0215	0.0120	0.0097	0.0090	0.0127	0.0065	0.0198	0.0534	0.0067
Polimorphic sites	55	48	32	17	14	22	13	8	38	22	10
Segregating sites	49	42	27	17	14	20	13	8	31	17	10
Gene diversity	0.949	0.981	0.985	0.941	0.714	0.914	0.800	0.572	0.934	1.000	0.633
Tajima's D	-0.700	-1.071	-0.971	-0.798	0.428	-0.932	0.227	0.968	-1.459	0.000	0.124
Tajima's D p-value	0.269	0.135	0.163	0.232	0.716	0.179	0.633	0.850	0.061	1.000	0.599
Fu's FS	**-24.065**	**-15.597**	**-3.447**	**-3.452**	0.297	**-5.135**	1.231	2.392	-2.205	3.091	0.255
Fu's FS p-value	0	0	0.039	0.039	0.602	0.028	0.745	0.853	0.132	0.599	0.6

Since our focus is relating environment and/or geography to genetics, we considered the two samples obtained from northern Brazil as a distinct population because we considered do not correct grouping these samples neither into Br_Uy nor CAB due to its geographic distances to both centroids. Ecological niche modeling data (do Amaral et al. 2015) and Moreno et al. (2005) suggested that individuals from northern Brazil could be not related to those from southeastern Brazil. Therefore, the higher values of molecular indexes obtained for this putative population were due to differences in the haplotypes recovered in these two samples, being necessary more samples to confirm the genetic diversity of dolphins from this area.

The neutrality tests estimated showed statistically significant and negative values of Fu's FS for the following putative populations: AZ, MAD, CAN, MAB and eGOM. Tajima's D was not statistically significant for all populations (Table 1).

Population differentiation

Pairwise F_{ST}, Φ_{ST} and Nei's dA are showed in Table 2. Overall, consistent differentiation between most putative populations was statiscally significant for F_{ST} and Φ_{ST}. Both fixation indexes were consistent in showing no differentiation at the population level between AZ, MAD and CAN. Significant differentiation between Br_Uy and BAH in relation to all other populations was recovered (except that the

F_{ST} value between Br_Uy and N_Br was not significant). Both fixation indexes were consistent showing differentiation at the population level between Western North Atlantic putative populations (except the F_{ST} value between eGOM and MAB was not significant). Western North Atlantic populations seems to be differentiated from AZ, MAD and CAN in relation to both indexes. However, the relationship between MAB and the eastern Atlantic Islands is not consistent between F_{ST} and Φ_{ST}. The relationship between CAB and the remaining putative population was also not consistent between F_{ST} and Φ_{ST}. However, both markers revealed consistent differentiation among CAB and SAB, BAH and Br_Uy; and were also consistent in showing no differentiation among CAB and CAN, MAB and N_Br. The highest F_{ST} value was 0.39 between BAH and Br_Uy; and the highest Φ_{ST} value was 0.64 between N_Br and BAH. The lowest significant F_{ST} value was 0.02 between CAB and MAD; and the lowest significant Φ_{ST} value was 0.04 between eGOM and MAB.

Nei's dA values obtained ranged from 0.000005 to 0.017. The lowest Nei's dA values were obtained between N_Br and Br_Uy; and the highest values between either N_Br and MAD or Br_Uy and MAD. Both N_Br and Br_Uy populations had Nei's dA values higher than 0.01 when compared to others populations (except in the pairwise comparison with SAB). The comparison between Nei's dA values and fixation indexes is not totally consistent because some pairwise comparison that were not significant at one or both fixation indexes had higher Nei's dA values (for instance, AZ and MAD); and some pairwise comparison with low Nei's dA values had significant fixation index values (for instance, Br_Uy and SAB).

AMOVA analysis showed significant genetic structure among three different combinations of populations based on geography proximity (Table 3). The highest FCT and significance was recovered with the following groups: Group 1 including AZ, MAD, CAN, MAB; Group 2 represented by SAB, Group 3 including eGOM and wGOM; Group 4 represented by BAH; and Group 5 including CAB and N_Br; and, finally Group 6 represented by Br_Uy (FCT = 0.12841; p-value < 0.000001).

Table 2. Pairwise fixation index and Nei's estimate of net divergence (dA) values obtained between Atlantic spotted dolphins putative populations for mtDNA control region marker. Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico. Statistical significant values are represented in bold.

		AZ	MAD	CAN	MAB	SAB	eGOM	wGOM	BAH	CAB	N_Br	Br_Uy
F_{ST}	AZ		0.402	0.387	0.007	0.000	0.000	0.000	0.000	0.013	0.283	0.000
	MAD	0.000		0.729	0.003	0.000	0.000	0.000	0.000	0.016	0.849	0.000
	CAN	0.002	-0.006		0.181	0.002	0.035	0.002	0.000	0.216	0.961	0.001
	MAB	**0.033**	**0.027**	0.018		0.004	0.182	0.007	0.000	0.245	0.258	0.000
	SAB	**0.091**	**0.122**	**0.114**	**0.087**		0.000	0.000	0.000	0.000	0.030	0.000
	eGOM	**0.050**	**0.044**	**0.036**	0.012	**0.125**		0.000	0.000	0.053	0.094	0.000
	wGOM	**0.109**	**0.097**	**0.110**	**0.085**	**0.228**	**0.115**		0.000	0.001	0.114	0.000
	BAH	**0.210**	**0.234**	**0.257**	**0.256**	**0.263**	**0.182**	**0.347**		0.000	0.033	0.000
	CAB	**0.032**	**0.025**	0.011	0.013	**0.158**	0.030	**0.125**	**0.269**		0.614	0.000
	N_Br	0.021	-0.019	-0.032	0.044	**0.206**	0.065	0.152	**0.357**	0.049		0.065
	Br_Uy	**0.120**	**0.149**	**0.178**	**0.231**	**0.266**	**0.195**	**0.302**	**0.388**	**0.211**	0.273	
Φ_{ST}	AZ		0.806	0.577	0.123	0.000	0.000	0.010	0.000	0.133	0.049	0.000
	MAD	-0.007		0.629	0.133	0.000	0.000	0.007	0.000	0.137	0.052	0.000
	CAN	-0.012	-0.012		0.157	0.014	0.003	0.018	0.000	0.093	0.096	0.011
	MAB	0.021	0.019	0.029		0.000	0.049	0.047	0.000	0.231	0.037	0.002
	SAB	**0.152**	**0.189**	**0.121**	**0.258**		0.000	0.000	0.000	0.000	0.001	0.000
	eGOM	**0.067**	**0.072**	**0.122**	**0.041**	**0.317**		0.002	0.000	0.004	0.004	0.000
	wGOM	**0.083**	**0.080**	**0.110**	**0.072**	**0.380**	**0.106**		0.000	0.054	0.094	0.000
	BAH	**0.190**	**0.244**	**0.293**	**0.316**	**0.403**	**0.174**	**0.324**		0.000	0.005	0.000
	CAB	0.021	0.021	0.049	0.016	**0.365**	**0.081**	0.049	**0.310**		0.116	0.000
	N_Br	**0.203**	0.223	0.164	**0.422**	**0.617**	**0.534**	0.367	**0.640**	0.151		0.012
	Br_Uy	**0.061**	**0.069**	**0.114**	**0.130**	**0.245**	**0.083**	**0.242**	**0.247**	**0.187**	**0.628**	
Nei's dA	AZ	0.0000										
	MAD	0.0044	0.0000									
	CAN	0.0013	0.0051	0.0000								
	MAB	0.0023	0.0003	0.0033	0.0000							
	SAB	0.0051	0.0088	0.0043	0.0069	0.0000						
	eGOM	0.0011	0.0068	0.0024	0.0037	0.0036	0.0000					
	wGOM	0.0012	0.0068	0.0025	0.0039	0.0054	0.0006	0.0000				
	BAH	0.0034	0.0069	0.0044	0.0051	0.0062	0.0043	0.0051	0.0000			
	CAB	0.0012	0.0050	0.0001	0.0032	0.0050	0.0026	0.0024	0.0050	0.0000		
	N_Br	0.0131	0.0172	0.0112	0.0154	0.0021	0.0103	0.0130	0.0133	0.0121	0.0000	
	Br_Uy	0.0130	0.0172	0.0111	0.0153	0.0020	0.0101	0.0129	0.0130	0.0120	0.0000047	0.0000

Table 3. Results from analysis of molecular variance (AMOVA) of population structure in Atlantic spotted dolphin. Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico. Statistical significant values are represented in bold

Groups	Source of variation	% variation	F-statistics
1 = {AZ, MAD, CAN}; 2 = {MAB, SAB, eGOM, wGOM, BAH}, 3 = {CAB, N_Br}, 4 = {Br_Uy]	Among groups	-0.240	FCT = -0.00242 (p-value > 0.05)
	Among populations within groups	15.060	
	Within populations	85.190	
1 = {AZ, MAD, CAN, MAB}; 2 = {SAB}, 3 = {eGOM, wGOM, CAB, N_Br}, 4 = {BAH], 5 = {Br_Uy}	Among groups	11.780	**FCT = 0.11784 (p-value < 0.005)**
	Among populations within groups	4.250	
	Within populations	83.970	
1 = {AZ, MAD, CAN, MAB}; 2 = {SAB}, 3 = {eGOM, wGOM}, 4 = {BAH}, 5 = {CAB, N_Br}, 6 = {Br_Uy}	Among groups	12.84	**FCT = 0.12841 (p-value < 0.000001)**
	Among populations within groups	3.19	
	Within populations	83.97	
1 = {AZ, MAD, CAN, MAB}; 2 = {SAB, eGOM, wGOM, BAH}, 3 = {Br_Uy}, 4 = {CAB, N_Br}	Among groups	0.32	FCT = 0.0032 (p-value > 0.05)
	Among populations within groups	14.62	
	Within populations	85.06	
1 = {AZ, MAD, CAN, MAB}; 2 = {SAB}, 3 = {BAH, eGOM, wGOM, CAB} 4 = {N_Br, Br_Uy}	Among groups	8.73	**FCT = 0.08726 (p-value < 0.01)**
	Among populations within groups	7.49	
	Within populations	83.79	
1 = {AZ, MAD, CAN, MAB}; 2 = {SAB}, 3 = {BAH, eGOM, wGOM, CAB, N_Br} 4 = {Br_Uy}	Among groups	8.84	**FCT = 0.08844 (p-value < 0.05)**
	Among populations within groups	7.4	
	Within populations	83.75	

Phylogenetic relationships

The median-joining network obtained was complex and revealed that all haplotypes are very closely related to each other, with a few of them showing higher differentiation by mutation steps (Fig. S1). Many haplotypes are shared among all populations and 71 are exclusive of different populations. AZ had the highest number of sequences analyzed, and 54 haplotypes were found in this population with 31 being exclusive. Taking into account the sample size, MAD, CAN, MAB and CAB had higher numbers of different haplotypes. AZ and MAD share several haplotypes between each other, but also with other populations. On the other hand, SAB, BAH and Br_Uy, that have more than 60 sequences analyzed, the number of haplotypes ranged from six to 11. SAB and Br_Uy had five and three haplotypes exclusives, respectively. BAH and N_Br do not have exclusive haplotyes. All populations shared a minimum of two haplotypes.

The phylogenetic tree of haplotypes revealed that Haplotype 52, which was recovered in one sample from La Guajira, diverged from all others haplotypes. Some clusters were recovered with posterior

probability higher than 0.9. In general, these clusters are mainly formed by haplotypes from North Atlantic at low frequencies (Fig. S2).

Procrustes analyses

   Despite our efforts to group samples into the most reasonable way taking into account geography and previous studies, we also analyzed samples at individual level analyses without any grouping of sequences *a priori* and further tested genetic data against both geography and environment through procrustes analyses.

   Procrustes analysis between genetic similarity and geographic distance suggested a significant correlation (t = 0.1719, p = 9.999e-05), while the analysis between genetic similarity and environmental space was also significant (t = 0.1015, p = 0.017098).

   The visual inspection of graphics (Fig. 2) suggested that those sequences from southern Atlantic (i.e., a Br_Uy and N_Br) have a strong relationship with those from northern Atlantic (Fig. 2a). When we analyze the graphic representing the relationship between genetic and environmental space (Figure 2b), we observe that AZ, MAD and CAN samples are further away from those from Western Atlantic; Br_Uy, SAB and MAB seem to be closest in relation to wGOM, eGOM, BAH, CAB and N_Br. The clusters observed in this graphic seem to be in agreement with our designation of groups *a priori*, since the colors used to represent *a priori* grouping of samples indicated that few individuals are dislocated in relation to its centroids and/or other individuals from the same group. Interesting, the most displaced sample from those collected in the Brazil-Uruguay region (blue color) is that sampled in Uruguay. This sample from Uruguay had the Haplotype 85, which is present in other 83 samples mainly from SAB (n= 59), AZ (n=18), BAH (n=15), and less often in others populations (CAN, eGOM, MAB, MAD). In Brazil, the Haplotype 85 was found only in three samples.

Fig. 2 Representation of procrustes analyses. a) The relationship between genetic and geography is showed. b) The relationship between the residuals of the previous procrustes and environment are represented in the environmental space. Samples were colored according to sampling area. Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico

Environmental analysis

Based on the correlation among environmental layers pre-selected, we chose six uncorrelated layers to represent the environmental information in this study: bathymetry, slope, Mean Annual SSS, Annual Range in SSS, Mean Annual SST, and Annual Range in SST. The exception was the inclusion of Annual Range in SST which is correlated with Mean Annual of SST, but we considered including this layer because its a very important variable to represent the ranges of oceanographic conditions.

Principal Component Analyses (PCA) was performed with six environmental variables and Principal component 1 (PC1) explained 92.2% of variation and PC2 explained 6.5%. Both PCs components explain almost 99% of variation. Bathymetry and Mean Annual SST were the environmental variables that mainly contribute to the first and second components of PCs, respectively. Plot of PCA (Fig. 3) revealed the heterogeneity along the study area. However, some regions seem to have similar conditions. Western North Atlantic, Eastern North Atlantic and Western South Atlantic seem to have similar conditions; the equatorial zone in coast from America and Africa also seem to be similar; Gulf of Mexico seem to exhibit distinct conditions not found in other regions of the Atlantic Ocean.

Fig. 3 Representation of Principal Component Analyses (PCA) performed with six environmental variables. Similar colours indicate regions with similar conditions

We also analyzed the differences of environmental variables among the putative populations represented by more than one geographic coordinate in our dataset. Kruskal-Wallis tests were significant for all environmental layers, revealing significant differences among all putative populations (Table S2). However, pairwise comparisons revealed that some populations were not differentiated in relation to some environmental layers (Table S3). The environmental characteristics of putative populations in relation to each environmental variable were represented in boxplots (Fig. 4).

In relation to geophysical layers, CAB had the highest median bathymetry (654 m depth) and Slope (20°). CAN had the lowest median bathymetry (5 m depth), probably because all samples from CAN are from stranded animals. Although MAB had median equal to 39 m depth equal to Br_Uy and similar to SAB, individuals from MAB were also recorded in waters of 2,000 m depth. Medians from eGOM and wGOM were 54 m depth and 92 m depth, respectively. Although almost of all populations had slope medians equal to 1 (except CAB), the slope of areas ranged considerable and CAN, CAB and MAB exhibited the highest differences. The remaining areas seem to be flatter and had lower values of slope.

In relation to SSS, CAN and N_Br had the highest median of Mean Annual SSS (more than 36 psu) as well as the lowest values of Annual Range in SSS (0.19 and 0.68 psu, respectively). Br_Uy, CAB, eGOM and SAB had medians of Mean Annual SSS around 35 psu, and Annual Range in SSS medians ranging from 0.90 psu to 3.65 in these areas. wGOM had Mean Annual of SSS equal to 34.7 psu and Annual Range in SSS equal to 3.7 psu. MAB had the lowest median of Mean Annual SSS (33 psu) and Annual Range in SSS equal to 2.24 psu.

In relation to SST, CAB and N_Br had the highest median of Mean Annual SST (more than 27°C) as well as the lowest values of Annual Range in SSS (3.2 and 2°C, respectively). Br_Uy and SAB had pretty similar median of Mean Annual SST (around 23°C), but medians were significantly different in relation to Annual Range in SST (Br_Uy = 5°C; SAB = 9°C). eGOM and wGOM were not differentiated in relation to both Mean Annual and Annual Range of SST, where median of these variables were closest to 24 – 25 °C and 8 – 10°C, respectively. CAN and MAB had the lowest values of Mean Annual SST (20.5°C and 18°C, respectively). MAB had the highest value of Annual Range in SST around 13°C.

Fig. 4 Boxplots representing ranges of the six environmental variables analyzed for each putative population (represented by more than one geographic record). Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico

Ecological Niche Modeling

In our study, the main aim of ecological niche modeling was to provide a map of suitability to estimate resistance matrix for further IBR analyses, thus we provided here a general description of models. We highlighted that these models were generated only taking into account the geographic coordinates analyzed in this study. Therefore, the models are most likely to just reflect environment suitability of these specific set of records and do not should be considered as a potential distribution model for the species in both contemporary and past conditions.

After filtering our geographic coordinates dataset representing the sequences analyzed, 98 presence records were used for training model. The best configuration model defined with the lowest AICc

value were feature class equals to Hinge (H) and regularization multiplier equals to 3.5. Environmental layers with the highest percentage of contribution to the model were bathymetry (85.9%) and Mean Annual SST. The model had AUC training equal to 0.967.

Prediction of the contemporary conditions (Fig. 5) suggested high environmental suitability along the continental shelf of Africa and around Oceanic Islands in the Eastern Atlantic; in the Western Atlantic, high suitability was recovered mainly in the Western North Atlantic shelf, Gulf of Mexico, and along the continental shelf from southeastern Brazil until Uruguay. In the Caribbean Sea, spots of high suitability were recovered in the continental shelf of Colombia and Venezuela. On the other hand, prediction in the LGM conditions (Figure 6) suggested high environmental suitability in specific areas along continental shelves of America and Africa. Since sea level reduced 120 m during the LGM, many areas of high suitability in the contemporary period were not available for the species in the LGM. Therefore, oceanic islands seems to exhibited more suitability at that time in relation to contemporary model.

Fig. 5 Maps representing the environmental suitability model for Atlantic spotted dolphin in (a) contemporary and (b) Last Glacial Maximum conditions Last Glacial Maximum conditions. Warm colours represent high environmental suitability; cold colours low environmental suitability; and, black zones representing continental shelf portions that were dry during 120 m sea level regression. Model was generated from 98 presence records analyzed in this study, therefore should not be considered as a potential distribution map for the species in both contemporary and past conditions

"Isolation by" analyses

Four least-cost distance matrices were generated as well as its respective maps representing least-cost paths (Fig. 6). Least-cost distance with no constraint considered the least-cost path ignoring bathymetry, while least-cost with constraints computed transition object with maximum depth constraint of 300 m, 600 m, and 900 m (i.e., path impossible in waters deeper than 300 m, 600 m, and 900 m, respectively). The least-cost distance with maximum depth constraints were similar to each other and revealed interesting paths between populations.

Fig. 6 Least-cost distances maps. Lines represented the least-cost distances computed among putative populations of Atlantic spotted dolphins without and with a bathymetric constraint of 300 m, 600 m and 900 m

On the other hand, resistance distance analyses based on suitability models for contemporary conditions indicated multiple pathways among populations. In relation to resistance analyses based on suitability models for LGM conditions, only pathways including MAD, CAN, MAB, eGOM, wGOM and

CAB were considered because the remaining centroids were not represented in the LGM suitability map due to the decreasing of 120 m of sea level.

Simple linear regression between genetic matrices and resistance, geographic and environmental matrices are presented in Table 4. In the F_{ST} and Φ_{ST} matrices significant relationships were detected between both fixation indexes and three leas-cost distances with constraints as well as between both fixation indexes and resistance matrix based on suitability map of LGM condition. In relation to Nei's dA matrix, significant relationships were detected between Nei's dA and least-cost distance with no constraint as well as between resistance matrices based on suitability map of contemporary and LGM conditions. The highest R² were from linearized Nei's dA against resistance matrix based on suitability map of contemporary conditions (R² = 0.172); and, linearized Φ_{ST} against least-cost distances with maximum of 300 m constraint (R² = 0.172).

Table 4. Simple linear regression between linearized F_{ST}, Φ_{ST} and Nei's dA matrices and least-cost as well as resistance matrices. Abbreviations: ENM, Ecological Niche Model; LGM, Last Glacial Maximum. Significant values are high-lighted in bold.

		r (slope)	R^2	P-value
F_{ST}	No restriction	2680.6	0.02704	0.2303
	Max. 300m depth	**-315300000000**	0.1667	**0.001973**
	Max. 600m depth	**-299800000000**	0.1692	**0.001808**
	Max. 900m depth	**-280000000000**	0.1683	**0.001863**
	Resistance ENM contemporary	20.85	0.004281	0.635
	Resistance ENM LGM	**-95065**	0.1387	**0.005112**
Φ_{ST}	No restriction	-279.4	0.001906	0.752
	Max. 300m depth	**-125700000000**	0.172	**0.00165**
	Max. 600m depth	**-118600000000**	0.1718	**0.001653**
	Max. 900m depth	**-110800000000**	0.1709	**0.001706**
	Resistance ENM contemporary	-19823	0.02511	0.2479
	Resistance ENM LGM	**-31222**	0.09707	**0.0206**
Nei's dA	No restriction	**206707.8**	0.1535	**0.0031**
	Max. 300m depth	-1020000000000	0.001666	0.767
	Max. 600m depth	-1042000000000	0.001951	0.749
	Max. 900m depth	-1031000000000	0.002178	0.7351
	Resistance ENM contemporary	**4287.23**	0.1727	**0.0016**
	Resistance ENM LGM	**-2488333**	0.09069	**0.02547**

DbRDA testes conducted considering least cost distances, environmental variables separately or summarized as PC1 did not detect significant IBD or IBE neither in marginal tests (Table 5) nor conditional tests (Table 6) that taking into account for spatial variation.

Table 5. Results of marginal dbRDA tests. Abbreviations: BAT, Bathymetry; SSS, Sea Surface Salinity; SST, Sea Surface Temperature; PC1, Principal Component 1

Marginal dbRDA Tests			
Variable	F	P-value	% Variance
F_{ST}			
SST	-0.88	0.96	-2.9
BAT	-0.06	0.6	-0.19
SSS	0.53	0.53	1.45
PC1	-0.06	0.58	0.17
Least cost distance no restriction	26.29	0	0
Least cost distance Max 300m depth restriction	0.83	0.58	11.96
Least cost distance Max 600m depth restriction	-0.21	0.98	-4.37
Least cost distance Max 900m depth restriction	-0.21	0.97	-4.37
Φ_{ST}			
SST	-2.31	0.97	-44.6
BAT	-0.69	0.87	-10.73
SSS	0.16	0.54	2.3
PC1	-0.65	0.83	-10.06
Least cost distance no restriction	128.78	0	0
Least cost distance Max 300m depth restriction	-0.10	0.92	-14.58
Least cost distance Max 600m depth restriction	-0.44	0.99	-54.40
Least cost distance Max 900m depth restriction	-0.44	0.99	-54.40
Nei dA			
SST	2.98	0.12	0.01
BAT	0.33	0.53	0.001
SSS	0.22	0.71	0.001
PC1	0.31	0.56	0.001
Least cost distance no restriction	0	0	0.03
Least cost distance Max 300m depth restriction	0.69	0.72	0.013
Least cost distance Max 600m depth restriction	0.61	0.71	0.0001
Least cost distance Max 900m depth restriction	0.61	0.71	0.01

Table 6. Results of conditional dbRDA tests. Abbreviations: BAT, Bathymetry; SSS, Sea Surface Salinity; SST, Sea Surface Temperature; PC1, Principal Component 1

		Variables	F	P-value	% Variance
		Conditional dbRDA Tests			
F_{ST}	SST	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	-0.09	0.84	-0.32
		Least cost distance Max 600m depth restriction	-0.13	0.87	-0.82
		Least cost distance Max 900m depth restriction	-0.13	0.87	0.82
	BAT	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	0.87	0.46	2.45
		Least cost distance Max 600m depth restriction	-0.25	0.95	-1.61
		Least cost distance Max 900m depth restriction	-0.25	0.95	-1.61
	SSS	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	0.02	0.82	0.07
		Least cost distance Max 600m depth restriction	0.08	0.77	0.48
		Least cost distance Max 900m depth restriction	0.08	0.79	0.48
	PC1	Least cost distance no restriction	0,000	0,000	0,000
		Least cost distance Max 300m depth restriction	0.702	0.504	2.14
		Least cost distance Max 600m depth restriction	-0.11	0.9	0.68
		Least cost distance Max 900m depth restriction	-0.11	0.92	0.68
Φ_{ST}	SST	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	0.06	0.58	2.08
		Least cost distance Max 600m depth restriction	-0.28	0.8	-10.81
		Least cost distance Max 900m depth restriction	-0.28	0.82	-10.81
	BAT	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	2.65	0.21	57.18
		Least cost distance Max 600m depth restriction	0.44	0.56	14.83
		Least cost distance Max 900m depth restriction	0.44	0.54	14.83
	SSS	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	-0.73	0.92	-32.19
		Least cost distance Max 600m depth restriction	-0.9	0.94	-40.06
		Least cost distance Max 900m depth restriction	-0.9	0.94	-40.06
	PC1	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	1.95	0.25	46.9
		Least cost distance Max 600m depth restriction	0.52	0.51	17.43
		Least cost distance Max 900m depth restriction	0.52	0.51	17.43
Nei dA	SST	Least cost distance no restriction	0	0	0
		Least cost distance Max 300m depth restriction	2.25	0.2	0.01
		Least cost distance Max 600m depth restriction	1.53	0.3	0.005
		Least cost distance Max 900m depth restriction	1.53	0.3	0.005
	BAT	Least cost distance no restriction	0	0	0

	Least cost distance Max 300m depth restriction	-0.01	0.81	−0.00004
	Least cost distance Max 600m depth restriction	-0.27	0.97	−0.001
	Least cost distance Max 900m depth restriction	-0.27	0.97	−0.001
	Least cost distance no restriction	0	0	0
SSS	Least cost distance Max 300m depth restriction	10.64	0.06	0.01
	Least cost distance Max 600m depth restriction	2.56	0.16	0.01
	Least cost distance Max 900m depth restriction	2.56	0.17	0.01
	Least cost distance no restriction	0	0	0
PC1	Least cost distance Max 300m depth restriction	-0.46	0.95	−0.002
	Least cost distance Max 600m depth restriction	-0.42	0.98	−0.002
	Least cost distance Max 900m depth restriction	-0.42	0.98	−0.002

Mantel tests between Nei's dA genetic matrix and least-cost distance with no restriction was statistically significant (r = 0.39, p = 0.02), as well as between Nei's dA and resistance matrix based on suitability map of contemporary condition (r = 0.41, p = 0.04) (Table 7). All remaining comparisons including marginal tests considering least-cost distances, environmental Euclidean Distances, resistance distances (Table 7), and, conditional tests considering Euclidean distances controlling for least-cost distances were not statistically significant (Table 8).

Table 7. Results of marginal Mantel tests. Abbreviation: LGM, Last Glacial Maximum Significant results are typed in bold

	Marginal Mantel Tests		
	Variable	r	P-value
	Least cost distance no restriction	0.16	0.19
	Least cost distance Max 300m depth restriction	-0.41	0.99
	Least cost distance Max 600m depth restriction	-0.41	0.99
F_{ST}	Least cost distance Max 900m depth restriction	-0.41	0.99
	Environmental (Euclidean Distance)	-0.22	0.74
	Contemporary Ecological Niche Modeling (Resistance)	0.06	0.39
	LGM Ecological Niche Modeling (Resistance)	-0.37	0.98
	Least cost distance no restriction	-0.04	0.59
	Least cost distance Max 300m depth restriction	-0.41	0.99
	Least cost distance Max 600m depth restriction	-0.41	0.99
ΦST	Least cost distance Max 900m depth restriction	-0.41	0.99
	Environmental (Euclidean Distance)	-0.15	0.63
	Contemporary Ecological Niche Modeling (Resistance)	-0.16	0.74
	LGM Ecological Niche Modeling (Resistance)	-0.31	0.99
Nei dA	**Least cost distance no restriction**	**0.39**	**0.02**
	Least cost distance Max 300m depth restriction	-0.04	0.52
	Least cost distance Max 600m depth restriction	-0.04	0.55
	Least cost distance Max 900m depth restriction	-0.05	0.57
	Environmental (Euclidean Distance)	-0.1	0.51

Contemporary Ecological Niche Modeling (Resistance)		**0.41**	**0.04**
LGM Ecological Niche Modeling (Resistance)		-0.30	0.93

Table 8. Results of conditional Mantel tests.

	Conditional Mantel Tests			
	Variables		r	P-value
F_{ST}	Environmental (Euclidean Distance)	Least cost distance no restriction	-0.20	0.66
		Least cost distance Max 300m depth restriction	-0.29	0.90
		Least cost distance Max 600m depth restriction	-0.29	0.92
		Least cost distance Max 900m depth restriction	-0.29	0.91
Φ_{ST}	Environmental (Euclidean Distance)	Least cost distance no restriction	-0.16	0.65
		Least cost distance Max 300m depth restriction	-0.22	0.88
		Least cost distance Max 600m depth restriction	-0.22	0.90
		Least cost distance Max 900m depth restriction	-0.22	0.88
Nei dA	Environmental (Euclidean Distance)	Least cost distance no restriction	-0.03	0.47
		Least cost distance Max 300m depth restriction	-0.10	0.52
		Least cost distance Max 600m depth restriction	-0.10	0.51
		Least cost distance Max 900m depth restriction	-0.10	0.53

Discussion

In this study, we evaluated by the first time the genetic structure along the full species range of Atlantic spotted dolphins using mtDNA CR marker. Furthermore, we investigated the relationship among genetic structure, geography, and contemporary and past environment heterogeneity following a seascape genetics approach. Our results show that both geography and environment have a significant relationship with genetic structure, including past environment conditions. Although we do not detect a strong relationship, the results suggest that geographic distances, namely IBD, could have more influence in such larger great study scale, while environmental heterogeneity could be more influent in smaller scales as already proposed for the species (Viricel and Rosel 2014).

Genetic Structure

Recently, the importance of mtDNA CR marker in studies conducted with cetaceans species was highlighted even in the era of next generation sequencing due to great number of species that have this marker sequenced and easily available in data repositories such as GenBank (Rosel et al. 2017a, b). Several metrics and markers that could be used to improve taxonomic resolution among cetaceans species were reviewed, and Nei's dA net divergence and Φ_{ST} seemed to perform better in relation to other metrics in

119

studies conducted with mtDNA CR marker (Rosel et al. 2017b). Since traditionally, landscape genetic studies use linearized F_{ST} to test for the relationships between genetic and geographic distances (Rousset 1997), we decide to also estimate this statistics so that this study can be comparable with others that have been conducted following a seascape genetics approach. Therefore, we analyzed and present results in three different metrics to investigate genetic structure (F_{ST}, Φ_{ST} and Nei's dA) along Atlantic spotted dolphin distribution. Furthermore, our Nei's DA results were within the interval considered appropriate for population designation (0.00007 – 0.004) and, in some comparisons Nei's dA values were higher than 0.014, which is the threshold considered appropriate to subspecies differentiation (Rosel et al. 2017b).

In relation to studies previously conducted with Atlantic spotted dolphin, our ranges of fixation and molecular diversity indexes seems to be similar to other studies (Adams and Rosel 2006; Green et al. 2007; Quérouil et al. 2010; Caballero et al. 2013; Viricel and Rosel 2014). However, the pairwise relationship between populations changed in some cases, and putative populations considered not statically differentiated in previous studies were considered different populations here; for example, we obtained significant F_{ST} between MAB and Azores, and between BAH in relation to GOM and SAB. Probably, this happened due to the inclusion of samples never analyzed before and also due to changes in the frequency of haplotypes in each population analyzed previously (Green 2008; Viricel and Rosel 2014), because we are not able to include the exactly same set of samples analyzed previously.

We also highlight the importance of inclusion of areas never analyzed before to understand population structure along Atlantic spotted dolphin distribution. Samples from the Canary Islands, northern Brazil and from the southernmost limit of the species were analyzed for the first time. We also included a higher number of samples from southeastern Brazil and added more samples from Caribbean in the dataset. Unfortunately, we could not get samples from Western Africa, which should become indispensable in further studies. In relation to these new samples added, CAN seems to be more closely related to the AZ and MAD. Therefore, AZ, MAD and CAN could be considered one oceanic population with the inclusion of MAB based on mtDNA CR marker.

The analyses of samples obtained from south/ southeastern Brazil and Uruguay revealed lower diversity indexes and were statistically differentiated from almost all populations in relation to both fixation indexes as suggested previously by Caballero et al. (2013). Since we analyzed a higher number of samples

from Brazil, we highlighted the lowest number of haplotypes and nucleotide diversity recovered in this population, being 8 and 0.0067 respectively. Other populations such as CAB and AZ with a sample size of almost a fifth and quarter, respectively, than Brazil have extremely higher values of both indexes. Nei's dA values were higher than those considered to designate subspecies in almost all pairwise comparisons, indicating a strong evidence for both geographic and reproductive isolation of this population.

In relation to Atlantic spotted dolphins that are recorded in north Brazil it is difficult to determinate its relationship with other populations because a gap of approximately 1,500 km is recognized between 6°S and 18°S due to the narrow of continental shelf (Moreno et al. 2005) and low environmental suitability (do Amaral et al. 2015). Taking into account fluxing of the warm North Brazil Current (NBC), it seems reasonable proposing that North Brazil individuals are more related to the Caribbean. Costa et al. (2017) had already proposed that the north Brazil cetacean fauna seems more similar with those from southern Caribbean region rather than from southern Brazil. However, two samples analyzed here were not enough to elucidate the relationship of individuals from north Brazil with the remaining populations. Our results revealed that one sample had Haplotype 45 which was only recovered in 4 samples from Brazil collected off the 50 m isobath. The other sample had Haplotype 53 which was also found in 6, 3 and 1 samples from AZ, MAD and CAN, respectively.

The position of Uruguayan sample in the Brazil population is intriguing. Moreno et al. (2005) had suggested that dolphins from southeastern Brazil follow the displacement of Brazil Current southward in the summer months. Posteriorly, this idea was corroborated by ecological niche modeling (do Amaral et al. 2015). These factors led us to group samples from south Brazil (~ 29ºS) and Uruguay (~34ºS) with those collected in the SBB, therefore considering only one population (namely, Br_Uy) from approximately 22ºS to 34ºS in the Western Atlantic Ocean. The sample collected at south Brazil (~29ºS) had Haplotype 71, which is also found in eight samples from SBB and two samples from AZ and MAD. However, Haplotype 85 recovered in the Uruguayan sample has high frequency in North Atlantic, mainly in SAB. This result is surprising and we considered that individuals from Atlantic Spotted dolphin southernmost distribution deserve attention in future studies, and they are essential to provide a better understanding of dispersal of the species.

Caballero et al. (2013) highlighted the high number of haplotypes shared between Azores, Madeira and Brazilian samples and suggested, based on data available at that time, Atlantic spotted dolphins in the Central Atlantic (i.e. Azores–Madeira–southeastern Brazil) should be managed as one population for practical terms. However, since we analyzed a sample size six times higher than those analyzed by Caballero et al. (2013) and recovered low diversity indexes and statistically significant differences between Br_Uy and all putative populations along species range, including Caribbean, we suggested that individuals from southeastern/south South America should be considered as a different population for management purposes until more data becomes available.

In relation to the Caribbean, despite the relatively small number of samples analyzed here the genetic diversity found was high. In general, fixation indexes and Nei's DA values indicated no differentiation of the Caribbean in relation to Western North Atlantic populations and the Oceanic population (AZ, MAD, CAN and MAB). Caballero et al. (2013) founded no differentiation in pairwise comparisons of CAB with Azores and Madeira in relation to Φ_{ST}; but these comparisons were significant for F_{ST}. We considered that further studies should be carried out to better determinate the relationships of Caribbean individuals with other regions. At this point, we can just confirmed the hypothesis proposed by Caballero et al. (2013), which suggested that Caribbean and the southeastern Brazil belong each other to separate populations based on significant differentiation found at mtDNA CR marker.

In relation to Western North Atlantic, we recovered similar patterns already obtained (e.g., Adams and Rosel 2006; Viricel and Rosel 2014). Small differences in the values of fixation or molecular diversity index could be explained by different sample sizes included here, since we filtered sequences according to the availability of precise geographic coordinates. An interesting pattern recovered in our study was the significant differentiation of BAH population in relation to all putative populations analyzed, and mainly from closest populations such as eGOM and SAB. Previous genetic analysis showed that this population was connected with those from the GOM and SAB probably mediated by the Gulf Stream flow (Green 2008). However, we recovered significant and higher differentiation in relation to wGOM and SAB; and, slightly lower differences were recovered in relation to eGOM. We believed that these differences could be attributed to different sample size analyzed, since after the studies of Green (2008) more sequences from

Western North Atlantic become available through the studies of Kingston et al. (2009) and Viricel and Rosel (2014).

Despite the inherent difficulty to group cetacean individuals in populations, we believe that our results are in agreement with previous studies that tested differentiation among putative populations of Atlantic spotted dolphins based on both mtDNA and nuclear data. Furthermore, the Procrustes analyses conducted at individual level and with no determination of groups *a priori* indicated significant relationships between genetic, geography and environment. Although these analyses are not commonly conducted with mtDNA data, we believe that the results obtained were very interesting and allowed a better understanding of our data. The relationships between geography and environment were not so evident in the "isolation by" analysis (see discussion below), but the patterns of grouping in the procrustes showed a pattern that corroborated other analyses. For example, the strong influence of North Atlantic haplotypes in South Hemisphere is evident in Fig. 2a. In the analysis of genetic versus environment the clusters showed in the Figure 2b are in agreement with our division of putative populations *a priori* with few individuals far from its counterparts. Such pattern is easily explained by the mobility of this top predator species. Furthermore, this last procrustes analysis brings light to similarities of clusters in the environmental space that would not be expected taking into account the great environmental heterogeneity of the Atlantic Ocean; for example the proximity of Br_Uy with SAB and MAB populations. These similarities and differences were also better visualized in the boxplot analyzes. As pointed out by (Massatti et al. (2017), procrustes analyses in a genetic context are interpreted under the assumption that individuals from populations closer in geographic proximity will be more closely related (i.e., IBD), and deviations from this pattern bring to light interesting biological and/or historical phenomena.

"Isolation by"

Taking as a whole, the regression of genetic distances against least-cost distances and resistance matrices were significant and suggested that both contemporary conditions and least-cost distance with a maximum of 300 m depth constraint could be considered as a predictor of genetic differentiation. Although we have significant values in our tests, our coefficient values are relatively low, suggesting that just 17% of genetic variation could be explained by environment or geography. These results are in agreement with procrustes analyses that also suggested a significant correlation between geography and environment with

genetics, being the correlation between geography and genetic slightly higher than environment and genetic.

Although we find some level of explanations in the models, we considered that many factors play role to explain these pattern such as 1) the scale of study, 2) the environmental similarity of some unexpected regions, 3) the utilization of one pair of geographic coordinates (i.e, centroids) to represent each population, 4) the methods utilized (dbRDA and Mantel tests), 5) the utilization of mtDNA CR, and 6) the possibility of social structure driving genetic patterns at some regions (e.g. BAH).

The formers four factors could be considered complementary to each other to explain our results. Environmental analyses (see Figs. 2b, 3 and 4) revealed similarities between regions unexpected to be similar, for example Western North Atlantic and Western South Atlantic. Since we used environmental information sampled for a centroid of each regions in both dbRDA and Mantel tests, maybe the environmental information extracted from just one point (i.e, centroid) was not able to capture properly the environmental differences among regions. Although is noteworthy to point out that the ecological similarities from Western North Atlantic and Western South Atlantic were recovered by a principal Component Analyses (PCA) performed with six environmental variables (see Fig. 3).

At large scale study conducted with the grey wolf (*Canis lupus*), a top predator widely distributed and with high dispersal capabilities, Geffen et al. (2004) recovered significant IBD and, when spatial variation was taken into account, a significant relationship between genetic variation and climate were detected in the case of microsatelite Nei's distance or F_{ST} based on mtDNA RFLP profiles. However, the authors did not detected significant relationships of a set of environmental variables with genetic structure even considering longitude and latitude as covariables. The authors attributed these results to a lack of power of the analyses due to the low number of populations analyzed at that time. Despite the inherent differences between terrestrial and marine environment, we considered some similarities between our study and the grey wolf' study because both used dbRDA tests that failed to detect isolation by environment in top predators with high dispersal abilities at large scales. Perhaps these relationship between environment and genetic should be tested at regional scales, where with a reduced dataset environmental data could be included in a more comprehensive way. For example, Viricel and Rosel (2014) had already suggested that

environment play a role to explain genetic differences between adjacent regions (eGOM and wGOM) in Western North Atlantic.

Mantel tests is widely used in landscape/seascape studies (Manel 2003; Storfer et al. 2007; Balkenhol et al. 2009; Mendez et al. 2010, 2011; Amaral et al. 2012; He et al. 2013). Despite several criticisms and discussions about the statistical performance of Mantel tests have been made, and other sophisticated and complex approaches to analyze spatial multivariate data are available the Mantel test is still widely used (Diniz-Filho et al. 2013). The authors suggested that Mantel tests provide a simple and useful tool for multivariate analysis of spatial patterns of genetic divergence mainly if the ecological or evolutionary hypotheses are expressed as pairwise distances or similarities and considering a careful application and interpretation of Mantel tests (Diniz-Filho et al. 2013).

The analysis conducted by Mendez et al. (2011) for humpback dolphins along Western Indian Ocean did not show patterns of IBD or IBE. The authors suggested that lack of IBD is not uncommon among cetaceans, and has been attributed to a negligible influence of geography in the presence of other factors, such as behavioral constraints or environmental discontinuities (Mendez et al. 2010). However, Mendez et al. (2011) observed a strong and seemingly overlapping genetic and environmental breaks and suggested that these environmental breaks could have some influence in the genetic structure of humpback dolphin populations in a non-linear or proportional manner. Indeed, the most part of analyzes performed here and the Mantel tests assuming a linear correlation among data. However, water resources data frequently exhibit features resulting in a deviation from linearity: assymetry or skewness, outliers and heavy tails (symmetric data with more observations at both extremes) (Helsel and Hirsch 2002). Thus, we hypothesized that the non-linearity of our data could also have an impact in the analyses performed here.

 In general, we observed that our different methods (Simple linear regression, Mantel tests and even procrustes analyses) are in agreement at some level. All of them suggested a pattern of IBD with some influence of IBE. Therefore, we believe that the large spatial scale of our study and the methods that assuming a linearity of data could play a role to explain the low levels of correlation among genetic structure, geography and environment.

In relation to use of mtDNA CR, we believed that the inclusion of more samples and more nuclear markers certainly will improve the power of analyses. But we advocate in defense of mtDNA CR because

this marker provides an exceptional opportunity to investigate the genetic structure along almost the total distribution of a cetacean species. As pointed out by Holderegger and Wagner (2008), mtDNA often do not provide enough genetic variation among individuals at the spatial scales of landscape genetic studies. However, if there is enough mtDNA variation among the individuals within a landscape, dispersal can be readily determined (Holderegger and Wagner 2008). At this point, we cannot determine dispersal among populations and this is not our goal, but certainly mtDNA CR analyses revealed some interesting patterns along Atlantic spotted dolphin distribution that should be further investigated.

In relation to social structure, our aim was neither looking for phylopatry within populations or within sexes nor identifying sex-biased dispersal - already investigated by Adams and Rosel (2006) and Quérouil et al. (2010). However, we observed that BAH population exhibit a high degree of pairwise differentiation even among the closest populations such as eGOM and SAB. The population of Bahamas is long-term studied, and social clusters and some level of cluster phylopatry for both sexes were found (Elliser and Herzing 2012). Furthermore, this community of Atlantic spotted dolphins in Bahamas has long-term affiliations that are often correlated with age, sex, and reproduction factors, being mating strategies and sex the primary factors shaping social structure. Reproduction and social familiarity strongly influence female associations, whereas age and alliance formation strongly affect male associations (Elliser and Herzing 2014). In Gulf of Mexico, indirect evidences of some kind of site fidelity was reported by (Viricel and Rosel 2014), since one of the samples was collected twice in the same region with a difference of five years. In Brazil, a relatively higher number of individuals from SBB was photo-identified in the same place in a interval of two years (Santos et al. 2018). Therefore, we hypothesized that social structure could have some impact in our analyses, probably confusing our matrices analyses because great genetic differentiation was recovered at short distances, for example, in the pairwise comparison between BAH and SAB.

Regression analyses detected a significant relationship between genetic structure and resistance matrices based on past conditions. Much of continental shelf was exposed when sea level reduced 120 m along Atlantic Ocean, therefore many areas that are occupied today by dolphins were lost during LGM (see Fig. 6). Some populations analyzed here (SAB, BAH, N_Br, Br_UY) were not considered in the resistance matrix based on environmental suitability of LGM, because its respective centroids were not represented in the LGM map. Transgression and regression of sea during glacial cycles had also a great impact in the

126

southeastern Brazil shelf (Mahiques et al. 2010). Despite a high environmental suitability was recovered in the LGM model, the continental shelf suffered a substantial narrowing in the SBB, for example. Furthermore, it was suggested that the western South Atlantic could be colder than at present, within the expected range for a glacial interval (Laprida et al. 2011). Overall, haplotype network indicated a strong connection of all haplotypes with those from AZ and MAD, and also a high genetic diversity and signal of population expansion were recovered in the Oceanic population (AZ, MAD, CAN, MAB). Procrustes analyses also revealed a strong relationship between southern hemisphere and northern hemisphere populations. Thus, we hypothesized that Oceanic population were environmentally more stable during the LGM, and since these individuals may were not present in the continental shelf, having an oceanic distribution, they were able to recolonize posteriorly different areas of the continental shelf that have similar environmental characteristicas (e.g., Western North Atlantic and Western South Atlantic) are not adapted to continental shelf they were able to recolonize different areas of the continental shelf. Viricel and Rosel (2014) had already proposed the influence of sea level changes to explain the genetic differences within GOM.

General considerations

In comparison to other studies that analyzed population structure using mtDNA CR marker in *Stenella* dolphins, we observed that our values were similar from those comparisons within oceanic basins (Escorza-Treviño et al. 2005; Andrews et al. 2013). However, in some pairwise comparisons between populations of Atlantic spotted dolphin we recovered higher values of Φ_{ST} than those values obtained in pairwise comparisons between ecotypes and/or subspecies described for Spinner dolphin (*Stenella longirostris*) and Pantropical spotted dolphin (*S. attenuta*). In relation to Clymene dolphin (*S. clymene*), which is also endemic of the Atlantic Ocean, a preliminary study also indicated that individuals from Brazil are distinct from those of North Atlantic, mainly from oceanic waters of North Atlantic, while and some level of connection was recovered between South Atlantic individuals and those from Gulf of Mexico (Nara et al. 2017) .

In general, information of genetic structure of *Stenella* dolphins in Atlantic Ocean and the relationships of dolphins from the Atlantic Coast of South America with those from North Atlantic is scarce

or even nonexistent for most species of *Stenella*. Therefore, based on our results and those obtained for Clymene dolphin (Nara et al. 2017), we suggested that other species of *Stenella* could also have exhibit some level of population differentiation in south Atlantic waters and deserve attention in future studies.

Conclusion

In this study, we revealed that Atlantic spotted dolphin exhibits significant population structure along its distribution based on mtDNA CR marker. Since we analyzed a short fragment of mtDNA CR, we highlighted that the patterns recovered here deserve further investigation.

We also pointed out the distinctiveness of those individuals from southeastern Brazil and the complex relationships among populations evidenced by our results. Nevertheless, we reinforce the necessity of more information to determine the status of individuals recorded in Brazil and also to provide a better understanding of the relationship of individuals recorded along the Atlantic Coast of South America. These aims should be addressing through the analyze of nuclear markers such as SNPs and the inclusion of more samples. Specifically, we recommend more sampling effort in the north and south Brazil as well as Uruguay, and from individuals recorded in the outer continental shelf. We believed that the inclusion of more genetic markers and individuals from the outer continental shelf should bring some light regarding the connections among populations. Beyond that, it is crucial the inclusion of individuals from Eastern Africa in further studies.

Despite we had detected some level of IBD and IBR including contemporary and past conditions, we also hypothesized that different process could play role to explain the genetic patterns recovered here such as social structure and some level of phylopatry within populations.

References

Adams LD, Rosel PE (2006) Population differentiation of the Atlantic spotted dolphin (*Stenella frontalis*) in the western North Atlantic, including the Gulf of Mexico. Mar Biol 148:671–681. doi: 10.1007/s00227-005-0094-2

Amaral AR, Beheregaray LB, Bilgmann K, Boutov D, Freitas L, Robertson KM, Sequeira M, Stockin KA, Coelho MM, Möller LM (2012) Seascape genetics of a globally distributed, highly mobile marine mammal: The short-beaked common dolphin (genus *Delphinus*). PLoS One. doi: 10.1371/journal.pone.0031482

Andrews KR, Perrin WF, Oremus M, Karczmarski L, Bowen BW, Puritz JB, Toonen RJ (2013) The evolving male: Spinner dolphin (*Stenella longirostris*) ecotypes are divergent at y chromosome but not mtDNA or autosomal markers. Mol Ecol 22:2408–2423. doi: 10.1111/mec.12193

Archer FI, Adams PE, Schneiders BB (2016) strataG: An R package for manipulating, summarizing, and analyzing population genetic data. Mol Ecol Resour doi: 10.1111/1755-0998.12559

Balkenhol N, Waits LP, Dezzani RJ (2009) Statistical approaches in landscape genetics: An evaluation of methods for linking landscape and genetic data. Ecography (Cop) 32:818–830. doi: 10.1111/j.1600-0587.2009.05807.x

Bandelt HJ, Forster P, Röhl A (1999) Median-joining networks for inferring intraspecific phylogenies. Mol Biol Evol 16:37–48. doi: 10.1093/oxfordjournals.molbev.a026036

Baumgartner MF, Mullin KD, May LN, Leming TD (2001) Cetacean habitats in the northern Gulf of Mexico. Fish Bull 99:219–239. doi: 10.1016/S0967-0637(01)00035-8

Benestan L, Quinn BK, Maaroufi H, Laporte M, Clark FK, Greenwood SJ, Rochette R, Bernatchez L (2016) Seascape genomics provides evidence for thermal adaptation and current-mediated population structure in American lobster (*Homarus americanus*). Mol Ecol 25:5073–5092. doi: 10.1111/mec.13811

Bierne N, Bonhomme F, David P (2003) Habitat preference and the marine-speciation paradox. Proc R Soc B Biol Sci 270:1399–1406. doi: 10.1098/rspb.2003.2404

Caballero S, Santos MCO, Sanches A, Mignucci-Giannoni AA (2013) Initial description of the phylogeography, population structure and genetic diversity of Atlantic spotted dolphins from Brazil and the Caribbean, inferred from analyses of mitochondrial and nuclear DNA. Biochem Syst Ecol 48:263–270. doi: 10.1016/j.bse.2012.12.016

Costa AF, Siciliano S, Emin-Lima R, Martins BML, Sousa MEM, Giarrizzo T, Júnior J de SES (2017) Stranding survey as a framework to investigate rare cetacean records of the north and north-eastern Brazilian coasts. Zookeys 111–134. doi: 10.3897/zookeys.688.12636

Davis RW, Ortega-Ortiz JG, Ribic CA, Evans WE, Biggs DC, Ressler PH, Cady RB, Leben RR, Mullin KD, Würsig B (2002) Cetacean habitat in the northern oceanic Gulf of Mexico. Deep Res Part I Oceanogr Res Pap 49:121–142. doi: 10.1016/S0967-0637(01)00035-8

Davis RW, Worthy GAJ, Würsig B, Lynn SK, Townsend FI (2006) Diving behavior and at-sea movements of an Atlantic spotted dolphin in the Gulf of Mexico. Mar Mammal Sci 12:569–581. doi: 10.1111/j.1748-7692.1996.tb00069.x

Diniz-Filho JAF, Soares TN, Lima JS, Dobrovolski R, Landeiro VL, Telles MP de C, Rangel TF, Bini LM (2013) Mantel test in population genetics. Genet. Mol. Biol. 36:475–485

Dinno A (2017) Dunn's Test of Multiple Comparisons Using Rank Sums. R package version 1.3.5

do Amaral KB, Alvares DJ, Heinzelmann L, Borges-Martins M, Siciliano S, Moreno IB (2015) Ecological niche modeling of *Stenella* dolphins (Cetartiodactyla: Delphinidae) in the southwestern Atlantic Ocean. J Exp Mar Bio Ecol 472:166–179. doi: 10.1016/j.jembe.2015.07.013

Dray S, Dufour AB (2007) The ade4 Package: Implementing the Duality Diagram for Ecologists. R package version 1.7-13

Elliser CR, Herzing DL (2012) Community structure and cluster definition of Atlantic spotted dolphins, *Stenella frontalis*, in the Bahamas. Mar Mammal Sci. doi: 10.1111/j.1748-7692.2012.00576.x

Elliser CR, Herzing DL (2014) Long-term social structure of a resident community of Atlantic spotted dolphins, *Stenella frontalis*, in the Bahamas 1991-2002. Mar Mammal Sci 30:308–328. doi: 10.1111/mms.12039

Escorza-Treviño S, Archer FI, Rosales M, Lang A, Dizon AE (2005) Genetic differentiation and intraspecific structure of Eastern Tropical Pacific spotted dolphins, *Stenella attenuata*, revealed by DNA analyses. Conserv Genet 6:587–600. doi: 10.1007/s10592-005-9013-9

Excoffier L, Lischer H (2015) An Integrated Software Package for Population Genetics Data Analysis. Evol Bioinform Online 1:47–50

Fernández R, Santos MB, Carrillo M, Tejedor M, Pierce GJ (2009) Stomach contents of cetaceans stranded in the Canary Islands. J Mar Biol Assoc United Kingdom 89:873–883. doi: 10.1017/S0025315409000290

Foote AD, Vijay N, Ávila-Arcos MC, Baird RW, Durban JW, Fumagalli M, Gibbs RA, Hanson MB, Korneliussen TS, Martin MD, Robertson KM, Sousa VC, Vieira FG, Vinar T, Wade P, Worley KC, Excoffier L, Morin PA, Gilbert MTP, Wolf JBW (2016) Genome-culture coevolution promotes rapid divergence of killer whale ecotypes. Nat Commun. doi: 10.1038/ncomms11693

Freitas CMA, Dellinger T, Reiner F (1989) First record of Atlantic spotted dolphins, *Stenella frontalis* G. Cuvier, 1829 (Cetacea: Delphinidae) from the Archipelago of Madeira. Bocagiana 190:1–5

GBIF.org (2018) GBIF Occurrence Download https://doi.org/10.15468/dl.qlesop. Accessed 01 April 2017

Geffen E, Anderson MJ, Wayne RK (2004) Climate and habitat barriers to dispersal in the highly mobile grey wolf. Mol Ecol 13:2481–2490. doi: 10.1111/j.1365-294X.2004.02244.x

Green ML (2008) Assessment of genetic population structure, promiscuity, and paternity in free-ranging Atlantic spotted dolphins *Stenella frontalis*, in the Bahamas. Dissertation, Florida Atlantic University

Green ML, Herzing DL, Baldwin JD (2007) Noninvasive methodology for the sampling and extraction of DNA from free-ranging Atlantic spotted dolphins (*Stenella frontalis*). Mol Ecol Notes 7:1287–1292. doi: 10.1111/j.1471-8286.2007.01858.x

Hammond PS, Bearzi G, Bjørge A, Forney KA, Karkzmarski L, Kasuya T, Perrin WF, Scott MD, Wang JY, Wells RS, Wilson B (2012) *Stenella frontalis*. In: IUCN Red List Threat. Species 2012. e.T20732A17832795.http://dx.doi.org/10.2305/IUCN.UK.2012.RLTS.T20732A17832795.en. Accessed 9 Jun 2018

Hancock-Hanser BL, Frey A, Leslie MS, Dutton PH, Archer FI, Morin PA (2013) Targeted multiplex next-generation sequencing: Advances in techniques of mitochondrial and nuclear DNA sequencing for population genomics. Mol Ecol Resour 13:254–268. doi: 10.1111/1755-0998.12059

Hauser L, Carvalho GR (2008) Paradigm shifts in marine fisheries genetics: Ugly hypotheses slain by beautiful facts. Fish Fish 9:333–362. doi: 10.1111/j.1467-2979.2008.00299.x

He Q, Edwards DL, Knowles LL (2013) Integrative testing of how environments from the past to the present shape genetic structure across landscapes. Evolution 67:3386–3402. doi: 10.1111/evo.12159

Helsel DR, Hirsch RM (2002) Statistical Methods in Water Resources Techniques of Water Resources Investigations, Book 4, chapter A3. U.S. Geological Survey

Herzing DL (1997) The Life History of Free-Ranging Atlantic Spotted Dolphins (*Stenella frontalis*): Age Classes, Color Phases, and Female Reproduction. Mar Mammal Sci 13:576–595. doi: 10.1111/j.1748-7692.1997.tb00085.x

Hijmans RJ, Phillips S, Leathwick JR, Elith J (2011) dismo: Species Distribution Modeling. R package version 1.1-4

Hijmans RJ, Etten J van, Cheng J, Mattiuzzi M, Sumner M, Greenberg JA, Lamigueiro OP, Bevan A, Racine EB, Shortridge A, Ghosh A (2017) raster: Geographic Data Analysis and Modeling. R package 2.6-7

Holderegger R, Wagner HH (2008) Landscape Genetics. Bioscience 58:199. doi: 10.1641/B580306

Jefferson TA, Curry BE, Leatherwood S, Powell JA (1997) Dolphins and porpoises of West Africa: a review of records (Cetacea: Delphinidae, Phocoenidae). Mammalia 61:87–108. doi: 10.1515/mamm.1997.61.1.87

Kingston SE, Adams LD, Rosel PE (2009) Testing mitochondrial sequences and anonymous nuclear markers for phylogeny reconstruction in a rapidly radiating group: Molecular systematics of the Delphininae (Cetacea: Odontoceti: Delphinidae). BMC Evol Biol. doi: 10.1186/1471-2148-9-245

Laprida C, Chapori N, Chiessi C, Violante R (2011) Middle Pleistocene sea surface temperature in the Brazil-Malvinas Confluence Zone: Paleoceanographic implications based on planktonic foraminifera. Micropaleontology 57(2): 183:195

Legendre P, Andersson MJ (1999) Distance-based redundancy analysis: Testing multispecies responses in multifactorial ecological experiments. Ecol Monogr 69:1–24. doi: 10.1890/0012-9615(1999)069[0001:DBRATM]2.0.CO;2

Legendre P, Legendre L (1998) Numerical Ecology, 2nd edn. Elsevier, Amsterdam

Leutner B, Horning N (2016) Rstoolbox: Tools for Remote Sensing Data Analysis. R package version 0.2.3

Mahiques M, Sousa S, Furtado V (2010) The Southern Brazilian shelf: general characteristics, quaternary evolution and sediment distribution. Braz J Oceanogr 58: 25-34

Manel S, Schwartz MK, Luikart G, Taberlet P (2003) Landscape genetics: combining landscape ecology and population genetics. Trends Ecol Evol 18:189–197. doi: 10.1016/S0169-5347(03)00008-9

Massatti R, Doherty KD, Wood TE (2017) Resolving neutral and deterministic contributions to genomic structure in *Syntrichia ruralis* (Bryophyta, Pottiaceae) informs propagule sourcing for dryland restoration. Conserv. Genet. 1–13. doi:10.1007/s10592-017-1026-7

McRae BH (2006) Isolation by Resistance. Evolution 60:1551. doi: 10.1554/05-321.1

Méndez-Fernandez P, Taniguchi S, Santos MCO, Cascão I, Quérouil S, Martín V, Tejedor M, Carrillo M, Rinaldi C, Rinaldi R, Montone RC (2018) Contamination status by persistent organic pollutants of the Atlantic spotted dolphin (*Stenella frontalis*) at the metapopulation level. Environ Pollut 236:785–794. doi: 10.1016/J.ENVPOL.2018.02.009

Mendez M, Rosenbaum HC, Subramaniam A, Yackulic C, Bordino P (2010) Isolation by environmental distance in mobile marine species: molecular ecology of franciscana dolphins at their southern range. Mol Ecol 19:2212–2228. doi: 10.1111/j.1365-294X.2010.04647.x

Mendez M, Subramaniam A, Collins T, Minton G, Baldwin R, Berggren P, Särnblad A, Amir OA, Peddemors VM, Karczmarski L, Guissamulo A, Rosenbaum HC (2011) Molecular ecology meets remote sensing: Environmental drivers to population structure of humpback dolphins in the Western Indian Ocean. Heredity 107:349–361. doi: 10.1038/hdy.2011.21

Moreno IB, Zerbini AN, Danilewicz D, De Oliveira Santos MC, Simões-Lopes PC, Lailson-Brito J, Azevedo AF (2005) Distribution and habitat characteristics of dolphins of the genus *Stenella*

(Cetacea: Delphinidae) in the southwest Atlantic Ocean. Mar Ecol Prog Ser 300:229–240. doi: 10.3354/meps300229

Muscarella R, Galante PJ, Soley-Guardia M, Boria RA, Kass JM, Uriarte M, Anderson RP (2014) ENMeval: An R package for conducting spatially independent evaluations and estimating optimal model complexity for Maxent ecological niche models. Methods Ecol Evol 5:1198–1205. doi: 10.1111/2041-210X.12261

Möller L, Valdez FP, Allen S, Bilgmann K, Corrigan S, Beheregaray LB (2011) Fine-scale genetic structure in short-beaked common dolphins *(Delphinus delphis)* along the East Australian Current. Mar Biol, 158(1): 113–126. doi:10.1007/s00227-010-1546-x

Nara L, de Meirelles ACO, Souto LRA, Silva-Jr JM, Farro APC(2017). An initial population structure and genetic diversity analysis for *Stenella clymene* (Gray, 1850): Evidence of differentiation between the North and South Atlantic Ocean. Aquat Mamm 43(5): 507–516. doi:10.1578/AM.43.5.2017.507

Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlinn D, Minchin PR, O'Hara RB, Simpson GL, Solymos P, Stevens MHH, Szoecs E, Wagner HH (2017) Vegan: Community Ecology Package. R package version 2.4-3

Palacios D, Baumgartner M, Laidre K, Gregr E (2013) Beyond correlation: integrating environmentally and behaviourally mediated processes in models of marine mammal distributions. Endanger Species Res 22:191–203. doi: 10.3354/esr00558

Pante E, Simon-Bouhet B (2013) marmap: A Package for Importing, Plotting and Analyzing Bathymetric and Topographic Data in R. PLoS One doi: 10.1371/journal.pone.0073051

Paro AD, Rojas E, Wedekin LL (2014) Southernmost record of the Atlantic spotted dolphin, *Stenella frontalis* in the south-west Atlantic Ocean. Mar Biodivers Rec. doi: 10.1017/S1755267214000517

Perrin WF (2009) Atlantic spotted dolphin: *Stenella frontalis*. In: Perrin WF, Würsig B, Thewissen JGM (eds) Encyclopedia of Marine Mammals, 2nd ed. Academic Press, San Diego, pp 54–56

Perrin WF, Mitchell ED, Mead JG, Caldwell DK, Caldwell MC, van Bree PJH, Dawbin WH (1987) Revision of the Spotted dolphins, *Stenella* spp. Mar Mammal Sci 3:99–170. doi: 10.1111/j.1748-7692.1987.tb00158.x

Phillips SJ, Anderson RP, Schapire RE (2006) Maximum Entropy Modeling of Species geographic distribuions. Ecol Model 190: 231–259. doi:10.1016/j.ecolmodel.2005.03.026

Quérouil S, Freitas L, Cascão I, Alves F, Dinis A, Almeida JR, Prieto R, Borràs S, Matos JA, Mendonça D, Santos RS (2010) Molecular insight into the population structure of common and spotted dolphins inhabiting the pelagic waters of the Northeast Atlantic. Mar Biol 157:2567–2580. doi: 10.1007/s00227-010-1519-0

R Core Team (2017) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/. Accessed 1 April 2017

Redfern J V, Ferguson MC, Becker EA, Hyrenbach KD, Good CP, Barlow J, Kaschner K, Baumgartner MF, Forney KA, Ballance LT (2006) Techniques for cetacean–habitat modeling. Mar Ecol Progess Ser 310:271–295

Reilly SB (1990) Seasonal changes in distribution and habitat differences among dolphins in the eastern tropical Pacific. Mar Ecol Progess Ser 66:1–11. doi: 10.3354/meps066001

Riginos C, Liggins L (2013) Seascape Genetics: Populations, Individuals, and Genes Marooned and Adrift. Geogr Compass 7:197–216. doi:10.1111/gec3.12032

Ronquist F, Huelsenbeck J, Teslenko M (2011) MrBAYES 3.2: Bayesian phylogenetic inference under mixed models. Bioinformatics 19:1572–1574. doi: 10.1093/bioinformatics/btg180

Rosel PE, Taylor BL, Hancock-Hanser BL, Morin PA, Archer FI, Lang AR, Mesnick SL, Pease VL, Perrin WF, Robertson KM, Leslie MS, Berta A, Cipriano F, Parsons KM, Viricel A, Vollmer NL, Martien KK (2017a) A review of molecular genetic markers and analytical approaches that have been used for delimiting marine mammal subspecies and species. Mar Mammal Sci. doi: 10.1111/mms.12412

Rosel PE, Hancock-hanser BL, Archer FI, Robertson KM, Martien KK, Leslie MS, Berta A, Cipriano F, Viricel A, Viaud-Martinez KA, Taylor BL (2017b) Examining metrics and magnitudes of molecular genetic differentiation used to delimit cetacean subspecies based on mitochondrial DNA control region sequences. Mar Mammal Sci 33:76–100. doi: 10.1111/mms.12410

Rousset F (1997) Genetic differentiation and estimation of gene flow from F statistics under isolation by distance. Genetics 145:1219–1228. doi: 10.1006/bijl.1996.0127

Santos MCO, Figueiredo G, Van Bressem MF (2018) Cetaceans using the marine protected area of "Parque Estadual Marinho da Laje de Santos", Southeastern Brazil. Brazilian J Oceanogr 65:605–613. doi: 10.1590/s1679-87592017130606504

Sbrocco EJ, Barber PH (2013) MARSPEC: ocean climate layers for marine spatial ecology. Ecology 94:979–979. doi: 10.1890/12-1358.1

Schwartz MK, Boness DJ (2017) Marine mammal subspecies in the age of genetics: Introductory remarks from the Associate Editor and Editor-in-Chief of Marine Mammal Science. Mar Mammal Sci. 33:7–11. doi:10.1111/mms.12416

Selkoe K, Henzler CM, Gaines SD (2008) Seascape genetics and the spatial ecology of marine populations. Fish Fish 9:363–377. doi: 10.1111/j.1467-2979.2008.00300.x

Shah VB, McRae BH (2008) Circuitscape: A Tool for Landscape Ecology. Proc 7th Python Sci Conf (SciPy 2008) 62–66. doi: 10.1111/j.1523-1739.2008.00942.x

Silva MA, Prieto R, Magalhães S, Cabecinhas R, Cruz A, Gonçalves JM, Santos RS (2003) Occurrence and distribution of cetaceans in the waters around the Azores (Portugal), Summer and Autumn 1999–2000. Aquat Mamm 29:77–83. doi: 10.1578/016754203101024095

Spalding MD, Fox HE, Allen GR, Davidson N, Ferdaña ZA, Finlayson M, Halpern BS, Jorge MA, Lombana A, Lourie SA, Martin KD, McManus E, Molnar J, Recchia CA, Robertson J (2007) Marine Ecoregions of the World: A Bioregionalization of Coastal and Shelf Areas. Bioscience 57:573. doi: 10.1641/B570707

Spalding MD, Agostini VN, Rice J, Grant SM (2012) Pelagic provinces of the world: A biogeographic classification of the world's surface pelagic waters. Ocean Coast Manag 60:19–30. doi: 10.1016/j.ocecoaman.2011.12.016

Storfer A, Murphy MA, Evans JS, Goldberg CS, Robinson S, Spear SF, Dezzani R, Delmelle E, Vierling L, Waits LP (2007) Putting the "landscape" in landscape genetics. Heredity 98:128–142. doi:10.1038/sj.hdy.6800917

Tezanos-Pinto G, Baker CS, Russell K, Martien K, Baird RW, Hutt a., Stone G, Mignucci-Giannoni a. a., Caballero S, Endo T, Lavery S, Oremus M, Olavarria C, Garrigue C (2008) A Worldwide Perspective on the Population Structure and Genetic Diversity of Bottlenose Dolphins (*Tursiops truncatus*) in New Zealand. J Hered 100:11–24. doi: 10.1093/jhered/esn039

Thomas L, Kennington WJ, Stat M, Wilkinson SP, Kool JT, Kendrick GA (2015) Isolation by resistance across a complex coral reef seascape. Proc R Soc B Biol Sci 282:20151217. doi: 10.1098/rspb.2015.1217

Viricel A, Rosel PE (2014) Hierarchical population structure and habitat differences in a highly mobile marine species: The Atlantic spotted dolphin. Mol Ecol 23:5018–5035. doi: 10.1111/mec.12923

Wang C, Zöllner S, Rosenberg NA (2012) A Quantitative Comparison of the Similarity between Genes and Geography in Worldwide Human Populations. PLoS Genet 8:e1002886. doi: 10.1371/journal.pgen.1002886

Wang IJ, Bradburd GS (2014) Isolation by environment. Mol. Ecol. 23:5649–5662.

Wee AKS, Takayama K, Asakawa T, Thompson B, Onrizal, Sungkaew S, Tung NX, Nazre M, Soe KK, Tan HTW, Watano Y, Baba S, Kajita T, Webb EL (2014) Oceanic currents, not land masses, maintain the genetic structure of the mangrove *Rhizophora mucronata* Lam. (Rhizophoraceae) in Southeast Asia. J Biogeogr 41:954–964. doi: 10.1111/jbi.12263

Weir CR (2010) A review of cetacean occurrence in west African waters from the Gulf of Guinea to Angola. Mamm Rev 40:2–39. doi:10.1111/j.1365-2907.2009.00153.x

Wright S (1943) Isolation by Distance. Genetics 28:114–138

Figures

No constraint

300 m depth constraint

600 m depth constraint

900 m depth constraint
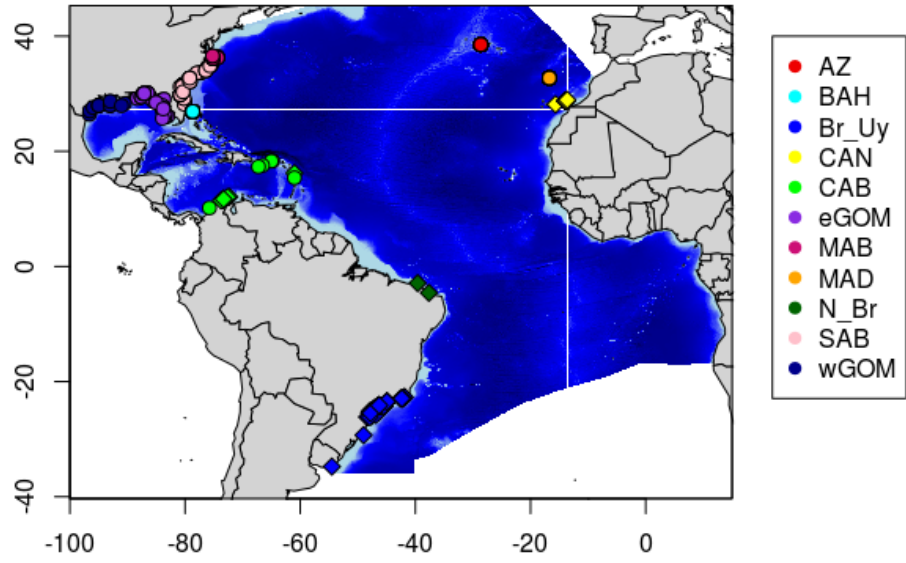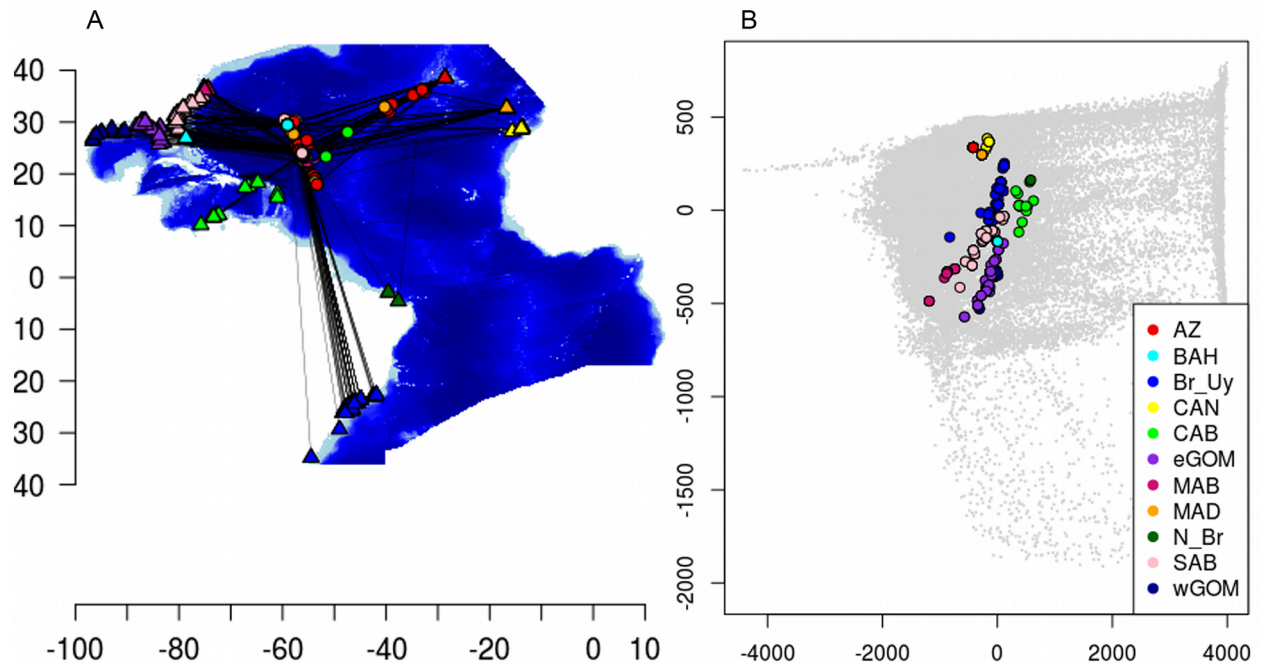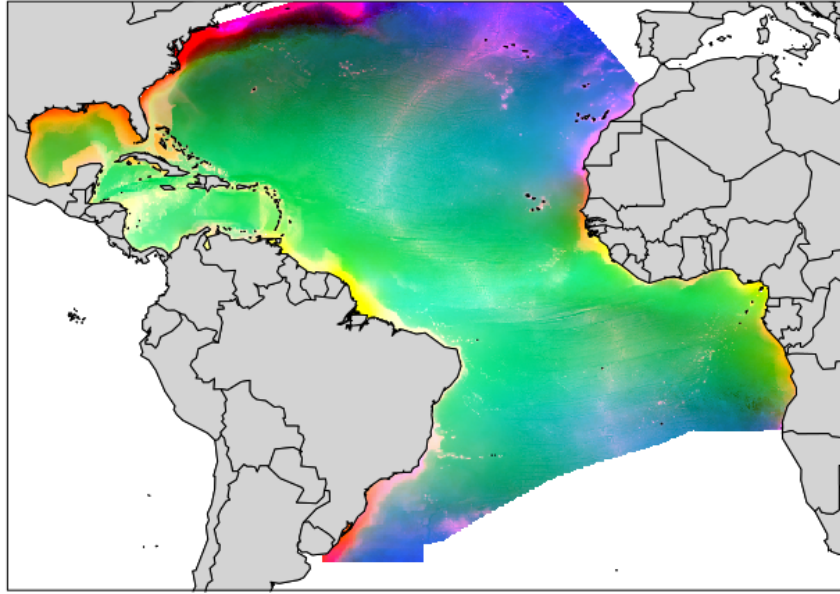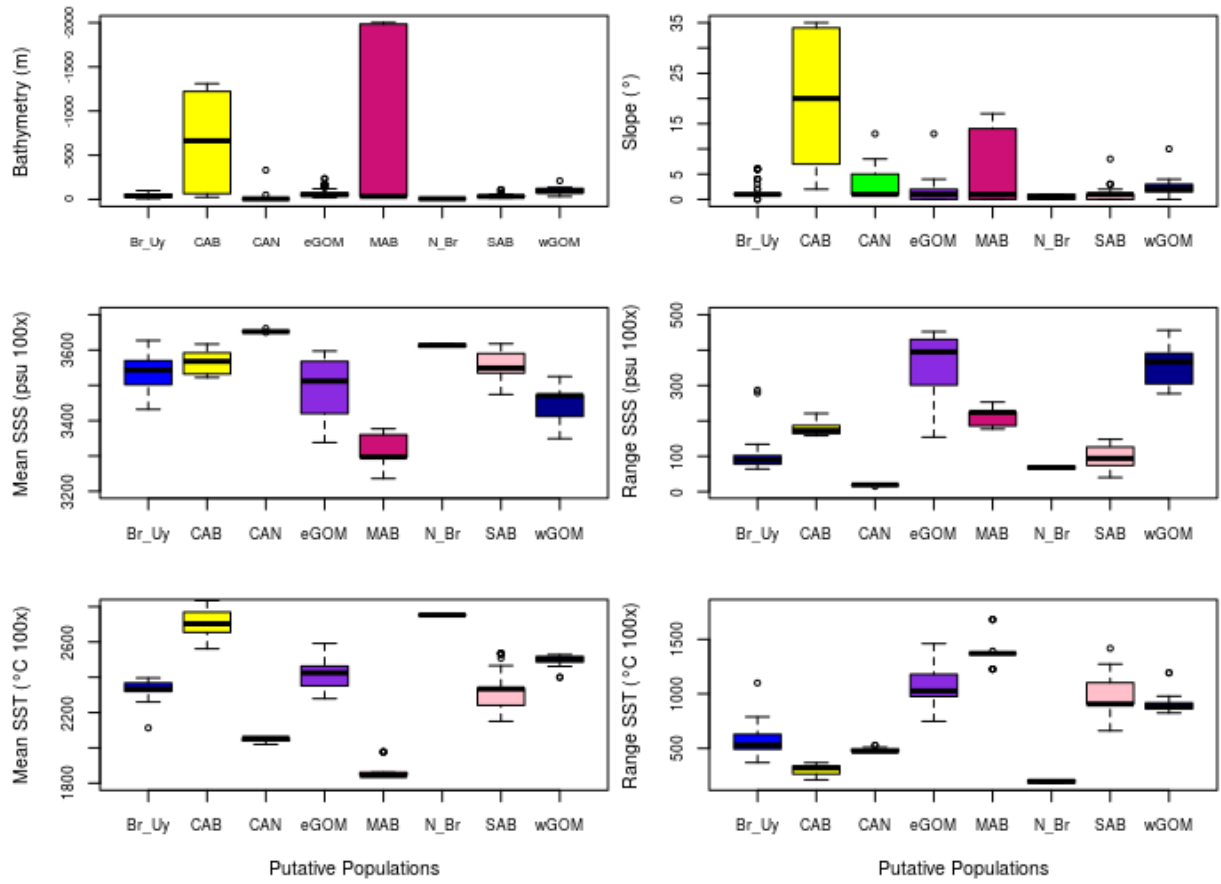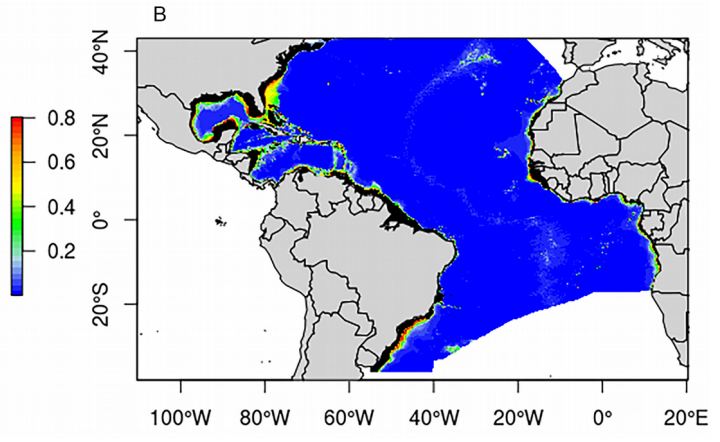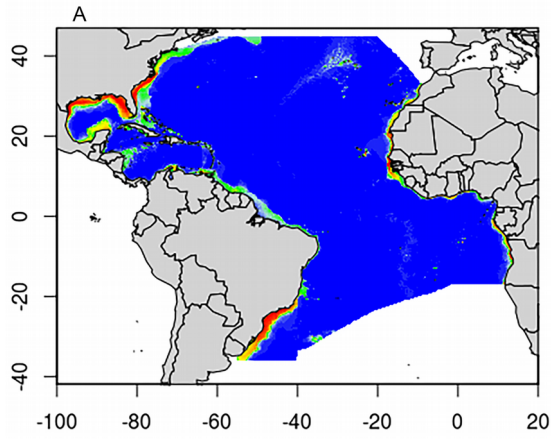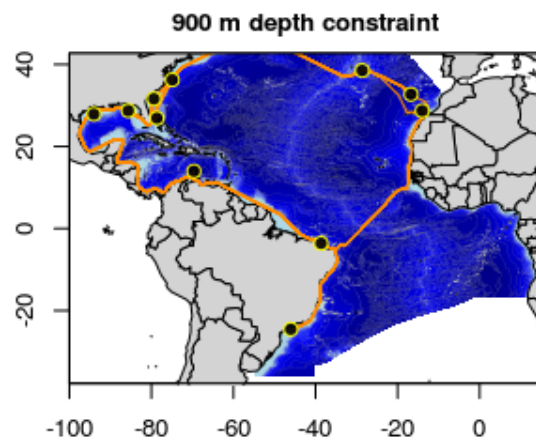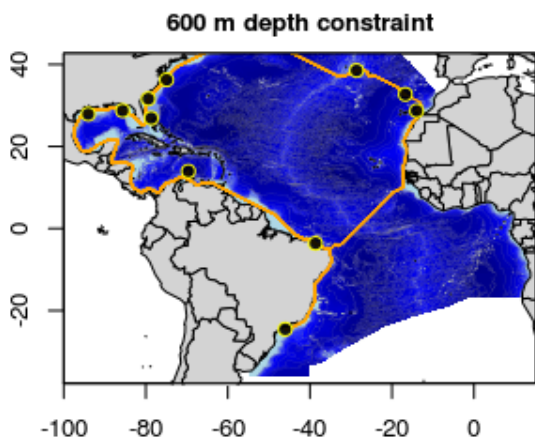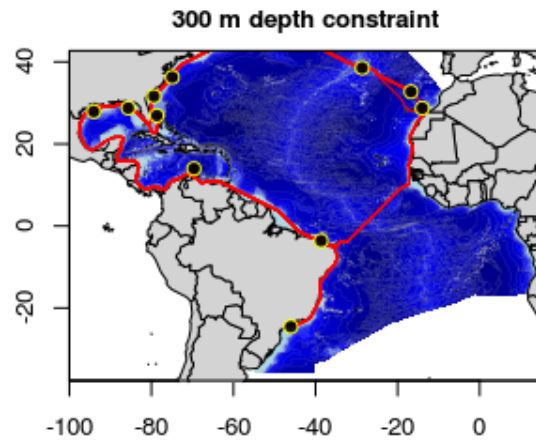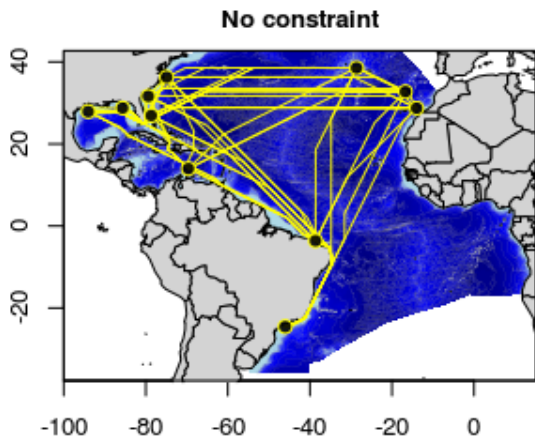
SUPPLEMENTARY TABLES

Table S1. Samples information and GenBank accession numbers. Abbreviations: AZ, Azores; BAH, Bahamas; Br_Uy, Brazil and Uruguay; CAB, Caribbean; CAN, Canary; eGOM, eastern Gulf of Mexico; MAB, Mid-Atlantic Bight; MAD, Madeira; N_Br, Northern Brazil; SAB, South Atlantic Bight; wGOM, western Gulf of Mexico

GenBank Accession Number	Lab_ID	Population	Haplotype	Longitude	Latitude	Reference
EF682734.1	AzMd_EF682734.1	AZ	hap61	-28.63	38.53	Quérouil et al. 2010
EF682794.1	AzMd_EF682794.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682776.1	AzMd_EF682776.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682709.1	AzMd_EF682709.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682692.1	AzMd_EF682692.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682663.1	AzMd_EF682663.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682763.1	AzMd_EF682763.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682685.1	AzMd_EF682685.1	AZ	hap5	-28.63	38.53	Quérouil et al. 2010
EF682659.1	AzMd_EF682659.1	AZ	hap53	-28.63	38.53	Quérouil et al. 2010
EF682658.1	AzMd_EF682658.1	AZ	hap57	-28.63	38.53	Quérouil et al. 2010
EF682661.1	AzMd_EF682661.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682748.1	AzMd_EF682748.1	AZ	hap57	-28.63	38.53	Quérouil et al. 2010
EF682655.1	AzMd_EF682655.1	AZ	hap38	-28.63	38.53	Quérouil et al. 2010
EF682676.1	AzMd_EF682676.1	AZ	hap33	-28.63	38.53	Quérouil et al. 2010
EF682778.1	AzMd_EF682778.1	AZ	hap5	-28.63	38.53	Quérouil et al. 2010
EF682710.1	AzMd_EF682710.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682772.1	AzMd_EF682772.1	AZ	hap37	-28.63	38.53	Quérouil et al. 2010
EF682652.1	AzMd_EF682652.1	AZ	hap69	-28.63	38.53	Quérouil et al. 2010
EF682693.1	AzMd_EF682693.1	AZ	hap70	-28.63	38.53	Quérouil et al. 2010
EF682727.1	AzMd_EF682727.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682690.1	AzMd_EF682690.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682760.1	AzMd_EF682760.1	AZ	hap34	-28.63	38.53	Quérouil et al. 2010
EF682696.1	AzMd_EF682696.1	AZ	hap5	-28.63	38.53	Quérouil et al. 2010
EF682691.1	AzMd_EF682691.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682781.1	AzMd_EF682781.1	AZ	hap38	-28.63	38.53	Quérouil et al. 2010
EF682736.1	AzMd_EF682736.1	AZ	hap9	-28.63	38.53	Quérouil et al. 2010
EF682664.1	AzMd_EF682664.1	AZ	hap30	-28.63	38.53	Quérouil et al. 2010
EF682775.1	AzMd_EF682775.1	AZ	hap43	-28.63	38.53	Quérouil et al. 2010
EF682756.1	AzMd_EF682756.1	AZ	hap70	-28.63	38.53	Quérouil et al. 2010
EF682657.1	AzMd_EF682657.1	AZ	hap32	-28.63	38.53	Quérouil et al. 2010
EF682746.1	AzMd_EF682746.1	AZ	hap5	-28.63	38.53	Quérouil et al. 2010
EF682679.1	AzMd_EF682679.1	AZ	hap38	-28.63	38.53	Quérouil et al. 2010
EF682750.1	AzMd_EF682750.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682666.1	AzMd_EF682666.1	AZ	hap98	-28.63	38.53	Quérouil et al. 2010
EF682712.1	AzMd_EF682712.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682671.1	AzMd_EF682671.1	AZ	hap71	-28.63	38.53	Quérouil et al. 2010
EF682682.1	AzMd_EF682682.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682785.1	AzMd_EF682785.1	AZ	hap84	-28.63	38.53	Quérouil et al. 2010
EF682688.1	AzMd_EF682688.1	AZ	hap94	-28.63	38.53	Quérouil et al. 2010
EF682744.1	AzMd_EF682744.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010

EF682650.1	AzMd_EF682650.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682786.1	AzMd_EF682786.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682788.1	AzMd_EF682788.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682714.1	AzMd_EF682714.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682769.1	AzMd_EF682769.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682678.1	AzMd_EF682678.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682701.1	AzMd_EF682701.1	AZ	hap103	-28.63	38.53	Quérouil et al. 2010
EF682684.1	AzMd_EF682684.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682677.1	AzMd_EF682677.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682721.1	AzMd_EF682721.1	AZ	hap53	-28.63	38.53	Quérouil et al. 2010
EF682705.1	AzMd_EF682705.1	AZ	hap53	-28.63	38.53	Quérouil et al. 2010
EF682779.1	AzMd_EF682779.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682791.1	AzMd_EF682791.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682755.1	AzMd_EF682755.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682743.1	AzMd_EF682743.1	AZ	hap99	-28.63	38.53	Quérouil et al. 2010
EF682767.1	AzMd_EF682767.1	AZ	hap86	-28.63	38.53	Quérouil et al. 2010
EF682752.1	AzMd_EF682752.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682675.1	AzMd_EF682675.1	AZ	hap101	-28.63	38.53	Quérouil et al. 2010
EF682761.1	AzMd_EF682761.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682718.1	AzMd_EF682718.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682790.1	AzMd_EF682790.1	AZ	hap90	-28.63	38.53	Quérouil et al. 2010
EF682764.1	AzMd_EF682764.1	AZ	hap63	-28.63	38.53	Quérouil et al. 2010
EF682681.1	AzMd_EF682681.1	AZ	hap60	-28.63	38.53	Quérouil et al. 2010
EF682667.1	AzMd_EF682667.1	AZ	hap64	-28.63	38.53	Quérouil et al. 2010
EF682753.1	AzMd_EF682753.1	AZ	hap62	-28.63	38.53	Quérouil et al. 2010
EF682698.1	AzMd_EF682698.1	AZ	hap53	-28.63	38.53	Quérouil et al. 2010
EF682789.1	AzMd_EF682789.1	AZ	hap53	-28.63	38.53	Quérouil et al. 2010
EF682716.1	AzMd_EF682716.1	AZ	hap59	-28.63	38.53	Quérouil et al. 2010
EF682715.1	AzMd_EF682715.1	AZ	hap58	-28.63	38.53	Quérouil et al. 2010
EF682656.1	AzMd_EF682656.1	AZ	hap61	-28.63	38.53	Quérouil et al. 2010
EF682703.1	AzMd_EF682703.1	AZ	hap57	-28.63	38.53	Quérouil et al. 2010
EF682777.1	AzMd_EF682777.1	AZ	hap57	-28.63	38.53	Quérouil et al. 2010
EF682699.1	AzMd_EF682699.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682792.1	AzMd_EF682792.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682749.1	AzMd_EF682749.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682751.1	AzMd_EF682751.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682737.1	AzMd_EF682737.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682765.1	AzMd_EF682765.1	AZ	hap46	-28.63	38.53	Quérouil et al. 2010
EF682766.1	AzMd_EF682766.1	AZ	hap50	-28.63	38.53	Quérouil et al. 2010
EF682713.1	AzMd_EF682713.1	AZ	hap29	-28.63	38.53	Quérouil et al. 2010
EF682702.1	AzMd_EF682702.1	AZ	hap24	-28.63	38.53	Quérouil et al. 2010
EF682680.1	AzMd_EF682680.1	AZ	hap50	-28.63	38.53	Quérouil et al. 2010
EF682771.1	AzMd_EF682771.1	AZ	hap50	-28.63	38.53	Quérouil et al. 2010
EF682662.1	AzMd_EF682662.1	AZ	hap50	-28.63	38.53	Quérouil et al. 2010
EF682686.1	AzMd_EF682686.1	AZ	hap49	-28.63	38.53	Quérouil et al. 2010
EF682774.1	AzMd_EF682774.1	AZ	hap49	-28.63	38.53	Quérouil et al. 2010
EF682722.1	AzMd_EF682722.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682668.1	AzMd_EF682668.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682787.1	AzMd_EF682787.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010

EF682759.1	AzMd_EF682759.1	AZ	hap73	-28.63	38.53	Quérouil et al. 2010
EF682742.1	AzMd_EF682742.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682700.1	AzMd_EF682700.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682793.1	AzMd_EF682793.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682683.1	AzMd_EF682683.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682730.1	AzMd_EF682730.1	AZ	hap54	-28.63	38.53	Quérouil et al. 2010
EF682770.1	AzMd_EF682770.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682726.1	AzMd_EF682726.1	AZ	hap11	-28.63	38.53	Quérouil et al. 2010
EF682729.1	AzMd_EF682729.1	AZ	hap46	-28.63	38.53	Quérouil et al. 2010
EF682733.1	AzMd_EF682733.1	AZ	hap11	-28.63	38.53	Quérouil et al. 2010
EF682773.1	AzMd_EF682773.1	AZ	hap16	-28.63	38.53	Quérouil et al. 2010
EF682762.1	AzMd_EF682762.1	AZ	hap12	-28.63	38.53	Quérouil et al. 2010
EF682747.1	AzMd_EF682747.1	AZ	hap70	-28.63	38.53	Quérouil et al. 2010
EF682711.1	AzMd_EF682711.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682670.1	AzMd_EF682670.1	AZ	hap6	-28.63	38.53	Quérouil et al. 2010
EF682707.1	AzMd_EF682707.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682654.1	AzMd_EF682654.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682739.1	AzMd_EF682739.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682673.1	AzMd_EF682673.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682689.1	AzMd_EF682689.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682784.1	AzMd_EF682784.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682783.1	AzMd_EF682783.1	AZ	hap18	-28.63	38.53	Quérouil et al. 2010
EF682725.1	AzMd_EF682725.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682738.1	AzMd_EF682738.1	AZ	hap17	-28.63	38.53	Quérouil et al. 2010
EF682694.1	AzMd_EF682694.1	AZ	hap35	-28.63	38.53	Quérouil et al. 2010
EF682708.1	AzMd_EF682708.1	AZ	hap70	-28.63	38.53	Quérouil et al. 2010
EF682754.1	AzMd_EF682754.1	AZ	hap16	-28.63	38.53	Quérouil et al. 2010
EF682757.1	AzMd_EF682757.1	AZ	hap7	-28.63	38.53	Quérouil et al. 2010
EF682758.1	AzMd_EF682758.1	AZ	hap81	-28.63	38.53	Quérouil et al. 2010
EF682740.1	AzMd_EF682740.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682728.1	AzMd_EF682728.1	AZ	hap6	-28.63	38.53	Quérouil et al. 2010
EF682745.1	AzMd_EF682745.1	AZ	hap68	-28.63	38.53	Quérouil et al. 2010
EF682768.1	AzMd_EF682768.1	AZ	hap68	-28.63	38.53	Quérouil et al. 2010
EF682660.1	AzMd_EF682660.1	AZ	hap7	-28.63	38.53	Quérouil et al. 2010
EF682697.1	AzMd_EF682697.1	AZ	hap2	-28.63	38.53	Quérouil et al. 2010
EF682653.1	AzMd_EF682653.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682717.1	AzMd_EF682717.1	AZ	hap2	-28.63	38.53	Quérouil et al. 2010
EF682704.1	AzMd_EF682704.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682687.1	AzMd_EF682687.1	AZ	hap26	-28.63	38.53	Quérouil et al. 2010
EF682732.1	AzMd_EF682732.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682672.1	AzMd_EF682672.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682741.1	AzMd_EF682741.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682665.1	AzMd_EF682665.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682674.1	AzMd_EF682674.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682719.1	AzMd_EF682719.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682706.1	AzMd_EF682706.1	AZ	hap76	-28.63	38.53	Quérouil et al. 2010
EF682651.1	AzMd_EF682651.1	AZ	hap88	-28.63	38.53	Quérouil et al. 2010
EF682695.1	AzMd_EF682695.1	AZ	hap88	-28.63	38.53	Quérouil et al. 2010
EF682735.1	AzMd_EF682735.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010

EF682731.1	AzMd_EF682731.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682782.1	AzMd_EF682782.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682669.1	AzMd_EF682669.1	AZ	hap85	-28.63	38.53	Quérouil et al. 2010
EF682724.1	AzMd_EF682724.1	AZ	hap20	-28.63	38.53	Quérouil et al. 2010
EF682780.1	AzMd_EF682780.1	AZ	hap89	-28.63	38.53	Quérouil et al. 2010
EF682720.1	AzMd_EF682720.1	AZ	hap82	-28.63	38.53	Quérouil et al. 2010
EF682723.1	AzMd_EF682723.1	AZ	hap82	-28.63	38.53	Quérouil et al. 2010
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008

DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060059.1	BAHAMAS_USA_DQ060059.1	BAH	hap41	-78.66	26.91	Green et al. 2007. Green 2008
DQ060057.1	BAHAMAS_USA_DQ060057.1	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
DQ060057.1	BAHAMAS_USA_DQ060057.1	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
DQ060057.1	BAHAMAS_USA_DQ060057.1	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF546440.2	Bahamas_USA_EF546440.2	BAH	hap5	-78.66	26.91	Green et al. 2007. Green 2008
EF971625.1	Bahamas_USA_FJ971625.1	BAH	hap20	-78.66	26.91	Green et al. 2007. Green 2008
EF971624.1	Bahamas_USA_FJ971624.1	BAH	hap51	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682752.1	BAHAMAS_USA_AzMd_EF682752.1	BAH	hap85	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008

EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
EF682726.1	BAHAMAS_USA_AzMd_EF682726.1	BAH	hap11	-78.66	26.91	Green et al. 2007. Green 2008
	Br_Uy_PA360	Br_Uy	hap71	-47.86	-25.14	This Study
	Br_Uy_SF28	Br_Uy	hap20	-46.26	-24.13	This Study
	Br_Uy_BC11	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_PA205	Br_Uy	hap20	-47.61	-26.03	This Study
	Br_Uy_SF09	Br_Uy	hap20	-46.26	-24.16	This Study
	Br_Uy_SF11	Br_Uy	hap74	-46.51	-24.23	This Study
	Br_Uy_GEMM219	Br_Uy	hap71	-41.9	-22.7	This Study
	Br_Uy_SF15	Br_Uy	hap71	-46.15	-24.33	This Study
	Br_Uy_PA198	Br_Uy	hap71	-47.66	-25.02	This Study
	Br_Uy_bc05	Br_Uy	hap71	-45.52	-24.24	This Study
	Br_Uy_MM42	Br_Uy	hap71	-49	-29.3	This Study
	Br_Uy_SF22	Br_Uy	hap71	-47.1	-24.68	This Study
	Br_Uy_bc16	Br_Uy	hap71	-45.52	-24.24	This Study
	Br_Uy_SF19	Br_Uy	hap71	-47.1	-24.68	This Study
	Br_Uy_SF16	Br_Uy	hap85	-46.15	-24.33	This Study
	Br_Uy_SFUY	Br_Uy	hap85	-54.5	-34.76	This Study
	Br_Uy_SF03	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_GEMM102	Br_Uy	hap72	-42.28	-22.93	This Study
	Br_Uy_GEMM59	Br_Uy	hap20	-42.56	-22.93	This Study
	Br_Uy_SF12	Br_Uy	hap20	-46.15	-24.33	This Study
	Br_Uy_bc03_frontalis	Br_Uy	hap85	-46.49	-25.36	This Study
	Br_Uy_PA165	Br_Uy	hap72	-48.31	-26.14	This Study
	Br_Uy_bc13	Br_Uy	hap10	-45.52	-24.24	This Study
	Br_Uy_bc04	Br_Uy	hap72	-46.49	-25.36	This Study
	Br_Uy_SF27	Br_Uy	hap20	-46.26	-24.13	This Study
	Br_Uy_SF01	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_PA164	Br_Uy	hap20	-48.31	-26.14	This Study
	Br_Uy_bc14	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_SF08	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_PA249	Br_Uy	hap20	-48.01	-25.37	This Study
	Br_Uy_SF30	Br_Uy	hap20	-46.26	-24.13	This Study
	Br_Uy_SF13	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_GEMM208	Br_Uy	hap20	-41.9	-22.7	This Study
	Br_Uy_SF20	Br_Uy	hap11	-47.1	-24.68	This Study
	Br_Uy_BC10	Br_Uy	hap45	-45.52	-24.24	This Study
	Br_Uy_bc02_frontalis_	Br_Uy	hap20	-46.86	-25.76	This Study
	Br_Uy_SF35	Br_Uy	hap20	-47.1	-24.68	This Study
	Br_Uy_bc08	Br_Uy	hap45	-45.52	-24.24	This Study
	Br_Uy_BC12	Br_Uy	hap45	-45.52	-24.24	This Study
	Br_Uy_bc06	Br_Uy	hap72	-45.52	-24.24	This Study
	Br_Uy_SF10	Br_Uy	hap20	-46.51	-24.23	This Study
	Br_Uy_SF02	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_SF04	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_SF05	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_PA199	Br_Uy	hap11	-47.66	-25.02	This Study
	Br_Uy_GEMM400_	Br_Uy	hap20	-42.21	-22.94	This Study
	Br_Uy_SF18	Br_Uy	hap20	-47.1	-24.68	This Study

	Br_Uy_GEMM305	Br_Uy	hap20	-42.42	-22.93	This Study
	Br_Uy_bc15	Br_Uy	hap45	-45.52	-24.24	This Study
	Br_Uy_SF14	Br_Uy	hap20	-46.15	-24.33	This Study
	Br_Uy_SF25	Br_Uy	hap20	-46.26	-24.13	This Study
	Br_Uy_PA209	Br_Uy	hap85	-47.9	-26.15	This Study
	Br_Uy_bc17	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_bc09	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_bc07	Br_Uy	hap20	-45.52	-24.24	This Study
	Br_Uy_SF32	Br_Uy	hap20	-46.26	-24.13	This Study
	Br_Uy_PA365	Br_Uy	hap20	-47.79	-25.48	This Study
	Br_Uy_SF07	Br_Uy	hap20	-44.95	-23.48	This Study
	Br_Uy_GEMM149	Br_Uy	hap20	-42.35	-22.93	This Study
	Br_Uy_SF26	Br_Uy	hap20	-46.26	-24.13	This Study
EF682654.1	NEPST366	CAB	hap76	-64.78	18.35	Caballero et al. 2013
KC204738.1	NEPST877	CAB	hap40	-65	18.3	Caballero et al. 2013
KC204737.1	GU01022801	CAB	hap27	-66.55	17.55	Caballero et al. 2013
KC204739.1	GU01030102	CAB	hap57	-67.25	17.33	Caballero et al. 2013
KC204740.1	STEN20010612	CAB	hap78	-61	15.4	Caballero et al. 2013
KC204736.1	SfronCCIR0103	CAB	hap65	-75.76	10.16	Caballero et al. 2013
	CAB_G40LG	CAB	hap5	-72.29	12.08	This Study
	CAB_G30LG	CAB	hap52	-73.28	11.78	This Study
	CAB_G28LG_	CAB	hap44	-73.36	11.55	This Study
	CAB_G24LG	CAB	hap5	-73.30	11.68	This Study
	CAB_G9LG	CAB	hap5	-73.75	11.48	This Study
	CAB_SFCARIBE	CAB	hap20	-61	16.2	This Study
	CAB_G38LG_	CAB	hap5	-72.61	12.04	This Study
	Cab_G10_Roosevelt	CAB	hap1	-73.30	11.69	This Study
	CI_2703	CAN	hap5	-13.86	28.74	This Study
	CI_0302	CAN	hap75	-13.83	28.70	This Study
	CI_2102	CAN	hap85	-13.86	28.74	This Study
	CI_0607	CAN	hap69	-13.74	28.89	This Study
	CI_230313	CAN	hap35	-15.66	28.15	This Study
	CI_1303	CAN	hap91	-13.86	28.74	This Study
	CI_2003	CAN	hap95	-13.86	28.74	This Study
	CI_0612	CAN	hap53	-13.95	28.73	This Study
	CI_2303	CAN	hap101	-13.86	28.74	This Study
	CI_2510	CAN	hap90	-13.95	28.73	This Study
	CI_0809	CAN	hap20	-13.86	28.74	This Study
	CI_0704	CAN	hap75	-13.63	28.92	This Study
	Sfro134	eGOM	hap85	-87.58	29.45	Viricel and Rosel 2014
	Sfro222	eGOM	hap85	-86.50	29.65	Viricel and Rosel 2014
	9673GOM	eGOM	hap85	-86.51	30.07	Viricel and Rosel 2014
	9793GOM	eGOM	hap56	-87.27	29.96	Viricel and Rosel 2014
	Sfro202	eGOM	hap11	-83.49	25.98	Viricel and Rosel 2014
	Sfro206	eGOM	hap11	-83.96	27.97	Viricel and Rosel 2014
	Sfro200	eGOM	hap11	-83.65	29.03	Viricel and Rosel 2014
	Sfro201	eGOM	hap11	-83.65	29.03	Viricel and Rosel 2014
	Sfro138	eGOM	hap11	-87.58	29.45	Viricel and Rosel 2014
	9672GOM	eGOM	hap11	-86.22	29.64	Viricel and Rosel 2014

Sfro223	eGOM	hap11	-86.50	29.65	Viricel and Rosel 2014
9786GOM	eGOM	hap11	-86.27	29.65	Viricel and Rosel 2014
9794GOM	eGOM	hap11	-87.27	29.96	Viricel and Rosel 2014
96102GOM	eGOM	hap11	-87.11	30.05	Viricel and Rosel 2014
Sfro217	eGOM	hap11	-86.50	30.11	Viricel and Rosel 2014
9778GOM	eGOM	hap20	-85.02	28.46	Viricel and Rosel 2014
9674GOM	eGOM	hap20	-86.51	30.07	Viricel and Rosel 2014
Sfro199	eGOM	hap5	-83.47	26.52	Viricel and Rosel 2014
Sfro219	eGOM	hap5	-85.48	28.75	Viricel and Rosel 2014
9768GOM	eGOM	hap5	-88.30	29.18	Viricel and Rosel 2014
Sfro135	eGOM	hap5	-87.58	29.45	Viricel and Rosel 2014
9787GOM	eGOM	hap5	-86.53	29.65	Viricel and Rosel 2014
9698GOM	eGOM	hap5	-87.11	30.05	Viricel and Rosel 2014
9675GOM	eGOM	hap5	-86.51	30.07	Viricel and Rosel 2014
9490GOM	eGOM	hap100	-83.77	27.37	Viricel and Rosel 2014
512-02	eGOM	hap100	-86.36	29.40	Viricel and Rosel 2014
Sfro214	eGOM	hap20	-83.86	27.55	Viricel and Rosel 2014
Sfro213	eGOM	hap4	-83.86	27.55	Viricel and Rosel 2014
9796GOM	eGOM	hap4	-87.27	29.96	Viricel and Rosel 2014
9695GOM	eGOM	hap4	-87.11	30.05	Viricel and Rosel 2014
9697GOM	eGOM	hap4	-87.11	30.05	Viricel and Rosel 2014
Sfro136	eGOM	hap14	-87.58	29.45	Viricel and Rosel 2014
Sfro137	eGOM	hap14	-87.58	29.45	Viricel and Rosel 2014
Sfro220	eGOM	hap55	-85.48	28.75	Viricel and Rosel 2014
9495GOM	eGOM	hap1	-82.99	26.09	Viricel and Rosel 2014
Sfro210	eGOM	hap1	-83.47	26.52	Viricel and Rosel 2014
9489GOM	eGOM	hap1	-83.77	27.37	Viricel and Rosel 2014
Sfro212	eGOM	hap1	-83.86	27.55	Viricel and Rosel 2014
Sfro215	eGOM	hap1	-83.86	27.55	Viricel and Rosel 2014
512-01	eGOM	hap1	-86.36	29.40	Viricel and Rosel 2014
9696GOM	eGOM	hap1	-87.11	30.05	Viricel and Rosel 2014
Sfro216	eGOM	hap1	-86.50	30.11	Viricel and Rosel 2014
Sfro218	eGOM	hap1	-86.50	30.11	Viricel and Rosel 2014
Sfro203	eGOM	hap8	-83.49	25.98	Viricel and Rosel 2014
Sfro204	eGOM	hap8	-83.49	25.98	Viricel and Rosel 2014
Sfro209	eGOM	hap8	-83.85	26.98	Viricel and Rosel 2014
Sfro207	eGOM	hap83	-83.96	27.97	Viricel and Rosel 2014
Sfro208	eGOM	hap22	-83.85	26.98	Viricel and Rosel 2014
Sfro211	eGOM	hap3	-83.47	26.52	Viricel and Rosel 2014
9494GOM	eGOM	hap41	-83.92	25.73	Viricel and Rosel 2014
Sfro205	eGOM	hap41	-83.86	27.55	Viricel and Rosel 2014
9779GOM	eGOM	hap41	-85.02	28.46	Viricel and Rosel 2014
Sfro133	eGOM	hap41	-87.58	29.45	Viricel and Rosel 2014
96100GOM	eGOM	hap41	-87.11	30.05	Viricel and Rosel 2014
9699GOM	eGOM	hap41	-87.11	30.05	Viricel and Rosel 2014
9491GOM	eGOM	hap79	-83.77	27.37	Viricel and Rosel 2014
9792GOM	eGOM	hap79	-87.27	29.96	Viricel and Rosel 2014
96101GOM	eGOM	hap79	-87.11	30.05	Viricel and Rosel 2014
96103GOM	eGOM	hap42	-87.11	30.05	Viricel and Rosel 2014

	99232ATL	MAB	hap85	-75.13	36.14	Viricel and Rosel 2014
	98080701	MAB	hap85	-75.01	36.14	Viricel and Rosel 2014
	99238ATL	MAB	hap21	-75.21	36.56	Viricel and Rosel 2014
	99237ATL	MAB	hap11	-75.13	36.14	Viricel and Rosel 2014
	99230ATL	MAB	hap92	-75.13	36.14	Viricel and Rosel 2014
	99231ATL	MAB	hap13	-75.13	36.14	Viricel and Rosel 2014
	99267ATL	MAB	hap38	-74.35	36.25	Viricel and Rosel 2014
	99268ATL	MAB	hap38	-74.35	36.25	Viricel and Rosel 2014
	99266ATL	MAB	hap38	-74.38	36.52	Viricel and Rosel 2014
	99269ATL	MAB	hap27	-74.35	36.25	Viricel and Rosel 2014
	99271ATL	MAB	hap77	-74.35	36.25	Viricel and Rosel 2014
	99235ATL	MAB	hap5	-75.13	36.14	Viricel and Rosel 2014
	99236ATL	MAB	hap5	-75.13	36.14	Viricel and Rosel 2014
	99234ATL	MAB	hap4	-75.13	36.14	Viricel and Rosel 2014
	99233ATL	MAB	hap1	-75.13	36.14	Viricel and Rosel 2014
	99239ATL	MAB	hap1	-75.21	36.56	Viricel and Rosel 2014
	99240ATL	MAB	hap1	-75.21	36.56	Viricel and Rosel 2014
EF682817.1	AzMd_EF682817.1	MAD	hap34	-16.73	32.73	Quérouil et al. 2010
EF682824.1	AzMd_EF682824.1	MAD	hap34	-16.73	32.73	Quérouil et al. 2010
EF682832.1	AzMd_EF682832.1	MAD	hap48	-16.73	32.73	Quérouil et al. 2010
EF682829.1	AzMd_EF682829.1	MAD	hap61	-16.73	32.73	Quérouil et al. 2010
EF682835.1	AzMd_EF682835.1	MAD	hap23	-16.73	32.73	Quérouil et al. 2010
EF682828.1	AzMd_EF682828.1	MAD	hap5	-16.73	32.73	Quérouil et al. 2010
EF682834.1	AzMd_EF682834.1	MAD	hap15	-16.73	32.73	Quérouil et al. 2010
EF682799.1	AzMd_EF682799.1	MAD	hap31	-16.73	32.73	Quérouil et al. 2010
EF682838.1	AzMd_EF682838.1	MAD	hap31	-16.73	32.73	Quérouil et al. 2010
EF682796.1	AzMd_EF682796.1	MAD	hap33	-16.73	32.73	Quérouil et al. 2010
EF682801.1	AzMd_EF682801.1	MAD	hap38	-16.73	32.73	Quérouil et al. 2010
EF682820.1	AzMd_EF682820.1	MAD	hap39	-16.73	32.73	Quérouil et al. 2010
EF682798.1	AzMd_EF682798.1	MAD	hap71	-16.73	32.73	Quérouil et al. 2010
EF682810.1	AzMd_EF682810.1	MAD	hap66	-16.73	32.73	Quérouil et al. 2010
EF682807.1	AzMd_EF682807.1	MAD	hap90	-16.73	32.73	Quérouil et al. 2010
EF682800.1	AzMd_EF682800.1	MAD	hap90	-16.73	32.73	Quérouil et al. 2010
EF682811.1	AzMd_EF682811.1	MAD	hap66	-16.73	32.73	Quérouil et al. 2010
EF682808.1	AzMd_EF682808.1	MAD	hap97	-16.73	32.73	Quérouil et al. 2010
EF682827.1	AzMd_EF682827.1	MAD	hap95	-16.73	32.73	Quérouil et al. 2010
EF682815.1	AzMd_EF682815.1	MAD	hap85	-16.73	32.73	Quérouil et al. 2010
EF682812.1	AzMd_EF682812.1	MAD	hap53	-16.73	32.73	Quérouil et al. 2010
EF682805.1	AzMd_EF682805.1	MAD	hap85	-16.73	32.73	Quérouil et al. 2010
EF682804.1	AzMd_EF682804.1	MAD	hap53	-16.73	32.73	Quérouil et al. 2010
EF682825.1	AzMd_EF682825.1	MAD	hap53	-16.73	32.73	Quérouil et al. 2010
EF682795.1	AzMd_EF682795.1	MAD	hap50	-16.73	32.73	Quérouil et al. 2010
EF682826.1	AzMd_EF682826.1	MAD	hap46	-16.73	32.73	Quérouil et al. 2010
EF682831.1	AzMd_EF682831.1	MAD	hap47	-16.73	32.73	Quérouil et al. 2010
EF682816.1	AzMd_EF682816.1	MAD	hap49	-16.73	32.73	Quérouil et al. 2010
EF682803.1	AzMd_EF682803.1	MAD	hap20	-16.73	32.73	Quérouil et al. 2010
EF682809.1	AzMd_EF682809.1	MAD	hap46	-16.73	32.73	Quérouil et al. 2010
EF682806.1	AzMd_EF682806.1	MAD	hap20	-16.73	32.73	Quérouil et al. 2010
EF682833.1	AzMd_EF682833.1	MAD	hap19	-16.73	32.73	Quérouil et al. 2010

EF682836.1	AzMd_EF682836.1	MAD	hap2	-16.73	32.73	Quérouil et al. 2010
EF682839.1	AzMd_EF682839.1	MAD	hap76	-16.73	32.73	Quérouil et al. 2010
EF682802.1	AzMd_EF682802.1	MAD	hap25	-16.73	32.73	Quérouil et al. 2010
EF682814.1	AzMd_EF682814.1	MAD	hap6	-16.73	32.73	Quérouil et al. 2010
EF682818.1	AzMd_EF682818.1	MAD	hap81	-16.73	32.73	Quérouil et al. 2010
EF682830.1	AzMd_EF682830.1	MAD	hap15	-16.73	32.73	Quérouil et al. 2010
EF682821.1	AzMd_EF682821.1	MAD	hap76	-16.73	32.73	Quérouil et al. 2010
EF682813.1	AzMd_EF682813.1	MAD	hap20	-16.73	32.73	Quérouil et al. 2010
EF682822.1	AzMd_EF682822.1	MAD	hap50	-16.73	32.73	Quérouil et al. 2010
EF682819.1	AzMd_EF682819.1	MAD	hap20	-16.73	32.73	Quérouil et al. 2010
EF682840.1	AzMd_EF682840.1	MAD	hap26	-16.73	32.73	Quérouil et al. 2010
EF682797.1	AzMd_EF682797.1	MAD	hap87	-16.73	32.73	Quérouil et al. 2010
EF682823.1	AzMd_EF682823.1	MAD	hap76	-16.73	32.73	Quérouil et al. 2010
EF682837.1	AzMd_EF682837.1	MAD	hap80	-16.73	32.73	Quérouil et al. 2010
	Br_Uy_aq286_ceara	N_Br	hap53	-37.64	-45.8	This Study
	Br_Uy_aq78_ceara	N_Br	hap45	-39.6	-2.9	This Study
	9991ATL	SAB	hap85	-80.1	28.62	Viricel and Rosel 2014
	9993ATL	SAB	hap85	-80.1	28.62	Viricel and Rosel 2014
	9994ATL	SAB	hap85	-80.1	28.62	Viricel and Rosel 2014
	99100ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	99103ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	99105ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	99106ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	99108ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	9998ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	9999ATL	SAB	hap85	-80.81	29.84	Viricel and Rosel 2014
	99110ATL	SAB	hap85	-80.36	30.32	Viricel and Rosel 2014
	99114ATL	SAB	hap85	-80.36	30.32	Viricel and Rosel 2014
	99115ATL	SAB	hap85	-80.36	30.32	Viricel and Rosel 2014
	99314ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99315ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99316ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99317ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99318ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99319ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99325ATL	SAB	hap85	-80.29	31.09	Viricel and Rosel 2014
	99126ATL	SAB	hap85	-80.48	31.15	Viricel and Rosel 2014
	99128ATL	SAB	hap85	-80.48	31.15	Viricel and Rosel 2014
	99130ATL	SAB	hap85	-80.48	31.15	Viricel and Rosel 2014
	99134ATL	SAB	hap85	-80.48	31.15	Viricel and Rosel 2014
	99145ATL	SAB	hap85	-80.22	31.66	Viricel and Rosel 2014
	99146ATL	SAB	hap85	-80.22	31.66	Viricel and Rosel 2014
	99148ATL	SAB	hap85	-80.22	31.66	Viricel and Rosel 2014
	99150ATL	SAB	hap85	-80.22	31.66	Viricel and Rosel 2014
	99158ATL	SAB	hap85	-79.18	32.7	Viricel and Rosel 2014
	99159ATL	SAB	hap85	-79.18	32.7	Viricel and Rosel 2014
	99162ATL	SAB	hap85	-79.18	32.7	Viricel and Rosel 2014
	99174ATL	SAB	hap85	-77.4	33.6	Viricel and Rosel 2014
	99175ATL	SAB	hap85	-77.4	33.6	Viricel and Rosel 2014

99176ATL	SAB	hap85	-77.4	33.6	Viricel and Rosel 2014
99205ATL	SAB	hap85	-75.97	34.63	Viricel and Rosel 2014
99206AT	SAB	hap85	-75.97	34.63	Viricel and Rosel 2014
99207ATL	SAB	hap85	-75.97	34.63	Viricel and Rosel 2014
9992ATL	SAB	hap96	-80.1	28.62	Viricel and Rosel 2014
99104ATL	SAB	hap28	-80.81	29.84	Viricel and Rosel 2014
99109ATL	SAB	hap92	-80.81	29.84	Viricel and Rosel 2014
99313ATL	SAB	hap92	-80.29	31.09	Viricel and Rosel 2014
99177ATL	SAB	hap92	-77.93	33.59	Viricel and Rosel 2014
99178ATL	SAB	hap92	-77.93	33.59	Viricel and Rosel 2014
99306ATL	SAB	hap92	-76.39	34.03	Viricel and Rosel 2014
99309ATL	SAB	hap92	-76.39	34.03	Viricel and Rosel 2014
99208ATL	SAB	hap92	-75.97	34.63	Viricel and Rosel 2014
SD604-4	SAB	hap92	-75.35	35.06	Viricel and Rosel 2014
98072001	SAB	hap20	-80.15	28.19	Viricel and Rosel 2014
98071905	SAB	hap20	-80.18	29.17	Viricel and Rosel 2014
99149ATL	SAB	hap20	-80.22	31.66	Viricel and Rosel 2014
99213ATL	SAB	hap20	-75.66	34.84	Viricel and Rosel 2014
99125ATL	SAB	hap85	-80.48	31.15	Viricel and Rosel 2014
99132ATL	SAB	hap36	-80.48	31.15	Viricel and Rosel 2014
99156ATL	SAB	hap36	-79.18	32.7	Viricel and Rosel 2014
99160ATL	SAB	hap36	-79.18	32.7	Viricel and Rosel 2014
99161ATL	SAB	hap36	-79.18	32.7	Viricel and Rosel 2014
99307ATL	SAB	hap36	-76.39	34.03	Viricel and Rosel 2014
99139ATL	SAB	hap102	-80.22	31.66	Viricel and Rosel 2014
99210ATL	SAB	hap85	-75.66	34.84	Viricel and Rosel 2014
99320ATL	SAB	hap93	-80.29	31.09	Viricel and Rosel 2014
99326ATL	SAB	hap93	-80.29	31.09	Viricel and Rosel 2014
99327ATL	SAB	hap93	-80.29	31.09	Viricel and Rosel 2014
99328ATL	SAB	hap93	-80.29	31.09	Viricel and Rosel 2014
99330ATL	SAB	hap93	-80.29	31.09	Viricel and Rosel 2014
98071906	SAB	hap5	-80.18	29.17	Viricel and Rosel 2014
99116ATL	SAB	hap5	-80.36	30.32	Viricel and Rosel 2014
99211ATL	SAB	hap5	-75.66	34.84	Viricel and Rosel 2014
99101ATL	SAB	hap1	-80.81	29.84	Viricel and Rosel 2014
99102ATL	SAB	hap1	-80.81	29.84	Viricel and Rosel 2014
99107ATL	SAB	hap1	-80.81	29.84	Viricel and Rosel 2014
98072301	SAB	hap1	-80.86	31.23	Viricel and Rosel 2014
99129ATL	SAB	hap1	-80.48	31.15	Viricel and Rosel 2014
99131ATL	SAB	hap1	-80.48	31.15	Viricel and Rosel 2014
99133ATL	SAB	hap1	-80.48	31.15	Viricel and Rosel 2014
99310ATL	SAB	hap1	-79.12	32.08	Viricel and Rosel 2014
99312ATL	SAB	hap1	-79.12	32.08	Viricel and Rosel 2014
99157ATL	SAB	hap1	-79.18	32.7	Viricel and Rosel 2014
99308ATL	SAB	hap1	-76.39	34.03	Viricel and Rosel 2014
99113ATL	SAB	hap41	-80.36	30.32	Viricel and Rosel 2014
99311ATL	SAB	hap41	-79.12	32.08	Viricel and Rosel 2014
99155ATL	SAB	hap41	-79.18	32.7	Viricel and Rosel 2014
99212ATL	SAB	hap41	-75.66	34.84	Viricel and Rosel 2014

Sfro106	wGOM	hap21	-96.61	26.41	Viricel and Rosel 2014
Sfro097	wGOM	hap21	-96.54	27	Viricel and Rosel 2014
Sfro100	wGOM	hap21	-94.99	27.95	Viricel and Rosel 2014
9972GOM	wGOM	hap21	-92.99	27.96	Viricel and Rosel 2014
9973GOM	wGOM	hap21	-92.99	27.96	Viricel and Rosel 2014
Sfro104	wGOM	hap21	-92.97	28.68	Viricel and Rosel 2014
Sfro107	wGOM	hap76	-90.5	28.31	Viricel and Rosel 2014
98091401	wGOM	hap67	-95.94	27.6	Viricel and Rosel 2014
Sfro099	wGOM	hap67	-94.99	27.95	Viricel and Rosel 2014
Sfro103	wGOM	hap67	-94.99	27.95	Viricel and Rosel 2014
Sfro101	wGOM	hap4	-94.99	27.95	Viricel and Rosel 2014
Sfro102	wGOM	hap4	-94.99	27.95	Viricel and Rosel 2014
9974GOM	wGOM	hap4	-92.99	27.96	Viricel and Rosel 2014
Sfro105	wGOM	hap51	-92.97	28.68	Viricel and Rosel 2014
9969GOM	wGOM	hap1	-91.03	27.92	Viricel and Rosel 2014

References

Caballero S, Santos MCO, Sanches A, Mignucci-Giannoni AA (2013) Initial description of the phylogeography, population structure and genetic diversity of Atlantic spotted dolphins from Brazil and the Caribbean, inferred from analyses of mitochondrial and nuclear DNA. Biochem Syst Ecol 48:263–270. doi: 10.1016/j.bse.2012.12.016

Green ML (2008) Assessment of genetic population structure, promiscuity, and paternity in free-ranging Atlantic spotted dolphins *Stenella frontalis*, in the Bahamas. Florida Atlantic University

Green ML, Herzing DL, Baldwin JD (2007) Noninvasive methodology for the sampling and extraction of DNA from free-ranging Atlantic spotted dolphins (*Stenella frontalis*). Mol Ecol Notes 7:1287–1292. doi: 10.1111/j.1471-8286.2007.01858.x

Quérouil S, Freitas L, Cascão I, Alves F, Dinis A, Almeida JR, Prieto R, Borràs S, Matos JA, Mendonça D, Santos RS (2010) Molecular insight into the population structure of common and spotted dolphins inhabiting the pelagic waters of the Northeast Atlantic. Mar Biol 157:2567–2580. doi: 10.1007/s00227-010-1519-0

Viricel A, Rosel PE (2014) Hierarchical population structure and habitat differences in a highly mobile marine species: The Atlantic spotted dolphin. Mol Ecol 23:5018–5035. doi: 10.1111/mec.12923

Table S2. Kruskall-Wallis test among putative populations in relation to environmental variables. Significant values were typed in bold. Abbreviations: SSS, Sea Surface Salinity; SST, Sea Surface Temperature.

Kruskal-Wallis	X^2	P-value
Batrhymetry	72.654	0.000000000000429
Slope	73.935	0.0000000000002359
Mean Annual SSS	121.75	0.00000000000000022
Annual Range in SSS	205.25	0.00000000000000022
Mean Annual SST	166.52	0.00000000000000022
Annual Range in SST	198.79	0.00000000000000022

Table S3. Dunn test between putative populations in relation to environmental variables. P-values are in the below diagonal and significant values were typed in bold in the upper diagonal. Abbreviations: SSS, Sea Surface Salinity; SST, Sea Surface Temperature.

		Br_Uy	CAN	CAB	eGOM	MAB	N_Br	SAB	wGOM
Bathymetry	Br_Uy	-	**-3.43**	**3.82**	**3.06**	0.97	-2.06	-1.32	**3.67**
	Canary	0.008	-	**5.64**	**5.2**	**3.58**	-0.52	2.78	**5.54**
	Caribbean	0.002	0.0000	-	-1.93	-1.07	**-3.46**	**-4.7**	-0.2
	eGOM	0.03	0.0000	0.75	-	-1.07	-2.84	**-4.6**	1.73
	MAB	1	0.0047	0.22	1	-	-2.33	-1.84	2.24
	N_br	0.55	1	0.008	0.06	0.27	-	1.75	**3.37**
	SAB	1	0.0756	0.0000	0.0001	0.91	1	-	**4.58**
	wGOM	0.0033	0.0000	1	1	0.35	0.01	0.0001	-
Slope	Br_Uy	-	-1.01	**-4.6**	1.55	1.48	1.33	**4.49**	-1.46
	Canary	1	-	-2.66	1.91	1.92	1.67	**3.51**	-0.26
	Caribbean	0.0001	0.11	-	**5.55**	**4.91**	**3.07**	**7.36**	2.54
	eGOM	1	0.78	0.0000	-	0.44	0.93	**2.8**	-2.45
	MAB	1	0.76	0.0000	1	-	0.73	1.34	-2.34
	N_br	1	1	0.03	1	1	-	0.27	-1.83
	SAB	0.0001	0.006	0.0000	0.0001	1	1	-	**4.22**
	wGOM	1	1	0.2	0.2	0.27	0.94	0.0003	-
Mean Annual SSS	Br_Uy	-	**-5.08**	-1.61	2.16	**5.97**	-1.92	-2.02	**3.96**
	Canary	0.0000	-	2.87	**6.32**	**8.61**	0.29	**4.08**	**7.1**
	Caribbean	1	0.06	-	**2.94**	**5.87**	-1.2	0.46	**4.36**
	eGOM	0.43	0.0000	0.04	-	**4.51**	-2.48	**-4.34**	2.58
	MAB	0.0000	0.0000	0.0000	0.0001	-	-4.04	-7.44	-1.4
	N_br	0.76	1	1	0.18	0.0007	-	1.45	**3.36**
	SAB	0.6	0.001	1	0.0002	0.0000	1	-	**5.29**
	wGOM	0.001	0.0000	0.0002	0.14	1	0.01	0.0000	-
Annual Range in SSS	Br_Uy	-	**3.28**	**-3.71**	**-9.91**	**-4.72**	0.99	-0.15	**-6.27**
	Canary	0.01	-	**-5.44**	**-9.01**	**-6.19**	-0.43	**-3.44**	**-7.34**
	Caribbean	0.003	0.0000	-	-2.4	-0.54	2.4	**3.73**	-1.9
	eGOM	0.0000	0.0000	0.23	-	1.88	**3.51**	**10.5**	0.03
	MAB	0.0000	0.0000	1	0.83	-	2.68	**4.77**	-1.44
	N_br	1	1	1	0.01	0.1	-	-1.02	**-3.34**
	SAB	1	0.01	0.003	0.0000	0.0000	1	-	**-6.35**
	wGOM	0.0000	0.0000	0.81	1	1	0.01	0.0000	-
Mean Annual SST	Br_Uy	-	**3.83**	**-6.13**	**-4.75**	**5.11**	-2.55	0.71	**-4.63**
	Canary	0.02	-	**-7.71**	**-6.57**	0.51	**-3.99**	**-3.53**	**-6.58**
	Caribbean	0.0000	0.0000	-	**3.2**	**8.93**	-0.02	**6.71**	1.3
	eGOM	0.0000	0.0000	0.02	-	**8.26**	-1.34	**5.80**	-1.61
	MAB	0.0000	1	0.0000	0.0000	-	**-4.34**	**-4.82**	**-7.74**
	N_br	0.15	0.001	1	1	0.0002	-	2.73	0.66
	SAB	1	0.006	0.0000	0.0000	0.0000	0.09	-	**-5.2**
	wGOM	0.0001	0.0000	1	1	0.0000	1	0.0000	-
Annual Range in SST	Br_Uy	-	1.04	2.24	**-9.24**	**-9.18**	1.07	**-7.4**	**-3.65**
	Canary	1	-	0.85	**-6.389**	**-7.57**	0.58	**-5.13**	**-3.57**
	Caribbean	0.35	1	-	**-7.93**	**-8.83**	0.14	**-6.64**	**-4.62**
	eGOM	0.0000	0.0000	0.0000	-	**-3.02**	3.43	2.56	2.21
	MAB	0.0000	0.0000	0.0000	0.0358	-	**4.41**	**4.75**	**4.15**

N_br	1	1	1	0.009	0.0001	-	-2.83	-2.42
SAB	0.0000	0.0000	0.0000	0.15	0.0000	0.06	-	0.72
wGOM	0.004	0.005	0.0001	0.38	0.0005	0.21	1	-

SUPPLEMENTARY FIGURES
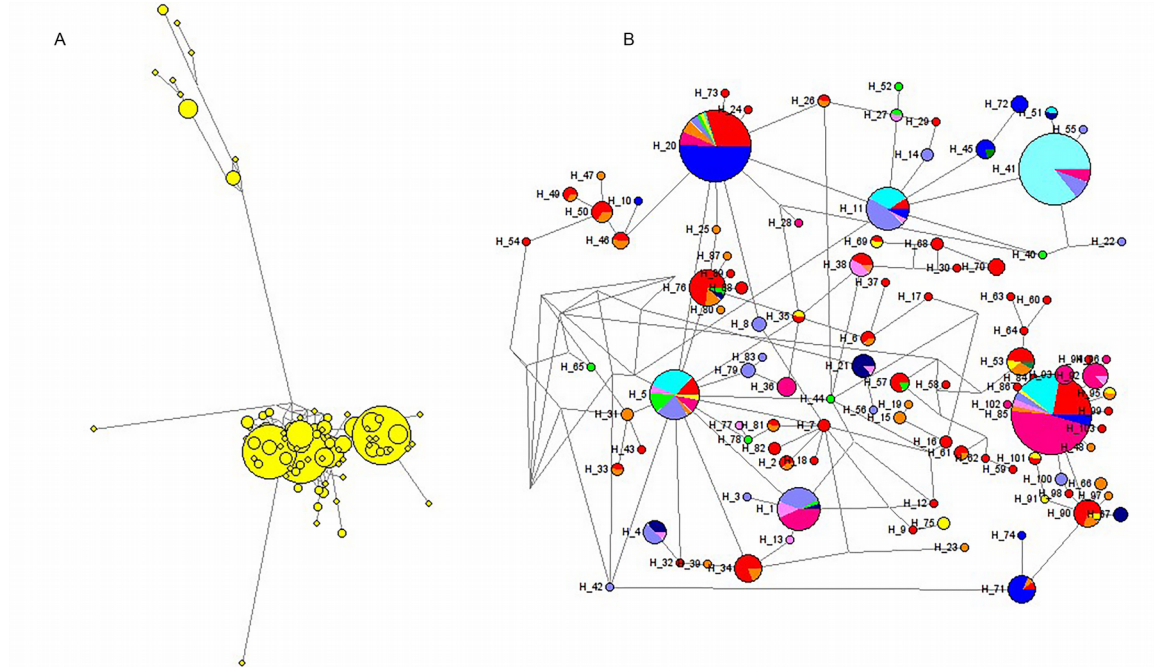
Fig. S1 Median-joining network of mtDNA control region haplotypes of Atlantic spotted dolphin. a) Circle size are proportional to the number of individuals exhibiting the corresponding haplotype and length of lines is proportional to the number of mutational steps separating haplotypes. b) Each haplotype is coloured according to the legend of Fig. 1 of main text

Fig. S2 Phylogenetic tree of mtDNA control region haplotypes of Atlantic spotted dolphin. Numbers above the branches indicate posterior probabilities

CAPÍTULO III

Population genomics of the Atlantic spotted dolphins (*Stenella frontalis*) across the Atlantic Ocean

Karina Bohrer do Amaral[1,2,][*], Dalia C. Barragán-Barrera[3,4], Nohelia Farías-Curtidor[3], Susana Caballero[4], Paula Mendéz-Fernandez [5], Marcos C. Oliveira Santos [6], Salvatore Siciliano [7], Vidal Martín [8], Manuel Carrillo [9], Ana Carolina de Meirelles[10], Valentina Franco-Trecu[11], Nelson Fagundes [2, 12], Ignacio Benites Moreno[1, 2, 13], Lacey Knowles[14], Ana Rita Amaral[15, 16]

[1]Laboratório de Sistemática e Ecologia de Aves e Mamíferos Marinhos (LABSMAR), Universidade Federal do Rio Grande do Sul, RS, Brazil.

[2] Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil.

[3] Fundación Macuáticos Colombia, Colombia, Medellín, Colombia.

[4] Laboratorio de Ecología Molecular de Vertebrados Acuáticos (LEMVA), Department of Biological Sciences, Universidad de los Andes, Bogotá, Colombia.

[5] Ingénieur de Recherche, Observatoire Pelagis, Université de La Rochelle, La Rochelle, France.

[6] Laboratório de Biologia da Conservação de Mamíferos Aquáticos (LABCMA), Departamento de Oceanografia Biológica, Instituto Oceanográfico, Universidade de São Paulo, São Paulo, SP, Brazil.

[7] Instituto Oswaldo Cruz/FIOCRUZ, Rio de Janeiro, RJ, Brazil.

[8] Sociedad para el Estudio de Cetáceos del Archipélago Canario (SECAC), Arrecife, Lanzarote, Spain.

[9] Tenerife Conservación, La Laguna, Tenerife, Canary Islands, Spain.

[10] AQUASIS – Associação de Pesquisa e Preservação de Ecossistemas Aquáticos, Caucaia, CE, Brazil.

[11] Laboratorio de Ecología Fisiológica y del Comportamiento, Facultad de Ciencias, Universidad de la República, Montevideo, Uruguay.

[12] Laboratório de Genética Médica e Evolução, Departamento de Genética, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil.

[13] Centro de Estudos Costeiros, Limnológicos e Marinhos (CECLIMAR), Campus Litoral Norte, Universidade Federal do Rio Grande do Sul, Imbé, RS, Brazil.

[14] Museum of Zoology, Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI, United States of America.

[15] Centre for Ecology, Evolution and Environmental Changes, Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal.

[16] Sackler Institute for Comparative Genomics, American Museum of Natural History, New York, United States of America.

Corresponding author:

* Karina Bohrer do Amaral, Laboratório de Sistemática e Ecologia de Aves e Mamíferos Marinhos (LABSMAR), Universidade Federal do Rio Grande do Sul, RS, Brazil and Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil. E-mail: karinabohrerdoamaral@gmail.com

Abstract

The Atlantic spotted dolphin (*Stenella frontalis*) is endemic to the Atlantic Ocean. It was hypothesized that the species has a complex geographical distribution, with a population structure suggestive of differing environmental requirements. We generated genomic data from individuals from the Southwestern Atlantic Ocean, Caribbean and Eastern Atlantic to test for patterns of genetic differentiation and evaluate specifically the genetic distinctiveness of Southwestern Atlantic Ocean populations. Our clustering results and population differentiation analyses suggests there may be at least three regional groups (a southeastern Brazil, Colombian Caribbean, and an oceanic group). However, with limited genetic differentiation, more samples are needed to confidently delineate regional population boundaries. Our results also revealed that genetic similarity is primarily correlated with geography, but that environment also has an effect on genetic structure. Lastly, our results support the hypothesized relative isolation of individuals in the southeastern Brazil. We detected a fine-scale population structure among southeastern Brazilian individuals. This substructure is comprised of a group that inhabits the inner and midshelf of the Southeast Brazilian Bight (SBB), which is geographically restricted to Brazil, and another group that occupies the outer shelf; members of this latter group have a similar genetic makeup to individuals from geographically distant locations, including the Canary Islands and Colombian Caribbean individuals. Our results are extremely important for informing the management and conservation of this species, especially in the Southeastern Brazil, individuals associated with inner and midshelf have a restricted geographic distribution that is suggestive of a demographically distinct population.

Key-words: Delphininae, genome-wide markers, SNPs, population genomics, DDRad-Seq

Running title: Population Genomics of Atlantic Spotted Dolphins

Introduction

The Atlantic spotted dolphin (*Stenella frontalis*) is a Delphinidae dolphin endemic to tropical, subtropical and warm temperate waters of the Atlantic Ocean (Perrin, 2009). The species ranges from Azores to at least Gabon in the Eastern Atlantic, and ranges from 45°N to 35°S in the Western Atlantic, where a distributional gap is recorded along South America (Moreno et al., 2005; Perrin, 2009). In general, Atlantic spotted dolphins inhabit the continental shelf along their distribution, but can also occur in oceanic waters (Baumgartner, Mullin, May, & Leming, 2001; Davis et al., 2002; Freitas, Dellinger, & Reiner, 1989;

Jefferson, Curry, Leatherwood & Powell, 1997; Moreno et al., 2005; Perrin, 2009; Weir, 2010). Despite its high dispersal capabilities, the Atlantic spotted dolphin presents a complex geographic distribution, where at least six distinct morphotypes were identified based on adult size, coloration and osteological measures of skulls (Perrin et al., 1987). Genetic studies conducted so far have also corroborated the evidence of geographic variation along its distribution (Adams & Rosel, 2006; Viricel & Rosel, 2014).

This species seems to challenge the long-standing assumptions about biodiversity in the marine environment, in which marine species maintain large and homogeneous populations (Bierne, Bonhomme, & David, 2003; Hauser & Carvalho, 2008). Vicariant and allopatric models for speciation are far less important in pelagic evolution than sympatric or parapatric speciation in which dispersal is not limiting (Norris, 2000).

Advances in genomic technology have been facilitating genomic studies in a range of marine species to answer questions about the process of both macroevolution and microevolution (Kelley, Brown, Therkildsen, & Foote, 2016). Moreover, population genomics have provided unprecedented resolution for addressing questions on population structure, speciation and adaptation in marine environments (e.g. Kelley, Brown, Therkildsen, & Foote, 2016).

Next generation sequencing is promising tool in phylogeography, because it allows different approaches to be used to discover, sequence, and genotype thousands of markers across any genome of interest in a fast and cost-effective way (Davey et al., 2011; McCormack, Hird, Zellmer, Carstens, & Brumfield, 2013). Single nucleotide polymorphisms (SNPs) have been successfully used to explore patterns of population differentiation over a significant part of the Atlantic spotted dolphin geographical range (Fernández et al., 2016; Foote et al., 2016; Leslie & Morin, 2016, 2018; Morin et al., 2015).

The application of genomic techniques to cetaceans as well as others non-model organisms has yielded numerous contributions to evolutionary biology and ecology, many of which would not have been possible with traditional genetic markers (Cammen et al., 2016; Hancock-Hanser et al., 2013). The advent of low-cost high throughput sequencing has led to dramatic increases in the number of neutral markers that can be evaluated even with low sample size, improving our power to resolve fine-scale or cryptic population structure in species with high dispersal capability such as Atlantic spotted dolphin. Recently, Leslie & Morin (2018) suggested that next generation techniques had enough statistical power to discern

163

closely related groups in others species of the *Stenella* genus (*S. attenuata* and *S. longirostris*) even using a relatively small dataset (n= 58 and n=72, respectively).

Atlantic spotted dolphins, as mentioned before, present a complex geographic pattern. In North Atlantic, several studies were conducted in order to investigate population structure of this species mainly along the Western North Atlantic coast (e.g. Adams & Rosel 2006, Viricel & Rosel 2014), as well as Bahamas (Green et al., 2007) and Azores and Madeira Archipelago (Quérouil et al., 2010). However, for a significant part of their distribution, little is known about population strutcture. Caballero et al. (2013) proposed that Atlantic spotted dolphins from southeastern Brazil and the Caribbean are distinct stocks based on levels of the genetic differentiation measures F_{ST} and Φ_{ST} based on a preliminary analysis of mitochondrial control region sequences (mtDNA CR) (Caballero, Santos, Sanches, & Mignucci-Giannoni, 2013). Moreno et al., (2005), based on a comprehensive review of sightings and captures of Atlantic spotted dolphins, had already suggested that individuals from Southwestern Atlantic Ocean were isolated.

do Amaral et al. (*in prep.*) analyzed 545 mtDNA CR sequences from individuals sampled along the entire species distribution, with exception of African waters. The results obtained confirmed different populations in the Western North Atlantic and Gulf of Mexico and also that individuals inhabiting oceanic islands in the Eastern Atlantic form one population connected with those from oceanic waters of the Western North Atlantic, corroborating previous studies (Adams & Rosel, 2006; Quérouil et al., 2010; Viricel & Rosel, 2014). Furthermore, the results indicated that individuals from southeastern Brazil represent one distinct population and exhibit low levels of genetic diversity based on the mtDNA CR marker.

Our main objective of this study is therefore to investigate the population structure of Atlantic spotted dolphins with emphasis in the Southwestern Atlantic Ocean individuals using genome-wide molecular markers. Based on previous studies, our main hypothesis is that those individuals found in the Southwestern Atlantic Ocean are isolated from others along Atlantic spotted dolphin range and represent at least a distinct population that deserves protection.

Material and Methods

Sample collection and DNA extraction

Samples consisted of different kinds of tissues (skin, muscle or liver) obtained from stranded individuals, remotely-darting biopsy or incidentally captured dolphins in pelagic drift gillnets collected from 1996 to 2016. These samples represent different regions of Atlantic Ocean: Southwestern Atlantic Ocean (Brazil, n=80; Uruguay, n=1), Caribbean (Colombia, n=7; Guadaloupe Island, n=1), and Eastern Atlantic (Canary Islands, n=19) (Table S1). A significant number of samples was collected along Brazilian coastal waters in the Southeast Brazilian Bight (hereafter, SBB) and these samples were treated as potentially two different groups: samples collected from stranded, incidentally captured and biopsied dolphins up to 100 km from coastline were considered   belonging to the "SBB inner/midshelf" group, and samples collected beyond 100 km of coastline were grouped in the "SBB outer shelf" group.

Samples were preserved in different ways, including ethanol, sodium chloride-saturated 20% dimethyl sulphoxide or lyophilized for long-term preservation. We extracted total genomic DNA from tissue samples using the DNeasy Blood and Tissue kit (Qiagen), following the manufacturer protocol. However, we extended the proteinase K digestion step overnight (Hancock-Hanser et al., 2013). We eluted DNA in lower volumes than recommended to avoid low concentrations of DNA mainly from samples obtained from stranded animals. DNA quality and concentration was verified using Qubit Fluorometric Quantition (Thermo Fisher Scientific Inc.).

Sex Determination

Molecular sexing techniques were used to determine the sex of specimens from which skin samples were collected using remotely-darting biopsies. We used a PCR-based method that consists in a multiplex reaction, which simultaneously targets the ZFX and SRY genes (Rosel, 2003). A fragment of 339 bp of the SRY gene was amplified through the primers TtSRYR (5' –ACCGGCTTTCCATTCGTGAACG-3') and PMSRYF (5'-CATTGTGTGGTCTCGTGATC-3'). A fragment of 382 bp of the ZFX gene was amplified using the primers ZFX0582F (5'-ATAGGTCTGCAGACTCTTCTA-3') and ZFX0923R (5'-AGAATATGGCGACTTAGAACG-3'). We performed an Exact Binomial Test to test if the true proportion of males and females in the dataset is 50%-50%. A two-tailed test was conducted in R 3.4.2 (R Core Team 2017) using the function binom.test in the package stats (R Core Team 2017) with 95% confidence level.

Double-Digest RAD-seq genomic data generation, processing and genotyping

165

We used Double Digest restriction-site-associated DNA sequencing (ddRAD-seq) (Peterson, Weber, Kay, Fisher, & Hoekstra, 2012), method to obtain genome-wide SNPs form the Atlantic spotted dolphin samples. Briefly, we built one library using two restriction enzymes (*Eco*RI and *Mse*I) to digest genomic DNA (~ 400 ng per sample). We performed a ligation step, where unique barcodes (10bp) and Illumina adapters were added to the digested DNA. After ligation, the product was cleaned and we selected fragments between 350 – 450 bp using a Pippin Prep platform (Sage Science). Fragments selected were amplified by PCR. The final library was sequenced for 100bp in one HiSeq2000 lane (Illumina, San Diego, CA, USA).

In order to analyse the raw data, we ran a pipeline through the University of Michigan flux, as described next. First, we demultiplex and processed the genomic sequences using Stacks version 1.45(Catchen, Hohenlohe, Bassham, Amores, & Cresko, 2013). One mismatch in the adapter sequence (--adapter_mm) and a barcode distance of two was used in process radtags to allow barcode rescue (--barcode_dist); adapter sites were also removed. In total, we obtained more than 155 million retained reads from 90 individuals (average of 1,733,002 ±1, 142,324 retained reads per individual).

After this initial step, we excluded eight samples from subsequent analyses, six samples collected from stranded animals, one sample collected from an incidental captured animal, and another one that was collected by remotely-darting biopsy. Therefore, samples with more than 500k retained reads were analyzed throughout the pipeline. Nevertheless, we chose to include 11 individuals with less than 500k retained reads (86,902 to 472,856) in the pipeline in order to increase representativity of some geographic areas, but almost all of these individuals ended up being excluded at some point along the pipeline due to the low number of retained reads. In general, samples obtained by remotely-darting biopsies or incidental capture had the highest DNA concentrations, a higher number of retained reads and good DNA quality, but we also observed that some samples from stranded specimens had enough DNA concentration and a high number of retained reads (higher than 1 million of retained reads in some cases).

We assembled the reads to the bottlenose dolphin genome (*Tursiops truncatus*, T_tru1.4, GenBank assembly accession: GCA_000151865.3 (latest), 2.5x Sanger; 3.5x 454; 30x Illumina coverage) with the short-read aligment tool BWA 0.7.15 (Li & Durbin, 2009). After alignment, we run PSTACKS with a

minimum depth of coverage to report a stack equal to three (-m 3), model type equal bounded (-
-model_type), and an error bound for ε of 0.1 (- -bound_high). We obtained a mean coverage of 9.6 (± 3.8).
A catalog of genomic sequences was built in CSTACKS using the aligned flag (--aligned), which base
catalog construction on alignment position, not sequence identity. Putative loci for each individual were
identified using SSTACKS using also the aligned flag (--aligned) in order to base matching on alignment
position, not sequence identity.

From SSTACKS output, we run a first POPULATIONS analyses with the following parameters: -r
0 -p 1 -m 3 --min_maf 0 --max_obs_het 0.5). We processed the resulting vcf file with a customized script in
R 3.4.2 (R Core Team 2017), in order to eliminate single-nucleotide polymorphisms (SNPs) from the last
five base pairs in the 3′- end of each locus, as well as loci with exceedingly high genetic diversity as such
high values are suggestive of sequencing and assembly errors (i.e., $\theta > 0.009$, representing loci in the upper
95% quantile of the distribution of genetic diversity; Fig. S1). In addition, we built a whitelist with
126,783 unique loci with 191,152 SNPs. We run POPULATIONS module again in order to keep only one
SNP per loci (--write_ random_snp). From this second POPULATIONS output, we used Plink v1.90b5
(Purcell et al., 2007) to filter missing data. Throughout Plink analyses, we excluded six individuals that had
more than 60% of missing data, and we also removed loci that had more 10% of missing data considering
all individuals. Therefore our resulting data set (hereafter referred as Dataset 1) contained a total of 9,450
SNPs in 9,450 loci, with a genotyping rate of 0.97, for 73 individuals. Dataset 1 was used as input for the
following analyses: the Principal Component Analysis (PCA) (Dray & Dufour, 2007), Discriminant
analysis of principal components (DAPC) (Jombart, Devillard, & Balloux, 2010) and Procrustes analyses
(Wang, Zöllner, & Rosenberg, 2012).

After Plink filtering, we created another list keeping loci/SNPs with a maximum of 50% of
missing data. We combined this list with the whitelist created previously resulting in a final whitelist with
87,322 SNPs in 49,187 loci . We used this final whitelist to run POPULATIONS both with and without the
--write_random_SNP flag.   The output of POPULATIONS without the flag (--write_ random_snp)
constitutes the Dataset 2, which in turn had 83,512 SNPs in 47,878 loci, with genotyping rate of 0.006.
Dataset 2 and was used to calculate genetic diversity summary statistics for each putative population and

F_{ST} values in Stacks version 1.45 (Catchen et al., 2013) and Arlequin 3.5.2.2 (Excoffier & Lischer, 2015), respectively. We also used this dataset to estimate heterozigosity and F_{ST} values using strataG package (Archer, Adams, & Schneiders, 2016). Furthermore, we used Dataset 2 to estimate co-ancestry proportions using sparse nonnegative matrix factorization implemented in the sNMF software (Frichot, Mathieu, Trouillon, Bouchard, & François, 2014). The output of POPULATIONS with the flag constituted the Dataset 3, in which we had 47,873 loci/SNPs. Dataset 3 was used as input for maximum likelihood analysis (see below).

Clustering of individuals and populations

We investigated levels of population structure using three different methods: discriminant analysis of principal components (DAPC), sNMF, and Procrustes analyses. Dataset 1 (see above) was used as input for the DAPC analyses with individuals grouped into seven putative populations based on sampling locations ("Canary Islands", " Caribbean (Isla Guadaloupe)", "Caribbean (La Guajira)", "SBB inner/midshelf", "SBB outer shelf", "Northern Brazil", and "Uruguay").

DAPC (Jombart et al., 2010) was performed using the *adegenet* (Jombart & Ahmed, 2011) package in R 3.4.2 (R Core Team 2017). This method aims to identify and describe genetic clusters using a few synthetic variables, which are constructed as linear combinations of the original variables (alleles), which have the largest between-group variance and the smallest within-group variance. Coefficients of the alleles used in the linear combination are called loadings, while the synthetic variables are themselves referred to as discriminant functions (Jombart & Ahmed, 2011; Jombart & Collins, 2015). Moreover, being based on the Discriminant Analysis, DAPC also provides membership probabilities of each individual for the different groups based on the retained discriminant functions (Jombart & Collins, 2015).

First, we used the function find.clusters (max.n.clust=8, i.e., maximum number of putative populations+1) to identify clusters without *a priori* assignment of individuals to groups based on sampling location (k-means clustering). Second, we ran the dapc function on the individuals when they were grouped by sampling location using the function optim.a.score as reccomented by authors (Jombart & Collins, 2015). However, optim.a.score function determined that the number of PC to retain was equal to 1. Therefore, we ran DAPC using the maximum number of PCs recommended, which is n/3 (Jombart &

Ahmed, 2011). We also used DAPC to assess membership probabilities of each individual for the different groups based on the retained discriminant functions. This analysis is useful for groups defined by an external criteria, i.e., defined biologically, as opposed to identified by k-means. We further investigate admixed individuals, which we define as those having no more than 0.6 probability of membership to any group (Jombart & Collins, 2015).

Since the delimitation of populations *a priori* based on geography is very questionable for cetaceans due to their great dispersal capabilities, an individual level analyses was performed through Procrustes approach to find an optimal transformation that maximizes the similarity between Principal Component Analysis (PCA) maps of genetic variation and geographic maps of population locations (Wang, Zöllner, & Rosenberg, 2012).

First, we applied Procrustes analysis to compare the individual-level coordinates of the first two components (PC1 and PC2) in the PCA performed on Dataset 1 to the geographic coordinates. PCA was perfomed using the function dudi.pca of ade4 package (Kopelman, Mayzel, Jakobsson, Rosenberg, & Mayrose, 2015). We used `procrustes` function in order to rotate a configuration to maximum similarity with another configuration, and the function `protest to` test the significance between two configurations. We used procrustes and protest functions from the vegan package (Oksanen et al., 2017). Posteriorly, we applied the residual function to estimate the residuals of the first procrustes, and performed a second one using a PCA of environmental rasters (see do Amaral et al. *in prep.*). The significance of the association statistic between the genetic PC values and geography or environmental space was evaluated based on 10 000 permutations.

We estimated individual ancestry coefficients based on sparse nonnegative matrix factorization algorithms using as input Dataset 2. This method is implemented in the computer program sNMF (Frichot et al., 2014). Like PCA, NMF algorithms are flexible approaches that are robust to departures from traditional population genetic model assumptions (Frichot et al., 2014).

Diversity estimates and population differentiation

An overall differentiation (F_{ST}) for each pairwise combination of population was estimated using the strataG package in R with 1,000 repetitions using Dataset 2.

Mean heterozygosity across SNPs for all individuals within a population is indicative of the overall genetic variation within the populations. Low heterozygosity could indicate smaller populations, inbred individuals, or poor sample quality that results in allelic dropout (Leslie & Morin, 2018). On the other hand, high hererozygosity could indicate outbred individuals, large historical population abundance, or be an artefact of the higher sample sizes for these populations (Leslie & Morin, 2018). Dataset 2 was used to calculate genetic diversity summary statistics for each putative population (only those with more than five individuals) in STACKS (i.e., π and heterozygosity expected averaged) across populations. From these summary statistics, estimates for long-term effective population sizes (N_e) can be approximated through the formula $\pi=4*N_e*\mu$ (Tajima, 1983), based on estimates of mutation rate per site for the bottlenose dolphin (0.84×10^{-9} mutations per site per generation (Fernández et al., 2016). F_{ST} values (distance matrix) and its significances were calculated in Arlequin 3.5.2.2 (Excoffier & Lischer, 2015) with 10, 000 replicates with a Bonferroni correction for multiple comparisons.

Phylogeographic Analysis

Dataset 3 was used as input for phylogenetic analysis in RAxML (Randomized Axelerated Maximum Likelihood) (Stamatakis, 2014). We performed the analysis in the CIPRES portal using RaxML-HPC2 on XSEDE (8.2.10) tool, in which provides a phylogenetic tree inference using maximum likelihood/rapid bootstraping algorithm to account for uncertainty in the estimation of the topology. Most of parameters were kept as default, with exception: we used GTRGAMMA model of sequence evolution, and we performed 1 000 bootstrap iterations.

Results

Genotyping and Sex determination

The genomic library was built with 90 individuals but after data processing we successfully genotyped 73 individuals representing the following geographical regions: "Canary Islands" (n= 7), "Caribbean (Isla Guadaloupe)" (n= 1), "Caribbean (La Guajira)" (n=5), "SBB inner/midshelf" (n=45), "SBB outer shelf (n=15)", "Northern Brazil (n=1)", and "Uruguay" (n=1) (Fig. 1). Due to a low number of retained reads, 17 individuals were not included in the analyses. Of these, 13 samples were obtained from stranded dolphins and 4 from biopsy darting. From these biopsy-collected samples, one sample was removed due to low retained reads (14,137); and the other three individuals (Sf39 from SBB

inner/midshelf; G24 and G40 from Caribbean - La Guajira) were removed because they were misplaced in early exploratory analyses.

From the 73 individuals genotyped, 57 samples were obtained from remotely biopsy darting, and sex identification in the field was not possible. Using molecular sexing methods, we identified 31 males, 19 females and 7 individuals remained undetermined. Samples obtained from stranded animals or incidentally captured, sexing identification was possible at the time of sampling collection (8 males and 8 females). In total, our dataset had 39 males, 27 females and seven unidentified samples. The exact binomial test did not reject the null hypothesis that the proportion of males and females in our dataset is 50% (p-value = 0.1753).

In the PCA (Fig. 2) estimated with Dataset 1, individuals are arranged in relatively well defined clusters, defined according to sampling location. Nevertheless, some individuals were grouped far from their geographical origin: a Guadaloupe Island sample grouped with individuals collected in southeastern Brazil; a Uruguayan sample was positioned closer to Northern Brazil and La Guajira samples; one sample from SBB inner/midshelf (Sf38) and another one from Caribbean - La Guajira (G38) were grouped with Canary Islands samples.

In the PCA performed with samples collected only in Brazil (Fig. 3) we observed two distinct groups, one of them including only individuals sampled in the inner/midshelf, and the other including both individuals collected in the inner/midshelf and in the outer shelf.

Clustering of individuals

The smallest BIC recovered throughout DAPC ran with the function find.cluster was with K=1 (K=1, BIC = 452.6768; k=2, BIC = 452.7890) (results not showed). We also ran a second DAPC with n.pca=50 and n.da=100 in order to apply the optim.a.score function, which by its turn determined that the number of PC to retain was equal to 1 (results not showed). Therefore we ran dapc function using n.da=100 and opted to set n.pca= 24, because n/3. Finally, we plotted the first two discriminat functions as two-dimensional scatters (Fig. 4). In general, the DAPC showed that genomic variation across individuals and populations was well represented by the first two eigenvalues of the DAPC although one eigenvalue was clearly dominant (inset Fig. 4). Graphical visualization suggests three separated groups along PC1 and PC2

axes. Southeastern Brazil (both inner/midshelf and outer shelf), Caribbean – La Guajira and Canary Islands individuals formed tight clusters at different portions of genomic space. As already observed in the PCA analyses, Uruguayan and Guadaloupe Island samples are genetically closer to Caribbean and southeastern Brazil clusters, respectively. The sample obtained from Northern Brazil was closer to Caribbean – La Guajira cluster. From this DAPC, we investigated the membership probability of each individual and presented this analysis as a STRUCUTURE-like graphic (Fig. 5). We identified three admixed individuals (BC_08, BC_16, PA_209), which having no more than 0.6 probability of membership to any group. (Fig. S2). However, these results show be interpreted with caution because they are probably overfitted.

In relation to southeastern Brazil individuals, we observed a partial overlapping among individuals collected in the inner/midshelf and those collected in the outer shelf. Therefore, we further investigated clustering within southeastern Brazil. The smallest BIC recovered throughout DAPC ran with the function find.cluster was also with K=1 (K=1, BIC = 354.9546; k=2, BIC = 357.0297). We also ran a DAPC with the optim.a.score function, which by its turn determined that the number of PC to retain was equal to 15. Since, a single discriminant function had been retained due to k=2, we plotted the densities of individuals on a given discriminant function with different colors for different groups (Fig. S3). Membership probabilities were investigated through DAPC conducted with the optim.a.score and results are showed in Fig. S4. Two individuals from the inner/midshelf group were identified as admixed (i.e., having no more than 0.6 probability of membership to any group) (Fig. S5).

We also analyzed samples at an individual level without any *a priori* clustering and further tested genetic data against both geography and environmental variables through Procrustes analyses. Procrustes analysis between genetic similarity and geographic distance suggested a significant correlation (t = 0.72, p= 9.999e-05), while the analysis between genetic similarity and environmental space was also significant (t = 0.31, p= 0.017198).

In general, the visual inspection of graphics (Fig. 6) suggested that those individuals from the Canary Islands had am intermediary position in relation to those from Caribbean and Southeastern Brazil (Fig. 6A). Uruguayan and Guadaloupe Island samples are genetically more similar to Caribbean and Brazilian samples, respectively. Despite surprising, the position of these samples is in agreement with PCA

and DAPC analyses. When we analyze the graphic representing the relationship between genetic and environmental space (Fig. 6B), we observe the clusters observed in this graphic seem to be in agreement with a designation of groups by sampling location, since individuals were positioned close with its counterparts (with exception of one individual from Canary Islands). The most displaced sample is those collected in Uruguay.

Analyses using sNMF across K-values raging from 1 to 8 indicated that K = 2 is the most likely number of populations according minimal cross-entropy criterion (Fig. S6). In the resulting graphic of K=2, an orange genomic axis is predominant on samples from southeastern Brazil (Fig. 7). We also provided a graphic of K=3 (Fig. S7), in which genetic variation broke down into three genomic axes, where a blue genomic axis predominated along individuals from Colombian and Canary Islands. Individuals sampled in the inner/midshelf showed a predominant orange genomic axis, that also appeared on Canary Islands samples. A pink genomic axis is spread out across almost all individuals, mainly in those from Brazilian outer shelf. Besides that, individuals sampled outside of these main areas, such as one sample from Northern Brazil and another one from Uruguay, were dominated by the blue genomic axis (Fig. S7).

Despite differences in the methods, we considered our clustering results consistent across analyses. Overall they suggested some level of differentiation among the main sampling locations: southeastern Brazil, Canary Islands and Caribbean. However, the differentiation among these three genetic clusters seems to be subtle. Individuals collected outside of these main locations (Northern Brazil and Uruguay) were positioned closer to Caribbean – La Guajira individuals, except one sample collected in Guadaloupe Island that clustered with southeastern Brazil samples. Both PCA conducted only with samples from southeastern Brazil and DAPC seem suggested a further subdivision within southeastern Brazil cluster.

Diversity estimates and population differentiation

We estimated diversity and population differentiation using two different methods and we considered in these analyses putative populations with at least five individuals. The results obtained with Dataset 2 are presented in Table 1 and Table 2.

Mean heterozygosity across SNPs for all individuals within a population is indicative of the overall genetic variation within the populations. Heterozygosity estimated in the Stacks, values ranged from

173

0.1251 to 0.1357 considering only variant positions, and from 0.0016 to 0.0017 considering variant and fixed positions of Dataset 2. Despite the disparity in sample sizes among putative populations heterozygosity values were similar across populations in each dataset (Table 1), however values ranged greatly using only variant positions or using variant and fixed positions.

We also observed that great variations in nucleotide diversity estimates, which by its turn had an impact in the effective size estimates (N_e) (Table 1). N_e estimated from nucleotide diversity calculated using just variant positions ranged from 392 697 to 413 764 individuals for SBB inner/midshelf and Canary group, respectively. However, N_e estimated from nucleotide diversity based on variant and fixed positions ranged from 4 775 to 5 056 individuals (Table 1).

In relation to the F_{ST} statistics, values estimated in StrataG were not possible to estimate its p-value, because the analysis was computally demanding. Anyway, we opted to present F_{ST} present the values (Table 2). The smallest F_{ST} value was equal to 0.01 between individuals sampled in the SBB outer shelf and inner/midshelf, and the highest F_{ST} value was equal to 0.1 between Caribbean - La Guajira and SBB inner/midshelf. In general, population differentiation calculated with the F_{ST} statistics in Arlequin indicated higher and significant differentiation (F_{ST} values between 0.07 and 0.12) between Caribbean - La Guajira and SBB inner/mid shelf, as well as between Caribbean - La Guajira and SBB outer shelf.

Phylogeographic Analysis

Phylogenetic analysis using Dataset 3 (47,873 SNPs/loci) produced a topology with low bootstrap support, mainly in the most external branches (Fig. 8). However in this analysis two main clades were recognized, being one clade almost exclusively formed by individuals collected in southeastern Brazilian waters, and another one formed by all remaining individuals from southeastern Brazil, Canary Islands and Caribbean – La Guajira.

The clade formed by individuals collected in southeastern Brazil included both individuals from SBB inner/midshelf and outer shelf, with exception of the individual sampled at Caribbean - Guadaloupe Island that is also presented in this clade. The clade is further structured in several small clades.

The second main clade included individuals collected at different sampling locations. This clade is also further structured in small clades, being one of them highly support formed by individuals from

174

Caribbean - La Guajira, Northern Brazil and Uruguay. With exception of one individual (SF_38) from SBB inner/midshelf, all individuals from southeastern Brazil presented in this clade were from SBB outer shelf. Individuals sampled at Canary Islands were spread across this clade.

Discussion

In this study, we used genome wide markers to understand population structure in Atlantic Spotted dolphin across a significant part of its geographic distribution, including Eastern Atlantic, Caribbean and Western South Atlantic. The most part of samples included here were collected in areas never analyzed before in relation to nuclear markers, such as Canary Islands, northern Brazil and the southernmost limit of the species. Our results therefore represent a first genome-wide investigation of population structure along these areas, and bring light into many more interesting aspects about the connectivity and biology of the species that deserves further investigation.

In general, the clustering methods used in our study were congruent, identifying three main clusters: southeastern Brazil, Caribbean and Canary Islands. Although some of the methods indicated that differentiation among these genetic populations is subtle, pairwise genetic differentiation was statistically significant between these clusters. Our results also indicated a possible hierarchical structure within the Brazilian population. Finally, our analyses revealed interesting connection patterns, in which some speculations about dispersal routes could be hypothesized.

Overall genetic differentiation

Our results are limited in terms of comparisons with other genetic studies conducted with Atlantic Spotted dolphins due to the main following reasons: 1) sampling areas analyzed here were never analyzed before with nuclear markers; 2) the markers used here, namely SNPs, were never analyzed in this species before, thus the levels of heterozigosity and population differentiation could only be compared with studies conducted with related species of the Delphinidae family (Fernández et al., 2016; Foote et al., 2016; Leslie & Morin, 2018; Morin et al., 2015).

Previous studies that aimed to investigate population structure across Atlantic Spotted dolphins were conducted mainly in North Atlantic through the analyses of microsatelites and mtDNA (Adams &

Rosel, 2006; Quérouil et al., 2010; Viricel & Rosel, 2014). These studies identified clusters corresponding to previously described morphotypes inhabiting oceanic and shelf waters, as well as sub-structuring in the shelf cluster, possibly related to environmental distinctiveness (Viricel & Rosel, 2014). Interestingly, Azores individuals, even located as far as 4,500 km, were considered to belong to the oceanic cluster recovered by Viricel & Rosel (2014), where no further division was detected across Western and Eastern North Atlantic. Furthermore, no genetic structure was recovered between Atlantic spotted dolphins from the Azores and Madeira, neither between the archipelagos nor between groups of islands (Quérouil et al., 2010). In our study, we did not had the opportunity to include these populations in our dataset, being available for us just individuals stranded at Canary Islands.

do Amaral et al. (*in prep.*) analyzed by the first time the genetic structure along almost the full species range using mtDNA CR marker. Their results suggested that Canary Islands individuals seems to be more closely related to the Azores and Madeira Archipelagos than other regions, and, therefore individuals inhabiting these set of islands in the Eastern Atlantic could be part of one oceanic population. In this context, we observed that individuals from Canary Islands had an intermediary position in relation to Brazilian and Caribbean clusters based on nuclear SNPs, as well as the levels of differentiation were statistically insignificant or smaller across several pairwise comparisons both in relation to Caribbean and southeastern Brazilian individuals ($F_{ST} > 0.07$). Therefore, our results based on genome-wide markers seem to confirm this oceanic "status" of these individuals. Such individuals are most likely to perform cross-oceanic dispersal and, therefore, could maintain gene flow across distant populations.

Caballero et al. (2013) analyzed individuals collected along Caribbean Sea both in relation to mtDNA and nuDNA. At the nuclear level, a high number of alleles were shared between samples collected in Caribbean (Colombia and Puerto Rico) and southeastern Brazil. However, at that time, the authors considered that southeastern Brazil and Caribbean should be considered population units genetically isolated. And, they suggested that the higher number of alleles shared between these populations could be the result of not enough time for nuDNA to diverge between population units due to lower mutation rates in nuDNA when compared to mtDNA (Caballero et al., 2013). Recently, do Amaral et al. (*in prep.*) confirmed

the results of Caballero et al. (2013) and suggested that southeastern Brazil and Caribbean individuals should be considered different populations based on mtDNA CR.

The results obtained here based on genome-wide SNPs are interesting and have potential to help understand the complex relationship among Brazil and Caribbean both in relation to Atlantic spotted dolphin and also in relation to cetacean fauna connectivity (Costa et al., 2017). Our dataset included samples from Caribbean collected in different locations: one sample collected from a stranded female in the Guadaloupe Island, and five samples of individuals collected by biopsy-darting in La Guajira Peninsula in northern Colombia. Interestingly, the individual collected in Guadaloupe grouped together with southeastern Brazil individuals in all analyses performed here. In relation to mtDNACR marker, do Amaral et al. (*in prep.*) observed this same pattern. This sample had Haplotype 20, which was widely distributed along species range, being common in Azores and Western North Atlantic, but also found in high frequency in southeastern Brazil.

Taking into account these results, but also that we excluded in the first steps of our analyses two samples from La Guajira due to a weird positioning of these samples in our exploratory analysis of data, we suggest that the Caribbean individuals could represent a very diversified population. do Amaral et al. (*in prep.*) also found high levels of diversity across Caribbean samples based on mtDNA marker. However, we highlight that this conclusion should be taken with caution, because our dataset is limited to few individuals mainly collected at one restricted locality, La Guajira. Furthermore, the higher levels of differentiation observed here could be related to a fine-scale population structure in Colombian waters.

In relation to the overall structure recovered from our dataset, the Procrustes analyses suggested a significant correlation between genetic similarity and environmental space, but mainly between genetic similarity and geographic distance. Therefore, it is not surprising that the clusters that we recovered across different analyses reflected sampling locations. In relation to environment, Bathymetry and Mean Annual SST were the environmental variables that mainly contribute to the first component of PC and the second PC, respectively. Therefore, its seems reasonable propose that individuals were positioned along axes 1 and 2 according Bathymetry and SST, respectively. Although at about 7% of our data was obtained from stranded individuals, 78% of our samples were collected from biopsy and therefore the environmental

conditions that species occupied along its distribution could be thought to be represented in our dataset. In relation to axis 1, we assumed that individuals were predominantly collected at relatively shallow waters, which is typical of this *Stenella* species (Perrin, 2009). In relation to axis 2 there is a reasonable stratification of individuals in a "tropical" to "temperate" gradient.

The ecological niches of cetaceans seem to be defined by water temperature, water depth and factors that affect the distribution and abundance of their prey (topography, ocean currents and primary productivity) (Baumgartner et al., 2001; Palacios, Baumgartner, Laidre, & Gregr, 2013; Redfern et al., 2006). In relation to Delphinidae dolphins, several species present population structure related to habitat specializations (e.g. Morin et al., 2015). Viricel & Rosel (2014) detected significant differences in sea surface temperature, depth and turbidity among the four genetic clusters identified in the Western North Atlantic. Moreno et al. (2005) had proposed that Atlantic Spotted dolphin is found just up to 1,000 m isobath based on a comprehensive review of *Stenella* species records in the Western South Atlantic. The authors also suggested a gap in the Atlantic spotted dolphin distribution, due to the narrow of continental shelf between 6 and 18°S (Danilewicz, Ott, Secchi, Andriolo, & Zerbini, 2013; do Amaral et al., 2015; Moreno et al., 2005) . do Amaral et al. (2015) corroborated the existence of this distributional gap using ecological niche modeling.

Atlantic spotted dolphins from the Southeast Brazilian Bight

Based on the distributional gap between 6 and 18°S due to the narrow of continental shelf, Moreno et al. (2005) suggested the existence of two populations along Brazil. Furthermore, they hypothesized that dolphins distributed off the northern coast of Brazil (north of 6°S) may represent the southern range of a population that is connected to the Caribbean, Gulf of Mexico and North Atlantic(Moreno et al., 2005). According authors, a second, geographically and possibly reproductively isolated, population would be found in southern and southeastern Brazil (21 to 33° S). Compared the skull morphology and morphometrics of specimens from this area with those from the North Atlantic Ocean and Caribbean and found significant differences in shape, metric and meristic characters (Moreno, 2002).

Genomic-wide markers analyzes here added an important information to help understand the relationship between South America and Caribbean. However, this relation seems to be more complex in

178

relation to what has been proposed in other studies (Caballero et al., 2013; Costa et al., 2017; Moreno et al., 2005). Based on the haplotypes found in the analyzes of mtDNA CR (do Amaral et al. *in prep.*) and several studies that proposed different forms or ecotypes according habitat specialization, namely "inshore", "coastal" and "offshore" (Barragán-Barrera et al., 2017; Möller et al., 2011; Sellas, Wells, & Rosel, 2005), we opted to threat those individuals collected along southeastern Brazil as two different sampling sites: one of them consisted of samples collected mainly in the inner/midshelf and approximately at 50 m isobath, and another group of samples were designated as SBB outer shelf, because these samples where collected more than 100 km from the coastline.

All samples analyzed here from southeastern Brazil, where collected up to 100 m of depth in the SBB and we opted to divided in these groups according the stratification of Brazilian Continental Shelf (Castro & Miranda, 1998). SBB had an inner shelf separated from the midshelf waters by a thermal front that changes seasonally, being closer to the coast (10 – 20 km) during summer and father offshore (40 – 50 km) during winter. The inner shelf is occupied mainly by Coastal Water, which has low salinities due to the mixture of land runoff from several estuaries and saline water. Inner shelf currents are highly dependents on the wind direction, in which northeasterly winds forces the currents southwestward, and vice versa (Castro & Miranda, 1998). Midshelf waters show a two-layer structure, and in the summer, there is a predominantly shallow and thin seasonal thermocline connected to the inner front. The waters of lower layers are classified as Cold South Atlantic Central Water (SACW, SST < 20°C and SSS < 36.4 psu). By its turn, midshelf waters are separated from outer shelf waters by a strong near-surface cross-shelf salinity front. The outer shelf front is located at 80 – 120 km from the coast. Salty waters of Tropical Water (SST > 20°C, SSS > 36.4 psu) are present in the upper layer, and SACW have a strong influence in the lower layer (Castro & Miranda, 1998).

Both SBB takes place between two prominent capes, namely, Cabo Frio (23°S) and Cabo de Santa Marta (~28°S) (Castro & Miranda, 1998), where upwelling induced by shelf break associated with the cyclonic meanders of the Brazil Current is recorded (Campos, Velhote, & Da Silveira, 2000). The upwelling plays an important role in the pumping of SACW from the slope region onto the continental shelf (Campos et al., 2000).

Although is not totally clear the distinction of these two "strata" in southeastern Brazil, we observed a pronounced differentiation among southeastern Brazil group in relation to La Guajira population. Furthermore, RaxML tree, DAPC and PCA conducted only with Brazilian samples suggested some level of differentiation of southeastern Brazil in relation to other sampling localities. As discussed above, habitat specializations could be triggered by environmental factors and indeed the SBB seem to have a great environmental heterogeneity along continental shelf. However, such differentiation could also be related to social structure, for example.

It is well-known that some coastal populations of bottlenose dolphins live in small populations characterized by low genetic diversity and low gene flow between neighboring populations, suggesting local founder events at least in some areas or recent isolation (Barragán-Barrera et al., 2017). Despite of the relatively higher number of samples analyzed here we also observed low levels of diversity in the SBB inner/midshelf cluster in relation to sampling locations analyzed with far fewer individuals. For example, SBB outer shelf individuals have slightly higher values of nucleotide diversity and heterozygosity (considering the different datasets analyzed) than SBB inner/midshelf cluster. It is important highlight that SBB outer shelf had a sample size of approximately one third in relation to the SBB inner/middle shelf.

Despite do Amaral et al. (*in prep.*) had not discriminated Brazilian samples in two distinct groups, their results also revealed very low levels of diversity and relatively high and significant differentiation in pairwise comparisons across populations based on mtDNA CR.

Statistical power of SNPs for population structure and conservation of Atlantic Spotted Dolphin

In our study, we used three different datasets, in which the number of SNPs were different. We included in some datasets independent SNPs (i.e., one SNP per locus) or more than one SNP per locus, and, finally, the number of allowed missing data across dataset was also variable. Moreover, we had a disparate sample size across sampling localities, being SBB inner/midshelf group with the highest sample size (n=43) and La Guajira group with the lowest sample size (n=5). The nature of our dataset and the sample size differences could have had an impact in our results (Huang & Knowles, 2016; Morin, Martien, & Taylor, 2009). Beyond that, we also did not have the opportunity to include in our dataset important samples from Western North Atlantic and Eastern Atlantic (Azores and Madeira) that were previously analyzed in relation to nuDNA (Adams & Rosel, 2006; Green et al., 2007; Quérouil et al., 2010; Viricel & Rosel, 2014).

Furthermore, dolphins from Eastern Africa remain genetically unknown. Probably if we have access to samples of these areas, our results could have been different (see Huang & Knowles, 2016).

Despite differences in methods used to estimate population differentiation, we observed that our results were consistent across analyses, resulting in F_{ST} values with almost the same order of magnitude. However, we obtained some negative values between some comparisons estimated in Arlequin software (e.g. SBB inner/midshelf and SBB outerh shelf, SBB outer shelf and Canary, and La Guajira and Canary), probably due to insufficient statistical power due to low sample size (Morin et al., 2009). Furthermore, it was not possible estimate p-values in StrataG, because the analysis was compute-intensive.

Comparisons in relation to the magnitude of values obtained here are difficult for different reasons. First, several factors could influence the final dataset such as the amount of sequence divergence among the individuals/taxa included in the study, the coverage and post-sequencing processing decisions about the reads, and the tolerance for missing data set by the researcher (Huang & Knowles, 2016). Second, our study analyzed by the first time genome-wide markers in the Atlantic spotted dolphin. Third, comparisons with similar studies using genome-wide markers in related species (e.g. Leslie & Morin, 2018) could provide some perspective about the magnitude of differentiation, but each species is unique in relation to, for example, small or large population size, behavior, demographic history that all together influence in the magnitude of results obtained (Morin et al., 2009)

For example, in relation to studies using SNPs in *Stenella* species, 4,381 SNPs were used to investigate population structure of Spinner dolphins (*Stenella longirostris*) across Eastern Tropical Pacific (ETP)(Leslie & Morin, 2016). In relation to heterozygosity values ranged from 0.25 to 0.27, and significant F_{ST} values ranged from 0.0009 to 0.0215, including comparisons between populations or subspecies. Population structure of Pantropical spotted dolphin (*Stenella attenuata*) across ETP was also investigated and the results obtained throughout the analyzes of 3,721 SNPs revealed a heterozygosity between 0.24 and 0.26 (Leslie & Morin, 2016). Significant F_{ST} values ranged from 0.0019 to 0.073 across populations or subspecies (Leslie & Morin, 2016). Population structure of Spinner and Pantropical spotted dolphins were also investigated across oceanic basins (Leslie & Morin, 2018). The authors recovered very similar values for heterozygosity and population differentiation in relation to their previous study (Leslie & Morin, 2016). In relation to Spinner dolphins, 3,340 SNPs were analyzed, heterozygosity ranged from 0.15 to 0.27; and

significant F_{ST} values ranged from 0.0035 to 0.0119 among subspecies, and from 0.0074 to 0.3 among populations across oceanic basins. In relation to Pantropical spotted dolphins, 3,524 SNPs were analyzed, heterozygosity ranged from 0.21 to 0.26; and significant F_{ST} values was equal to 0.05 between subspecies within the same oceanic basin (ETP), and from 0.012 to 0.1727 across populations (Leslie & Morin, 2018). Leslie & Morin (2018) observed that the disparity in sample sizes between ETP populations and those from the global sampling made comparisons of heterozygosity within both species difficult, and pairwise tests of population differentiation based on allele frequencies showed high levels of differentiation in both species despite high dispersal potential of individual animals.

Abundance estimates of Atlantic spotted dolphins in Western North Atlantic ranges from 14,438 to 37,611 individuals (Mullin & Fulling, 1998; Waring, Josephson, Maze-Foley, & Rosel, 2009). Although it is difficult to know whether our estimates of N_e were reliable, this information is provided by the first time for the Atlantic spotted dolphin across a considerable part of its distribution.

Although made comparison with the studies cited above are difficult due to biological features of each species and the complex and unresolved phylogenetic relationship of *Stenella* species (Amaral, Jackson, Möller, Beheregaray, & Coelho, 2012; Perrin, Rosel, & Cipriano, 2013), these studies were the only ones conducted with a relatively similar dataset and related species so far, specially those conducted with Spinner and Pantropical spotted dolphins (Leslie & Morin, 2016, 2018). We observed that our results of population differentiation among our main sampling locations were in the same order of magnitude of comparisons among subspecies of Spinner and Pantropical spotted dolphin.

More specifically for conservation purposes, we believed that our study could help to protect Brazilian individuals. Morin et al. (2009) assessed statistical power of SNPs for population structure and conservation studies of cetaceans based on the Endangered Species Act (ESA) and the Marine Mammal Protection Act (MMPA), that designated two different units-to-conserve. The authors suggested through a simulated dataset of whales that a F_{ST} equal 0.2 could be helpful to identify Distinct Population Segment (DPS), which are similar to the evolutionarily significant unit (ESU) (Moritz, 1994; Waples, 1991), while a F_{ST} equal to 0.0025 could be helpful to identify demographically independent population (DIP) (Taylor, 1997; Taylor et al., 2017). Whether we apply these thresholds in our dataset, we could conclude that at least individuals analyzed here from different sampling locations constitutes at least three demographically

independent populations (southeastern Brazil, Caribbean – La Guajira, and Canary Islands). Furthermore, southeastern Brazil is further structured in two independent populations.

Despite our levels of differentiation within southeastern Brazil were not consistent across all analyses, we observed that pairwise comparison performed with Dataset 2 in the StrataG reached a F_{ST} value equal to 0.01, although we cannot estimate the p-value. Taking into account, the findings of Méndez-Fernandez et al. (2018) in relation to POPs concentrations in individuals from southeastern Brazil (the most industrialized region in Brazil with a human population of more than 80 millions), we highlight the necessity to, at least, southern Brazilian dolphins be considered a different management unit due to constant threats faced by these individuals. Furthermore, we suggested that those individuals inhabiting the inner/midshelf waters also deserves some management actions, since they probably represent a lineage with some level of social structure and/or founder events (see Santos et al., 2018, Barregán-Barrera et al., 2017). Currently, the main source of human-induced mortality in the southern range of the species in Brazil is bycatch in fishing gear, particularly bottom set and drift gillnets (Reeves et al., 2013), habitat degradation, underwater noise and overfishing of prey species. Therefore, due to its unknown status, isolation and relatively limited range, further studies on abundance and trends, mortality and genetic structure should be viewed as a priority to assess the conservation status of this Atlantic spotted dolphin populations in Brazil. And, at least in the near future the conservation status of the species should be determined at the national level.

Conclusion

Our study provide an unprecedented information about population structure of Atlantic spotted dolphin across a significant part of its distribution that have never analyzed before in relation nuclear markers.   In general, we observed a subtle population structure among the main sampling locations analyzed here: Brazil, Colombian Caribbean and Canary Islands. We also investigated an additional level of structure in the southeastern Brazil population. Our analyses suggested a correlation of geography, and environment (bathymetry and sea surface temperature) with genetic similarities.

Taking into account we had a relatively large sample size in Brazilian cluster and based on the levels of differentiation observed here we proposed that the both populations should be treated as different

"units" and deserve protection due to constant anthropogenic threats faced by these individuals in the southeastern Brazil, mainly related to ship traffic, oil exploration, agricultural and industrial pollutants.

The status of the Canary Islands and Caribbean should be better investigate, including more samples, as well as we strongly recommend the inclusion of samples from Eastern Africa and the South America. Additional samples across all species distribution had the potential to elucidate dispersal routes and biogeographic history of this species along Atlantic Ocean.

REFERENCES

Adams, L. D., & Rosel, P. E. (2006). Population differentiation of the Atlantic spotted dolphin (Stenella frontalis) in the western North Atlantic, including the Gulf of Mexico. *Marine Biology*, *148*(3), 671–681. doi:10.1007/s00227-005-0094-2

Amaral, A. R., Jackson, J. A., Möller, L. M., Beheregaray, L. B., & Coelho, M. (2012). Species tree of a recent radiation: The subfamily Delphininae (Cetacea, Mammalia). *Molecular Phylogenetics and Evolution*, *64*(1), 243–253. doi:10.1016/j.ympev.2012.04.004

Archer, F. I., Adams, P. E., & Schneiders, B. B. (2016). strataG: An R package for manipulating, summarizing, and analyzing population genetic data. *Molecular Ecology Resources*, n/a-n/a. doi:10.1111/1755-0998.12559

Barragán-Barrera, D. C., May-Collado, L. J., Tezanos-Pinto, G., Islas-Villanueva, V., Correa-Cárdenas, C. A., & Caballero, S. (2017). High genetic structure and low mitochondrial diversity in bottlenose dolphins of the Archipelago of Bocas del Toro, Panama: A population at risk? *PLOS ONE*, *12*(12), e0189370. doi:10.1371/journal.pone.0189370

Baumgartner, M. F., Mullin, K. D., May, L. N., & Leming, T. D. (2001). Cetacean habitats in the northern Gulf of Mexico. *Fishery Bulletin*, *99*(2), 219–239. doi:10.1016/S0967-0637(01)00035-8

Bierne, N., Bonhomme, F., & David, P. (2003). Habitat preference and the marine-speciation paradox. *Proceedings of the Royal Society B: Biological Sciences*, *270*(1522), 1399–1406. doi:10.1098/rspb.2003.2404

Caballero, S., Santos, M. C. O., Sanches, A., & Mignucci-Giannoni, A. A. (2013). Initial description of the phylogeography, population structure and genetic diversity of Atlantic spotted dolphins from Brazil and the Caribbean, inferred from analyses of mitochondrial and nuclear DNA. *Biochemical Systematics and Ecology*, *48*, 263–270. doi:10.1016/j.bse.2012.12.016

Cammen, K. M., Andrews, K. R., Carroll, E. L., Foote, A. D., Humble, E., Khudyakov, J. I., … Van Cise, A. M. (2016). Genomic Methods Take the Plunge: Recent Advances in High-Throughput Sequencing of Marine Mammals. *Journal of Heredity*, *107*(6), 481–495. doi:10.1093/jhered/esw044

Campos, E. J. D., Velhote, D., & Da Silveira, I. C. A. (2000). Shelf break upwelling driven by Brazil current cyclonic meanders. *Geophysical Research Letters*, *27*(6), 751–754. doi:10.1029/1999GL010502

Castro, B. M., & Miranda, L. B. (1998). Physical oceanography of the western Atlantic continental shelf located between 4º N and 34º S. In A. Robinson & K. H. Brink (Eds.), *The sea* (pp. 209–251). John Wiley & Sons, Inc. Retrieved from https://scholar.google.com.br/scholar?q=castro+%26+miranda+1998+Physical+oceanography+of+the+western+Atlantic+continental+shelf+located+between+4+N+and+34+S&btnG=&hl=en&as_sdt=0%2C5

Catchen, J., Hohenlohe, P. A., Bassham, S., Amores, A., & Cresko, W. A. (2013). Stacks: an analysis tool set for population genomics. *Molecular Ecology*, *22*(11), 3124–3140. doi:10.1111/mec.12354

Costa, A. F., Siciliano, S., Emin-Lima, R., Martins, B. M. L., Sousa, M. E. M., Giarrizzo, T., & Júnior, J. de S. E. S. (2017). Stranding survey as a framework to investigate rare cetacean records of the north and north-eastern Brazilian coasts. *ZooKeys*, (688), 111–134. doi:10.3897/zookeys.688.12636

Danilewicz, D., Ott, P. H., Secchi, E., Andriolo, A., & Zerbini, A. (2013). Occurrence of the Atlantic spotted dolphin, Stenella frontalis, in southern Abrolhos Bank, Brazil. *Marine Biodiversity Records*, *6*. doi:10.1017/S1755267212000929

Davey, J. W., Hohenlohe, P. A., Etter, P. D., Boone, J. Q., Catchen, J. M., & Blaxter, M. L. (2011). Genome-wide genetic marker discovery and genotyping using next-generation sequencing. *Nature Reviews Genetics*, *12*(7), 499–510. doi:10.1038/nrg3012

Davis, R. W., Ortega-Ortiz, J. G., Ribic, C. A., Evans, W. E., Biggs, D. C., Ressler, P. H., … Würsig, B. (2002). Cetacean habitat in the northern oceanic Gulf of Mexico. *Deep-Sea Research Part I: Oceanographic Research Papers*, *49*(1), 121–142. doi:10.1016/S0967-0637(01)00035-8

do Amaral, K. B., Alvares, D. J., Heinzelmann, L., Borges-Martins, M., Siciliano, S., & Moreno, I. B. (2015). Ecological niche modeling of Stenella dolphins (Cetartiodactyla: Delphinidae) in the southwestern Atlantic Ocean. *Journal of Experimental Marine Biology and Ecology*, *472*, 166–179. doi:10.1016/j.jembe.2015.07.013

Dray, S., & Dufour, A.-B. (2007). The ade4 Package: Implementing the Duality Diagram for Ecologists. *JSS Journal of Statistical Software*, *22*(4). Retrieved from http://www.jstatsoft.org/

Excoffier, L., & Lischer, H. (2015). An Integrated Software Package for Population Genetics Data Analysis. *Evolutionary Bioinformatics Online*, *1*(1), 47–50. Retrieved from http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=2658868&tool=pmcentrez&rendertype=abstract

Fernández, R., Schubert, M., Vargas-Velázquez, A. M., Brownlow, A., Víkingsson, G. A., Siebert, U., … Orlando, L. (2016). A genomewide catalogue of single nucleotide polymorphisms in white-beaked and Atlantic white-sided dolphins. *Molecular Ecology Resources*, *16*(1), 266–276. doi:10.1111/1755-0998.12427

Foote, A. D., Vijay, N., Ávila-Arcos, M. C., Baird, R. W., Durban, J. W., Fumagalli, M., … Wolf, J. B. W. (2016). Genome-culture coevolution promotes rapid divergence of killer whale ecotypes. *Nature Communications*, *7*. doi:10.1038/ncomms11693

Freitas, C. M. A., Dellinger, T., & Reiner, F. (1989). First record of Atlantic spotted dolphins, Stenella frontalis G. Cuvier, 1829 (Cetacea: Delphinidae) from the Archipelago of Madeira. *Bocagiana*, *190*, 1–5.

Frichot, E., Mathieu, F., Trouillon, T., Bouchard, G., & François, O. (2014). Fast and efficient estimation of individual ancestry coefficients. *Genetics*, *196*(4), 973–83. doi:10.1534/genetics.113.160572

Green, M. L., Herzing, D. L., & Baldwin, J. D. (2007). Noninvasive methodology for the sampling and extraction of DNA from free-ranging Atlantic spotted dolphins (Stenella frontalis). *Molecular Ecology Notes*, *7*(6), 1287–1292. doi:10.1111/j.1471-8286.2007.01858.x

Hancock-Hanser, B. L., Frey, A., Leslie, M. S., Dutton, P. H., Archer, F. I., & Morin, P. A. (2013). Targeted multiplex next-generation sequencing: Advances in techniques of mitochondrial and nuclear DNA sequencing for population genomics. *Molecular Ecology Resources*, *13*(2), 254–268. doi:10.1111/1755-0998.12059

Hauser, L., & Carvalho, G. R. (2008). Paradigm shifts in marine fisheries genetics: Ugly hypotheses slain by beautiful facts. *Fish and Fisheries*. doi:10.1111/j.1467-2979.2008.00299.x

Huang, H., & Knowles, L. L. (2016). Unforeseen Consequences of Excluding Missing Data from Next-Generation Sequences: Simulation Study of RAD Sequences. *Systematic Biology*, *65*(3), 357–365. doi:10.1093/sysbio/syu046

Jefferson, T. A., Curry, B. E., Leatherwood, S., & Powell, J. A. (1997). Dolphins and porpoises of West Africa: a review of records (Cetacea: Delphinidae, Phocoenidae). *Mammalia*, *61*(1), 87–108. doi:10.1515/mamm.1997.61.1.87

Jombart, T., & Ahmed, I. (2011). adegenet 1.3-1: new tools for the analysis of genome-wide SNP data. *Bioinformatics*, *27*(21), 3070–3071. doi:10.1093/bioinformatics/btr521

Jombart, T., & Collins, C. (2015). A tutorial for Discriminant Analysis of Principal Components (DAPC) using adegenet 2.0.0. Retrieved from http://adegenet.r-forge.r-project.org/files/tutorial-dapc.pdf

Jombart, T., Devillard, S., & Balloux, F. (2010). Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. *BMC Genetics*, *11*(1), 94. doi:10.1186/1471-2156-11-94

Kelley, J. L., Brown, A. P., Therkildsen, N. O., & Foote, A. D. (2016). The life aquatic: advances in marine vertebrate genomics. *Nature Reviews Genetics*, *17*(9), 523–534. doi:10.1038/nrg.2016.66

Kjerfve, B., Ribeiro, C. H. A., Dias, G. T. M., Filippo, A. M., & Quaresma, V. D. S. (1997). Oceanographic characteristics of an impacted coastal bay: Baía de Guanabara, Rio de Janeiro, Brazil. *Continental Shelf Research*, *17*(13), 1609–1643. doi:10.1016/S0278-4343(97)00028-9

Kopelman, N. M., Mayzel, J., Jakobsson, M., Rosenberg, N. A., & Mayrose, I. (2015). Clumpak : a program for identifying clustering modes and packaging population structure inferences across *K*. *Molecular Ecology Resources*, *15*(5), 1179–1191. doi:10.1111/1755-0998.12387

Leslie, M. S., & Morin, P. A. (2016). Using Genome-Wide SNPs to Detect Structure in High-Diversity and Low-Divergence Populations of Severely Impacted Eastern Tropical Pacific Spinner (Stenella longirostris) and Pantropical Spotted Dolphins (S. attenuata). *Frontiers in Marine Science*, *3*, 253. doi:10.3389/fmars.2016.00253

Leslie, M. S., & Morin, P. A. (2018). Structure and phylogeography of two tropical predators, spinner (*Stenella longirostris*) and pantropical spotted (*S. attenuata*) dolphins, from SNP data. *Royal Society Open Science*, *5*(4), 171615. doi:10.1098/rsos.171615

Li, H., & Durbin, R. (2009). Fast and accurate short read alignment with Burrows-Wheeler transform. *Bioinformatics*, *25*(14), 1754–1760. doi:10.1093/bioinformatics/btp324

McCormack, J. E., Hird, S. M., Zellmer, A. J., Carstens, B. C., & Brumfield, R. T. (2013). Applications of next-generation sequencing to phylogeography and phylogenetics. *Molecular Phylogenetics and Evolution*, *66*(2), 526–538. doi:10.1016/J.YMPEV.2011.12.007

Méndez-Fernandez, P., Taniguchi, S., Santos, M. C. O., Cascão, I., Quérouil, S., Martín, V., … Montone, R. C. (2018). Contamination status by persistent organic pollutants of the Atlantic spotted dolphin (Stenella frontalis) at the metapopulation level. *Environmental Pollution*, *236*, 785–794. doi:10.1016/J.ENVPOL.2018.02.009

Möller, L., Valdez, F. P., Allen, S., Bilgmann, K., Corrigan, S., & Beheregaray, L. B. (2011). Fine-scale genetic structure in short-beaked common dolphins (Delphinus delphis) along the East Australian Current. *Marine Biology*, *158*(1), 113–126. doi:10.1007/s00227-010-1546-x

Moreno, I. B., Zerbini, A. N., Danilewicz, D., De Oliveira Santos, M. C., Simões-Lopes, P. C., Lailson-Brito, J., & Azevedo, A. F. (2005). Distribution and habitat characteristics of dolphins of the genus Stenella (Cetacea: Delphinidae) in the southwest Atlantic Ocean. *Marine Ecology Progress Series*, *300*, 229–240. doi:10.3354/meps300229

Morin, P. A., Martien, K. K., & Taylor, B. L. (2009). Assessing statistical power of SNPs for population structure and conservation studies. *Molecular Ecology Resources*, *9*(1), 66–73. doi:10.1111/j.1755-0998.2008.02392.x

Morin, P. A., Parsons, K. M., Archer, F. I., Ávila-Arcos, M. C., Barrett-Lennard, L. G., Dalla Rosa, L., … Foote, A. D. (2015). Geographic and temporal dynamics of a global radiation and diversification in the killer whale. *Molecular Ecology*, *24*(15), 3964–3979. doi:10.1111/mec.13284

Moritz, C. (1994). Defining "Evolutionarily Significant Units" for conservation. *Trends in Ecology & Evolution*, *9*, 373–375. Retrieved from https://ac-els-cdn-

com.proxy.lib.umich.edu/0169534794900574/1-s2.0-0169534794900574-main.pdf?_tid=16d06d4f-2e6f-4a10-b957-65d1d00c95f0&acdnat=1528573864_ee655a74f03ad45f85738ca041bc9fe4

Mullin, K. D., & Fulling, G. L. (1998). Abundance of cetaceans in the southern U.S. North Atlantic Ocean during summer. *Fishery Bulletin*, *101*(3), 603–613. Retrieved from http://aquaticcommons.org/15150/

Norris, R. D. (2000). Pelagic species diversity, biogeography, and evolution. *Paleobiology*, *26*, 236–258. doi:10.1666/0094-8373(2000)26[236:PSDBAE]2.0.CO;2

Palacios, D., Baumgartner, M., Laidre, K., & Gregr, E. (2013). Beyond correlation: integrating environmentally and behaviourally mediated processes in models of marine mammal distributions. *Endangered Species Research*, *22*(3), 191–203. doi:10.3354/esr00558

Perrin, W. F. (2009). Atlantic spotted dolphin: Stenella frontalis. In *Encyclopedia of Marine Mammals* (pp. 54–56). doi:10.1016/B978-0-12-373553-9.00014-6

Perrin, W. F., Mitchell, E. D., Mead, J. G., Caldwell, D. K., Caldwell, M. C., van Bree, P. J. H., & Dawbin, W. H. (1987). Revision of the Spotted dolphins, Stenella spp. *Marine Mammal Science*, *3*(2), 99–170. doi:10.1111/j.1748-7692.1987.tb00158.x

Perrin, W. F., Rosel, P. E., & Cipriano, F. (2013). How to contend with paraphyly in the taxonomy of the delphinine cetaceans? *Marine Mammal Science*, *29*(4), n/a-n/a. doi:10.1111/mms.12051

Peterson, B. K., Weber, J. N., Kay, E. H., Fisher, H. S., & Hoekstra, H. E. (2012). Double Digest RADseq: An Inexpensive Method for De Novo SNP Discovery and Genotyping in Model and Non-Model Species. *PLoS ONE*, *7*(5), e37135. doi:10.1371/journal.pone.0037135

Purcell, S., Neale, B., Todd-Brown, K., Thomas, L., Ferreira, M. A. R., Bender, D., … Sham, P. C. (2007). PLINK: A Tool Set for Whole-Genome Association and Population-Based Linkage Analyses. *The American Journal of Human Genetics*, *81*(3), 559–575. doi:10.1086/519795

Quérouil, S., Freitas, L., Cascão, I., Alves, F., Dinis, A., Almeida, J. R., … Santos, R. S. (2010). Molecular insight into the population structure of common and spotted dolphins inhabiting the pelagic waters of the Northeast Atlantic. *Marine Biology*, *157*(11), 2567–2580. doi:10.1007/s00227-010-1519-0

Redfern, J. V, Ferguson, M. C., Becker, E. A., Hyrenbach, K. D., Good, C. P., Barlow, J., … Ballance, L. T. (2006). Techniques for cetacean–habitat modeling.

Reeves, R., McClellan, K., & Werner, T. (2013). Marine mammal bycatch in gillnet and other entangling net fisheries, 1990 to 2011. *Endangered Species Research*, *20*(1), 71–97. doi:10.3354/esr00481

Rosel, P. E. (2003). PCR-based sex determination in Odontocete cetaceans. *Conservation Genetics*, *4*, 647–649. Retrieved from https://link-springer-com.proxy.lib.umich.edu/content/pdf/10.1023/A:1025666212967.pdf

Santos, M. C. de O., Figueiredo, G. C. e, Bressem, M.-F. Van, Santos, M. C. de O., Figueiredo, G. C. e, & Bressem, M.-F. Van. (2018). Cetaceans using the marine protected area of "Parque Estadual Marinho da Laje de Santos", Southeastern Brazil. *Brazilian Journal of Oceanography*, *65*(4), 605–613. doi:10.1590/s1679-87592017130606504

Sellas, A. B., Wells, R. S., & Rosel, P. E. (2005). Mitochondrial and nuclear DNA analyses reveal fine scale geographic structure in bottlenose dolphins (Tursiops truncatus) in the Gulf of Mexico. *Conservation Genetics*, *6*(5), 715–728. doi:10.1007/s10592-005-9031-7

Stamatakis, A. (2014). RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, *30*(9), 1312–1313. doi:10.1093/bioinformatics/btu033

Tajima, F. (1983). Evolutionary relationship of DNA sequences in finite populations. *Genetics*, *105*(2).

Taylor, B. L. (1997). Defining "populations" to meet management objectives for marine mammals. *Marine Mammal Science*, *Special Pu*, 49–65. Retrieved from https://pdfs.semanticscholar.org/43f0/d33d1b5e1ac10959d9021989b0b9ff6fe6a2.pdf

Taylor, B. L., Archer, F. I., Martien, K. K., Rosel, P. E., Hancock-Hanser, B. L., Lang, A. R., … Baker, C. S. (2017). Guidelines and quantitative standards to improve consistency in cetacean subspecies and species delimitation relying on molecular genetic data. *Marine Mammal Science*, *33*, 132–155. doi:10.1111/mms.12411

Viricel, A., & Rosel, P. E. (2014). Hierarchical population structure and habitat differences in a highly mobile marine species: The Atlantic spotted dolphin. *Molecular Ecology*, *23*(20), 5018–5035. doi:10.1111/mec.12923

Wang, C., Zöllner, S., & Rosenberg, N. A. (2012). A Quantitative Comparison of the Similarity between Genes and Geography in Worldwide Human Populations. *PLoS Genetics*, *8*(8), e1002886. doi:10.1371/journal.pgen.1002886

Waples, R. (1991). Pacific Salmon, Oncorhynchus spp., and the Definition of "Species" Under the Endangered Species Act. *Marine Fisheries Review*, *53*(3), 11–22. Retrieved from http://aquaticcommons.org/9917/

Waring, G. T., Josephson, E., Maze-Foley, K., & Rosel, P. E. (2009). Altantic and Gulf of Mexico Marine Mammal Stock Assessments 2009. NOAA Tech Memo NMFS NE. Retrieved from https://www.nefsc.noaa.gov/nefsc/publications/tm/tm213/

Weir, C. R. (2010). A review of cetacean occurrence in west African waters from the Gulf of Guinea to Angola. *Mammal Review*. doi:10.1111/j.1365-2907.2009.00153.x

Tables

Table 1. Population genetics summary statistics for Atlantic spotted dolphin based Dataset 2 (83,512 SNPs in 47,878 loci). Numbers of females (F), males (M) and I (indetermined sex) is provided by putative population.

Putative populations	Samples	Sex (M/F/I)	Private Alleles
SBB outer shelf	15	10/4/1	4047
SBB inner/midshelf	43	21/18/4	14700
Caribbean – La Guajira	5	2/1/2	4815
Canary Islands	7	4/3/0	8163

Variant positions			
Putative populations	**Exp Het (mean/var/SE)**	**Obs Het (mean/var/SE)**	**π (mean/var/SE)**	**N_e**
SBB outer shelf	0.1355/0.0267/0.0006	0.1303/0.028/0.0006	0.1412/0.0292/0.0006	396629
SBB inner/midshelf	0.1374/0.025/0.0005	0.1251/0.0215/0.0005	0.1398/0.0259/0.0006	392697
Caribbean – La Guajira	0.1197/0.0296/0.0006	0.1284/0.0445/0.0007	0.1382/0.0406/0.0007	388202
Canary Islands	0.1229/0.0282/0.0006	0.1357/0.0476/0.0008	0.1473/0.0461/0.0008	413764

Variant and Fixed positions			
Putative populations	**Exp Het (mean/var/SE)**	**Obs Het (mean/var/SE)**	**π (mean/var/SE)**	**N_e**
SBB outer shelf	0.0017/0.0006/0	0.0016/0.0006/0	0.0018/0.0006/0	5056
SBB inner/midshelf	0.0017/0.0005/0	0.0016/0.0005/0	0.0017/0.0006/0	4775
Caribbean – La Guajira	0.0015/0.0005/0	0.0016/0.0008/0	0.0017/0.0007/0	4775
Canary Islands	0.0015/0.0005/0	0.0017/0.0008/0	0.0018/0.0008/0	5056

Table 2. Pairwise population genetic differentiation statistics for Atlantic spotted dolphin calculated in StrataG. F_{ST} is below the diagonal and p-value is above. Comparisons significantly different from zero ($p < 0.05$) are in bold. SBB, Southeast Brazilian Bight. ***** p-values were not calculated.

	Arlequin Analysis			
Putative Populations	SBB outer shelf	SBB inner/midshelf	Caribbean – La Guajira	Canary Islands
SBB outer shelf	-	0.9651	0.0001	0.9999
SBB inner/midshelf	-0.01629	-	0.000001	0.52916
Caribbean – La Guajira	**0.07902**	**0.11795**	-	0.72527
Canary Islands	-0.09723	-0.00263	-0.2581	-
	StrataG Analysis			
Putative Populations	SBB outer shelf	SBB inner/midshelf	Caribbean – La Guajira	Canary Islands
SBB outer shelf	-	*	*	*
SBB inner/midshelf	0.0102	-	*	*
Caribbean – La Guajira	0.0651	0.0696	-	*
Canary Islands	0.1026	0.10976		-

Figures
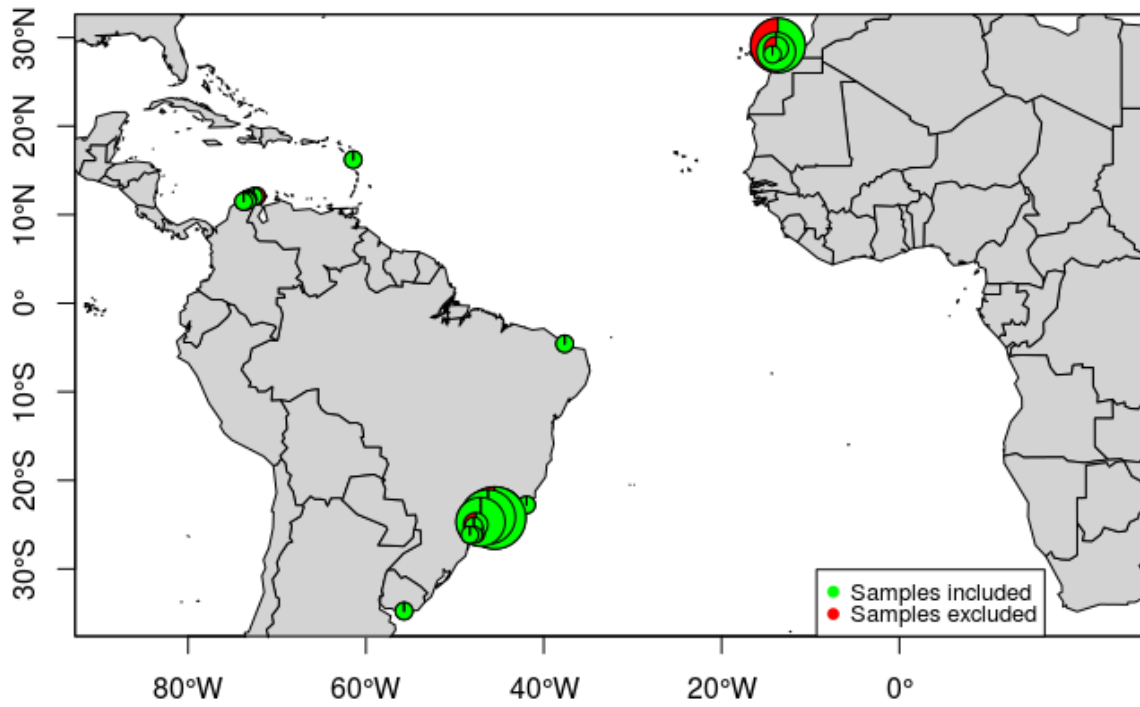
Figure 1. Sampling of Atlantic spotted dolphin. The size of circle is proportional of number of samples collected at each geographic coordinates. Green circles indicated samples that were used throughout analyses, while red circles represent samples that were removed at some point of the bioinformatic pipeline or that were not considered in the analysis due to low DNA quantity/quality.
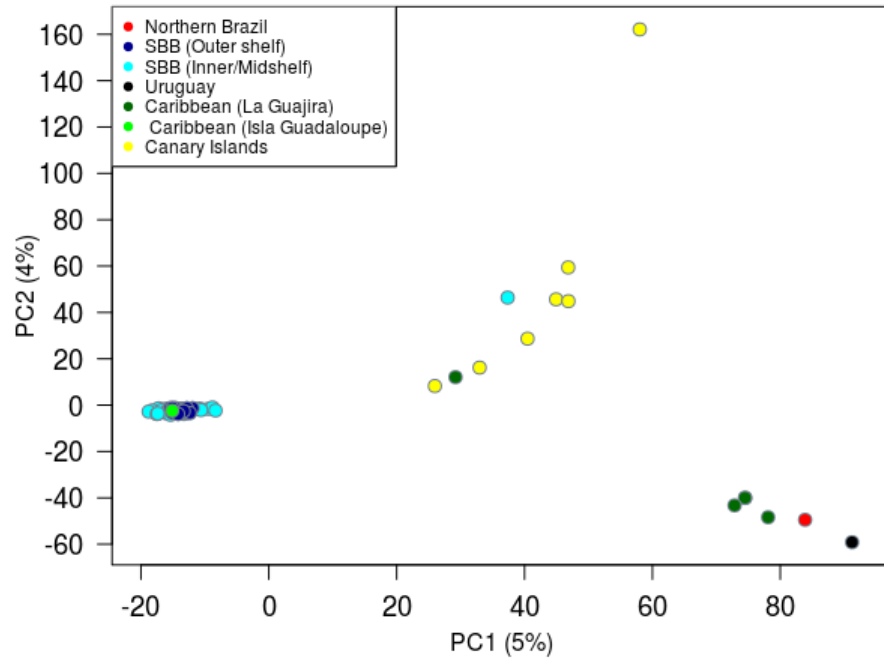
Figure 2. Principal Component Analysis (PCA) performed with Dataset 1. Different colours representing the different sampling locations as described in the figure insert.
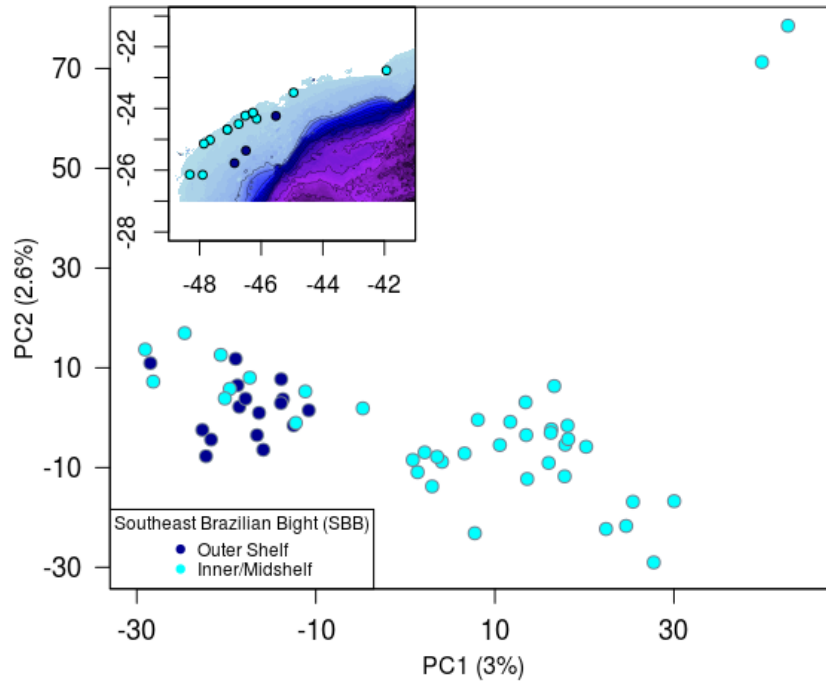
Figure 3. Principal Component Analysis (PCA) performed with Dataset 1, but just considering samples from southeastern Brazil. Different colours represent sampling locations. Insert map indicates the sampling locations in the Southeast Brazilian Bight (SBB).
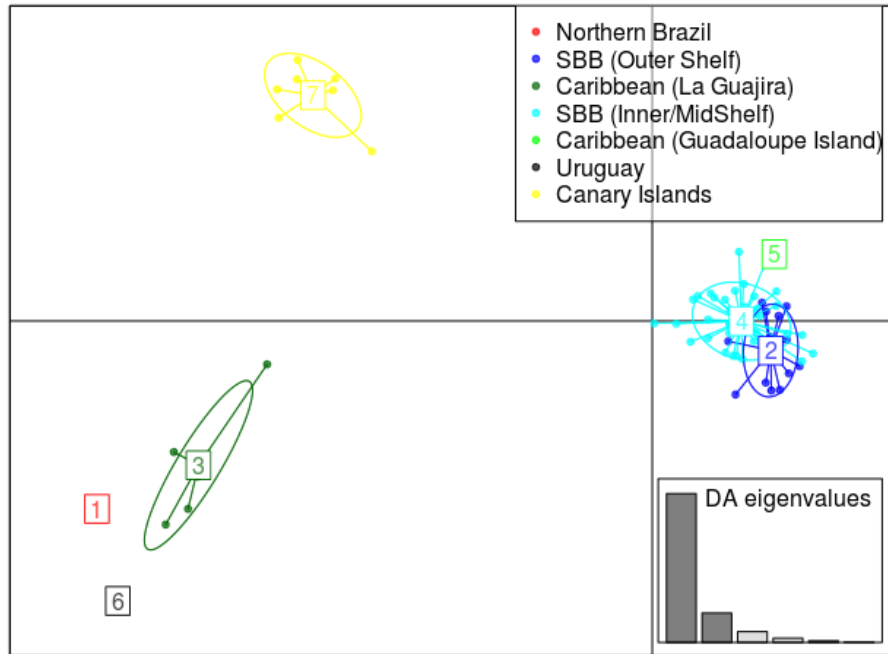
Figure 4. Genomic variation across individuals and populations of Atlantic spotted dolphin across Atlantic Ocean. Scatter plot of individuals based on the first two eigenvalues (created from n/3) of the DAPC. The obtained graph represents the individuals as dots and the groups as inertia ellipses, which are colored by sampling locations. Eigenvalues of the analysis are displayed in inset.
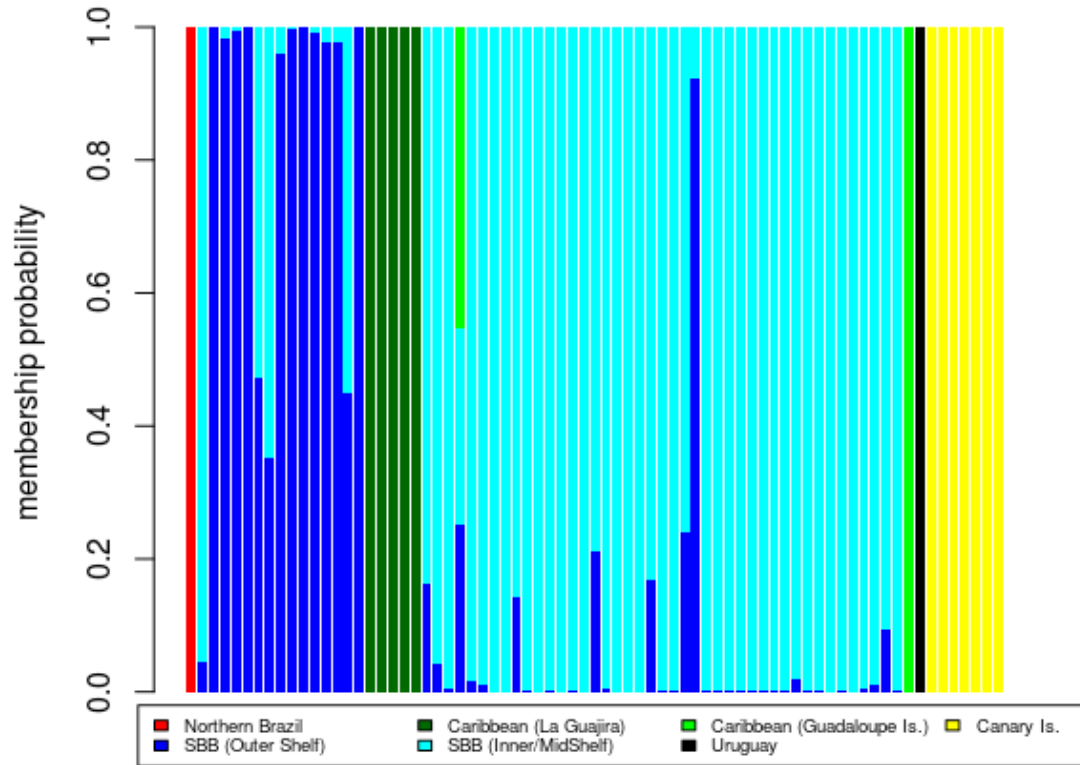
Figure 5. A STRUCTURE-like graphical representation of membership probabilities to have a global picture of the clusters composition based on the DAPC performed with n.pca equal to 24 (n/3). Colors represent sampling locations and individuals were colored according the probability to belong to determined cluster.
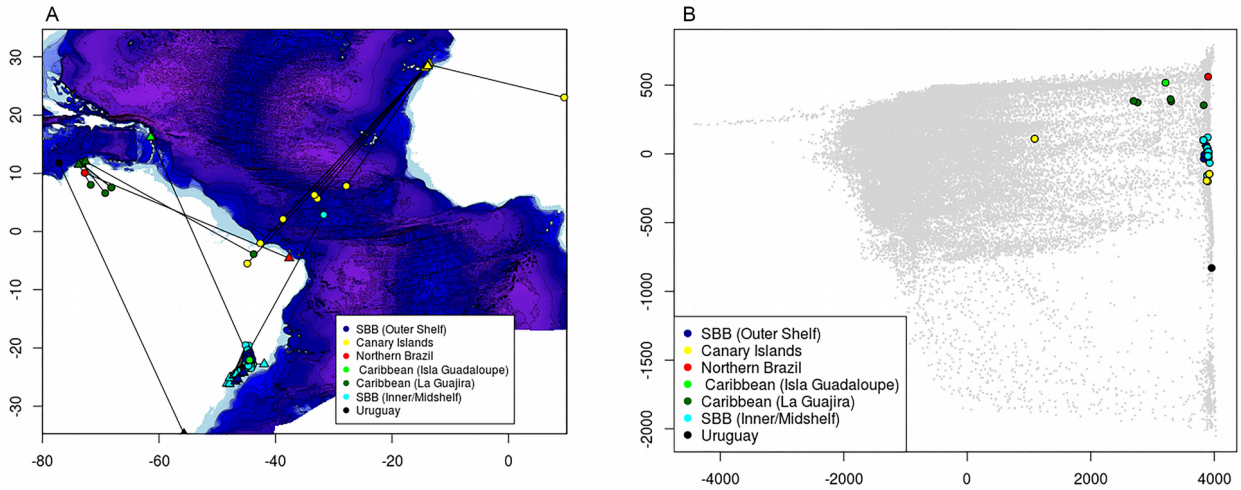
Figure 6. Representation of Procrustes analyses. A, the relationship between genetic and geography is showed. B, the relationship between the residuals of the previous Procrustes and environment are represented in the environmental space by the first two PCs. Bathymetry and Mean Annual STT were the environmental variables that mainly contribute to the PC 1 and PC 2, respectively. Samples were colored according to sampling area.
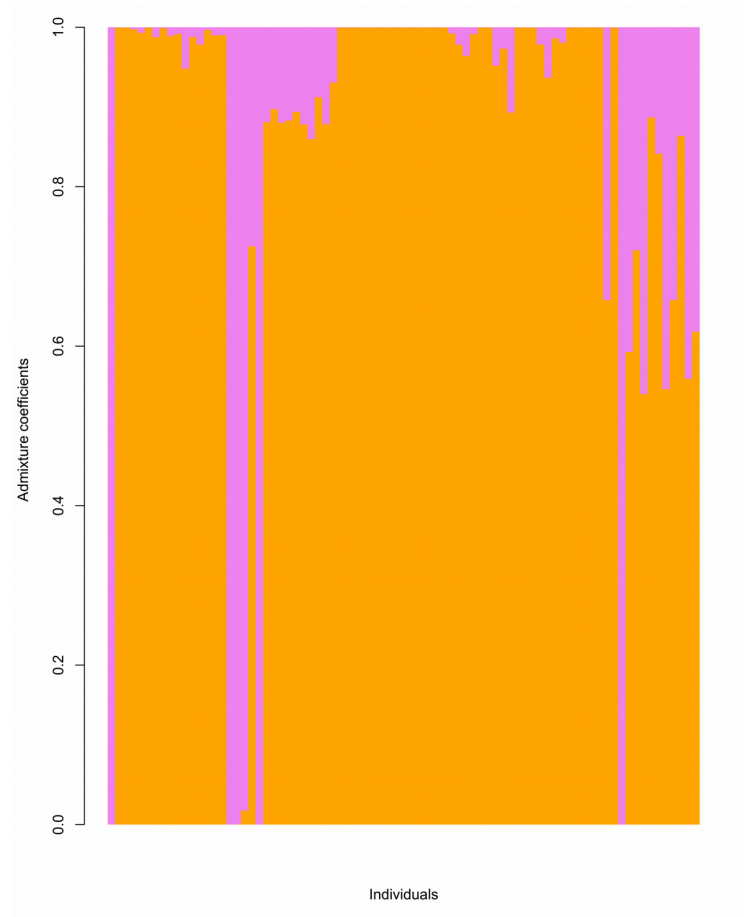
Figure 7. Graphical representation of ancestry estimates obtained for Atlantic spotted dolphin Dataset 2 (K = 8). Shown are estimated ancestry coefficients using sNMF with K=2 (cross-entropy = 0.44).
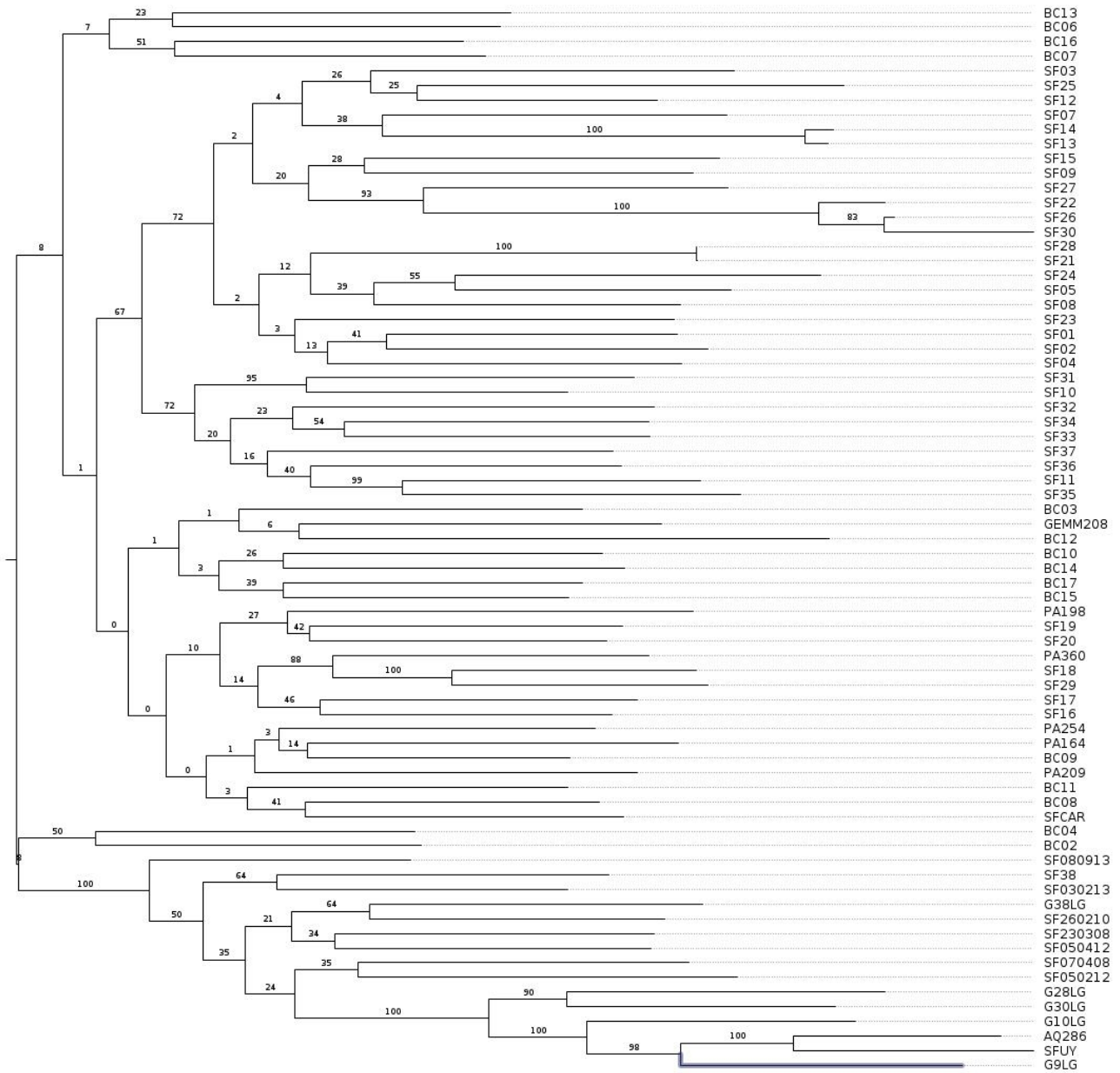
Figure 8. Topology of Maximum Likelihood tree for Atlantic spotted dolphin using Dataset 3. Nodes are labelled with bootstrap support. SBB inner/midshelf individuals: codes GEMM, PA and SF; SBB outher shelf invidtuals: BC; Caribbean – La Guajira individuals: G; Caribbean – Guadaloupe Island individual: SFCAR; Canary Islands: SF followed by six digits.

Supplementary Tables

Table S1. Sampling and genomic sequences per individual pre- and post-processing in STACKS; * marks individuals removed from the analysis because of poor sequence quality, large numbers of missing loci, or a single individual in that population.

Sampling Location	Sample	Type	Sex	Longitude	Latitude	Retained Reads	Mean Coverage	Maximum Coverage
Northern Brazil	AQ_286	Strading	M	-37.6450917	-4.58076944	3253358	15.95	446
SBB Outer Shelf	BC_02	Biopsy	M	-46.86639	-25.76958	3624265	17.67	348
SBB Outer Shelf	BC_03	Biopsy	M	-46.49609	-25.365	2565923	16.11	406
SBB Outer Shelf	BC_04	Biopsy	M	-46.49609	-25.365	2912275	12.81	643
SBB Outer Shelf	BC_05*	Biopsy	F	-45.5231	-24.2412	14137	-	-
SBB Outer Shelf	BC_06	Biopsy	M	-45.5231	-24.2412	2812841	16.73	65
SBB Outer Shelf	BC_07	Biopsy	M	-45.5231	-24.2412	3068807	18.27	576
SBB Outer Shelf	BC_08	Biopsy	F	-45.5231	-24.2412	2275299	12.72	581
SBB Outer Shelf	BC_09	Biopsy	M	-45.5231	-24.2412	2828635	15.61	118
SBB Outer Shelf	BC_10	Biopsy	M	-45.5231	-24.2412	3113945	14.59	309
SBB Outer Shelf	BC_11	Biopsy	F	-45.5231	-24.2412	3096880	12.57	645
SBB Outer Shelf	BC_12	Biopsy	M	-45.5231	-24.2412	3728673	15.05	549
SBB Outer Shelf	BC_13	Biopsy	M	-45.5231	-24.2412	3150348	14.82	488
SBB Outer Shelf	BC_14	Biopsy	M	-45.5231	-24.2412	2936780	13.38	851
SBB Outer Shelf	BC_15	Biopsy	F	-45.5231	-24.2412	2775644	13.93	631
SBB Outer Shelf	BC_16	Biopsy	F	-45.5231	-24.2412	2734000	13.51	398
SBB Outer Shelf	BC_17	Biopsy	I	-45.5231	-24.2412	2959420	14.55	1251
Caribbean – La Guajira	G10_LG	Biopsy	F	-73.3044444	11.6972222	3608553	13.8	589
Caribbean – La Guajira	G24_LG*	Biopsy	I	-73.3027778	11.6880556	3246338	-	-
Caribbean – La Guajira	G28_LG	Biopsy	I	-73.3602778	11.5563889	3698714	13.77	790
Caribbean – La Guajira	G30_LG	Biopsy	I	-73.2808333	11.7858333	3248635	11.17	302
Caribbean – La Guajira	G38_LG	Biopsy	M	-72.6161111	12.0461111	513954	5.19	936
Caribbean – La Guajira	G40_LG*	Biopsy	I	-72.2988889	12.0894444	1317152	-	-
Caribbean – La Guajira	G9_LG	Biopsy	M	-73.7586111	11.4844444	2839831	12.35	220
SBB Inner/Midshelf	GEMM_027*	Strading	M	-42.026081	-22.957836	289	-	-
SBB Inner/Midshelf	GEMM_149*	Strading	F	-42.35629	-22.935298	95779	3.32	788
SBB Inner/Midshelf	GEMM_208	Strading	M	-41.93508	-22.765293	556674	4.95	768
SBB Inner/Midshelf	GEMM_219*	Strading	F	-41.94424	-22.7562	12570	-	-
SBB Inner/Midshelf	GEMM_305*	Strading	F	-42.422808	-22.93448	119272	3.47	695
SBB Inner/Midshelf	GEMM_316*	Strading	M	-40.57866	-21.20285	271	-	-
SBB Inner/Midshelf	PA_164	Incidental Capture	F	-48.3180556	-26.1411111	443314	4.69	653
SBB Inner/Midshelf	PA_198	Incidental Capture	F	-47.6630556	-25.0205556	555510	4.82	622
SBB Inner/Midshelf	PA_199*	Incidental Capture	M	-47.6630556	-25.0205556	257186	4	486
SBB Inner/Midshelf	PA_205*	Incidental Capture	M	-47.6108333	-26.0386111	220039	3.88	1088
SBB Inner/Midshelf	PA_209	Incidental Capture	F	-47.9	-26.15	796756	6.03	825
SBB Inner/Midshelf	PA_254	Incidental Capture	M	-	-	337896	4.39	799
SBB Inner/Midshelf	PA_360	Incidental Capture	F	-47.8612	-25.1417	1331820	7.52	244
SBB Inner/Midshelf	PA_365*	Incidental Capture	F	-47.793367	-25.485133	9397	-	-
SBB Inner/Midshelf	SF_01	Biopsy	M	-44.95	-23.483333	2137870	9.93	502
SBB Inner/Midshelf	SF_02	Biopsy	F	-44.95	-23.483333	1775982	9.32	539

SBB Inner/Midshelf	SF_03	Biopsy	M	-44.95	-23.483333	2292796	10.28	959
SBB Inner/Midshelf	SF_04	Biopsy	M	-44.95	-23.483333	2926753	11.86	668
SBB Inner/Midshelf	SF_05	Biopsy	M	-44.95	-23.483333	1666950	9.13	642
SBB Inner/Midshelf	SF_07	Biopsy	M	-44.95	-23.483333	1937426	9.58	934
SBB Inner/Midshelf	SF_08	Biopsy	M	-44.95	-23.483333	2664738	11.56	152
SBB Inner/Midshelf	SF_09	Biopsy	M	-46.266667	-24.166667	2332106	10.66	609
SBB Inner/Midshelf	SF_10	Biopsy	F	-46.516667	-24.233333	2731074	10.26	937
SBB Inner/Midshelf	SF_11	Biopsy	M	-46.516667	-24.233333	2315917	10.1	777
SBB Inner/Midshelf	SF_12	Biopsy	F	-46.15	-24.333333	1531863	8.81	655
SBB Inner/Midshelf	SF_13	Biopsy	M	-46.15	-24.333333	1708493	10.7	378
SBB Inner/Midshelf	SF_14	Biopsy	F	-46.15	-24.333333	1657911	7.56	223
SBB Inner/Midshelf	SF_15	Biopsy	F	-46.15	-24.333333	2008064	10.16	851
SBB Inner/Midshelf	SF_16	Biopsy	F	-46.15	-24.333333	2749312	12	608
SBB Inner/Midshelf	SF_17	Biopsy	M	-46.15	-24.333333	2212398	12.31	889
SBB Inner/Midshelf	SF_18	Biopsy	F	-47.1	-24.683333	1827654	10.91	1066
SBB Inner/Midshelf	SF_19	Biopsy	F	-47.1	-24.683333	1599277	9.72	470
SBB Inner/Midshelf	SF_20	Biopsy	F	-47.1	-24.683333	2373687	11.45	407
SBB Inner/Midshelf	SF_21	Biopsy	F	-47.1	-24.683333	1167902	7.63	185
SBB Inner/Midshelf	SF_22	Biopsy	F	-47.1	-24.683333	1779901	10.11	643
SBB Inner/Midshelf	SF_23	Biopsy	M	-46.266667	-24.133333	740521	5.3	611
SBB Inner/Midshelf	SF_24	Biopsy	M	-46.266667	-24.133333	1331604	7.6	81
SBB Inner/Midshelf	SF_25	Biopsy	I	-46.266667	-24.133333	472856	4.57	1060
SBB Inner/Midshelf	SF_26	Biopsy	M	-46.266667	-24.133333	1353557	8.76	935
SBB Inner/Midshelf	SF_27	Biopsy	I	-46.266667	-24.133333	1559278	9.88	627
SBB Inner/Midshelf	SF_28	Biopsy	I	-46.266667	-24.133333	2711463	12.09	722
SBB Inner/Midshelf	SF_29	Biopsy	M	-46.266667	-24.133333	2059179	10.69	825
SBB Inner/Midshelf	SF_30	Biopsy	M	-46.266667	-24.133333	468914	4.73	819
SBB Inner/Midshelf	SF_31	Biopsy	M	-46.266667	-24.133333	1660803	9.27	450
SBB Inner/Midshelf	SF_32	Biopsy	I	-46.266667	-24.133333	1437312	8.69	179
SBB Inner/Midshelf	SF_33	Biopsy	M	-47.1	-24.683333	2091192	9.63	697
SBB Inner/Midshelf	SF_34	Biopsy	F	-47.1	-24.683333	1603593	8.42	351
SBB Inner/Midshelf	SF_35	Biopsy	F	-47.1	-24.683333	2578140	10.57	690
SBB Inner/Midshelf	SF_36	Biopsy	F	-46.733333	-24.5	1948266	9.22	707
SBB Inner/Midshelf	SF_37	Biopsy	M	-46.733333	-24.5	1784997	8.83	504
SBB Inner/Midshelf	SF_38	Biopsy	M	-46.733333	-24.5	823137	5.61	1415
SBB Inner/Midshelf	SF_39*	Biopsy	F	-46.733333	-24.5	3435868	-	-
Caribbean – Isla Guadaloupe	SF_CAR	Strading	F	-61.43091	16.20633	3457297	15.07	18
Uruguay	SF_UY	Strading	M	-55.71	-34.76	3467961	12.76	991
Canary Islands	SF030213	Strading	F	-13.835876	28.705264	876024	6.58	24
Canary Islands	SF050212	Strading	M	-	-	501974	5.31	586
Canary Islands	SF050412	Strading	M	-14.276971	28.094439	1184084	6.58	154
Canary Islands	SF060711	Strading	M	-13.743676	28.898027	86902	3.41	856
Canary Islands	SF061212	Strading	M	-13.609064	28.935655	156725	3.85	595
Canary Islands	SF070408	Strading	M	-13.633615	28.927775	1033526	6.89	24
Canary Islands	SF080913	Strading	F	-13.868572	28.745253	931994	6.51	444
Canary Islands	SF200315*	Strading	M	-	-	133	-	-
Canary Islands	SF200708*	Strading	F	-13.663621	29.111105	197477	3.55	703
Canary Islands	SF210212*	Strading	F	-	-	231	-	-
Canary Islands	SF230308	Strading	F	-	-	818156	6.54	1524

201

Canary Islands	SF260210	Strading	M	-13.861742	28.43191	737841	5.94	1193

Supplementary Figures
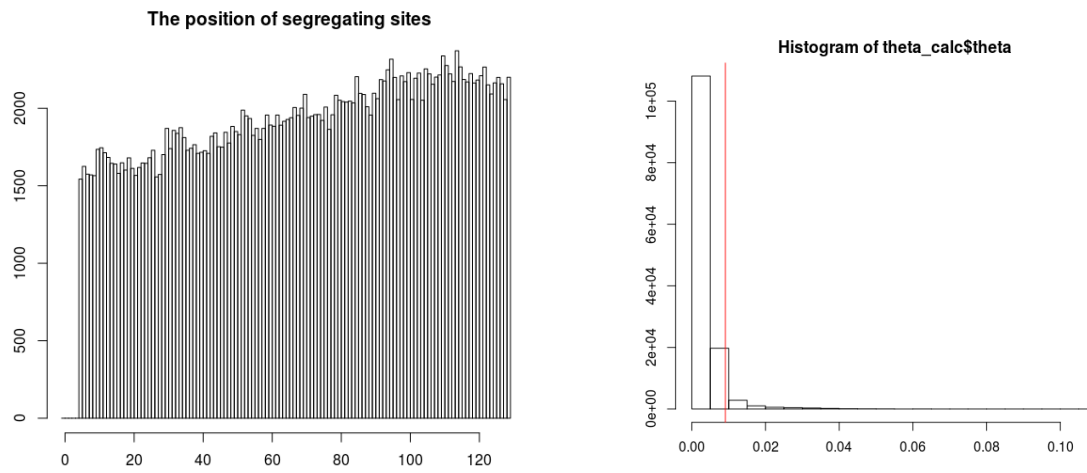
Figure S1. Summary of the frequency of segregating sites for each base-pair position of a locus (A), and the distribution of theta, θ, per loci (B), with the red line marking the θ-values in the 95 percentile that were excluded from analyses to avoid including variation likely reflective of sequencing and/or assembly errors.
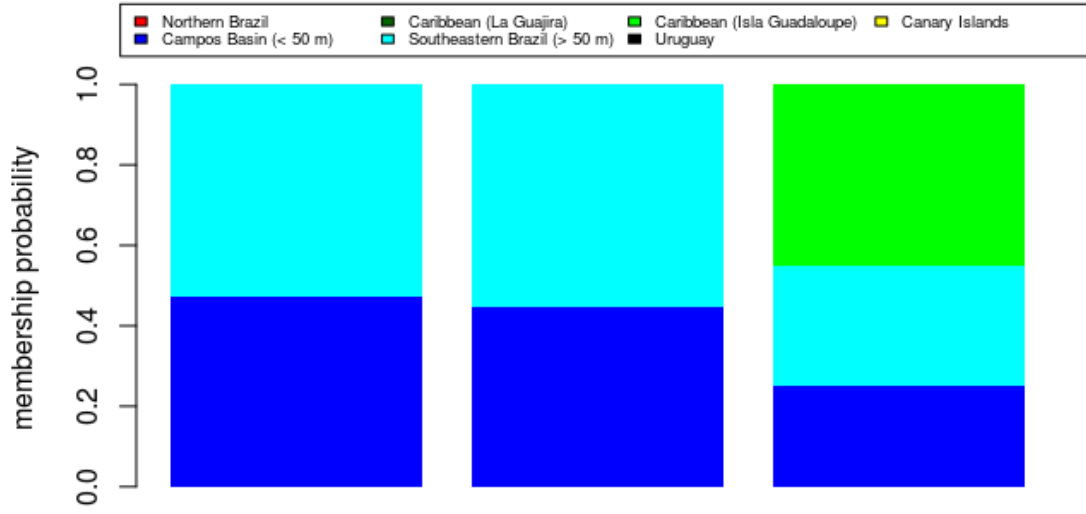
Figure S2. Representation of admixed individuals. Admixed individuals having no more than 60% of probability of membership in a single cluster.
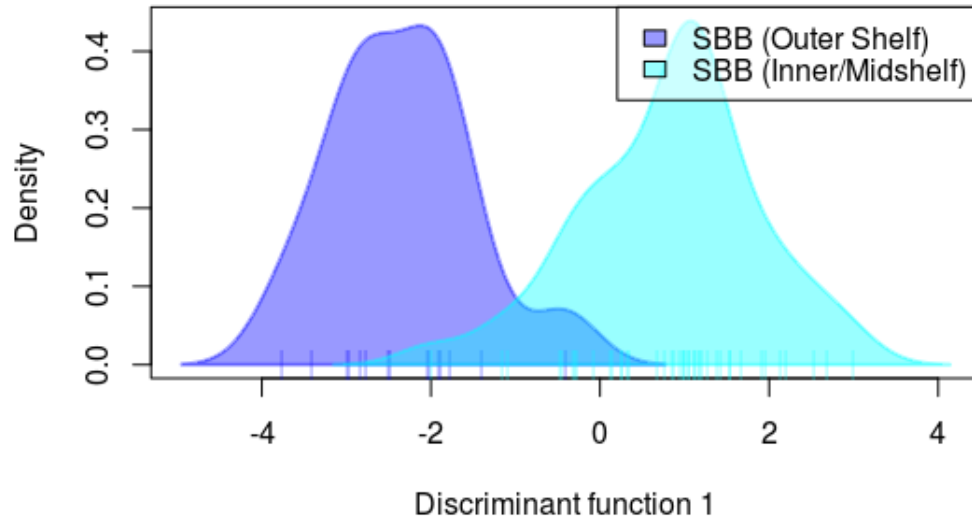
Figure S3. Genomic variation across individuals and populations of Atlantic spotted dolphin across Atlantic Ocean. Scatter plot of individuals based on the first two eigenvalues (created from optimum 15 principal components) of the DAPC. The obtained graph represents the individuals as dots and the groups as inertia ellipses, which are colored by sampling locations. Eigenvalues of the analysis are displayed in inset.
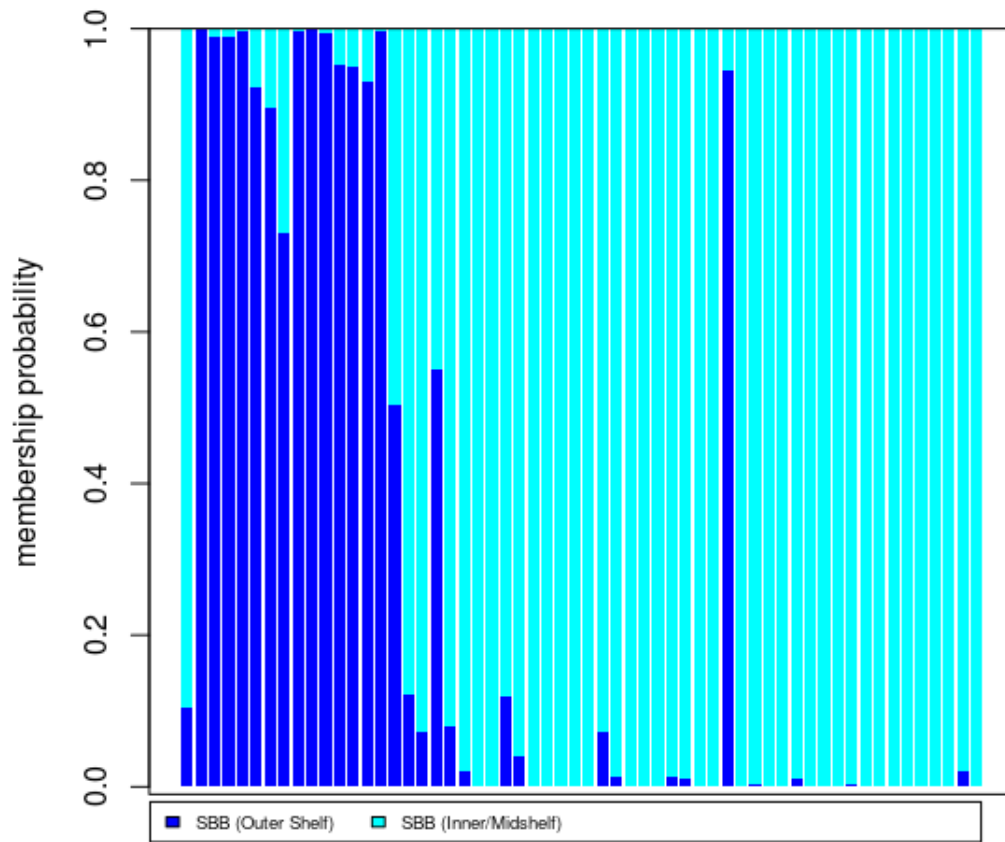
Figure S4. A STRUCTURE-like graphical representation of membership probabilities to have a global picture of the clusters composition based on the DAPC performed with optimum 15 principal components (n.pca=15). Colors represent sampling locations and individuals were colored according the probability to belong to determined cluster.
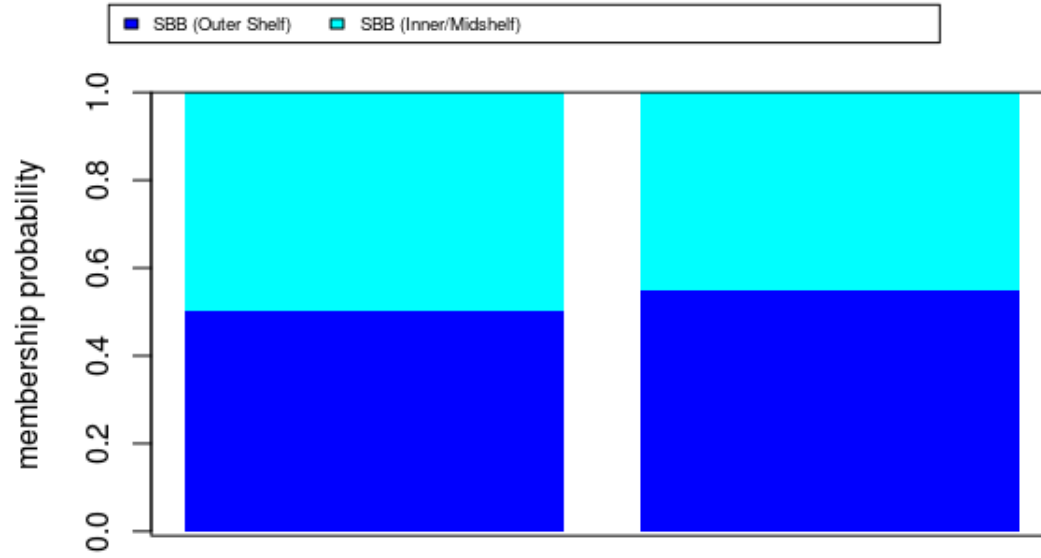
Figure S5. Representation of admixed individuals in the southeastern Brazil group. Admixed individuals having no more than 60% of probability of membership in a single cluster.
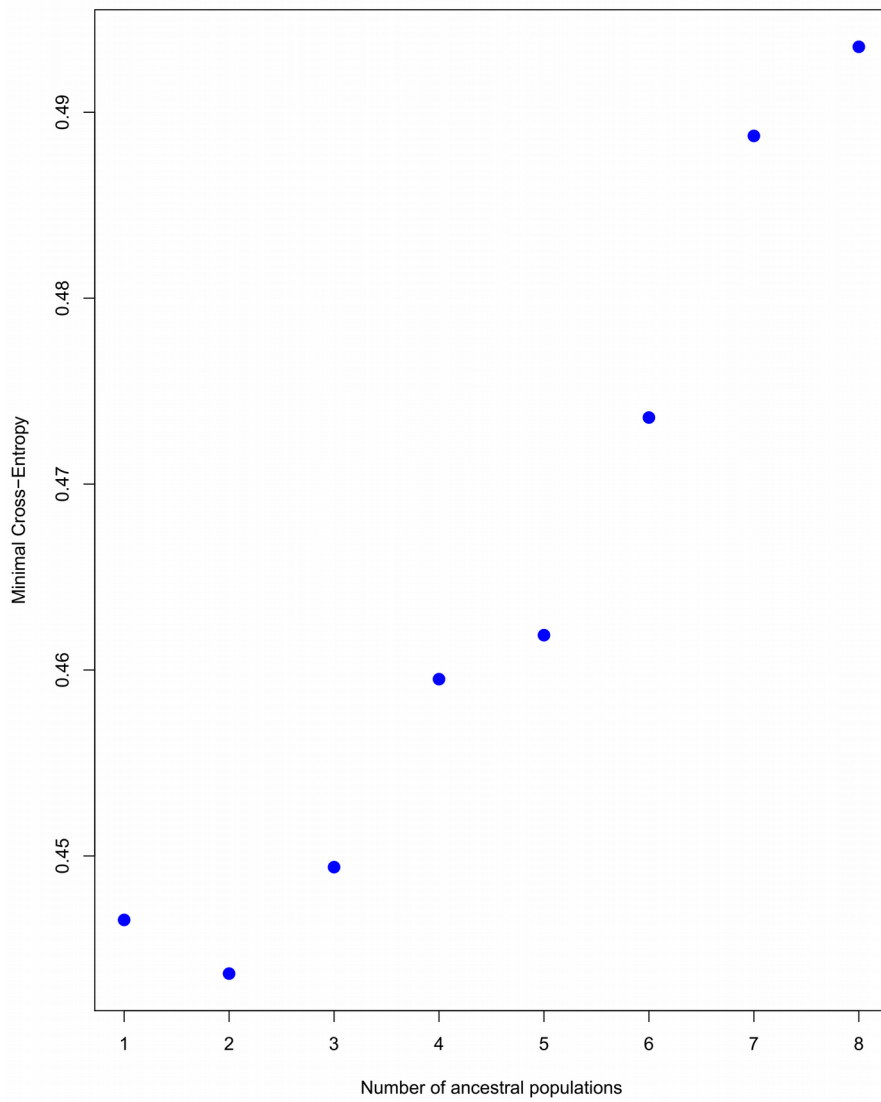
Figure S6. The most likely number of ancestral populations according minimal cross-entropy.
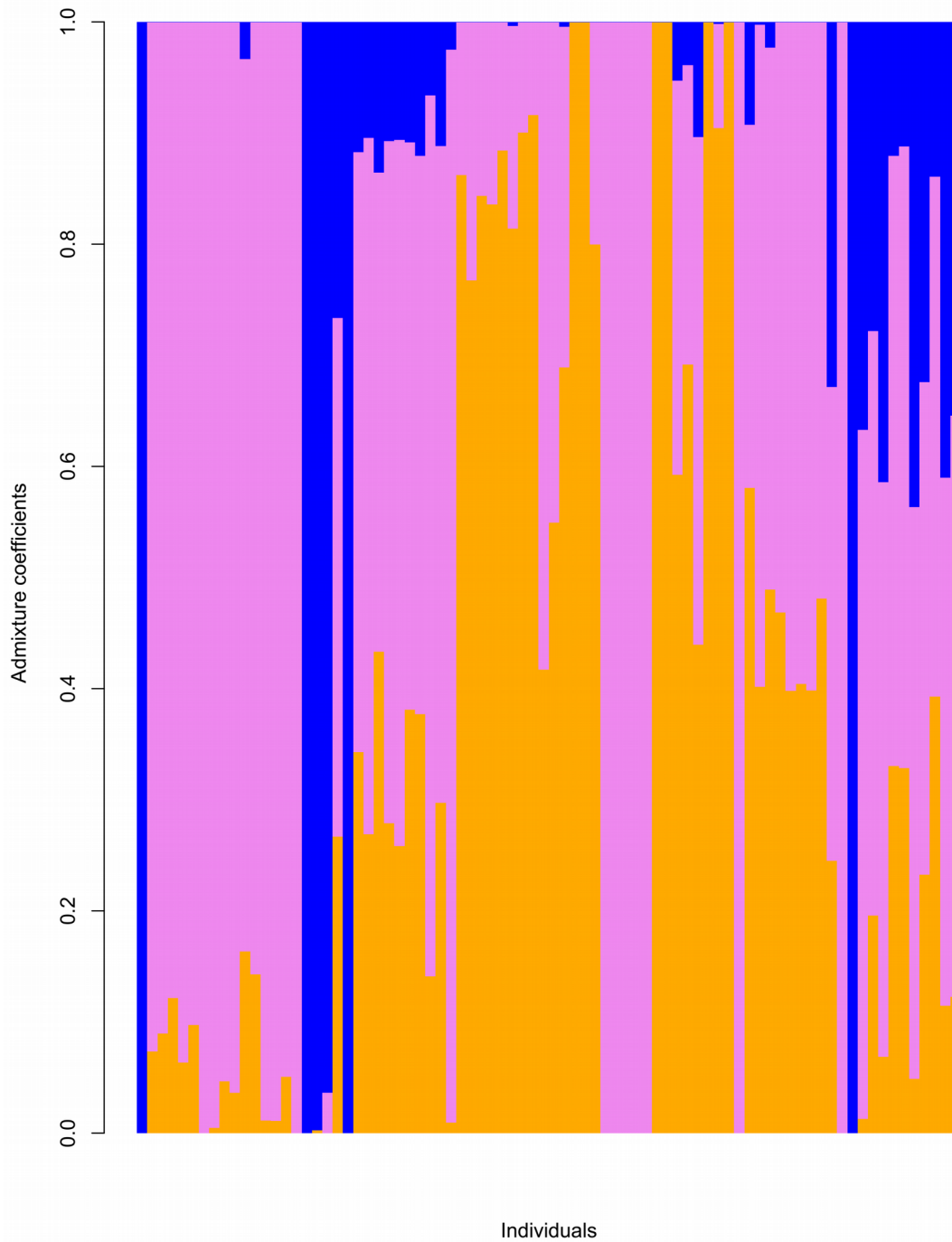
Figure S7. Graphical representation of ancestry estimates obtained for Atlantic spotted dolphin Dataset 2 (K = 8). Shown are estimated ancestry coefficients using sNMF with K=3 (cross-entropy = 0.45).

O padrão de distribuição de uma espécie é resultado de complexas interações biológicas com o ambiente. Em espécies que vivem no ambiente marinho é especialmente difícil reconhecer os fatores que limitam a distribuição de uma espécie, porque o ambiente parece ser 3-dimensionalmente contínuo e, portanto, as barreiras não são tão óbvias como no ambiente terrestre. No entanto, características hidrográficas (salinidade, turbidez, temperatura e produtividade) e topográficas (relevo, declividade, e extensão da plataforma continental) parecem ser os principais delimitadores da distribuição de espécies de cetáceos, que por serem predadores de topo de cadeia exibem grande capacidade de dispersão.

Nesta tese, a influência do ambiente marinho no padrão de distribição e estrutura genética de duas espécies de cetáceos com histórias de vida bastante diferentes, mas endêmicas do Oceano Atlântico e com padrões de distribuição semelhantes (i.e., a presença de hiatos de distribuição) no Brasil foram investigados. As espécies analisadas foram a franciscana (*Pontoporia blainvellei*) e o golfinho-pintado-do-Atlântico (*Stenella frontalis*). E, em relação as variáveis ambientais analisou-se especificamente batimetria, declividade, temperatura, salinindade, produtividade e turbidez da água. De forma geral, os estudos foram conduzidos em duas escalas: uma mais restrita, onde analisou-se toda distribuição da franciscana ao longo da costa brasileira, enquanto que os estudos com o golfinho-pintado-do-Atlântico incluiram praticamente toda distribuição da espécie no Oceano Atlântico.

Embora a distribuição da franciscana seja bem conhecida, os fatores ambientais que poderiam explicar a ausência da espécies em duas porções do sudeste do Brasil ainda

eram alvo de debate entre os especialistas. No Capítulo I, demonstramos que embora os hiatos apresentam condições que poderiam favorecer a presença da espécie, uma vez que fazem parte do seu nicho fundamental; no entanto, o hiato sul apresenta uma plataforma continental muita estreita, e o hiato norte embora não seja tão restrito em termos de extensão longitudinal da plataforma continental, exibe níveis de salinidade um pouco mais elevado do que os ambientes ocupados, de fato, pela franciscana. A plataforma estreita longitudinalmente pode intensificar relações ecológicas (como competição e predação), mas pode ter tido um efeito mais importante durante as trangressões e regressões marinhas registradas no Pleistoceno, por exemplo. Uma vez que a espécie exibe estruturação genética ao longo da sua distribuição e também diferenças morfológicas/ecológicas, pode-se concluir que este padrão de distribuição na costa brasileira seja também resultado da história evolutiva da espécie ao longo dos últimos 10 milhões de anos.

Em relação ao golfinho-pintado-do-Atlântico, embora muito se conheça sobre a distribuição da espécie e sua estruturação genética no Atlântico Norte, pouco ainda se sabia sobre a espécie no Atlântico Sul. No Brasil, a espécie não é registrada em uma faixa muito estreita da plataforma continental entre 6 e 18°S, e possivelmente, os golfinhos registrados ao sul de 18°S poderiam estar isolados dos demais que ocorrem ao longo do Oceano Atlântico.

No Capítulo II, sequências da região controle do mtDNA de praticamente toda a distribuição da espécie foram analisadas e confirmaram a existência de várias populações. Embora a utilização de apenas um marcador não seja o suficiente para determinar com confiabilidade a existência e limites destas populações, este estudo investigou pela

primeira vez um número grande de amostras obtidas em regiões de pouco conhecimento da espécie (Atlântico Sul Ocidental, Caribe e Ilhas Canárias). Além disso, observou-se que embora de forma não muito expressiva, esta estruturação genética pode ser resultado da influência de fatores como distâncias geográficas e ecológicas. No Capítulo III, marcadores genômicos foram utilizados para investigar a estrutura genética entre três áreas: Atlântico Sul Ocidental, Caribe e Canárias. No geral, os resultados indicam diferenciação ao longo destas áreas e, possivelmente, dois grupos distintos podem existir numa escala muito fina dentro do "*Southeast Brazilian Bight*", sendo um grupo associado a parte interna/média da plataforma continental e o outro associado a plataforma mais externa.

Os resultados obtidos ao longo destes três capítulos trazem à luz informações de grande relevância para futuros estudos sobre cetáceos no Brasil, podendo ser utilizadas principalmente em planos de manejo e conservação da franciscana e do golfinho-pintado-do-Atlântico, ao menos, em escala regional.

ANEXOS

JOURNAL OF EXPERIMENTAL MARINE BIOLOGY AND ECOLOGY

TABLE OF CONTENTS

DESCRIPTION

The *Journal of Experimental Marine Biology and Ecology* provides a forum for experimental ecological research on marine organisms in relation to their environment. Topic areas include studies that focus on biochemistry, physiology, behavior, genetics, and ecological theory. The main emphasis of the Journal lies in hypothesis driven experimental work, both from the laboratory and the field. Natural experiments or descriptive studies that elucidate fundamental ecological processes are welcome. Submissions should have a broad ecological framework beyond the specific study organism or geographic region.

Short communications that highlight emerging issues and exciting discoveries within five printed pages will receive a rapid turnaround. Papers describing important new analytical, computational, experimental and theoretical techniques and methods are encouraged and will be highlighted as *Methodological Advances*. We welcome proposals for *Review Papers* synthesizing a specific field within marine ecology. Finally, the journal aims to publish Special Issues at regular intervals synthesizing a particular field of marine science. All printed papers undergo a peer review process before being accepted and will receive a first decision within three months.

AUDIENCE

Marine Physiologists and Biochemists, Marine Ecologists and Marine Biologists.

IMPACT FACTOR

2016: 1.937 © Clarivate Analytics Journal Citation Reports 2017

ABSTRACTING AND INDEXING

Aquatic Sciences and Fisheries Abstracts
EMBiology
Aquabiology
BIOSIS
Current Contents/Agriculture, Biology & Environmental Sciences
Ecological Abstracts
Engineering Village - GEOBASE
Marine Science Contents Tables
Oceanography Abstracts
PASCAL/CNRS
PASCAL M and S (Earth Sciences)
Selected Water Resources Abstracts
Scopus

EDITORIAL BOARD

GUIDE FOR AUTHORS

Your Paper Your Way

We now differentiate between the requirements for new and revised submissions. You may choose to submit your manuscript as a single Word or PDF file to be used in the refereeing process. Only when your paper is at the revision stage, will you be requested to put your paper in to a 'correct format' for acceptance and provide the items required for the publication of your article.
To find out more, please visit the Preparation section below.

INTRODUCTION

Types of contribution

1. Original research articles (Regular Articles)
2. Essay Reviews
3. Monographs
4. Book Reviews

Original research articles should report the results of original research. The material should not have been previously published elsewhere, except in a preliminary form.

Essay Reviews will deal with tightly defined topics which are highly novel and of particular interest to the readership of the Journal. These Reviews discuss topics that may be rather dispersed in the literature, but with a high degree of relevance to experimental marine ecology.

Monographs are comprehensive and integrated studies (up to 100 pages) that represent a detailed synthesis and in-depth interpretation of results. A small number (2 to 4) of monographs will be published a year, authors should consult with the editors regarding possible contributions prior to submission of full monographs.

Book Reviews will be included in the journal on a range of relevant books which are not more than 2 years old. Book reviews will be solicited by the Book Review Editor. Unsolicited reviews will not usually be accepted, but suggestions for appropriate books for review may be sent to the Book Review Editor:

I. Marsden,
University of Canterbury,
Private Bag 4800,
Christchurch,
New Zealand

BEFORE YOU BEGIN

Ethics in publishing

Please see our information pages on Ethics in publishing and Ethical guidelines for journal publication.

Declaration of interest

All authors must disclose any financial and personal relationships with other people or organizations that could inappropriately influence (bias) their work. Examples of potential competing interests include employment, consultancies, stock ownership, honoraria, paid expert testimony, patent applications/registrations, and grants or other funding. Authors must disclose any interests in two places: 1. A summary declaration of interest statement in the title page file (if double-blind) or the manuscript file (if single-blind). If there are no interests to declare then please state this: 'Declarations of interest: none'. This summary statement will be ultimately published if the article is accepted. 2. Detailed disclosures as part of a separate Declaration of Interest form, which forms part of the journal's official records. It is important for potential interests to be declared in both places and that the information matches. More information.

Submission declaration and verification

Submission of an article implies that the work described has not been published previously (except in the form of an abstract, a published lecture or academic thesis, see 'Multiple, redundant or concurrent publication' for more information), that it is not under consideration for publication elsewhere, that its publication is approved by all authors and tacitly or explicitly by the responsible authorities where the work was carried out, and that, if accepted, it will not be published elsewhere in the same form, in English or in any other language, including electronically without the written consent of the copyright-holder. To verify originality, your article may be checked by the originality detection service Crossref Similarity Check.

Preprints
Please note that preprints can be shared anywhere at any time, in line with Elsevier's sharing policy. Sharing your preprints e.g. on a preprint server will not count as prior publication (see 'Multiple, redundant or concurrent publication' for more information).

Contributers
Each author is required to declare his or her individual contribution to the article: all authors must have materially participated in the research and/or article preparation, so roles for all authors should be described. **The statement that all authors have approved the final article should be true and included in the disclosure.**

Changes to authorship

Authors are expected to consider carefully the list and order of authors **before** submitting their manuscript and provide the definitive list of authors at the time of the original submission. Any addition, deletion or rearrangement of author names in the authorship list should be made only **before** the manuscript has been accepted and only if approved by the journal Editor. To request such a change, the Editor must receive the following from the **corresponding author**: (a) the reason for the change in author list and (b) written confirmation (e-mail, letter) from all authors that they agree with the addition, removal or rearrangement. In the case of addition or removal of authors, this includes confirmation from the author being added or removed.

Only in exceptional circumstances will the Editor consider the addition, deletion or rearrangement of authors **after** the manuscript has been accepted. While the Editor considers the request, publication of the manuscript will be suspended. If the manuscript has already been published in an online issue, any requests approved by the Editor will result in a corrigendum.

Article transfer service
This journal is part of our Article Transfer Service. This means that if the Editor feels your article is more suitable in one of our other participating journals, then you may be asked to consider transferring the article to one of those. If you agree, your article will be transferred automatically on your behalf with no need to reformat. Please note that your article will be reviewed again by the new journal. More information.

Copyright

Upon acceptance of an article, authors will be asked to complete a 'Journal Publishing Agreement' (see more information on this). An e-mail will be sent to the corresponding author confirming receipt of the manuscript together with a 'Journal Publishing Agreement' form or a link to the online version of this agreement.

Subscribers may reproduce tables of contents or prepare lists of articles including abstracts for internal circulation within their institutions. Permission of the Publisher is required for resale or distribution outside the institution and for all other derivative works, including compilations and translations. If excerpts from other copyrighted works are included, the author(s) must obtain written permission from the copyright owners and credit the source(s) in the article. Elsevier has preprinted forms for use by authors in these cases.

For gold open access articles: Upon acceptance of an article, authors will be asked to complete an 'Exclusive License Agreement' (more information). Permitted third party reuse of gold open access articles is determined by the author's choice of user license.

Author rights
As an author you (or your employer or institution) have certain rights to reuse your work. More information.

Elsevier supports responsible sharing
Find out how you can share your research published in Elsevier journals.

Role of the funding source

You are requested to identify who provided financial support for the conduct of the research and/or preparation of the article and to briefly describe the role of the sponsor(s), if any, in study design; in the collection, analysis and interpretation of data; in the writing of the report; and in the decision to submit the article for publication. If the funding source(s) had no such involvement then this should be stated.

Funding body agreements and policies
Elsevier has established a number of agreements with funding bodies which allow authors to comply with their funder's open access policies. Some funding bodies will reimburse the author for the gold open access publication fee. Details of existing agreements are available online.

Open access
This journal offers authors a choice in publishing their research:

Subscription
• Articles are made available to subscribers as well as developing countries and patient groups through our universal access programs.
• No open access publication fee payable by authors.
• The Author is entitled to post the accepted manuscript in their institution's repository and make this public after an embargo period (known as green Open Access). The published journal article cannot be shared publicly, for example on ResearchGate or Academia.edu, to ensure the sustainability of peer-reviewed research in journal publications. The embargo period for this journal can be found below.

Gold open access
• Articles are freely available to both subscribers and the wider public with permitted reuse.
• A gold open access publication fee is payable by authors or on their behalf, e.g. by their research funder or institution.

Regardless of how you choose to publish your article, the journal will apply the same peer review criteria and acceptance standards.

For gold open access articles, permitted third party (re)use is defined by the following Creative Commons user licenses:

Creative Commons Attribution (CC BY)
Lets others distribute and copy the article, create extracts, abstracts, and other revised versions, adaptations or derivative works of or from an article (such as a translation), include in a collective work (such as an anthology), text or data mine the article, even for commercial purposes, as long as they credit the author(s), do not represent the author as endorsing their adaptation of the article, and do not modify the article in such a way as to damage the author's honor or reputation.

Creative Commons Attribution-NonCommercial-NoDerivs (CC BY-NC-ND)
For non-commercial purposes, lets others distribute and copy the article, and to include in a collective work (such as an anthology), as long as they credit the author(s) and provided they do not alter or modify the article.

The gold open access publication fee for this journal is **USD 3000**, excluding taxes. Learn more about Elsevier's pricing policy: https://www.elsevier.com/openaccesspricing.

Green open access
Authors can share their research in a variety of different ways and Elsevier has a number of green open access options available. We recommend authors see our green open access page for further information. Authors can also self-archive their manuscripts immediately and enable public access from their institution's repository after an embargo period. This is the version that has been accepted for publication and which typically includes author-incorporated changes suggested during submission, peer review and in editor-author communications. Embargo period: For subscription articles, an appropriate amount of time is needed for journals to deliver value to subscribing customers before an article becomes freely available to the public. This is the embargo period and it begins from the date the article is formally published online in its final and fully citable form. Find out more.

This journal has an embargo period of 24 months.

Elsevier Researcher Academy
Researcher Academy is a free e-learning platform designed to support early and mid-career researchers throughout their research journey. The "Learn" environment at Researcher Academy offers several interactive modules, webinars, downloadable guides and resources to guide you through the process of writing for research and going through peer review. Feel free to use these free resources to improve your submission and navigate the publication process with ease.

Language Services

Manuscripts should be written in English. Authors who are unsure of correct English usage should have their manuscript checked by someone proficient in the language. Manuscripts in which the English is difficult to understand may be returned to the author for revision before scientific review.

Authors who require information about language editing and copyediting services pre- and post-submission please visit http://www.elsevier.com/languagepolishing for more information. Please note Elsevier neither endorses nor takes responsibility for any products, goods or services offered by outside vendors through our services or in any advertising. For more information please refer to our Terms & Conditions: http://www.elsevier.com/termsandconditions.

Submission

Our online submission system guides you stepwise through the process of entering your article details and uploading your files. The system converts your article files to a single PDF file used in the peer-review process. Editable files (e.g., Word, LaTeX) are required to typeset your article for final publication. All correspondence, including notification of the Editor's decision and requests for revision, is sent by e-mail.

Please submit your article via https://www.evise.com/profile/api/navigate/JEMBE

Referees

Please submit, with the manuscript, the names and addresses of four potential referees.

Page charges

Journal of Experimental Marine Biology and Ecology has no page charges.

PREPARATION

Peer review

This journal operates a single blind review process. All contributions will be initially assessed by the editor for suitability for the journal. Papers deemed suitable are then typically sent to a minimum of two independent expert reviewers to assess the scientific quality of the paper. The Editor is responsible for the final decision regarding acceptance or rejection of articles. The Editor's decision is final. More information on types of peer review.

Use of word processing software

It is important that the file be saved in the native format of the word processor used. The text should be in single-column format. Keep the layout of the text as simple as possible. Most formatting codes will be removed and replaced on processing the article. In particular, do not use the word processor's options to justify text or to hyphenate words. However, do use bold face, italics, subscripts, superscripts etc. When preparing tables, if you are using a table grid, use only one grid for each individual table and not a grid for each row. If no grid is used, use tabs, not spaces, to align columns. The electronic text should be prepared in a way very similar to that of conventional manuscripts (see also the Guide to Publishing with Elsevier: http://www.elsevier.com/guidepublication). Note that source files of figures, tables and text graphics will be required whether or not you embed your figures in the text. See also the section on Electronic artwork.

To avoid unnecessary errors you are strongly advised to use the 'spell-check' and 'grammar-check' functions of your word processor.

Use of word processing software

Regardless of the file format of the original submission, at revision you must provide us with an editable file of the entire article. Keep the layout of the text as simple as possible. Most formatting codes will be removed and replaced on processing the article. The electronic text should be prepared in a way very similar to that of conventional manuscripts (see also the Guide to Publishing with Elsevier). See also the section on Electronic artwork.

To avoid unnecessary errors you are strongly advised to use the 'spell-check' and 'grammar-check' functions of your word processor.

LaTeX

You are recommended to use the Elsevier article class elsarticle.cls to prepare your manuscript and BibTeX to generate your bibliography.

Our LaTeX site has detailed submission instructions, templates and other information.

Article structure

Subdivision

Manuscripts should be typewritten with numbered lines, with wide margins and double spacing throughout, i.e. also for abstracts, footnotes and references. **Every page of the manuscript, including the title page, references, tables, etc., should be numbered in the upper right-hand corner.** However, in the text no reference should be made to page numbers; if necessary, one may refer to sections. Avoid excessive usage of italics to emphasize part of the text.

Subdivision - numbered sections
Divide your article into clearly defined and numbered sections. Subsections should be numbered 1.1 (then 1.1.1, 1.1.2, ...), 1.2, etc. (the abstract is not included in section numbering). Use this numbering also for internal cross-referencing: do not just refer to 'the text'. Any subsection may be given a brief heading. Each heading should appear on its own separate line.

Introduction
State the objectives of the work and provide an adequate background, avoiding a detailed literature survey or a summary of the results.

Material and methods
Provide sufficient detail to allow the work to be reproduced. Methods already published should be indicated by a reference: only relevant modifications should be described.

Theory/calculation
A Theory section should extend, not repeat, the background to the article already dealt with in the Introduction and lay the foundation for further work. In contrast, a Calculation section represents a practical development from a theoretical basis.

Results
Results should be clear and concise.

Discussion
This should explore the significance of the results of the work, not repeat them. Only in exceptional circumstances should results and discussion be combined and this should be discussed with the editor prior to submission. Avoid extensive citations and discussion of published literature.

Conclusions
The main conclusions of the study may be presented in a short Conclusions section, which may stand alone or form a subsection of a Discussion or Results and Discussion section.

Glossary
Please supply, as a separate list, the definitions of field-specific terms used in your article.

Essential title page information

• **Title.** Concise and informative. Titles are often used in information-retrieval systems. Avoid abbreviations and formulae where possible.
• **Author names and affiliations.** Please clearly indicate the given name(s) and family name(s) of each author and check that all names are accurately spelled. You can add your name between parentheses in your own script behind the English transliteration. Present the authors' affiliation addresses (where the actual work was done) below the names. Indicate all affiliations with a lower-case superscript letter immediately after the author's name and in front of the appropriate address. Provide the full postal address of each affiliation, including the country name and, if available, the e-mail address of each author.
• **Corresponding author.** Clearly indicate who will handle correspondence at all stages of refereeing and publication, also post-publication. This responsibility includes answering any future queries about Methodology and Materials. **Ensure that the e-mail address is given and that contact details are kept up to date by the corresponding author.**
• **Present/permanent address.** If an author has moved since the work described in the article was done, or was visiting at the time, a 'Present address' (or 'Permanent address') may be indicated as a footnote to that author's name. The address at which the author actually did the work must be retained as the main, affiliation address. Superscript Arabic numerals are used for such footnotes.

Abstract

A concise and factual abstract is required. The abstract should state briefly the purpose of the research, the principal results and major conclusions. An abstract is often presented separate from the article, so it must be able to stand alone. For this reason, References should be avoided, but if essential, they must be cited in full, without reference to the reference list. Also, non-standard or uncommon abbreviations should be avoided, but if essential they must be defined at their first mention in the abstract itself. Abstracts must not be longer than 400 words.

Graphical abstract
Although a graphical abstract is optional, its use is encouraged as it draws more attention to the online article. The graphical abstract should summarize the contents of the article in a concise, pictorial form designed to capture the attention of a wide readership. Graphical abstracts should be submitted as a separate file in the online submission system. Image size: Please provide an image with a minimum of 531 × 1328 pixels (h × w) or proportionally more. The image should be readable at a size of 5 × 13 cm using a regular screen resolution of 96 dpi. Preferred file types: TIFF, EPS, PDF or MS Office files. You can view Example Graphical Abstracts on our information site.
Authors can make use of Elsevier's Illustration Services to ensure the best presentation of their images and in accordance with all technical requirements.

Highlights
Highlights are mandatory for this journal. They consist of a short collection of bullet points that convey the core findings of the article and should be submitted in a separate editable file in the online submission system. Please use 'Highlights' in the file name and include 3 to 5 bullet points (maximum 85 characters, including spaces, per bullet point). You can view example Highlights on our information site.

Keywords

Immediately after the abstract, provide a maximum of 6 keywords, using American spelling and avoiding general and plural terms and multiple concepts (avoid, for example, 'and', 'of'). Be sparing with abbreviations: only abbreviations firmly established in the field may be eligible. These keywords will be used for indexing purposes.

Abbreviations
Define abbreviations that are not standard in this field in a footnote to be placed on the first page of the article. Such abbreviations that are unavoidable in the abstract must be defined at their first mention there, as well as in the footnote. Ensure consistency of abbreviations throughout the article.

Acknowledgements
Collate acknowledgements in a separate section at the end of the article before the references and do not, therefore, include them on the title page, as a footnote to the title or otherwise. List here those individuals who provided help during the research (e.g., providing language help, writing assistance or proof reading the article, etc.).

Formatting of funding sources
List funding sources in this standard way to facilitate compliance to funder's requirements:

Funding: This work was supported by the National Institutes of Health [grant numbers xxxx, yyyy]; the Bill & Melinda Gates Foundation, Seattle, WA [grant number zzzz]; and the United States Institutes of Peace [grant number aaaa].

It is not necessary to include detailed descriptions on the program or type of grants and awards. When funding is from a block grant or other resources available to a university, college, or other research institution, submit the name of the institute or organization that provided the funding.

If no funding has been provided for the research, please include the following sentence:

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Nomenclature and Units

1. Authors and editors are, by general agreement, obliged to accept the rules governing biological nomenclature, as laid down in the International Code of Botanical Nomenclature, the International Code of Nomenclature of Bacteria, and the International Code of Zoological Nomenclature.
2. All biota (crops, plants, insects, birds, mammals, etc.) should be identified by their scientific names when the English term is used, with the exception of common domestic animals.

3. The first mention of the scientific names of the species used in the work - in title or text - should be accompanied by the taxonomic authority unless they can all be referred to a general work in which the authorities are given. Scientific names of species referred to in other studies need no authority. Generic names should only be abbreviated when immediately preceded in the text by the mention of the same species or another of the same genus.

4. All biocides and other organic compounds must be identified by their Geneva names when first used in text. Active ingredients of all formulations should be likewise identified.

5. For chemical nomenclature, the conventions of the International Union of Pure and Applied Chemistry and the official recommendations of the IUPAC-IUB Combined Commission on Biochemical Nomenclature should be followed.

6. When referring to salinity, please do not use units i.e. no PSU or 0/00. Please use "a salinity of X, or salinity" instead of adding units.

Genbank

GenBank/DNA sequence linking. DNA sequences and GenBank Accession numbers Many Elsevier journals cite "gene accession numbers" in their running text and footnotes. Gene accession numbers refer to genes or DNA sequences about which further information can be found in the databases at the National Center for Biotechnical Information (NCBI) at the National Library of Medicine. Elsevier authors wishing to enable other scientists to use the accession numbers cited in their papers via links to these sources, should type this information in the following manner:

For each and every accession number cited in an article, authors should type the accession number in **bold, underlined text**. Letters in the accession number should always be capitalised. (See Example 1 below). This combination of letters and format will enable Elsevier's typesetters to recognize the relevant texts as accession numbers and add the required link to GenBank's sequences.

Example 1: "GenBank accession nos. AI631510, AI631511, AI632198, and BF223228), a B-cell tumor from a chronic lymphatic leukemia (GenBank accession no. BE675048), and a T-cell lymphoma (GenBank accession no. AA361117)".

Authors are encouraged to check accession numbers used very carefully. An error in a letter or number can result in a dead link.

In the final version of the printed article, the accession number text will not appear bold or underlined (see Example 2 below).

Example 2: "GenBank accession nos. AI631510, AI631511, AI632198, and BF223228), a B-cell tumor from a chronic lymphatic leukemia (GenBank accession no. BE675048), and a T-cell lymphoma (GenBank accession no. AA361117)".

In the final version of the electronic copy, the accession number text will be linked to the appropriate source in the NCBI databases enabling readers to go directly to that source from the article (see Example 3 below).

Example 3: "GenBank accession nos. AI631510, AI631511, AI632198, and BF223228), a B-cell tumor from a chronic lymphatic leukemia (GenBank accession no. BE675048), and a T-cell lymphoma (GenBank accession no. AA361117)".

Formulae

1. Formulae should be typewritten, if possible. Leave ample space around the formulae.

2. Subscripts and superscripts should be clear.

3. Greek letters and other non-Latin or handwritten symbols should be explained in the margin where they are

Footnotes

Footnotes should be used sparingly. Number them consecutively throughout the article. Many word processors can build footnotes into the text, and this feature may be used. Otherwise, please indicate the position of footnotes in the text and list the footnotes themselves separately at the end of the article. Do not include footnotes in the Reference list.

Artwork

Electronic Artwork

•Make sure you use uniform lettering and sizing of your original artwork.

- Save text in illustrations as 'graphics' or enclose the font.
- Only use the following font in your illustrations: Arial.
- Number the illustrations according to their sequence in the text.
- When labelling composite figures, please label as A,B,C, etc. in Arial font, positioned on the upper left corner, on the panel whenever possible. Please do not include any periods, parentheses, etc.
- Use a logical naming convention for your artwork files.
- Provide captions to illustrations separately.
- Produce images near to the desired size of the printed version.
- Submit each figure as a separate file.
- Extra frames and boxes around figures should be eliminated.

Please include only X and Y (and Z if applicable) axes. Background lines on figures should only be included when absolutely necessary.
- Legend material and explanations of symbols, etc. should be on the panel, not hanging off to the side of the figure. No frame is necessary. If this material does not fit on the panel, it should be included in the actual figure legend.
- Submitting figures as they are printed from Excel or other spread sheets is not acceptable formatting for publication.

A detailed guide on electronic artwork is available on our website:
http://www.elsevier.com/artworkinstructions
You are urged to visit this site; some excerpts from the detailed information are given here.
Formats
Regardless of the application used, when your electronic artwork is finalised, please 'save as' or convert the images to one of the following formats (note the resolution requirements for line drawings, halftones, and line/halftone combinations given below):
EPS: Vector drawings. Embed the font or save the text as 'graphics'.
TIFF: Color or grayscale photographs (halftones): always use a minimum of 300 dpi.
TIFF: Bitmapped line drawings: use a minimum of 1000 dpi.
TIFF: Combinations bitmapped line/half-tone (color or grayscale): a minimum of 500 dpi is required.
If your electronic artwork is created in a Microsoft Office application (Word, PowerPoint, Excel) then please supply 'as is'.**Please do not:**
- Supply files that are optimised for screen use (e.g., GIF, BMP, PICT, WPG); the resolution is too low;
- Supply files that are too low in resolution;
- Submit graphics that are disproportionately large for the content.

Color Artwork
Please make sure that artwork files are in an acceptable format (TIFF, EPS or MS Office files) and with the correct resolution. If, together with your accepted article, you submit usable color figures then Elsevier will ensure, at no additional charge, that these figures will appear in color on the Web (e.g., ScienceDirect and other sites) regardless of whether or not these illustrations are reproduced in color in the printed version. **For color reproduction in print, you will receive information regarding the costs from Elsevier after receipt of your accepted article**. Please indicate your preference for color: in print or on the Web only. For further information on the preparation of electronic artwork, please see http://www.elsevier.com/artworkinstructions.
Please note: Because of technical complications which can arise by converting color figures to 'gray scale' (for the printed version should you not opt for color in print) please submit in addition high-resolution black and white versions of all the color illustrations. Simply printing color as black and white is not acceptable.

Figure captions
Ensure that each illustration has a caption. Supply captions separately, not attached to the figure. A caption should comprise a brief title (**not** on the figure itself) and a description of the illustration. Keep text in the illustrations themselves to a minimum but explain all symbols and abbreviations used.

Tables
Please submit tables as editable text and not as images. Tables can be placed either next to the relevant text in the article, or on separate page(s) at the end. Number tables consecutively in accordance with their appearance in the text and place any table notes below the table body. Be sparing in the use of tables and ensure that the data presented in them do not duplicate results described elsewhere in the article. Please avoid using vertical rules and shading in table cells.

References

Citation in text
Please ensure that every reference cited in the text is also present in the reference list (and vice versa). Any references cited in the abstract must be given in full. Unpublished results and personal communications are not recommended in the reference list, but may be mentioned in the text. If these references are included in the reference list they should follow the standard reference style of the journal and should include a substitution of the publication date with either 'Unpublished results' or 'Personal communication'. Citation of a reference as 'in press' implies that the item has been accepted for publication.

Web references
As a minimum, the full URL should be given and the date when the reference was last accessed. Any further information, if known (DOI, author names, dates, reference to a source publication, etc.), should also be given. Web references can be listed separately (e.g., after the reference list) under a different heading if desired, or can be included in the reference list.

Data references
This journal encourages you to cite underlying or relevant datasets in your manuscript by citing them in your text and including a data reference in your Reference List. Data references should include the following elements: author name(s), dataset title, data repository, version (where available), year, and global persistent identifier. Add [dataset] immediately before the reference so we can properly identify it as a data reference. The [dataset] identifier will not appear in your published article.

References in a special issue
Please ensure that the words 'this issue' are added to any references in the list (and any citations in the text) to other articles in the same Special Issue.

Reference management software
Most Elsevier journals have their reference template available in many of the most popular reference management software products. These include all products that support Citation Style Language styles, such as Mendeley and Zotero, as well as EndNote. Using the word processor plug-ins from these products, authors only need to select the appropriate journal template when preparing their article, after which citations and bibliographies will be automatically formatted in the journal's style. If no template is yet available for this journal, please follow the format of the sample references and citations as shown in this Guide. If you use reference management software, please ensure that you remove all field codes before submitting the electronic manuscript. More information on how to remove field codes.

Users of Mendeley Desktop can easily install the reference style for this journal by clicking the following link:
http://open.mendeley.com/use-citation-style/journal-of-experimental-marine-biology-and-ecology
When preparing your manuscript, you will then be able to select this style using the Mendeley plug-ins for Microsoft Word or LibreOffice.

Reference style
Text: All citations in the text should refer to:
1. *Single author:* the author's name (without initials, unless there is ambiguity) and the year of publication;
2. *Two authors:* both authors' names and the year of publication;
3. *Three or more authors:* first author's name followed by 'et al.' and the year of publication.
Citations may be made directly (or parenthetically). Groups of references should be listed first alphabetically, then chronologically.
Examples: 'as demonstrated (Allan, 2000a, 2000b, 1999; Allan and Jones, 1999). Kramer et al. (2010) have recently shown ....'
List: References should be arranged first alphabetically and then further sorted chronologically if necessary. More than one reference from the same author(s) in the same year must be identified by the letters 'a', 'b', 'c', etc., placed after the year of publication.
Examples:
Reference to a journal publication:
Van der Geer, J., Hanraads, J.A.J., Lupton, R.A., 2010. The art of writing a scientific article. J. Sci. Commun. 163, 51–59.
Reference to a book:
Strunk Jr., W., White, E.B., 2000. The Elements of Style, fourth ed. Longman, New York.

Reference to a chapter in an edited book:

Mettam, G.R., Adams, L.B., 2009. How to prepare an electronic version of your article, in: Jones, B.S., Smith , R.Z. (Eds.), Introduction to the Electronic Age. E-Publishing Inc., New York, pp. 281–304.

Reference to a website:

Cancer Research UK, 1975. Cancer statistics reports for the UK. http://www.cancerresearchuk.org/aboutcancer/statistics/cancerstatsreport/ (accessed 13 March 2003).

Reference to a dataset:

[dataset] Oguro, M., Imahiro, S., Saito, S., Nakashizuka, T., 2015. Mortality data for Japanese oak wilt disease and surrounding forest compositions. Mendeley Data, v1. https://doi.org/10.17632/xwj98nb39r.1.

Video

Elsevier accepts video material and animation sequences to support and enhance your scientific research. Authors who have video or animation files that they wish to submit with their article are strongly encouraged to include links to these within the body of the article. This can be done in the same way as a figure or table by referring to the video or animation content and noting in the body text where it should be placed. All submitted files should be properly labeled so that they directly relate to the video file's content. . In order to ensure that your video or animation material is directly usable, please provide the file in one of our recommended file formats with a preferred maximum size of 150 MB per file, 1 GB in total. Video and animation files supplied will be published online in the electronic version of your article in Elsevier Web products, including ScienceDirect. Please supply 'stills' with your files: you can choose any frame from the video or animation or make a separate image. These will be used instead of standard icons and will personalize the link to your video data. For more detailed instructions please visit our video instruction pages. Note: since video and animation cannot be embedded in the print version of the journal, please provide text for both the electronic and the print version for the portions of the article that refer to this content.

AudioSlides

The journal encourages authors to create an AudioSlides presentation with their published article. AudioSlides are brief, webinar-style presentations that are shown next to the online article on ScienceDirect. This gives authors the opportunity to summarize their research in their own words and to help readers understand what the paper is about. More information and examples are available. Authors of this journal will automatically receive an invitation e-mail to create an AudioSlides presentation after acceptance of their paper.

Data visualization

Include interactive data visualizations in your publication and let your readers interact and engage more closely with your research. Follow the instructions here to find out about available data visualization options and how to include them with your article.

Supplementary material

Supplementary material such as applications, images and sound clips, can be published with your article to enhance it. Submitted supplementary items are published exactly as they are received (Excel or PowerPoint files will appear as such online). Please submit your material together with the article and supply a concise, descriptive caption for each supplementary file. If you wish to make changes to supplementary material during any stage of the process, please make sure to provide an updated file. Do not annotate any corrections on a previous version. Please switch off the 'Track Changes' option in Microsoft Office files as these will appear in the published version.

Research data

This journal encourages and enables you to share data that supports your research publication where appropriate, and enables you to interlink the data with your published articles. Research data refers to the results of observations or experimentation that validate research findings. To facilitate reproducibility and data reuse, this journal also encourages you to share your software, code, models, algorithms, protocols, methods and other useful materials related to the project.

Below are a number of ways in which you can associate data with your article or make a statement about the availability of your data when submitting your manuscript. If you are sharing data in one of these ways, you are encouraged to cite the data in your manuscript and reference list. Please refer to the "References" section for more information about data citation. For more information on depositing, sharing and using research data and other relevant research materials, visit the research data page.

Data linking

If you have made your research data available in a data repository, you can link your article directly to the dataset. Elsevier collaborates with a number of repositories to link articles on ScienceDirect with relevant repositories, giving readers access to underlying data that gives them a better understanding of the research described.

There are different ways to link your datasets to your article. When available, you can directly link your dataset to your article by providing the relevant information in the submission system. For more information, visit the database linking page.

For supported data repositories a repository banner will automatically appear next to your published article on ScienceDirect.

In addition, you can link to relevant data or entities through identifiers within the text of your manuscript, using the following format: Database: xxxx (e.g., TAIR: AT1G01020; CCDC: 734053; PDB: 1XFN).

Mendeley Data

This journal supports Mendeley Data, enabling you to deposit any research data (including raw and processed data, video, code, software, algorithms, protocols, and methods) associated with your manuscript in a free-to-use, open access repository. During the submission process, after uploading your manuscript, you will have the opportunity to upload your relevant datasets directly to *Mendeley Data*. The datasets will be listed and directly accessible to readers next to your published article online.

For more information, visit the Mendeley Data for journals page.

MethodsX

You have the option of converting relevant protocols and methods into one or multiple MethodsX articles, a new kind of article that describes the details of customized research methods. Many researchers spend a significant amount of time on developing methods to fit their specific needs or setting, but often without getting credit for this part of their work. MethodsX, an open access journal, now publishes this information in order to make it searchable, peer reviewed, citable and reproducible. Authors are encouraged to submit their MethodsX article as an additional item directly alongside the revised version of their manuscript. If your research article is accepted, your methods article will automatically be transferred over to MethodsX where it will be editorially reviewed. Please note an open access fee is payable for publication in MethodsX. Full details can be found on the MethodsX website. Please use this template to prepare your MethodsX article.

Data statement

To foster transparency, we encourage you to state the availability of your data in your submission. This may be a requirement of your funding body or institution. If your data is unavailable to access or unsuitable to post, you will have the opportunity to indicate why during the submission process, for example by stating that the research data is confidential. The statement will appear with your published article on ScienceDirect. For more information, visit the Data Statement page.

Submission Checklist

The following list will be useful during the final checking of an article prior to sending it to the journal for review. Please consult this Guide for Authors for further details of any item.
Ensure that the following items are present:
One author has been designated as the corresponding author with contact details:
• E-mail address
• Full postal address
• Phone numbers
The cover letter must include a declaration that all authors agree to the submission
All necessary files have been uploaded, and contain:
• Keywords
• All figure captions
• All tables (including title, description, footnotes)
Further considerations:
• Manuscript has been 'spell-checked' and 'grammar-checked'
• References are in the correct format for this journal
• All references mentioned in the Reference list are cited in the text, and vice versa

• Permission has been obtained for use of copyrighted material from other sources (including the Web)
• Color figures are clearly marked as being intended for color reproduction on the Web (free of charge) and in print, or to be reproduced in color on the Web (free of charge) and in black-and-white in print
• If only color on the Web is required, black-and-white versions of the figures are also supplied for printing purposes
For any further information please visit our customer support site at http://support.elsevier.com

AFTER ACCEPTANCE

Online proof correction

Corresponding authors will receive an e-mail with a link to our online proofing system, allowing annotation and correction of proofs online. The environment is similar to MS Word: in addition to editing text, you can also comment on figures/tables and answer questions from the Copy Editor. Web-based proofing provides a faster and less error-prone process by allowing you to directly type your corrections, eliminating the potential introduction of errors.

If preferred, you can still choose to annotate and upload your edits on the PDF version. All instructions for proofing will be given in the e-mail we send to authors, including alternative methods to the online version and PDF.

We will do everything possible to get your article published quickly and accurately. Please use this proof only for checking the typesetting, editing, completeness and correctness of the text, tables and figures. Significant changes to the article as accepted for publication will only be considered at this stage with permission from the Editor. It is important to ensure that all corrections are sent back to us in one communication. Please check carefully before replying, as inclusion of any subsequent corrections cannot be guaranteed. Proofreading is solely your responsibility.

Offprints

The corresponding author will, at no cost, receive a customized Share Link providing 50 days free access to the final published version of the article on ScienceDirect. The Share Link can be used for sharing the article via any communication channel, including email and social media. For an extra charge, paper offprints can be ordered via the offprint order form which is sent once the article is accepted for publication. Both corresponding and co-authors may order offprints at any time via Elsevier's Webshop. Corresponding authors who have published their article gold open access do not receive a Share Link as their final published version of the article is available open access on ScienceDirect and can be shared through the article DOI link.

Author's Discount

Contributors to Elsevier journals are entitled to a 30% discount on most Elsevier books, if ordered directly from Elsevier.

AUTHOR INQUIRIES

Visit the Elsevier Support Center to find the answers you need. Here you will find everything from Frequently Asked Questions to ways to get in touch.
You can also check the status of your submitted article or find out when your accepted article will be published.

Springer

Ecology

Home > Life Sciences > Ecology

SUBDISCIPLINES   JOURNALS   BOOKS   SERIES   TEXTBOOKS   REFERENCE WORKS

Marine Biology

International Journal on Life in Oceans and Coastal Waters

Editor-in-Chief: Ulrich **Sommer**

ISSN: 0025-3162 (print version)
ISSN: 1432-1793 (electronic version)

Journal no. 227

Read Online

$199.00   Personal Rate e-only for the Americas

Get Subscription

Online subscription, valid from January through December of current calendar year

Immediate access to this year's issues via SpringerLink

1 Volume(-s) with 12 issue(-s) per annual subscription

Automatic annual renewal

More information: >> FAQs // >> Policy

ABOUT THIS JOURNAL   EDITORIAL BOARD   ETHICS & DISCLOSURES   INSTRUCTIONS FOR AUTHORS

Instructions for Authors

GENERAL INFORMATION

Please note that the journal does not offer pre-evaluation. Therefore please directly submit your manuscript to EditorialManager at the link below. The Editors will then contact you.

Important notes:

Formatting as per journal instructions is essential, otherwise the manuscript will be returned without review.
Please follow the "Instructions for Authors" as well as the "Specific requirements" when preparing your manuscript, There are some regulations which apply specifically to Marine Biology. Carefully prepared manuscripts which are conform to the regulations of the journal have a higher chance to be considered for publication.

For questions please contact the Managing Editor at marinebiology@geomar.de

TYPES OF PAPERS

* Original papers:

These are the most important components of Marine Biology. They report on original research in all fields of marine biology and conform to the accepted standards of scientific quality. Interim reports and papers with inconclusive results will usually not be published. In the latter case, exceptions can be made if the inconclusiveness is a robust and important result with relation to widely debated theory.

⁙ Reviews, concepts, and syntheses:

Review papers are invited on any topic related to the focus of Marine Biology. These reviews may either summarize recently terminated research areas of wide importance, provide an up-to-date account of the present status of active research areas, or set the perspective for future research. They are not meant to be mere literature surveys. These reviews are meant to be in-depth and comprehensive efforts, and authors should have demonstrated expertise in the topic area. Very high standards of assessment with respect to quality and importance are applied to these reviews.

⁙ Methods:

Method articles may describe methods developed by the authors or a compendium of methods from the "grey" literature, if these methods deserve the attention of a wider community. Application examples demonstrating the usefulness of the method are welcome.

⁙ Rapid communications:

Rapid communications are reports of important new research results or discoveries which deserve to be published more rapidly than usual articles. The reasons for the special urgency have to be given in the cover letter. The articles have to conform to the highest priority criteria in respect to originality and importance. They can only be accepted, if no major revision of the original manuscript is needed. Rejected rapid communications cannot be submitted as regular manuscripts.

⁙ Short notes

Short notes are brief papers that contain significant observations that do not warrant full-length papers or important experimental results that are not sufficiently elaborated or developed as to justify an original paper. They may also present opinions or novel interpretation of existing ideas. Short Notes must be of considerable potential significance for a wide readership, preliminary work will not be considered. Short notes could combine the results and discussion.

⁙ Comments and replies:

Comments relate to articles in Marine Biology not older than one year. Their intention has either to be a substantial critique of the original article or the clarification of a major misunderstanding that could have been caused by the original article. The authors of the criticized articles have the right to write a reply. Comment and reply will be published together. The comment will be reviewed externally, while the reply will only be edited for clarity.

⁙ Highlight articles:

Outstanding papers of all categories may be selected as highlight articles. These articles must be exceptional in respect to the originality of the study, the importance to a diverse group of marine biologists and to the robustness of the methods. The specific importance of the article is emphasized by an accompanying comment of the responsible Editor. Highlight articles are promoted in social media.

MANUSCRIPT SUBMISSION

Manuscript Submission

Submission of a manuscript implies: that the work described has not been published before; that it is not under consideration for publication anywhere else; that its publication has been approved by all co-authors, if any, as well as by the responsible authorities – tacitly or explicitly – at the institute where the work has been carried out. The publisher will not be held legally responsible should there be any claims for compensation.

Permissions

Authors wishing to include figures, tables, or text passages that have already been published elsewhere are required to obtain permission from the copyright owner(s) for both the print and online format and to include evidence that such permission has been granted when submitting their papers. Any material received without such evidence will be assumed to originate from the authors.

Online Submission

Please follow the hyperlink "Submit online" on the right and upload all of your manuscript files following the instructions given on the screen.

TITLE PAGE

Title Page

The title page should include:

- The name(s) of the author(s)
- A concise and informative title
- The affiliation(s) and address(es) of the author(s)
- The e-mail address, and telephone number(s) of the corresponding author
- If available, the 16-digit ORCID of the author(s)

Abstract

Please provide an abstract of 150 to 250 words. The abstract should not contain any undefined abbreviations or unspecified references.

TEXT

Text Formatting

Manuscripts should be submitted in Word.

- Use a normal, plain font (e.g., 10-point Times Roman) for text.
- Use italics for emphasis.
- Use the automatic page numbering function to number the pages.
- Do not use field functions.
- Use tab stops or other commands for indents, not the space bar.
- Use the table function, not spreadsheets, to make tables.
- Use the equation editor or MathType for equations.
- Save your file in docx format (Word 2007 or higher) or doc format (older Word versions).

Manuscripts with mathematical content can also be submitted in LaTeX.

  LaTeX macro package (zip, 182 kB)

Headings

Please use no more than three levels of displayed headings.

Abbreviations

Abbreviations should be defined at first mention and used consistently thereafter.

Footnotes

Footnotes can be used to give additional information, which may include the citation of a reference included in the reference list. They should not consist solely of a reference citation, and they should never include the bibliographic details of a reference. They should also not contain any figures or tables.

Footnotes to the text are numbered consecutively; those to tables should be indicated by superscript lower-case letters (or asterisks for significance values and other statistical data). Footnotes to the title or the authors of the article are not given reference symbols.

Always use footnotes instead of endnotes.

Acknowledgments

Acknowledgments of people, grants, funds, etc. should be placed in a separate section on the title page. The names of funding organizations should be written in full.

Important note:

Contrary to the above text, the journal does not encourage the use of footnotes.

SCIENTIFIC STYLE

Genus and species names should be in italics.

REFERENCES

Citation

Cite references in the text by name and year in parentheses. Some examples:

Negotiation research spans many disciplines (Thompson 1990).
This result was later contradicted by Becker and Seligman (1996).
This effect has been widely studied (Abbott 1991; Medvec et al. 1993; Barakat et al. 1995; Kelso and Smith 1998).

Reference List

The list of references should only include works that are cited in the text and that have been published or accepted for publication. Personal communications and unpublished works should only be mentioned in the text. Do not use footnotes or endnotes as a substitute for a reference list.

Reference list entries should be alphabetized by the last names of the first author of each work.

- Journal article

Gamelin FX, Baquet G, Berthoin S, Thevenet D, Nourry C, Nottin S, Bosquet L (2009) Effect of high intensity intermittent training on heart rate variability in prepubescent children. Eur J Appl Physiol 105:731-738. doi: 10.1007/s00421-008-0955-8 Ideally, the names of all authors should be provided, but the usage of "et al" in long author lists will also be accepted: Smith J, Jones M Jr, Houghton L et al (1999) Future of health insurance. N Engl J Med 965:325–329

- Article by DOI

Slifka MK, Whitton JL (2000) Clinical implications of dysregulated cytokine production. J Mol Med. doi:10.1007/s001090000086

- Book

South J, Blass B (2001) The future of modern genomics. Blackwell, London

- Book chapter

Brown B, Aaron M (2001) The politics of nature. In: Smith J (ed) The rise of modern genomics, 3rd edn. Wiley, New York, pp 230-257

- Online document

Cartwright J (2007) Big stars have weather too. IOP Publishing PhysicsWeb. http://physicsweb.org/articles/news/11/6/16/1. Accessed 26 June 2007

- Dissertation

Trent JW (1975) Experimental acute renal failure. Dissertation, University of California

Always use the standard abbreviation of a journal's name according to the ISSN List of Title Word Abbreviations

   ISSN List of Title Word Abbreviations

For authors using EndNote, Springer provides an output style that supports the formatting of in-text citations and reference list.

   EndNote style (zip, 2 kB)

TABLES

- All tables are to be numbered using Arabic numerals.

- Tables should always be cited in text in consecutive numerical order.

- For each table, please supply a table caption (title) explaining the components of the table.

- Identify any previously published material by giving the original source in the form of a reference at the end of the table caption.

- Footnotes to tables should be indicated by superscript lower-case letters (or asterisks for significance values and other statistical data) and included beneath the table body.

ARTWORK

For the best quality final product, it is highly recommended that you submit all of your artwork – photographs, line drawings, etc. – in an electronic format. Your art will then be produced to the highest standards with the greatest accuracy to detail. The published work will directly reflect the quality of the artwork provided.

Electronic Figure Submission

- Supply all figures electronically.

- Indicate what graphics program was used to create the artwork.

- For vector graphics, the preferred format is EPS; for halftones, please use TIFF format. MS Office files are also acceptable.

- Vector graphics containing fonts must have the fonts embedded in the files.

- Name your figure files with "Fig" and the figure number, e.g., Fig1.eps.

Line Art
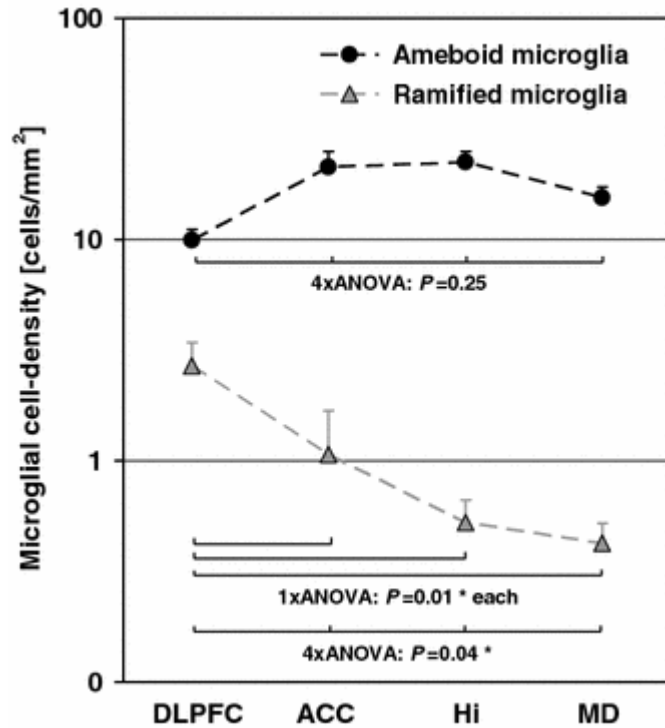
- Definition: Black and white graphic with no shading.
- Do not use faint lines and/or lettering and check that all lines and lettering within the figures are legible at final size.
- All lines should be at least 0.1 mm (0.3 pt) wide.
- Scanned line drawings and line drawings in bitmap format should have a minimum resolution of 1200 dpi.
- Vector graphics containing fonts must have the fonts embedded in the files.

Halftone Art

Definition: Photographs, drawings, or paintings with fine shading, etc.
If any magnification is used in the photographs, indicate this by using scale bars within the figures themselves.
Halftones should have a minimum resolution of 300 dpi.
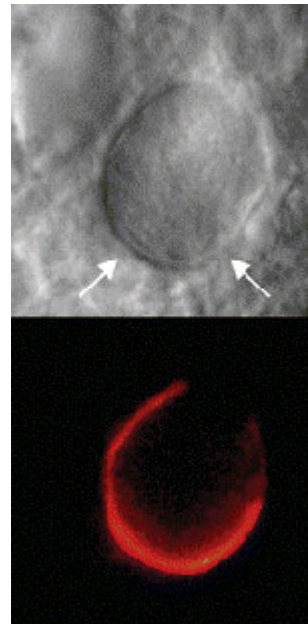
Combination Art
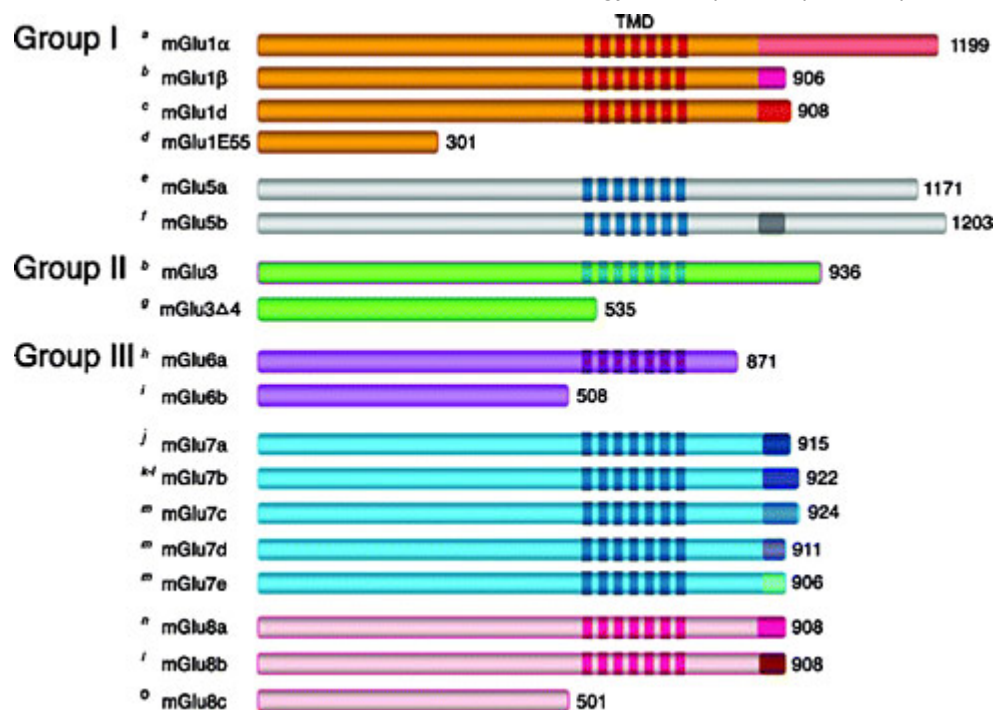
Definition: a combination of halftone and line art, e.g., halftones containing line drawing, extensive lettering, color diagrams, etc.
Combination artwork should have a minimum resolution of 600 dpi.

Color Art

Color art is free of charge for print and online publication.
Color illustrations should be submitted as RGB.

Figure Lettering

- To add lettering, it is best to use Helvetica or Arial (sans serif fonts).
- Keep lettering consistently sized throughout your final-sized artwork, usually about 2–3 mm (8–12 pt).
- Variance of type size within an illustration should be minimal, e.g., do not use 8-pt type on an axis and 20-pt type for the axis label.
- Avoid effects such as shading, outline letters, etc.
- Do not include titles or captions within your illustrations.

Figure Numbering

All figures are to be numbered using Arabic numerals.
Figures should always be cited in text in consecutive numerical order.
Figure parts should be denoted by lowercase letters (a, b, c, etc.).
If an appendix appears in your article and it contains one or more figures, continue the consecutive numbering of the main text. Do not number the appendix figures, "A1, A2, A3, etc." Figures in online appendices (Electronic Supplementary Material) should, however, be numbered separately.

Figure Captions

- Each figure should have a concise caption describing accurately what the figure depicts. Include the captions in the text file of the manuscript, not in the figure file.

- Figure captions begin with the term Fig. in bold type, followed by the figure number, also in bold type.
- No punctuation is to be included after the number, nor is any punctuation to be placed at the end of the caption.
- Identify all elements found in the figure in the figure caption; and use boxes, circles, etc., as coordinate points in graphs.
- Identify previously published material by giving the original source in the form of a reference citation at the end of the figure caption.

Figure Placement and Size

When preparing your figures, size figures to fit in the column width.
For most journals the figures should be 39 mm, 84 mm, 129 mm, or 174 mm wide and not higher than 234 mm.
For books and book-sized journals, the figures should be 80 mm or 122 mm wide and not higher than 198 mm.

Permissions

If you include figures that have already been published elsewhere, you must obtain permission from the copyright owner(s) for both the print and online format. Please be aware that some publishers do not grant electronic rights for free and that Springer will not be able to refund any costs that may have occurred to receive these permissions. In such cases, material from other sources should be used.

Accessibility

In order to give people of all abilities and disabilities access to the content of your figures, please make sure that

All figures have descriptive captions (blind users could then use a text-to-speech software or a text-to-Braille hardware)
Patterns are used instead of or in addition to colors for conveying information (color-blind users would then be able to distinguish the visual elements)
Any figure lettering has a contrast ratio of at least 4.5:1

Important note:

In addition to the submission of the figure source files without captions (as mentioned above), the figures with their captions should also be included into the running text.

ELECTRONIC SUPPLEMENTARY MATERIAL

Springer accepts electronic multimedia files (animations, movies, audio, etc.) and other supplementary files to be published online along with an article or a book chapter. This feature can add dimension to the author's article, as certain information cannot be printed or is more convenient in electronic form.

Before submitting research datasets as electronic supplementary material, authors should read the journal's Research data policy. We encourage research data to be archived in data repositories wherever possible.

Submission

Supply all supplementary material in standard file formats.

Please include in each file the following information: article title, journal name, author names; affiliation and e-mail address of the corresponding author.
To accommodate user downloads, please keep in mind that larger-sized files may require very long download times and that some users may experience other problems during downloading.

Audio, Video, and Animations

Aspect ratio: 16:9 or 4:3
Maximum file size: 25 GB
Minimum video duration: 1 sec
Supported file formats: avi, wmv, mp4, mov, m2p, mp2, mpg, mpeg, flv, mxf, mts, m4v, 3gp

Text and Presentations

Submit your material in PDF format; .doc or .ppt files are not suitable for long-term viability.
A collection of figures may also be combined in a PDF file.

Spreadsheets

Spreadsheets should be submitted as .csv or .xlsx files (MS Excel).

Specialized Formats

Specialized format such as .pdb (chemical), .wrl (VRML), .nb (Mathematica notebook), and .tex can also be supplied.

Collecting Multiple Files

It is possible to collect multiple files in a .zip or .gz file.

Numbering

If supplying any supplementary material, the text must make specific mention of the material as a citation, similar to that of figures and tables.
Refer to the supplementary files as "Online Resource", e.g., "... as shown in the animation (Online Resource 3)", "... additional data are given in Online Resource 4".
Name the files consecutively, e.g. "ESM_3.mpg", "ESM_4.pdf".

Captions

For each supplementary material, please supply a concise caption describing the content of the file.

Processing of supplementary files

Electronic supplementary material will be published as received from the author without any conversion, editing, or reformatting.

Accessibility

In order to give people of all abilities and disabilities access to the content of your supplementary files, please make sure that

The manuscript contains a descriptive caption for each supplementary material
Video files do not contain anything that flashes more than three times per second (so that users prone to seizures caused by such effects are not put at risk)

RESEARCH DATA POLICY

A submission to the journal implies that materials described in the manuscript, including all relevant raw data, will be freely available to any researcher wishing to use them for non-commercial purposes, without breaching participant confidentiality.

The journal strongly encourages that all datasets on which the conclusions of the paper rely should be available to readers. We encourage authors to ensure that their datasets are either deposited in publicly available repositories (where available and appropriate) or presented in the main manuscript or additional supporting files whenever possible. Please see Springer Nature's information on recommended repositories.

List of Repositories

Research Data Policy

General repositories - for all types of research data - such as figshare and Dryad may be used where appropriate.

Where a widely established research community expectation for data archiving in public repositories exists, submission to a community-endorsed, public repository is mandatory. Persistent identifiers (such as DOIs and accession numbers) for relevant datasets must be provided in the paper.

For more information:

Research Data Policy Frequently Asked Questions

Data availability

All original articles must include a Data availability statement. Data availability statements should include information on where data supporting the results reported in the article can be found including, where applicable, hyperlinks to publicly archived datasets analysed or generated during the study. By data we mean the minimal dataset that would be necessary to interpret, replicate and build upon the findings reported in the article. We recognise it is not always possible to share research data publicly, for instance when individual privacy could be compromised, and in such instances data availability should still be stated in the manuscript along with any conditions for access. Data Availability statements can take one of the following forms (or a combination of more than one if required for multiple datasets):

1. The datasets generated during and/or analysed during the current study are available in the [NAME] repository, [PERSISTENT WEB LINK TO DATASETS]

2. The datasets generated during and/or analysed during the current study are not publicly available due [REASON WHY DATA ARE NOT PUBLIC] but are available from the corresponding author on reasonable request.

3. The datasets during and/or analysed during the current study are available from the corresponding author on reasonable request.

4. Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

All data generated or analysed during this study are included in this published article [and its supplementary information files].

5. The data that support the findings of this study are available from [third party name] but restrictions apply to the availability of these data, which were used under license for the current study, and so are not publicly available. Data are however available from the authors upon reasonable request and with permission of [third party name].

More examples of template data availability statements, which include examples of openly available and restricted access datasets, are available:

Data availability statements

The journal also requires that authors cite any publicly available data on which the conclusions of the paper rely in the manuscript. Data citations should include a persistent identifier (such as a DOI) and should ideally be included in the reference list. Citations of datasets, when they appear in the reference list, should include the minimum information recommended by DataCite and follow journal style. Dataset identifiers including DOIs should be expressed as full URLs.

Helpdesk

Research data and peer review

Peer reviewers are encouraged to check the manuscript's Data availability statement, where applicable. They should consider if the authors have complied with the journal's policy on the availability of research data, and whether reasonable effort has been made to make the data that support the findings of the study available for replication or reuse by other researchers. Peer reviewers are entitled to request access to underlying data (and code) when needed for them to perform their evaluation of a manuscript.

Springer Nature provides a research data policy support service for authors and editors, which can be contacted at researchdata@springernature.com.

This service provides advice on research data policy compliance and on finding research data repositories. It is independent of journal, book and conference proceedings editorial offices and does not advise on specific manuscripts.

Helpdesk

ETHICAL RESPONSIBILITIES OF AUTHORS

This journal is committed to upholding the integrity of the scientific record. As a member of the Committee on Publication Ethics (COPE) the journal will follow the COPE guidelines on how to deal with potential acts of misconduct.

Authors should refrain from misrepresenting research results which could damage the trust in the journal, the professionalism of scientific authorship, and ultimately the entire scientific endeavour. Maintaining integrity of the research and its presentation can be achieved by following the rules of good scientific practice, which include:

- The manuscript has not been submitted to more than one journal for simultaneous consideration.
- The manuscript has not been published previously (partly or in full), unless the new work concerns an expansion of previous work (please provide transparency on the re-use of material to avoid the hint of text-recycling ("self-plagiarism")).
- A single study is not split up into several parts to increase the quantity of submissions and submitted to various journals or to one journal over time (e.g. "salami-publishing").
- No data have been fabricated or manipulated (including images) to support your conclusions
- No data, text, or theories by others are presented as if they were the author's own ("plagiarism"). Proper acknowledgements to other works must be given (this includes material that is closely copied (near verbatim), summarized and/or paraphrased),

quotation marks are used for verbatim copying of material, and permissions are secured for material that is copyrighted.

Important note: the journal may use software to screen for plagiarism.

- Consent to submit has been received explicitly from all co-authors, as well as from the responsible authorities - tacitly or explicitly - at the institute/organization where the work has been carried out, **before** the work is submitted.

- Authors whose names appear on the submission have contributed sufficiently to the scientific work and therefore share collective responsibility and accountability for the results.

- Authors are strongly advised to ensure the correct author group, corresponding author, and order of authors at submission. Changes of authorship or in the order of authors are **not** accepted **after** acceptance of a manuscript.

- Adding and/or deleting authors and/or changing the order of authors **at revision stage** may be justifiably warranted. A letter must accompany the revised manuscript to explain the reason for the change(s) and the contribution role(s) of the added and/or deleted author(s). Further documentation may be required to support your request.

- Requests for addition or removal of authors as a result of authorship disputes after acceptance are honored after formal notification by the institute or independent body and/or when there is agreement between all authors.

- Upon request authors should be prepared to send relevant documentation or data in order to verify the validity of the results. This could be in the form of raw data, samples, records, etc. Sensitive information in the form of confidential proprietary data is excluded.

If there is a suspicion of misconduct, the journal will carry out an investigation following the COPE guidelines. If, after investigation, the allegation seems to raise valid concerns, the accused author will be contacted and given an opportunity to address the issue. If misconduct has been established beyond reasonable doubt, this may result in the Editor-in-Chief's implementation of the following measures, including, but not limited to:

If the article is still under consideration, it may be rejected and returned to the author. If the article has already been published online, depending on the nature and severity of the infraction, either an erratum will be placed with the article or in severe cases complete retraction of the article will occur. The reason must be given in the published erratum or retraction note. Please note that retraction means that the paper is **maintained on the platform**, watermarked "retracted" and explanation for the retraction is provided in a note linked to the watermarked article. The author's institution may be informed.

COMPLIANCE WITH ETHICAL STANDARDS

To ensure objectivity and transparency in research and to ensure that accepted principles of ethical and professional conduct have been followed, authors should include information regarding sources of funding, potential conflicts of interest (financial or non-financial), informed consent if the research involved human participants, and a statement on welfare of animals if the research involved animals.

Authors should include the following statements (if applicable) in a separate section entitled "Compliance with Ethical Standards" when submitting a paper:

Disclosure of potential conflicts of interest
Research involving Human Participants and/or Animals
Informed consent

Please note that standards could vary slightly per journal dependent on their peer review policies (i.e. single or double blind peer review) as well as per journal subject discipline. Before submitting your article check the instructions following this section carefully.

The corresponding author should be prepared to collect documentation of compliance with ethical standards and send if requested during peer review or after publication.

The Editors reserve the right to reject manuscripts that do not comply with the above-mentioned guidelines. The author will be held responsible for false statements or failure to fulfill the above-mentioned guidelines.

DISCLOSURE OF POTENTIAL CONFLICTS OF INTEREST

Authors must disclose all relationships or interests that could have direct or potential influence or impart bias on the work. Although an author may not feel there is any conflict, disclosure of relationships and interests provides a more complete and transparent process, leading to an accurate and objective assessment of the work. Awareness of a real or perceived conflicts of interest is a perspective to which the readers are entitled. This is not meant to imply that a financial relationship with an organization that sponsored the research or compensation received for consultancy work is inappropriate. Examples of potential conflicts of interests **that are directly or indirectly related to the research** may include but are not limited to the following:

- Research grants from funding agencies (please give the research funder and the grant number)
- Honoraria for speaking at symposia
- Financial support for attending symposia
- Financial support for educational programs
- Employment or consultation
- Support from a project sponsor
- Position on advisory board or board of directors or other type of management relationships
- Multiple affiliations
- Financial relationships, for example equity ownership or investment interest
- Intellectual property rights (e.g. patents, copyrights and royalties from such rights)
- Holdings of spouse and/or children that may have financial interest in the work

In addition, interests that go beyond financial interests and compensation (non-financial interests) that may be important to readers should be disclosed. These may include but are not limited to personal relationships or competing interests directly or indirectly tied to this research, or professional interests or personal beliefs that may influence your research.

The corresponding author collects the conflict of interest disclosure forms from all authors. In author collaborations where formal agreements for representation allow it, it is sufficient for the corresponding author to sign the disclosure form on behalf of all authors. Examples of forms can be found

here:

The corresponding author will include a summary statement in the text of the manuscript in a separate section before the reference list, that reflects what is recorded in the potential conflict of interest disclosure form(s).

See below examples of disclosures:

Funding: This study was funded by X (grant number X).

Conflict of Interest: Author A has received research grants from Company A. Author B has received a speaker honorarium from Company X and owns stock in Company Y. Author C is a member of committee Z.

If no conflict exists, the authors should state:

Conflict of Interest: The authors declare that they have no conflict of interest.

RESEARCH INVOLVING HUMAN PARTICIPANTS AND/OR ANIMALS

1) Statement of human rights

When reporting studies that involve human participants, authors should include a statement that the studies have been approved by the appropriate institutional and/or national research ethics committee and have been performed in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards.

If doubt exists whether the research was conducted in accordance with the 1964 Helsinki Declaration or comparable standards, the authors must explain the reasons for their approach, and demonstrate that the independent ethics committee or institutional review board explicitly approved the doubtful aspects of the study.

The following statements should be included in the text before the References section:

Ethical approval: "All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards."

For retrospective studies, please add the following sentence:

"For this type of study formal consent is not required."

2) Statement on the welfare of animals

The welfare of animals used for research must be respected. When reporting experiments on animals, authors should indicate whether the international, national, and/or institutional guidelines for the care and use of animals have been followed, and that the studies have been approved by a research ethics committee at the institution or practice at which the studies were conducted (where such a committee exists).

For studies with animals, the following statement should be included in in the text before the References section:

Ethical approval: "All applicable international, national, and/or institutional guidelines for the care and use of animals were followed."

If applicable (where such a committee exists): "All procedures performed in studies involving animals were in accordance with the ethical standards of the institution or practice at which the studies were conducted."

If articles do not contain studies with human participants or animals by any of the authors, please select one of the following statements:

"This article does not contain any studies with human participants performed by any of the authors."

"This article does not contain any studies with animals performed by any of the authors."

"This article does not contain any studies with human participants or animals performed by any of the authors."

AFTER ACCEPTANCE

Upon acceptance of your article you will receive a link to the special Author Query Application at Springer's web page where you can sign the Copyright Transfer Statement online and indicate whether you wish to order OpenChoice and offprints.

Once the Author Query Application has been completed, your article will be processed and you will receive the proofs.

Copyright transfer

Authors will be asked to transfer copyright of the article to the Publisher (or grant the Publisher exclusive publication and dissemination rights). This will ensure the widest possible protection and dissemination of information under copyright laws.

Offprints

Offprints can be ordered by the corresponding author.

Color illustrations

Publication of color illustrations is free of charge.

Proof reading

The purpose of the proof is to check for typesetting or conversion errors and the completeness and accuracy of the text, tables and figures. Substantial changes in content, e.g., new results, corrected values, title and authorship, are not allowed without the approval of the Editor.

After online publication, further changes can only be made in the form of an Erratum, which will be hyperlinked to the article.

OPEN CHOICE

Open Choice allows you to publish open access in more than 1850 Springer Nature journals, making your research more visible and accessible immediately on publication.

Benefits:

Increased researcher engagement: Open Choice enables access by anyone with an internet connection, immediately on publication.
Higher visibility and impact: In Springer hybrid journals, OA articles are accessed 4 times more often on average, and cited 1.7 more times on average*.

Easy compliance with funder and institutional mandates: Many funders require open access publishing, and some take compliance into account when assessing future grant applications.

It is easy to find funding to support open access – please see our funding and support pages for more information.

*) Within the first three years of publication. Springer Nature hybrid journal OA impact analysis, 2018.

Open Choice
Funding and Support pages

Copyright and license term – CC BY

Open Choice articles do not require transfer of copyright as the copyright remains with the author. In opting for open access, the author(s) agree to publish the article under the Creative Commons Attribution License.

Find more about the license agreement

ENGLISH LANGUAGE EDITING

For editors and reviewers to accurately assess the work presented in your manuscript you need to ensure the English language is of sufficient quality to be understood. If you need help with writing in English you should consider:

Asking a colleague who is a native English speaker to review your manuscript for clarity.

Visiting the English language tutorial which covers the common mistakes when writing in English.

Using a professional language editing service where editors will improve the English to ensure that your meaning is clear and identify problems that require your review. Two such services are provided by our affiliates Nature Research Editing Service and American Journal Experts. Springer authors are entitled to a 10% discount on their first submission to either of these services, simply follow the links below.

English language tutorial

Nature Research Editing Service

American Journal Experts

Please note that the use of a language editing service is not a requirement for publication in this journal and does not imply or guarantee that the article will be selected for peer review or accepted.

If your manuscript is accepted it will be checked by our copyeditors for spelling and formal style before publication.

.

为便于编辑和评审专家准确评估您稿件中陈述的研究工作，您需要确保您的英语语言质量足以令人理解。如果您需要英文写作方面的帮助，您可以考虑：

● 请一位以英语为母语的同事审核您的稿件是否表意清晰。

● 查看一些有关英语写作中常见语言错误的教程。

● 使用专业语言编辑服务，编辑人员会对英语进行润色，以确保您的意思表达清晰，并识别需要您复核的问题。我们的附属机构 Nature Research Editing Service 和合作伙伴 American Journal Experts 即可提供此类服务。

教程

Nature Research Editing Service

American Journal Experts

请注意，使用语言编辑服务并非在期刊上发表文章的必要条件，同时也并不意味或保证文章将被选中进行同行评议或被接受。

如果您的稿件被接受，在发表之前，我们的文字编辑会检查您的文稿拼写是否规范以及文体是否正式。

.

エディターと査読者があなたの論文を正しく評価するには、使用されている英語の質が十分に高いことが必要とされます。英語での論文執筆に際してサポートが必要な場合には、次のオプションがあります：

・英語を母国語とする同僚に、原稿で使用されている英語が明確であるかをチェックしてもらう。

・英語で執筆する際のよくある間違いに関する英語のチュートリアルを参照する。

・プロの英文校正サービスを利用する。校正者が原稿の意味を明確にしたり、問題点を指摘し、英語の質を向上させます。Nature Research Editing Service とAmerican Journal Experts の2つは弊社と提携しているサービスです。Springer の著者は、いずれのサービスも初めて利用する際には10%の割引を受けることができます。以下のリンクを参照ください。

英語のチュートリアル

Nature Research Editing Service

American Journal Experts

英文校正サービスの利用は、投稿先のジャーナルに掲載されるための条件ではないこと、また論文審査や受理を保証するものではないことに留意してください。

原稿が受理されると、出版前に弊社のコピーエディターがスペルと体裁のチェックを行います。

.

영어 원고의 경우, 에디터 및 리뷰어들이 귀하의 원고에 실린 결과물을 정확하게 평가할 수 있도록, 그들이 충분히 이해할 수 있을 만한 수준으로 작성되어야 합니다. 만약 영작문과 관련하여 도움을 받기를 원하신다면 다음의 사항들을 고려하여 주십시오:

• 귀하의 원고의 표현을 명확히 해줄 영어 원어민 동료를 찾아서 리뷰를 의뢰합니다.

• 영어 튜토리얼 페이지에 방문하여 영어로 글을 쓸 때 자주하는 실수들을 확인합니다.

• 리뷰에 대비하여, 원고의 의미를 명확하게 해주고 리뷰에서 요구하는 문제점들을 식별해서 영문 수준을 향상시켜주는 전문 영문 교정 서비스를 이용합니다. Nature Research Editing Service와 American Journal Experts에서 저희와 협약을 통해 서비스를 제공하고 있습니다. Springer 저자들이 본 교정 서비스를 첫 논문 투고를 위해 사용하시는 경우 10%의 할인이 적용되며, 아래의 링크를 통하여 확인이 가능합니다.

영어 튜토리얼 페이지

Nature Research Editing Service

American Journal Experts

영문 교정 서비스는 게재를 위한 요구사항은 아니며, 해당 서비스의 이용이 피어 리뷰에 논문이 선택되거나 게재가 수락되는 것을 의미하거나 보장하지 않습니다.

원고가 수락될 경우, 출판 전 저희측 편집자에 의해 원고의 철자 및 문체를 검수하는 과정을 거치게 됩니다.

READ THIS JOURNAL ON SPRINGERLINK

All Volumes & Issues

♦ Special Issues in Ecology ♦

FOR AUTHORS AND EDITORS

2017 Impact Factor **2.215**

Aims and Scope

Submit Online

Open Choice - Your Way to Open Access

Instructions for Authors

Instructions for Authors - specific requ...

SERVICES FOR THE JOURNAL

Contacts

Download Product Flyer

Shipping Dates

Order Back Issues

Article Reprints

Bulk Orders

ALERTS FOR THIS JOURNAL

Get the table of contents of every new issue published in
Marine Biology.

<div align="center">LOGIN</div>

☐ Please send me information on new Springer
publications in Freshwater & Marine Ecology.

Wiley Online Library

≡ Journal   ⚏ Articles   Actions ⌄

MOLECULAR ECOLOGY

Author Guidelines

Sections

1. SUBMISSION

Authors should note that submission implies that the content has not been published or submitted for publication elsewhere except as a brief abstract in the proceedings of a scientific meeting or symposium.
Once the submission materials have been prepared in accordance with the Author Guidelines, manuscripts should be submitted online at https://mc.manuscriptcentral.com/mec

- The submission system will prompt authors to use an ORCID iD (a unique author identifier) to help distinguish their work from that of other researchers. Click here to find out more.
- **_Conflict of Interest Statement_**
  - o   Upon submission, authors will be asked to affirm a conflict of interest statement. For guidance, see the 'Conflict of Interest' section in the Editorial Policies and Ethical Considerations section below. Authors should ensure they liaise with all co-authors to confirm agreement with the statement.
- Click here for more details on how to use the ScholarOne submission database.
- For help with submissions, please contact: molecol@wiley.com

2. AIMS AND SCOPE

Molecular Ecology publishes papers that utilize molecular genetic techniques to address consequential questions in ecology, evolution, behaviour and conservation.

Studies may employ neutral markers for inference about ecological and evolutionary processes or examine ~~ecologically important genes and their products~~

≡ Journal   ⋮⋮ Articles   Actions ⌄

---

broadly applicable in ecology are not appropriate for *Molecular Ecology* and should instead be submitted to a more specialized journal or to *Ecology & Evolution.*

If your work addresses technical methods, computer programs and genomic resource development, please submit these to our companion journal, *Molecular Ecology Resources.*

**
各位作者， 请在投稿信中陈述您的科研成果与Molecular Ecology 的办刊宗旨和报道领域的契合之处。
Molecular Ecology 刊载的科研论文致力于运用分子基因技术解决生态，进化，行为和生态保护的相关问题。
研究包括采用中性标记监测生态和进化过程的干扰，以及对生态重要基因和相应产 物的检验。
若投稿论文仅描述并适用于当前正在研究的类群，而不涉及解决更广泛的生态学问 题，此类文章不在*Molecular Ecology* 的报道领域内，建议向更专业的期刊投稿，或*Ecology & Evolution.*
如果您的研究成果专注于技术方法，电脑程序和基因组资源开发，请向我们的姐妹 刊*Molecular Ecology Resources* 投稿。
**

Research areas of interest to *Molecular Ecology* include:

- ecological, evolutionary, and population genomics
- population structure and phylogeography
- landscape genomics
- community ecology and coevolution
- reproductive strategies
- relatedness and kin selection
- sex allocation
- population genetic theory
- analytical methods development
- conservation genetics
- speciation and hybridization
- microbial biodiversity
- evolutionary dynamics of ecologically important genes or QTLs
- ecological interactions
- molecular adaptation and environmental genomics
- impact of genetically modified organisms

Authors, please include a statement in your Cover Letter describing how your work fits the Aims and Scope of

3. MANUSCRIPT CATEGORIES AND REQUIREMENTS

Original Articles (Primary Research Papers)

Our principal function is to publish primary research papers. Such papers are reports of research projects that are complete to the extent that they yield valuable insights into topics within the Aims and Scope of *Molecular Ecology.*

- limit of 8000 words per paper, excluding references
- manuscripts generally contain in this order:

☰ Journal   ⠿ Articles   Actions ⌄

- o Introduction
- o Materials and Methods
- o Results
- o Discussion
- o Acknowledgements
- o References
- o Data Accessibility
- o Author Contributions
- o Tables and Figures (with captions)

'From the Cover' Papers

Primary Research Papers of exceptional interest to a wide audience within the Aims and Scope of *Molecular Ecology*.

- Accepted articles will be highlighted on the cover and in the table of contents, and will frequently be featured in commentaries and press alerts.
- From the Cover submissions that are judged not to merit this designation may still be considered as regular Original Articles.
- From the Cover submissions can include papers previously reviewed by other high impact journals. In these instances, we can utilize all documents associated with the previous review process. The use of these review materials does not guarantee acceptance or that the manuscript will not receive external review.
    - o To increase the probability that further external review will not be necessary, authors in these cases should revise the manuscript according to reviewers' comments and submit a cover letter describing these changes and explaining why their paper would be appropriate for publication as a From the Cover article in Molecular Ecology.
- limit of 8000 words per paper, excluding references
- N.B. We appreciate that authors of From the Cover papers are looking for rapid publication, and hence we will consider initial submissions tha g6t are not in standard *Molecular Ecology* format.
- e.g., manuscripts in Nature, PNAS or Science format are welcome.
- Please note that articles deemed suitable for publication will need to be changed to Molecular Ecology format prior to final acceptance

Invited Reviews and Syntheses
Invited Reviews

- Invited by the Reviews Editor from individuals who have major contributions to make to the field of molecular ecology. We will consider unsolicited review papers, but authors wishing to submit such manuscripts

2000 words each)

o   Color figures in these articles are published in print free of charge

Syntheses

- These papers bring together data from many different studies to address an important hypothesis in ecology or evolution. They are not commissioned by an editor and can be directly submitted to the journal.
- Specifics:

≡ Journal   Articles   Actions ⌄

o   Colour figures in these articles are published in print free of charge.

Opinions

These papers present points of view, that are relevant and potentially controversial, as a means of encouraging debate. Such manuscripts may present speculative and provocative viewpoints, although they must be conditioned by the normal standards of scientific objectivity, and they will be subject to peer review. Opinion Articles should be shorter than 6000 words, excluding references.

Comments

Comments on published papers, principally those published in, Molecular Ecology, will be considered by the editors and published after consultation or peer review. Such manuscripts should be as brief as possible. A rebuttal by the original author(s) may also be solicited and published alongside the Comment.

Meeting Reviews

These papers describe the theme, notable presentations and conclusions of a scientific meeting of interest to the molecular ecology community. Meeting Reviews are only published after peer review, and should not present new data. They should be shorter than 6000 words, excluding references.

4. PREPARING THE SUBMISSION

Cover Letters
Your cover letter should contain a clear statement of how your manuscript fits the scope of the journal.
As submission implies that the content has not been published or submitted for publication elsewhere except as a brief abstract in the proceedings of a scientific meeting or symposium, it is not necessary to reiterate this information.

Response to Reviewers
If your paper is a revision or resubmission, please prepare a detailed response to the previous set of editor and reviewer comments.

- The manuscript submission system removes text highlighting, bold type or text colours, so the most robust approach is to copy the decision letter into a Word document and insert your responses beneath each comment, starting your text with ">>>".
- Uploading a copy of the manuscript with changes tracked also assists with the review process, particularly for papers given a 'reconsider after revision' or an 'accept, minor revisions' decision.

Formatting

- Double-spaced text
- Single-spacing for:
  - o   Table and figure captions
  - o   References
  - o   Appendices
  - o   Supporting/Supplemental information
- Side margins 2.5 cm side margins, top and bottom margins 3 cm

Manuscripts failing to include any of these elements will be returned without review.

File Types

For initial submission, manuscripts can be:

- Microsoft Word with tables and figures either embedded in the document or uploaded as separate files
- A single pdf containing the text, tables and figures
- LaTeX
  - o   please use the LaTeX 'article' class
  - o   do not add coding to 'force' line breaks or the positioning of 'floats', as this coding will need to be removed in the conversion of the file to XML
  - o   To submit your manuscript to ScholarOne Manuscripts, please combine all of your LaTeX and EPS (figure) source files into a single PDF and upload this file as your designated 'Main Document'. (This will be used as a reference file.)
  - o   Please then upload all LaTeX and EPS (figure) source files as a single zipped folder designated as a 'TeX/LaTeX Source Folder'.
- Keep files as small as possible to facilitate information transmission (max 50 MB)
- With the exception of LaTeX support files as outlined above, do not use any form of compression or zipping, as these can interfere with our upload process.

Note: If accepted you must supply the manuscript in an editable format (Word, LaTeX), separate files for each figure and tables in an editable format (Word or Excel).

Tables and Figures

- Tables and figures should appear after the main text.
- Captions should appear with their respective table or figure.
- Footnotes for tables should be given below the table.
- Colour images are welcome, but authors are charged for colour production in print (see Final MS Preparation).
- In the full-text online edition of the journal, figure captions may be truncated in abbreviated links to the full screen version. Therefore, the first 100 characters of any caption should inform the reader of key aspects of the figure.

Preparation of Figures

- Almost all figures submitted to Molecular Ecology should be vector graphics, as these are clear at all magnifications and reproduce well both in print and online.
- Graphs should always be saved directly as .eps or .pdf files from a professional graphics program (e.g., R)

possible.

- • Photographic images can be in a pixel-based format, but please ensure that these are saved as .tif files with at least 300 dpi, or (failing that) a .jpg with no compression.
- • Prepare figures such that, after reduction to fit across one column, two-thirds page width, or two columns (80 mm, 112 mm, or 169 mm, respectively) as required, all lettering and symbols will be clear and easy to read,
  - o i.e., no axes labels should be too large or too small. Further details are available at http://authorservices.wiley.com/bauthor/illustration.asp.

≡ Journal   ⠿ Articles   Actions ⌄

media from the publisher of the original source, and for supplying Wiley with that permission

Charges for Colour

- • Figures published in Molecular Ecology will appear in colour in the online version of the article, at no cost to authors.
  - o In the 'Colour Online Only' option, figures have the colour saturation of the original version reduced to zero for print. As a result, we recommend authors consider paying for colour printing if their figures and captions do not convey the same information in greyscale as they do in colour. More information on making figures that are legible in colour online and greyscale in print can be found at http://www.molecularecologist.com/figure-guidelines/.
- • It is journal policy that authors pay the full cost for any print reproduction of colour artwork.
  - o Please contact the production staff for current colour figure charges: mec@wiley.com

To learn about options for help with figure preparation, please seeWiley Editing Services.

Related Manuscripts

Reviewers and Editors often ask to see unpublished manuscripts (i.e. 'in press', 'in review' or 'submitted') that appear to be related to the submitted paper. As obtaining these during the review process adds unnecessary delays, we request that these related manuscripts are uploaded as 'supplemental files for review only' at the submission stage.

Parts of the Manuscript

Separate files should be uploaded for the main text and for each figure.

Original Articles and From the Cover Articles include, in this order:

Main Text File

The text file should be presented in the following order:

Title Page

1. A short, informative title containing the major key words within the first 65 characters.
    a. see Wiley's best practice SEO tips
    b. The title should not contain abbreviations
    c. It's fun to present your work with a really clever title, but don't let it compromise the discoverability of the work. The best way to have some fun and make sure your work will be found on internet searches is to present the title this way:

Accurate Scientific Title With Keywords: Fun, Catchy, Clever Title

2. A short running title of less than 45 characters;
3. The full names and affiliation of the authors;
    a. The author's institutional affiliations should be that where the work was conducted

c.   Please refer to the journal's Authorship policy in the Editorial Policies and Ethical Considerations section for details on author listing eligibility.

Abstract

Please provide an abstract of no more than 250 words containing the major keywords.

Keywords

Please provide four to six keywords.

☰ Journal   ⠿ Articles   Actions ⌄

~~Results (separate from Discussion)~~

Discussion

Acknowledgements

- These should briefly give credit to other people who have made a contribution to the study.
- Ensure that all relevant grant numbers are listed.

References

Please see this document for examples of implementing APA Reference Style.

Data Accessibility Statement

Authors are required to archive their data in a publicly accessible repository such as Dryad, FigShare, GenBank, etc. (not a laboratory homepage).

- Upon submission, this statement must be included, but can describe curation plans prior to data having been thus archived.
- Upon acceptance, data must be archived and the Data Accessibility statement completed including database and information such as accession numbers or DOI (as available) for all data from the manuscript.
- Note: if data, scripts, or other artefacts used to generate the analyses presented in the paper are available via a publicly available data repository, authors should include a reference to the location of the material within their paper.

Example:

"Data Accessibility:
- DNA sequences: Genbank accessions F234391-F234402; NCBI SRA: SRX0110215
- Final DNA sequence assembly uploaded as online
- Climate data and MaxEnt input files: Dryad doi:10.5521/dryad.12311
- Sampling locations, morphological data and microsatellite genotypes: Dryad doi:10.5521/dryad.12311"

Manuscripts lacking a Data Accessibility section will not be passed through to an editor. Please note that reviewers will be asked to comment on the completeness of this section.

Author Contributions

Authors should include a brief Author Contributions statement at the end of the paper in which they describe their specific contributions to the published work. Contributions could include, but are not limited to:

- designed research
- performed research
- contributed new reagents or analytical tools

Tables

Tables should be self-contained and complement, not duplicate, information contained in the text. Specifics:

- Must be editable files, not pasted as images
- Legends should be concise but comprehensive – the table, legend, and footnotes must be understandable without reference to the text
- All abbreviations must be defined in footnotes.

≡ Journal   ⸬ Articles   Actions ⌄

- Statistical measures such as SD or SEM should be identified in the headings.

Figure Legends

Legends should be concise but comprehensive – the figure and its legend must be understandable without reference to the text. Include definitions of any symbols used and define/explain all abbreviations and units of measurement.

Figures

Although authors are encouraged to send the highest-quality figures possible, for peer-review purposes, a wide variety of formats, sizes, and resolutions are accepted. Click here for the basic figure requirements for figures submitted with manuscripts for initial peer review, as well as the more detailed post-acceptance figure requirements.

Figures submitted in colour

- will be reproduced in colour online free of charge
- Please note, however, that it is preferable that line figures (e.g. graphs and charts) are supplied in black and white so that they are legible if printed by a reader in black and white
- to have figures printed in colour in hard copies of the journal, a fee will be charged by the Publisher.

Additional Files

Supporting/Supplemental Information

Supporting information is information that is not essential to the article, but provides greater depth and background. It is hosted online and appears without editing or typesetting. It may include tables, figures, videos, datasets, etc. Click here for Wiley's FAQs on supporting information.

- If feasible, consider using this branded Molecular Ecology Supporting Information Document.
- Place article title and all author names at the beginning of the Supplemental document
- Consolidate Supplemental information into as few files as possible
- If the document becomes very large, prepare a Table of Contents for the document
- Supporting Information should be uploaded in a separate file and given the file designation 'Supporting Information for online publication.'
- This material will not appear in the PDF Version of Record

Supporting Information will not be edited or altered from its original format during the production process, and therefore proofs of your Supporting Information will not be available. Supporting Information will appear online when your article is published.

Cover Image

- Specs for the Cover Image must be:
  - o   Height - 148.51 mm
  - o   Width - 156.13 mm
  - o   Dpi - 150 dpi

Wiley Author Resources
*Manuscript Preparation Tips:*Wiley has a range of resources for authors preparing manuscripts for submission

≡ Journal   ⠿ Articles   Actions ⌄

Editing, Translation, and Formatting Support:Wiley Editing Services greatly improve the chances of a manuscript being accepted. Offering expert help in English language editing, translation, manuscript formatting, and figure preparation, Wiley Editing Services ensures that the manuscript is ready for submission.

5. EDITORIAL POLICIES AND ETHICAL CONSIDERATIONS

Editorial Review and Acceptance
The acceptance criteria for all papers are the quality and originality of the research, its fit to the scope of the journal and significance to journal readership.
Wiley's policy on confidentiality of the review process is available here.

Referrals to the Open Access Journal Ecology and Evolution
 For rapid publication of quality research that we are unable to accept, select authors of declined manuscripts will be offered the option of having the paper, along with any related reviews, automatically transferred for consideration by Ecology and Evolution.Ecology and Evolution is a Wiley Open Access journal and article publication fees apply.

- •   Authors will not need to reformat or rewrite their manuscript at this stage, and publication decisions will be made a short time after the transfer takes place.
- •   Once the referral is made, the manuscript will be held in a secure Wiley FTP site that is not accessed until authors request to transfer their manuscript.
- •   The Editor of Ecology and Evolution will accept submissions reporting well-conducted research that reaches the standard acceptable for publication. Accepted papers can be published rapidly, typically within 15 days of acceptance.

Data Storage and Documentation

Archiving of data in a publicly accessible repository is mandatory for publication in *Molecular Ecology.*

We require that authors include a 'Data Accessibility' section after the References (see 'Preparing the Submission' Section for details). This section must be present at initial submission, and data archiving must be completed before final acceptance.

Data are important products of the scientific enterprise, and they should be preserved and usable for decades in the future. As such, Molecular Ecology requires authors to archive the data supporting their results and conclusions along with sufficient details so that a third party can interpret them correctly. Papers with exemplary data and code archiving are more valuable for future research, and, all else being equal, will be given higher priority for publication.

TreeBASE, Dryad, FigShare, the Knowledge Network for Biocomplexity, your own institutional or funder repository, or as Supporting Information on the Molecular Ecology web site.

- The utility of archived data is greatly enhanced when the scripts and input files used in the analyses are also made available.
- Given that scripts may be a mix of proprietary and freely available code, their deposition is not compulsory, but we nonetheless strongly encourage authors to make these scripts available whenever possible.
- Software and documentation may be made accessible from a long-term server (e.g., github), however, at

continued access to these resources is ensured.

- Whitlock et al. (2010), states that accurate interpretation of data will likely "require a short additional text document, with details specifying the meaning of each column in the data set. The preparation of such shareable data sets will be easiest if these files are prepared as part of the data analysis phase of the preparation of the paper, rather than after acceptance of a manuscript."
- For additional guidelines on data deposition best practice, please visit http://datadryad.org/depositing.

Data must be publicly available at time of publication. Embargos may be granted in exceptional instances at the discretion of the Managing Editor. Exemptions to this policy may also be granted, especially for sensitive information such as human subject data or the location of endangered species.

Preprints

Molecular Ecology will consider submissions that have previously been made available online, either on a preprint server like arXiv, bioRxiv, or PeerJ PrePrints, or on the authors' own website. However, any such submissions must not have been published in a scientific journal, book or other venue that could be considered formal publication. Authors must inform the editorial office at submission if their paper has been made available as a preprint.

- Authors of accepted papers that were made available as preprints must be able to assign copyright to Molecular Ecology, or agree to the terms of the Wiley Open Access agreement and pay the associated fee.
- Given that the measurable impact of the article is diminished when citations are split between the preprint and the published article, authors are required to:
    - o update the entry on the preprint server so that it links to and cites the DOI for the published version
    - o cite only the published article themselves.

Independent Peer Review Services

Molecular Ecology will consider referrals from independent review services. However such manuscripts may be subject to additional external review by Molecular Ecology. If appropriate, we will invite authors to submit a revision of their manuscript to Molecular Ecology. Note that we cannot guarantee a positive decision for referred manuscripts.

Human Studies and Subjects

For manuscripts reporting medical studies that involve human participants, a statement identifying the ethics committee that approved the study and confirmation that the study conforms to recognized standards is required, for example: Declaration of Helsinki; US Federal Policy for the Protection of Human Subjects; or European Medicines Agency Guidelines for Good Clinical Practice.

Images and information from individual participants will only be published where the authors have obtained the individual's free prior informed consent. Authors do not need to provide a copy of the consent form to the publisher; however, in signing the author license to publish, authors are required to confirm that consent has been obtained.

A statement indicating that the protocol and procedures employed were ethically reviewed and approved, as well as the name of the body giving approval, must be included in the Methods section of the manuscript. Authors are encouraged to adhere to animal research reporting standards, for example the ARRIVE reporting guidelines for reporting study design and statistical analysis; experimental procedures; experimental animals and housing and husbandry. Authors should also state whether experiments were performed in accordance with relevant institutional and national guidelines for the care and use of laboratory animals:

- US authors should cite compliance with the US National Research Council's Guide for the Care and Use of

☰ Journal   ⠿ Articles   Actions ⌄

- UK authors should conform to UK legislation under the Animals (Scientific Procedures) Act 1986 Amendment Regulations (SI 2012/3039).
- European authors outside the UK should conform to Directive 2010/63/EU.

Compliance with International Conventions and Regulations on Biological Diversity and Endangered Species
We strongly recommend that papers submitted to Molecular Ecology comply with the Convention on Biological Diversity and the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CBD and CITES). Within the CBD, we ask that authors follow the Access to Benefit Sharing (ABS) guidelines, and give credit and equal access to benefits to countries, academic institutions and scientists that participated in the collection and analysis of data. Under the CITES convention, we request that authors observe the need for permits for the import and export of specimens that fall under CITES guidelines.

Compliance with Laws on Animal Experimentation and Sampling from Natural Populations
We expect that papers submitted to Molecular Ecology comply with the ARRIVE guidelines for the use of animals in research (http://www.nc3rs.org.uk/ARRIVE), as well as any other legal requirements of the countries where the work was conducted. Sampling procedures for natural populations must be properly described and should be designed to minimize their impact on the taxa involved and their habitat. They must also comply with any international and national legal requirements.

Research Reporting Guidelines
Accurate and complete reporting enables readers to fully appraise research, replicate it, and use it. Authors are encouraged to adhere to the following research reporting standards.

- ARRIVE guidelines

Species Names
Upon its first use in the title, abstract, and text, the common name of a species should be followed by the scientific name (genus, species, and authority) in parentheses. For well-known species, however, scientific names may be omitted from article titles. If no common name exists in English, only the scientific name should be used.

Number of Loci, Populations and Individuals
Sampling strategies and marker choices should be designed to best address the question motivating the study. Unless there are exceptional circumstances, authors of single species phylogeographic studies must base their inferences on multiple loci: our editors and reviewers often question the reliability of inferences based on a single locus and such manuscripts will typically not be sent out for review.
We have been reluctant to formulate guidelines regarding the minimum number of independent loci, populations and individuals needed for publication in Molecular Ecology, largely because such guidelines would depend on the

guidance on this policy.

Data Analysis Best Practice
Molecular Ecology expects that statistical and molecular tools used in submitted papers should meet a high standard of rigor. All analytical approaches have inherent limitations, and authors should therefore attempt to identify the limitations of their chosen approach and corroborate their interpretations when possible.

examples of acceptable nomenclature are provided.

Sequence Data
Nucleotide sequence data can be submitted in electronic form to any of the three major collaborative databases: DDBJ, EMBL, or GenBank. It is only necessary to submit to one database as data are exchanged between DDBJ, EMBL, and GenBank on a daily basis. The suggested wording for referring to accession-number information is: 'These sequence data have been submitted to the DDBJ/EMBL/GenBank databases under accession number U12345'. Addresses are as follows:

- DNA Data Bank of Japan (DDBJ) www.ddbj.nig.ac.jp
- EMBL Nucleotide Archive: ebi.ac.uk/ena
- GenBank www.ncbi.nlm.nih.gov/genbank

Proteins sequence data should be submitted to either of the following repositories.
- Protein Information Resource (PIR): pir.georgetown.edu
- SWISS-PROT: expasy.ch/sprot/sprot-top

Structural Data
For papers describing structural data, atomic coordinates and the associated experimental data should be deposited in the appropriate databank (see below). **Please note that the data in databanks must be released, at the latest, upon publication of the article.** We trust in the cooperation of our authors to ensure that atomic coordinates and experimental data are released on time.

- **Organic and organometallic compounds**: Crystallographic data should not be sent as Supporting Information, but should be deposited with the Cambridge Crystallographic Data Centre (CCDC) at ccdc.cam.ac.uk/services/structure_deposit.
- **Inorganic compounds**: Fachinformationszentrum Karlsruhe (FIZ; fiz-karlsruhe.de).
- **Proteins and nucleic acids:** Protein Data Bank (rcsb.org/pdb).
- **NMR spectroscopy data**: BioMagResBank (bmrb.wisc.edu).

Use of RAPD/ISSR Markers
The appropriateness of RAPD or ISSR markers for population genetic inference is increasingly questioned by our reviewers and editors because of concerns about reproducibility, dominance, and homology. Given these worries, and the ready availability of other kinds of markers that do not suffer from all of these problems, studies based primarily on RAPD/ISSR rarely pass the scrutiny of peer review in Molecular Ecology.

Reporting Stable Isotope Data
For papers using stable isotopes, we recommend that authors follow the guidelines developed by the IUPAC

Software and Programming Code

Software and documentation may be made accessible from a long-term server (e.g., github), however, at least a snapshot of these resources must be posted on Dryad, CRAN (or similar academic/publishing archiving sites) with a link to a long-term server where software development and future releases can be found such that continued access to these resources is ensured.

Conflict of Interest

The journal requires that all authors disclose any potential sources of conflict of interest. Any interest or relationship

describe in their manuscript. Potential sources of conflict of interest include, but are not limited to: patent or stock ownership, membership of a company board of directors, membership of an advisory board or committee for a company, and consultancy for or receipt of speaker's fees from a company. The existence of a conflict of interest does not preclude publication. If the authors have no conflict of interest to declare, they must also state this at submission. It is the responsibility of the corresponding author to review this policy with all authors and collectively to disclose with the submission ALL pertinent commercial and other relationships.

Funding

Authors should list all funding sources in the Acknowledgments section. Authors are responsible for the accuracy of their funder designation. If in doubt, please check the Open Funder Registry for the correct nomenclature: https://www.crossref.org/services/funder-registry/

Authorship

The list of authors should accurately illustrate who contributed to the work and how. All those listed as authors should qualify for authorship according to the following criteria:
1. Have made substantial contributions to conception and design, or acquisition of data, or analysis and interpretation of data;
2. Been involved in drafting the manuscript or revising it critically for important intellectual content;
3. Given final approval of the version to be published. Each author should have participated sufficiently in the work to take public responsibility for appropriate portions of the content; and
4. Agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Contributions from anyone who does not meet the criteria for authorship should be listed, with permission from the contributor, in an Acknowledgments section (for example, to recognize contributions from people who provided technical help, collation of data, writing assistance, acquisition of funding, or a department chairperson who provided general support). Prior to submitting the article all authors should agree on the order in which their names will be listed in the manuscript.

Additional Authorship Options: Joint first or senior authorship: In the case of joint first authorship, a footnote should be added to the author listing, e.g. 'X and Y should be considered joint first author' or 'X and Y should be considered joint senior author.'

ORCID

As part of the journal's commitment to supporting authors at every step of the publishing process, the journal requires the submitting author (only) to provide an ORCID iD when submitting a manuscript. This takes around 2 minutes to complete. Find more information here.

Publishing Ethics Tips for Authors here. Wiley's Publication Ethics Guidelines can be found at authorservices.wiley.com/ethics-guidelines/index.html.

Privacy

By submitting a manuscript to or reviewing for this publication, your name, email address, and affiliation, and other contact details the publication might require, will be used for the regular operations of the publication, including, when necessary, sharing with the publisher (Wiley) and partners for production and publication. The publication and the publisher recognize the importance of

☰ Journal   ⋮⋮ Articles   Actions ⌄

personal data collected and processed. You can learn more at https://authorservices.wiley.com/statements/data-protection-policy.html.

6. AUTHOR LICENSING

If a paper is accepted for publication, the author identified as the formal corresponding author will receive an email prompting them to log in to Author Services, where via the Wiley Author Licensing Service (WALS) they will be required to complete a copyright license agreement on behalf of all authors of the paper.

Authors may choose to publish under the terms of the journal's standard copyright agreement, or OnlineOpen under the terms of a Creative Commons License.

General information regarding licensing and copyright is available here. To review the Creative Commons License options offered under OnlineOpen, please click here. (Note that certain funders mandate a particular type of CC license be used; to check this please click here.)
Self-Archiving Definitions and Policies: Note that the journal's standard copyright agreement allows for self-archiving of different versions of the article under specific conditions. Please click here for more detailed information about self-archiving definitions and policies.
Open Access fees: Authors who choose to publish using OnlineOpen will be charged a fee. A list of Article Publication Charges for Wiley journals is available here.
Funder Open Access: Please click here for more information on Wiley's compliance with specific Funder Open Access Policies.

7. PUBLICATION PROCESS AFTER ACCEPTANCE
Accepted Article Received in Production
When an accepted article is received by Wiley's production team, the corresponding author will receive an email asking them to login or register with Wiley Author Services. The author will be asked to sign a publication license at this point.
Accepted Articles
The journal offers Wiley's Accepted Articles service for all manuscripts. This service ensures that accepted 'in press' manuscripts are published online shortly after acceptance, prior to copy-editing or typesetting. Accepted Articles are published online a few days after final acceptance, appear in PDF format only, and are given a Digital Object Identifier (DOI), which allows them to be cited and tracked. After publication of the final version article (the article of record), the DOI remains valid and can still be used to cite and access the article.
Proofs
Once the paper is typeset, the author will receive an email notification with the URL to download a PDF typeset page

receipt of first proof.

Publication Charges

Colour figures. Colour figures may be published online free of charge; however, the journal charges for publishing figures in colour in print. If the author supplies colour figures, they will be sent a Colour Work Agreement once the accepted paper moves to the production process. If the Colour Work Agreement is not returned by the specified date, figures will be converted to black and white for print publication.

Early View

≡ Journal   ⠿ Articles   Actions ⌄

DOI for citations.

8. POST PUBLICATION

Video Abstracts

A video abstract can be a quick way to make the message of your research accessible to a much larger audience. Wiley and its partner Research Square offer a service of professionally produced video abstracts, available to authors of articles accepted in this journal. You can learn more about it at www.wileyauthors.com/videoabstracts . If you have any questions, please direct them to videoabstracts@wiley.com .

Access and Sharing

When the article is published online:

- The author receives an email alert (if requested).
- The link to the published article can be shared through social media.
- The author will have free access to the paper (after accepting the Terms & Conditions of use, they can view the article).
- The corresponding author and co-authors can nominate up to ten colleagues to receive a publication alert and free online access to the article.

Email offprint@cosprinters.com

To find out how to best promote an article, click here.

Measuring the Impact of an Article

Wiley also helps authors measure the impact of their research through specialist partnerships with Kudos and Altmetric.

9. EDITORIAL OFFICE CONTACT DETAILS

Administration: molecol@wile.ycom

Managing Editor: manager.molecol@wiley.com

Author Guidelines updated May, 2017

Tools

✍ **Submit an Article**

📄 **Browse free sample issue**

👍 **Recommend to a librarian**

✪ **Subscribe to this journal**

More from this journal

☰ Journal   Articles   Actions ⌄

Wiley Job Network
Jobs

Related Title

MOLECULAR ECOLOGY RESOURCES

ISSUE

Volume 18, Issue 3
Pages: **381-728**
May 2018

About Wiley Online Library   ⌄

Help & Support   ⌄

Opportunities   ⌄

Connect with Wiley   ⌄
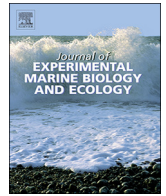
Reassessment of the franciscana *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) distribution and niche characteristics in Brazil

Karina Bohrer do Amaral[a,b,*], Daniel Danilewicz[c,d,e], Alexandre Zerbini[d,f,g], Ana Paula Di Beneditto[h], Artur Andriolo[d,i], Diego Janisch Alvares[b,j], Eduardo Secchi[k], Emanuel Ferreira[l], Federico Sucunza[c,d,i], Márcio Borges-Martins[b,c,j], Marcos César de Oliveira Santos[m], Marta Cremer[n], Pablo Denuncio[o], Paulo Henrique Ott[c,p], Ignacio Benites Moreno[a,b,q]

[a] *Laboratório de Sistemática e Ecologia de Aves e Mamíferos Marinhos (LABSMAR), Universidade Federal do Rio Grande do Sul, RS, Brazil*
[b] *Programa de Pós-Graduação em Biologia Animal, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, 91501-70 Porto Alegre, RS, Brazil*
[c] *Grupo de Estudos de Mamíferos Aquáticos do Rio Grande do Sul (GEMARS), Rua Bento Gonçalves, 165/1002, 95560-000 Torres, RS, Brazil*
[d] *Instituto Aqualie, Av. Dr. Paulo Japiassu Coelho, 714, Sala 206, 36033-310 Juiz de Fora, MG, Brazil*
[e] *Programa de Pós-Graduação em Zoologia, Universidade Estadual de Santa Cruz, Rodovia Ilhéus/Itabuna, km 16, 45662-900 Ilhéus, BA, Brazil*
[f] *National Marine Mammal Laboratory, Alaska Fisheries Science Center/NOAA, 7600 Sand Point Way NE, Seattle, WA 98115-649, USA*
[g] *Cascadia Research Collective, 2181/2 W 4th Ave, Olympia, WA 98501, USA*
[h] *Laboratório de Ciências Ambientais (CBB), Universidade Estadual do Norte Fluminense, Avenida Alberto Lamego, 2000, 28013-602 Campos, RJ, Brazil*
[i] *Laboratório de Ecologia Comportamental e Bioacústica, Departamento de Zoologia, Instituto de Ciências Biológicas, Universidade Federal de Juiz de Fora, Rua José Lourenço Kelmer, s/n - Campus Universitário, 36036-900 Juiz de Fora, MG, Brazil*
[j] *Laboratório de Herpetologia, Departamento de Zoologia, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Avenida Bento Gonçalves, 9500, Bloco IV, Prédio 43435, sala 102, Porto Alegre, RS 91501-70, Brazil*
[k] *Laboratório de Ecologia e Conservação da Megafauna Marinha-EcoMega, Instituto de Oceanografia, Universidade Federal do Rio Grande-FURG, Caixa Postal 474, 96203-900 Rio Grande, RS, Brazil*
[l] *Associação R3 Animal, Rodovia João Gualberto Soares – Entrada do Parque Estadual do Rio Vermelho, 88061-500 Florianópolis, SC, Brazil*
[m] *Laboratório de Biologia da Conservação de Mamíferos Aquáticos (LABCMA), Departamento de Oceanografia Biológica, Instituto Oceanográfico, Universidade de São Paulo, Praça do Oceanográfico, 191, Sala 145-A, 05508-120 São Paulo, SP, Brazil*
[n] *Projeto Toninhas, Universidade da Região de Joinville (UNIVILLE), São Francisco do Sul, SC, Brazil*
[o] *Instituto de Investigaciones Marinas y Costeras (IIMyC-CONICET), Departamento de Ciencias Marinas, Universidad Nacional de Mar del Plata (UNMDP), Casilla de Correo 1260, Mar del Plata, Argentina*
[p] *Universidade Estadual do Rio Grande do Sul (Uergs), Unidade do Litoral Norte, Rua Machado de Assis, 1456, 95520-000 Osório, RS, Brazil*
[q] *Centro de Estudos Costeiros, Limnológicos e Marinhos (CECLIMAR/UFRGS), Campus Litoral Norte, Universidade Federal do Rio Grande do Sul, Avenida Tramandaí, 976, 95625-000 Imbé, RS, Brazil*

ARTICLE INFO

ABSTRACT

The franciscana (*Pontoporia blainvillei*) is the most threatened small cetacean of South America. The species is endemic to coastal waters of the western South Atlantic Ocean, where it is distributed from Itaúnas (Brazil) to Golfo San Matias (Argentina). Its range was divided in four Franciscana Management Areas (FMAs) for conservation purposes. However, the distribution of the franciscana is not continuous along its range, with two hiatuses proposed in southeastern Brazilian coast. The absence of franciscana records in these regions has been confirmed by multiple years of research, however the reasons for this discontinuous distribution is not well understood. In this study, information on the distribution of the franciscana in south and southeastern Brazil is updated and new limits for FMAs are proposed. NicheA 3.0 software was used to investigate the environmental suitability of distributional gaps in relation to four weakly correlated, allegedly relevant descriptors of franciscana's distribution. In total, 788 records from dedicated aerial and boat surveys and bycatch were used to verify and to confirm the new FMAs limits proposed by franciscana's experts previously. The distributional gaps were reshaped and defined as following: Gap I from Piraquê-Açu River Mouth, Santa Cruz (19°57′S) in the state of Espírito Santo to Barra de Itabapoana (21°18′S) in the state of Rio de Janeiro; and Gap II from Armação dos

Búzios (22°44′S) to Piraquara de Dentro (22°59′S) in Rio de Janeiro. The ecological niche model indicated that distributional gaps are inside franciscana's fundamental niche, and are relatively suitable in terms of salinity, temperature, diffuse attenuation and bathymetry. However, the narrow of continental shelf seems to be the main factor explaining the absence of franciscanas in the distributional gaps as well as for the differentiation of some of the FMAs proposed. Narrowness of continental shelf seems to be intensifying the dynamics of biotic interactions promoting food competition for example, and/or causing geographic limitation to maintain minimal viable population size in present or past times periods.

1. Introduction

The franciscana *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) is the most threatened small cetacean of South America (Secchi et al., 2003a). Mortality in gillnets have been impacting franciscana dolphins throughout their range for at least 50 years (e.g. Ott et al., 2002; Prado et al., 2013, 2016; Secchi et al., 2003a, 2003b), compromising the viability of its populations (Kinas, 2002; Secchi, 2006). The franciscana faces a high risk of extinction and is listed as "Vulnerable" on a global scale by IUCN (Zerbini et al., 2017), while regionally in Brazil it is officially listed as "Critically Endangered" (MMA, 2014).

The franciscana is endemic to coastal waters of Brazil, Uruguay, and Argentina. Currently, the species occurs from Itaúnas (18°25′S), in the state of Espírito Santo, southeastern Brazil (Siciliano et al., 2002) to Golfo San Matias (41°10′S), Rio Negro, Argentina (Crespo et al., 1998). Early studies showed evidence that franciscana is not continuously distributed along its range in Brazil (Siciliano et al., 2002). Many years

of bycatch monitoring, beach surveys for stranded animals and aerial surveys confirms the existence of two distributional gaps: (1) from Regência (19°40′S), in Espírito Santo, to Barra do Itabapoana (21°18′S), in the state of Rio de Janeiro, namely northern distributional gap (Gap I); and (2) from Macaé (22°25′S) to Ilha Grande (23°09′S), in Rio de Janeiro, namely southern distributional gap (Gap II) (e.g. Azevedo et al., 2002; Danilewicz et al., 2012; de Moura et al., 2009). Systematic and long-term monitoring has confirmed the absence of franciscanas, mainly in the central portion of these gaps (e.g. de Moura et al., 2009). However, there is no consensus about the exact boundaries of the gaps (e.g. Azevedo et al., 2002; Siciliano et al., 2015) which play an important role in the delineation of management units for the species (Secchi et al., 2003a).

Previous studies revealed the existence of geographical population structure based on external morphology and genetic markers (e.g. Higa et al., 2002; Ott, 2002; Pinedo, 1995; Ramos et al., 2002; Secchi et al., 1998). After applying a multi-methodological approach for identifying
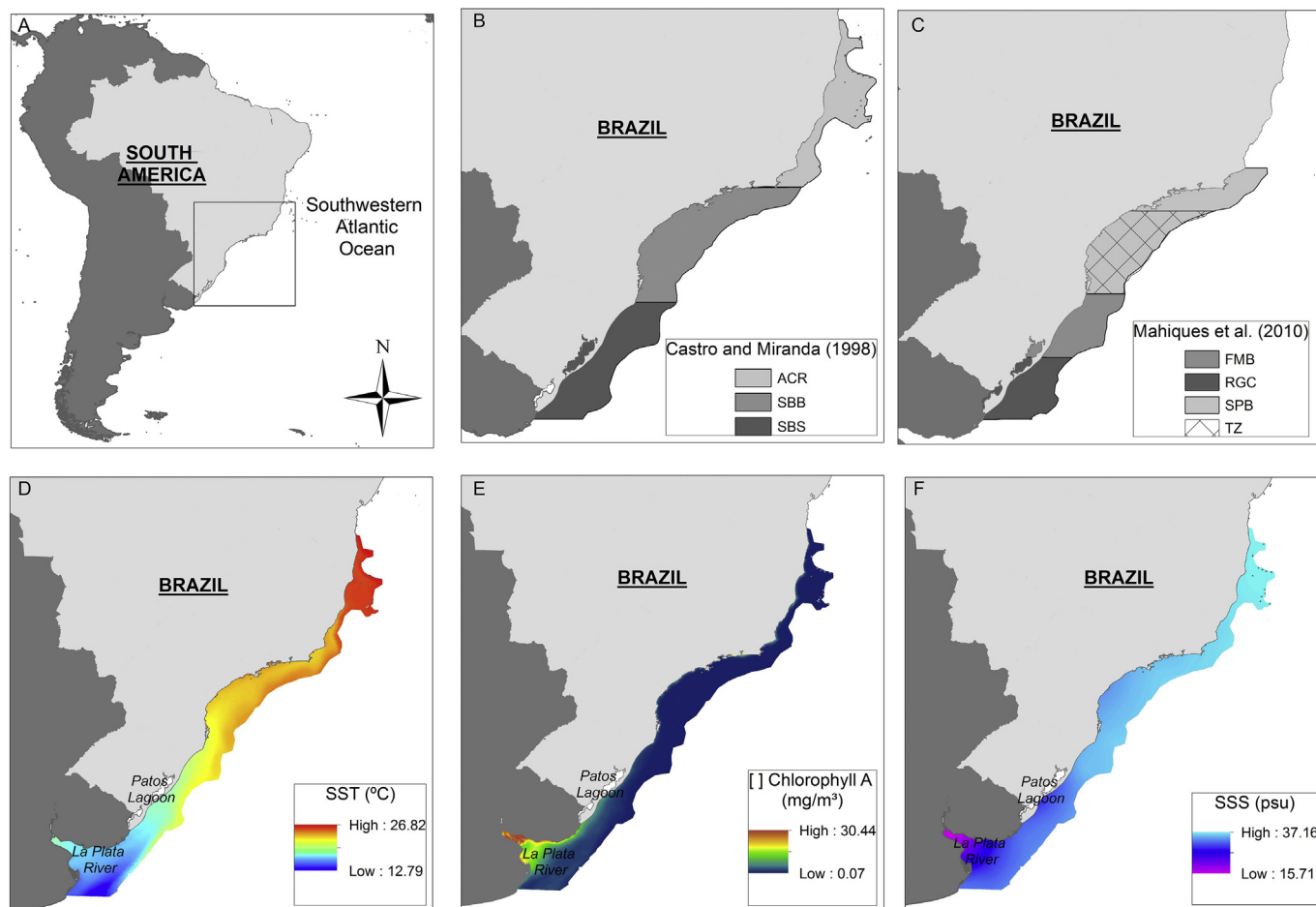

Fig. 1. A) Study area of franciscana dolphin distribution. Brazilian continental shelf zones proposed by B) Castro and Miranda (1998) and C) Mahiques et al. (2010). Representation of annual means of D) Mean Annual Sea Surface Temperature (SST), E) Mean Annual Concentration of Chlorophyll A, and F) Mean Annual Sea Surface Salinity (SSS). Abbreviations: ACR, Abrolhos – Campos Region; SBB, South Brazilian Bight; SBS, Southern Brazilian Shelf; SPB, São Paulo Bight; FMB, Florianópolis – Mostardas Bight (FMB); RGC, Rio Grande Cone; TZ, Transitional Zone.

stock discreteness, Secchi et al. (2003a) divided the franciscana's range into four Franciscana Management Areas (FMAs) (please see Fig. 1 of the refereed article). FMA I and FMA II are located exclusively in southeastern and southern Brazil, FMA III includes southern Brazil and Uruguay, and FMA IV encompasses the range of the species in Argentina. These management divisions are supported by recent data on pollutant loads, diet, external morphology and parasites (e.g. Alonso et al., 2012; Barbato et al., 2012; Costa-Urrutia et al., 2012; de la Torre et al., 2012; Hoss et al., 2017). New studies have suggested the need of reformulation of the former FMA's subdivisions (e.g. Gariboldi et al., 2015, 2016; Mendez et al., 2010), including the separation of FMA I in two distinct management units (FMA Ia and FMA Ib) separated by the northern distributional gap (Anonymous, 2015; Cunha et al., 2014).

The increased effort from properly designed aerial surveys to estimate franciscana's abundance (e.g. Danilewicz et al., 2010, 2012; Zerbini et al., 2011) and long-term projects evaluating franciscana bycatch (see Material and Methods) have provided many georeferenced at-sea records for the species. These data have been useful to characterise the distributional ecology of franciscanas' populations in a comprehensive manner and can be used to perform ecological niche modeling in order to investigate factors that influence their distribution.

Correlative species distribution models are based on algorithms that estimate ecological niches and explore potential distributional areas by assessing relationships between species occurrences and environmental information (Qiao et al., 2016). Niche modeling approaches dramatically expanded in recent years and currently several techniques and toolkits are available (Phillips et al., 2006; Qiao et al., 2016). In addition, these techniques have been widely used in studies on the distribution of cetaceans (e.g. do Amaral et al., 2015; Palacios et al., 2013; Rossi-Santos and Oliveira, 2016), including estimates of the potential franciscana distribution (Gomez and Cassini, 2015).

Given the high risk faced by the franciscana, especially the extremely high risk of extinction observed regionally in Brazil (Rocha-Campo et al., 2010), and the importance of distributional ecology to either the process of risk assessment and conservation planning, the aim of this study is (1) to update information on the franciscana distribution in Brazil, including a review of FMAs I, II and III as well as the distributional gaps between them, and (2) to investigate the factors that potentially explain the existence of gaps in the range of the franciscana.

2. Materials and methods

2.1. Study area

The study area includes the Brazilian continental shelf from 18°S to 34°S, including only those waters up to the 50 m isobath (Fig. 1A). The area is characterized by different physical oceanographic processes. Castro and Miranda (1998) therefore proposed a segmentation of the Brazilian continental shelf into six zones, of which three zones are encompassed by the study area: Abrolhos – Campos Region (15°S – 23°S), South Brazilian Bight (23°S – 28°30′S) and Southern Brazilian Shelf (28°30′S – 34°S) (Fig. 1B). These areas are characterized by different features in relation to topography, productivity, sea surface temperatures and salinity due to upwelling, land runoff from several estuaries and convergence of currents (Fig. 1D–F). Conversely to Castro and Miranda (1998), who proposed a division of the Brazilian continental shelf for practical reasons, Mahiques et al. (2010) suggest a division in terms of geology, bathymetry, declivities and the presence of canyons and channels (Fig. 1C).

2.2. Franciscana dataset

Franciscana records used in the present analyses corresponded to observations of live animals in situ through dedicated aerial and boat surveys or to specimen entangled in coastal gillnets fisheries in Brazil

(for which precise location data were available). Only data from the marine environment were considered, therefore franciscana records previously observed in estuarine areas such as Babitonga Bay (Cremer and Simões-Lopes, 2005, 2008) and Paranaguá Bay (Santos et al., 2009) in southern Brazil were not included. Only sightings data from dedicated surveys and georeferenced data from bycatch were used in the present analysis in order to estimate franciscana's fundamental ecological niche. Sampling effort and potential biases associated with non-uniform sampling effort, especially those related to fishery monitoring, have not been considered in this study.

Data from aerial surveys were obtained through dedicated line transect studies designed to assess franciscana distribution and to estimate abundance (details in Danilewicz et al., 2010, 2012; Zerbini et al., 2011). Bycatch data were obtained directly by some of the authors via onboard surveys or logbook information provided by reliable and well known captains of fishing vessels operating along the Brazilian coast from 1992 to 2004 (Danilewicz, 2007; Danilewicz et al., 2009; Ott, 1998; Secchi et al., 1997, 2004). Additional records were obtained from peer-reviewed literature (Di Beneditto, 2003; Di Beneditto et al., 2001; Flores, 2009; Moreno et al., 2003; Santos and Netto, 2005; Santos et al., 2002, 2009; Siciliano et al., 2002).

2.3. Environmental dataset

Ten environmental variables that are considered to influence cetaceans distributions (e.g. Baumgartner et al., 2001; Palacios et al., 2013; Redfern et al., 2006) and specifically franciscanas (Gomez and Cassini, 2015; Siciliano et al., 2002) were initially selected to describe the characteristics of the franciscana's habitats and distributional gaps (Table 1).

Environmental information was obtained from Bio-Oracle (Tyberghein et al., 2012) and MARSPEC (Sbrocco and Barber, 2013). These public databases provide a set of user-friendly and high-resolution GIS data layers of the ocean and were designed for species distribution modeling applications (Sbrocco and Barber, 2013; Tyberghein et al., 2012). The layers consist of global coverage satellite-based and in situ measured data interpolated and assembled at an annual temporal resolution and at different spatial resolutions (1 km and 9 km from MARSPEC and Bio-Oracle datasets, respectively). Geophysical layers were derived from the SRTM30_PLUS high resolution bathymetry dataset (Sbrocco and Barber, 2013), and bioclimatic layers were derived from a long term dataset from NOAA's World Ocean Atlas and NASA's MODIS satellite imagery (Sbrocco and Barber, 2013; Tyberghein et al., 2012; for more details about environmental dataset access: http://www.marspec.org/ and http://www.oracle.ugent.be/). All environmental layers were processed in ArcGIS 10.2.2 (ESRI, 2013) in datum WGS 84, using the same spatial extent (18°S

Table 1

List of environmental variables analyzed in this study and its respective source, resolution and unit.

Environmental variables	Source	Unit	Original resolution
Bathymetry (Depth of the seafloor)	MARSPEC	Meters	1 km
Distance to shore	MARSPEC	Kilometres	1 km
Bathymetric Slope	MARSPEC	Degrees	1 km
Mean Annual Concentration of Chlorophyll A	Bio-Oracle	mg/m^3	9 km
Annual Range in Concentration of Chlorophyll A	Bio-Oracle	mg/m^3	9 km
Mean Annual Diffuse Attenuation	Bio-Oracle	m^{-1}	9 km
Mean Annual Sea Surface Salinity	MARSPEC	Psu	1 km
Annual Range in Sea Surface Salinity	MARSPEC	Psu	1 km
Mean Annual Sea Surface Temperature	MARSPEC	degrees °C	1 km
Annual Range in Sea Surface Temperature	MARSPEC	degrees °C	1 km

to 34˚S at a 9 km resolution.

In order to assess the shelf habitat available for franciscanas in the study area, distance to shore data was obtained from distance to shore layer, in which we extracted its values at 0.5˚ latitudinal intervals along the 25 m and 50 m isobaths.

2.4. Environmental analyses

Non-independence of predictor variables is a well-known problem in ecology (e.g. Dormann et al., 2013), and it is recommended a preliminary selection of layers in order to avoid redundant data layers in ecological niche analysis (e.g. Qiao et al., 2016). Therefore, correlation of environmental layers was assessed, and factorial analyses were used to select variables with low multicollinearity. Collinearity analyses were conducted in R Statistical Software version 3.2.4 (R Development Core Team, 2016) using the corrplot package (Wei and Simko, 2016) on all variables presented in Table 1, with the exception of distance to shore.

Non-parametric tests (Kruskal-Wallis and Dunn tests) were conducted to provide a preliminary assessment of potential differences between occupied and unoccupied areas with respect to environmental variables selected by the factorial analysis. In order to comply with the assumptions of independence and randomization of sampling required by nonparametric tests, sample points randomly distributed throughout the study areas were used. In a first step, polygons were designed representing areas adjacent to the gaps (i.e. FMA Ia, FMA Ib and FMA II) and areas not occupied by franciscana (i.e. gaps). The polygons were constrained longitudinally by the 50 m isobath, and latitudinally by the limits for the new FMAs proposed here (see Results section). In a second step, a number of random points within each polygon were generated

taking into account the proportions of areas (100 points were created within the polygon with the smallest area and so forth). Data were then grouped as "Area occupied by Franciscana (AOF)", Gap I, and Gap II. AOF corresponded to the region between Itaúnas in Espírito Santo, and the center of Ilha de Santa Catarina in the state of Santa Catarina, without a discrimination of FMAs and the exclusion of distributional gaps. Finally, significant differences were tested among the medians of the variables identified by the factorial analysis using a Kruskal-Wallis test followed by the Dunn test. All statistical tests were performed in software R Statistical Software version 3.2.4 (R Development Core Team, 2016) using the nortest (Gross and Ligges, 2015) and the dunn (Dinno, 2017) packages. A significance level of $\alpha = 0.05$ was adopted and the p-value for multiple comparisons was adjusted using the Bonferroni method.

2.5. Ecological niche analysis

NicheA 3.0 (Qiao et al., 2016) was used to investigate if the distributional gaps are consistent with franciscana's fundamental ecological niche. NicheA software generates ecological niche models following the Hutchinsonian approach of an n-multidimensional space, and projects these models in geographic space in the form of continuous species suitability models (for details, see Qiao et al., 2016). NicheA assumes that a species' fundamental ecological is convex in shape, and thus can be operationalized as minimum-volume ellipsoids (MVE) (Qiao et al., 2016). Similar to others modeling approaches (e.g. Maxent – Phillips et al., 2006), MVE could be influenced by sampling biases; however, MVE is only influenced by bias in the periphery of the cloud points. If there are sampling biases that affect the concentrations of points in the interior of the cloud, those will have no effect (A. Townsend Peterson 2017, personal
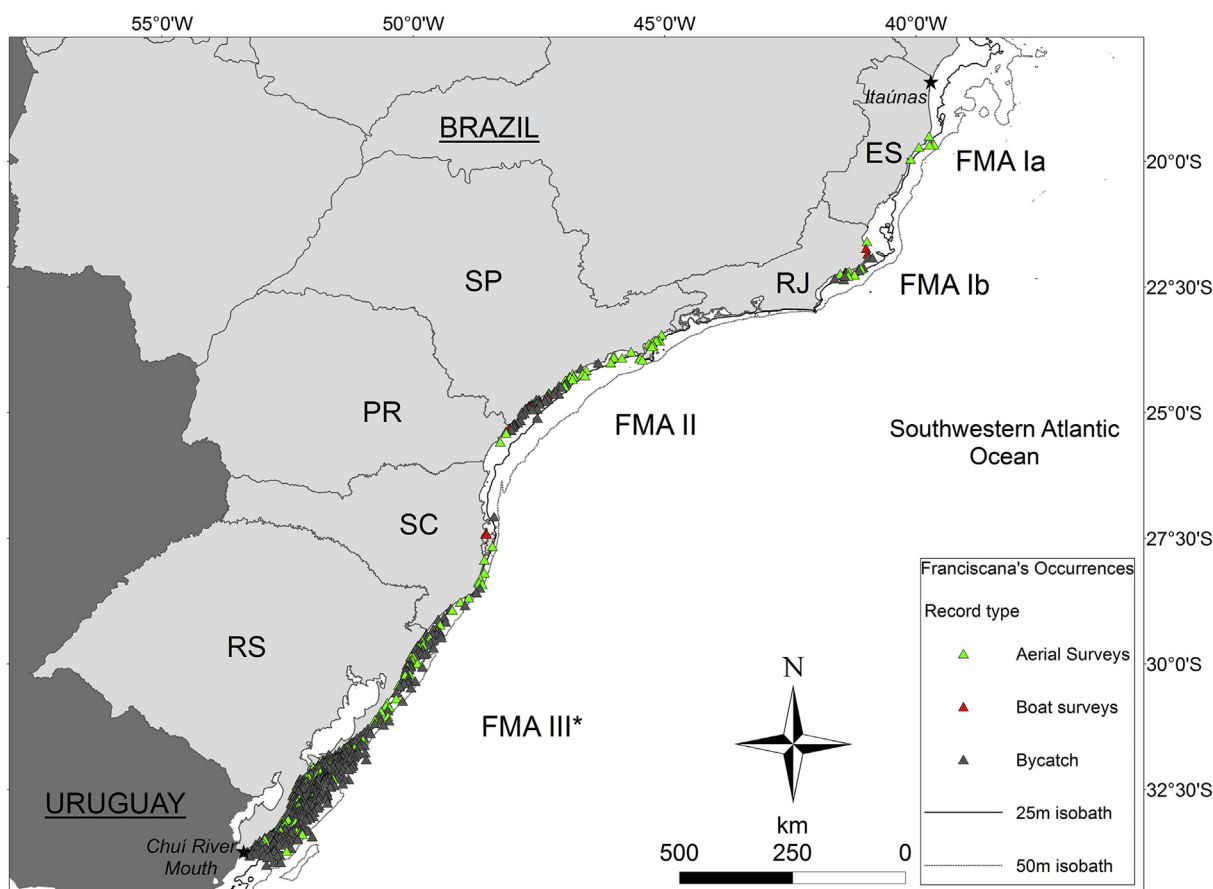

Fig. 2. Compiled records of franciscana dolphin along Brazilian coastal waters from Itaúnas (ES) to Chuí River Mouth (RS). Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SP, São Paulo; PR, Paraná; SC, Santa Catarina; RS, Rio Grande do Sul.

communication). This means that MVE is not influenced by the density of the points (Huiji Qiao 2017, personal communication). Considering the biased nature of the franciscana data set (e.g. uncorrected for effort), NicheA was deemed the most suitable tool to investigate the characteristics of the franciscana's distribution. In order to better represent the franciscana's fundamental niche, all types of records (bycatch, aerial and boats surveys) were pooled.

Finally, MVEs, representing the franciscanas' fundamental ecological niche, were projected to a habitat suitability map. For the MVE, continuous values of suitability were assessed as the Euclidean distance to the niche centroid (Qiao et al., 2016). The most suitable areas are those closest to the niche centroid (with values close to 1), while the most unsuitable are those areas further away from the niche centroid (with values close to 0); areas totally outside of species niche were set to −1 suitability.

3. Results

3.1. Franciscana distribution update

In total, 788 records of franciscanas in Brazil were compiled from Itaúnas (18°25′S) in Espírito Santo to Chuí River Mouth (33°44′S) in the state of Rio Grande do Sul, located on the Brazil-Uruguay border (Fig. 2). Most of the data were collected between 1992 and 2014. Bycatch data represented 78% of these records, sightings from aerial surveys represented 20.9% and sightings from boat surveys accounted for only 1.1% of the overall data (records for each FMA are summarized in Table 2).

Based on the records compiled, a reassessment of the limits of the FMAs and the distributional gaps were proposed (Table 2, Fig. 3). The distributional gaps were defined as following: Gap I is located from Santa Cruz (19°57′S) to Barra de Itabapoana (21°18′S) in Espírito Santo; Gap II is located from Armação dos Búzios (22°44′S) to Piraquara de Dentro (22°59′S) in Rio de Janeiro.

3.2. Environmental layers analyses

From nine layers initially considered to have some influence in the franciscana's distribution, four pairs of environmental layers exhibited correlation coefficient higher than 0.7 (Fig. 4). Therefore, the following environmental layers were selected based on the highest value of each factor of factorial analyses (Table 3): Mean Annual Diffuse Attenuation, Annual Range in Sea Surface Temperature, Mean Annual Sea Surface Salinity, and Bathymetry.

Polygons representing areas adjacent to the gaps (i.e. FMA Ia, FMA Ib and FMA II) and areas not occupied by franciscana (i.e. gaps) are presented in Fig. 5. Considering the proportions of areas, the number of random points created for each polygon is presented in Table 4.

Differences between AOF and Gap I were statistically significant for Mean Annual Sea Surface Salinity (Tables 5 and 6, Fig. 6A), and Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B). Differences between AOF and Gap II were statistically significant for Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B) and Mean Annual Diffuse Attenuation (Tables 5 and 6, Fig. 6C). Gap I and Gap II were statistically differentiated in relation to Mean Annual of Sea Surface Salinity (Tables 5 and 6, Fig. 6A), Annual Range in Sea Surface Temperature (Tables 5 and 6, Fig. 6B) and Mean Annual of Diffuse Attenuation (Tables 5 and 6, Fig. 6C). Bathymetry was not statistically different among the areas analyzed (Table 5, Fig. 6D). In general, Gap I had the highest median of Mean Annual of Sea Surface Salinity; Gap II had the highest median of Mean Annual of Diffuse Attenuation; and, AOF had the highest median of Annual Range in Sea Surface Temperature.

3.3. Ecological niche analysis

The franciscana's Minimum-Volume Ellipsoid (MVE, representing the franciscana's fundamental ecological niche) was estimated using 788 occurrence records in a three-dimensional environmental space represented by Mean Annual Diffuse Attenuation, Annual Range in Sea Surface Temperature, Mean Annual Sea Surface Salinity, and Bathymetry in NicheA.

The franciscana's distribution model (i.e. the MVE projected in geographic space) revealed that the waters in the continental shelf up to 25 m were closest to niche centroid (values close to 1), therefore these areas corresponded to the most suitable habitat for franciscanas (Fig. 7). On the other hand, water depths between 25 m and 50 m isobaths exhibited a progressive decrease of environmental suitability and were more distant from franciscana niche centroids (values close to 0). Gap I and Gap II exhibited values of distance to niche centroid lower than 0.75.

3.4. Shelf habitat availability

The 25 m isobath was very close to the shore in the areas corresponding to distributional gaps (i.e. very little area with shallow waters), while the areas suitable for franciscanas were characterized by shallow waters (up to the 50 m isobath) up to quite some distance from the coast. In the Gap I, 25 m isobaths were identified between 5 km and 30 km of coastal line. In the Gap II, the 25 m isobaths were at < 10 km from shore, as close as just 1 km from the coast line at 23°S (close to Arraial do Cabo in Rio de Janeiro; see Fig. 8).

The location of the 50 m isobath was similar to those of 25 m, being closest to shore in the areas corresponding to the gaps. In the Gap I, 50 m isobaths was > 30 km far from coast line. In the Gap II, 50 m isobaths was positioned closest to shore, being < 1 km far from shore at 23°S (Fig. 8). In addition to the distributional gaps, a marked narrowing of continental shelf is also observed around the Ilha de Santa Catarina (27°35′S) (Fig. 8).

Table 2

Summary of franciscanas' records by areas and data source and gaps limits. FMAs were established according to Cunha et al. (2014) and limits were updated.

Records summary information					
Areas	New limits	Aerial Surveys	Boat Surveys	Bycatch	Total
FMA Ia	Itaúnas, ES (18°25'S) to Santa Cruz, ES (19°57'S)	6	0	0	6
Gap I (north)	Piraquê-Açu River Mouth, Santa Cruz, ES (19°57'S) to Barra de Itabapoana, ES (21°18'S)				
FMA Ib	Barra de Itabapoana, RJ (21°18'S) to Armação de Búzios, RJ (22°44'S)	13	2	11	26
Gap II (south)	Armação dos Búzios, RJ (22°44'S) to Piraquara de Dentro, RJ (22°59'S)				
FMA II	Piraquara de Dentro, RJ (22°59'S) to Ilha de Santa Catarina, SC (27°35'S)	41	7	60	108
FMA III[a]	Ilha de Santa Catarina, SC (27°35'S) to Chuí River Mouth, RS (33°44'S)	105	0	543	648
TOTAL		165	9	614	788
Percentage (%)		20.9%	1.1%	78.0%	100.0%

Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SC, Santa Catarina; RS, Rio Grande do Sul.

[a] FMA III is partially represented, because it extends into Uruguay.
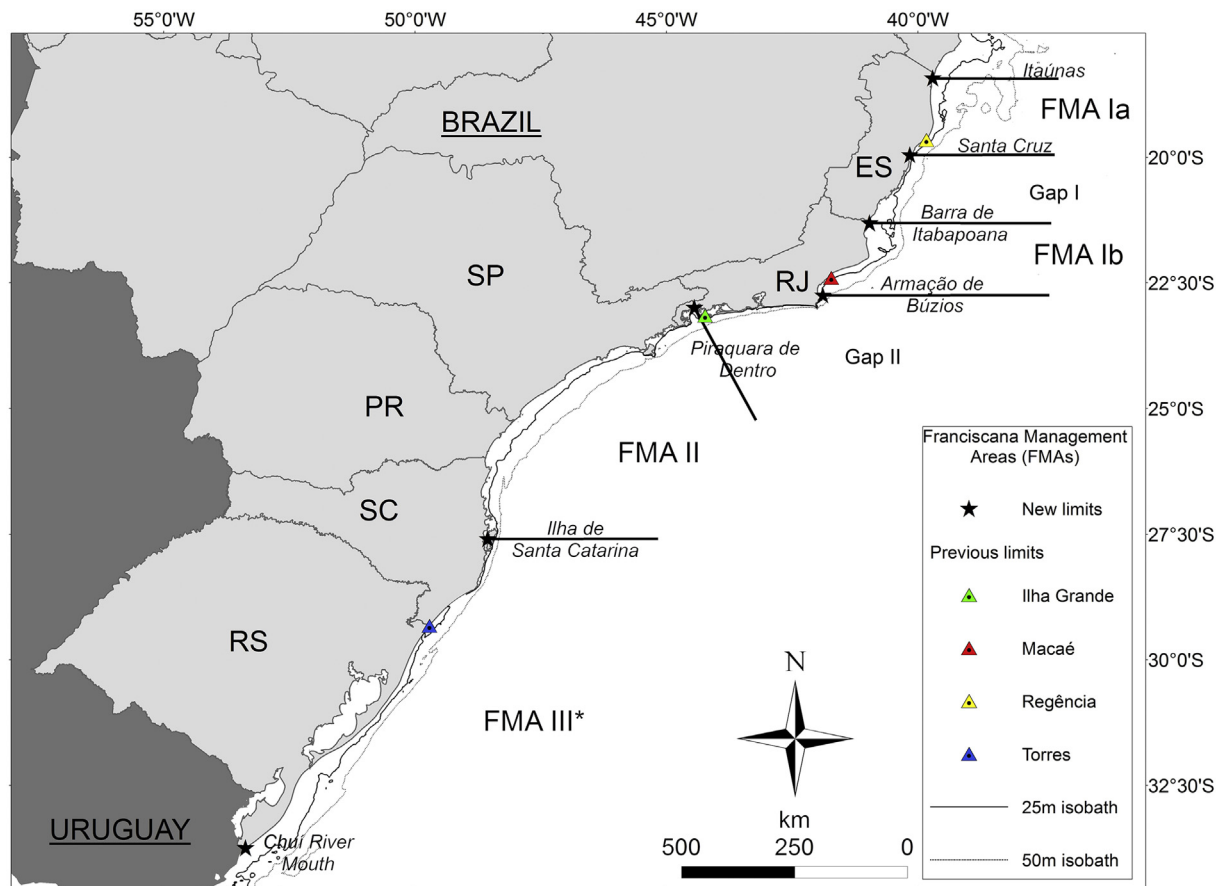
Fig. 3. New geographic ranges of the Franciscanas Management Areas (FMAs) and distributional gaps. Localities already considered limits are indicated in the map by triangles symbols. Abbreviations: ES, Espírito Santo; RJ, Rio de Janeiro; SP, São Paulo; PR, Paraná; SC, Santa Catarina; RS, Rio Grande do Sul. *FMAIII is partially represented because this management area extends further to the south to include the coast of Uruguay.

4. Discussion

The comprehensive review of the franciscana's occurrences along Brazilian coastal waters support the boundaries of FMAs as well as distributional gaps proposed by franciscanas' experts recently (see Anonymous, 2015; Ott et al., 2015). In relation to previous studies (for instance Secchi et al., 2003a; Siciliano et al., 2002), FMA Ia was extended further south from Regência (19°40′S) to Santa Cruz (19°57′S) in Espírito Santo; the southern limit of FMA Ib was relocated southward from Macaé (22°25′S) to Armação de Búzios (22°44′S) in Rio de Janeiro, due to the stranding of a live animal in the locality of Manguinhos, Armação de Búzios, reported by Siciliano et al. (2015). The northern limit of FMA II was established as Piraquara de Dentro (22°59′S) in Rio de Janeiro, while the southern limit was dislocated further northward from Torres (29°20′S) to the center of Ilha de Santa Catarina (27°35′S) in Santa Catarina, based on previous genetic studies (Cunha et al., 2014; Ott, 2002) (see Fig. 3). These changes on FMAs have impact direct on the extension of distributional gaps, which by its turn were reduced in relation to previous studies (Azevedo et al., 2002; Danilewicz et al., 2012; de Moura et al., 2009; Secchi et al., 2003a; Siciliano et al., 2002).

In general, the habitat suitability model presented here confirmed the well-known distribution of franciscanas (Danilewicz et al., 2009), indicating high environmental suitability for the species mainly up to the 25 m isobath (Fig. 7). However, this highly suitable environment could extend up to 50 m in the southernmost portion of franciscanas' distribution in Brazil as already indicated by Danilewicz et al. (2009). The ecological niche analyses also showed that both distributional gaps seem suitable for franciscanas at some level and they are inside of the

fundamental niche of species.

The resulting map of environmental suitability generated here is consistent with that proposed by Gomez and Cassini (2015), where habitat suitability map indicated high suitability for franciscanas in waters up to approximately 30 m depth from Brazil to Argentina (Gomez and Cassini, 2015). Even though Gomez and Cassini (2015) did not include bathymetry as a predictor, their resulting map agreed with the IUCN map. On the other hand, the franciscana's IUCN map was proposed by experts based on the 30 m isobath to establish the eastern border of franciscanas' distribution. In contrast to Gomez and Cassini (2015), the habitat suitability model proposed here indicated some level of suitability for franciscana in the gaps.

Bathymetry and distance to shore are considered important predictors of franciscanas' distribution (e.g. Danilewicz et al., 2009; Secchi and Ott, 1999), since individuals are rarely recorded beyond 50 m isobaths (Danilewicz et al., 2009). However, the present analysis did not indicate that bathymetry differs statistically among the area occupied by franciscanas and the gaps (Fig. 6D). On the other hand, the analysis of shelf habitat availability indeed revealed that the continental platform is extremely narrow in the gaps, reaching just 1 km of distance from shore at 23°S, for instance (Fig. 8). It was already suggested the narrowing of the continental shelf in the distributional gaps would limit habitat availability for franciscanas (Di Beneditto et al., 2001; Netto and Siciliano, 2007; Siciliano et al., 2002).

A similar example has been demonstrated in the western South Atlantic with the Atlantic spotted dolphin *Stenella frontalis* (G. Cuvier 1829). A gap in the distribution of this dolphin species exists where the Brazilian continental shelf narrows substantially between Abrolhos Bank (~18°S) and 6°S (Danilewicz et al., 2013; Moreno et al., 2005).

slope	**0.21**	**0.26**	**-0.15**	**-0.23**	**-0.23**	**-0.21**	**-0.21**	**-0.21**
	sss_mean	**0.85**	**-0.16**	**-0.36**	**-0.23**	**-0.22**	**-0.19**	**-0.13**
		sst_mean	**0.06**	**-0.55**	**-0.51**	**-0.44**	**-0.24**	**-0.17**
			bat	**-0.1**	**-0.11**	**-0.08**	**0.33**	**0.22**
				sss_range	**0.77**	**0.51**	**0.25**	**0.22**
					sst_range	**0.6**	**0.4**	**0.33**
						cl_range	**0.7**	**0.67**
							cl_mean	**0.94**
								da_mean

Fig. 4. Correlation matrix of nine environmental variables evaluated in the study. Abbreviations: bat, Bathymetry; slope, Bathymetric Slope; da_mean, Mean Annual Diffuse Attenuation; cl_mean, Mean Annual Concentration of Chlorophyll A; cl_rg, Annual Range in Concentration of Chlorophyll A; sss_mean, Mean Annual Sea Surface Salinity; sss_range, Annual Range in Sea Surface Salinity; sst_mean, Mean Annual Sea Surface Temperature; sst_range, Annual Range in Sea Surface Temperature.

Ecological niche modeling revealed lack of optimal environmental conditions for the species in the region of the coast where the continental shelf narrows (see do Amaral et al., 2015). In relation to franciscanas' distribution it is also interesting to note that a narrowing of continental shelf also exists around Ilha de Santa Catarina (27°35'S), where the limits between the FMA II and FMA III has been proposed (Ott, 2002; Cunha et al., 2014).

Analysis performed here showed that Gap II is located in a very restrict band of continental shelf, where the coastline orientation changes abruptly from NE-SW to E-W (Castro and Miranda, 1998). The continental shelf is almost nonexistent in the Gap II resulting in a drastic reduction of the shelf habitat even if other conditions such as temperature, salinity and productivity could support the existence of

Table 3
Factorial analysis of nine environmental variables used in this study.

	Factor1	Factor2	Factor3	Factor4	Factor5
bat	0.135			0.771	
slope	−0.143	−0.226	0.155		0.126
cl_mean	0.929	0.190		0.289	
cl_range	0.695	0.440	−0.169	−0.190	0.163
da_mean	0.959	0.122		0.128	−0.208
sss_mean		−0.121	0.976	−0.147	
sss_range	0.118	0.791	−0.271		
sst_mean	−0.123	−0.404	0.837	0.102	
sst_range	0.256	0.884	−0.129		
SS loadings	2.402	1.887	1.805	0.786	0.104
Proportion Var	0.267	0.210	0.201	0.087	0.012
Cumulative Var	0.267	0.477	0.677	0.765	0.776

Abbreviations: bat, Bathymetry; dist, Distance to Shore; slope, Bathymetric Slope; da_mean, Mean Annual Diffuse Attenuation; cl_mean, Mean Annual Concentration of Chlorophyll A; cl_range, Annual Mean in Concentration of Chlorophyll A; sss_mean, Mean Annual Sea Surface Salinity; sss_range, Annual Range in Sea Surface Salinity; sst_mean, Mean Annual Sea Surface Temperature; sst_range, Annual Range in Sea Surface Temperature.

species. In relation to Gap I, for example, the narrow shelf associated with higher levels of salinity could play a role to explain the absence of franciscanas in this area.

As suitability was projected in the distributional gaps, the absence of franciscana could be attributed to the reduction in the shelf habitat due to the narrowing of continental shelf. This environmental change could in turn intensify the biotic interactions such as competition by food and predation with other marine species. Guiana dolphin *Sotalia guianensis* (Van Bénedén, 1864) is a species with similar habitats (Da Silva et al., 2010) and could compete with franciscana by food and/or space. Furthermore, it was already observed a significant overlap in the diet of franciscana and largehead hairtail *Trichiurus lepturus* Linnaeus, 1758 (see Bittar and Di Benedetto, 2009; Di Benedetto et al., 2013). Also, the reduction of shelf habitat could enhance the vulnerability to predation by other cetaceans such as killer whale *Orcinus orca* (Linnaeus, 1758) (e.g. Ott and Danilewicz, 1998; Santos and Netto, 2005). Besides the likely strengthening of these biotic interactions, intrinsic factors, as minimal viable population size, also can play an important role in this very reduced range of habitat. In fact, a synergistic effect of biotic and abiotic factors can be determinant for the absence of franciscanas in the distributional gaps.

As top predators, cetacean distribution is limited by different factors (for example, productivity, temperature and salinity) that constraint both its prey and predator's distributions (e.g. Baumgartner et al., 2001; Palacios et al., 2013; Redfern et al., 2006). Temperature is a well-recognised factor delimiting species distribution (e.g. Jeffree and Jeffree, 1994) and salinity is also a well-known factor that have influence on cetacean distribution due to importance of physiological mechanisms to maintain the water and salt balance in cetaceans (e.g. Xu et al., 2013). Therefore, both salinity and temperature seem to impose physiological constraints for franciscanas, including stress triggered by high salt levels (e.g. São Pedro et al., 2015; Xu et al., 2013) and offspring resistance to cold environment (e.g. Danilewicz, 2003).

The presence of franciscana in FMA Ia appears to be associated with the plume of Doce River that probably maintains the levels of salinity more favorable to franciscanas or its prey species (Netto and Siciliano, 2007; Siciliano et al., 2002). Therefore, higher levels of salinity in Gap I could be a potential explanation for the absence of franciscanas in this area, once this gap has the highest median of Sea Surface Salinity. Since the franciscana seems to be associated with areas with great salinity ranges such as estuaries or river mouths (Cremer and Simões-Lopes, 2005; Santos et al., 2009; Siciliano et al., 2002), a higher constant salinity could impose some physiological constraint to the franciscana and/or to its prey species. Although franciscana has a fairly opportunist behavior in terms of prey abundance and occurrence (Bassoi, 2005) and shifts in prey composition overtime were already detected in southern Brazil (Secchi et al., 2003b), sciaenid fishes and long-finned squid *Doryteuthis sanpaulensis* (Brakoniecki, 1984) are very representative in the diet of the franciscana along its geographic range (Bassoi, 2005; Danilewicz et al., 2002). Sciaenid species are mainly present in tropical to warm and temperate environments over sandy and muddy bottoms in brackish, estuarine and low-salinity coastal regions (Martins and Haimovici, 2016).

Gomez and Cassini (2015) also suggested based on their ecological niche analysis that temperature and salinity could be considered the environmental predictors of franciscana along its entire range as well as the potential distribution of the stripped weakfish *Cynoscion guatucupa* (Cuvier, 1830). They also highlighted the physiological constrains imposed by Sea Surface Temperature and Salinity, and they did not find any support for previous statements that turbidity could be an important ocean determinant of franciscana distribution.

Diffuse attenuation is an indicator of the water turbidity and it is directly related to the presence of scattering particles in the water column. The analysis performed here indicated that the Gap II has the highest median of Diffuse Attenuation. This finding is a little bit controversial in relation to previous studies that suggested a preference for
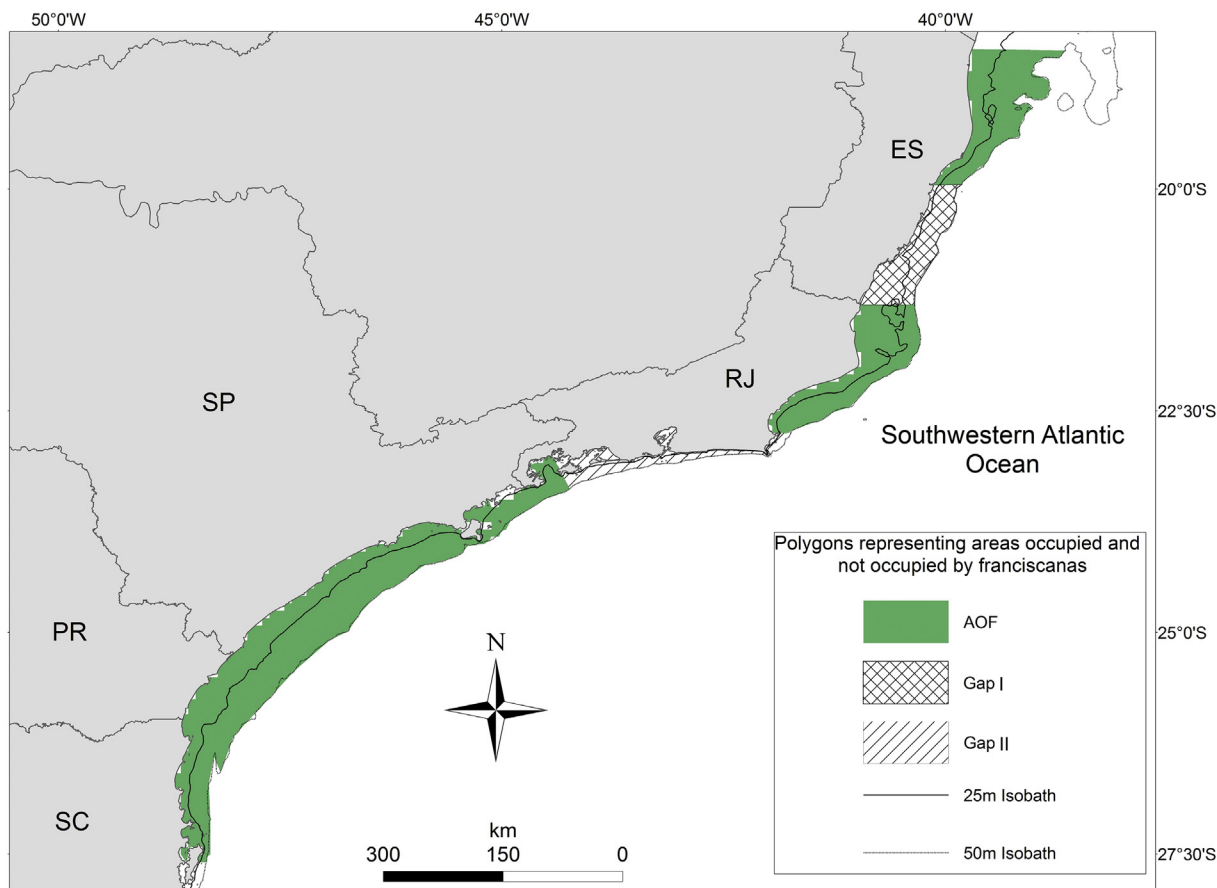
Fig. 5. Map of polygons used to create random points in order to represent the area occupied by franciscana and those not occupied. Abbreviation: AOF, Area occupied by Franciscana.

Table 4

Information used to investigate the environmental distinctiveness of areas occupied and not occupied by franciscanas. Abbreviation: AOF, Area Occupied by Franciscana.

Areas		Proportion in relation to the smallest polygon (i.e., Gap II)	Number of random points generated
AOF	Area Ia	2.44	244
	Area Ib	2.34	234
	Area II	7.72	772
Gap I		1.48	148
Gap II		1	100

Table 5

Medians comparisons through Kruskal-Wallis.

Environmental layer	Kruskal-Wallis Test	
Mean Annual Sea Surface Salinity	$\chi2 = 106.45$	***p*-value $< .05$**
Annual Range in Sea Surface Temperature	$\chi2 = 164.01$	***p*-value $< .05$**
Mean Annual Diffuse Attenuation	$\chi2 = 19.799$	***p*-value $< .05$**
Bathymetry	$\chi2 = 4.0115$	p-value $=.13$

Statistically significant values are in bold.

Table 6

Areas medians comparisons through Dunn tests for each environmental layer, which Kruskal -Wallis test was significant.

Dunn test			
	AOF	Gap I	
Mean Annual Sea Surface Salinity	**−10.229 (0.00001)**	–	Gap I
	0.433967 (0.9965)	7.218 (0.00001)	Gap II
Annual Range in Sea Surface Temperature	**12.13 (0.0001)**	–	Gap I
	5.165059 (0.00001)	**−3.999 (0.0001)**	Gap II
Mean Annual Diffuse Attenuation	−0.664 (0.7597)	–	Gap I
	−4.441 (0.00001)	**−3.119 (0.0027)**	Gap II

Abbreviation: AOF, Area Occupied by Franciscana. P-value is indicated among parenthesis and statistically significant values are in bold.

turbid waters by franciscanas (Siciliano et al., 2002). The reader should be aware that these results are influenced by the Guanabara Bay, Rio de Janeiro, and its discharge, which in turn could be increasing the values of Diffuse Attenuation, not reflecting a condition along the entire Gap II.

Finally, it is important to highlight that distributional gaps have an immediate impact on the population structure of franciscanas mainly in relation to franciscanas from FMA I. Genetic evidences based on mtDNA suggests that franciscanas are divided in two evolutionary lineages, franciscanas from FMA I (i.e. those northward Gap II) being a distinct lineage in comparison to all remaining franciscanas (Cunha et al., 2014). Furthermore, FMA I is further sub-structured into FMA Ia and FMA Ib (Cunha et al., 2014). From these findings it is possible to assume that both northern and southern distributional gaps have been acting as a barrier long enough to have an impact on the population structure in the areas adjacent to the gaps. Considering the importance of shelf habitat for franciscanas, it seems to be reasonable to suppose that historical factors such as sea level oscillations and, consequently, fragmentation of coastal platform during glacial and interglacial cycles,
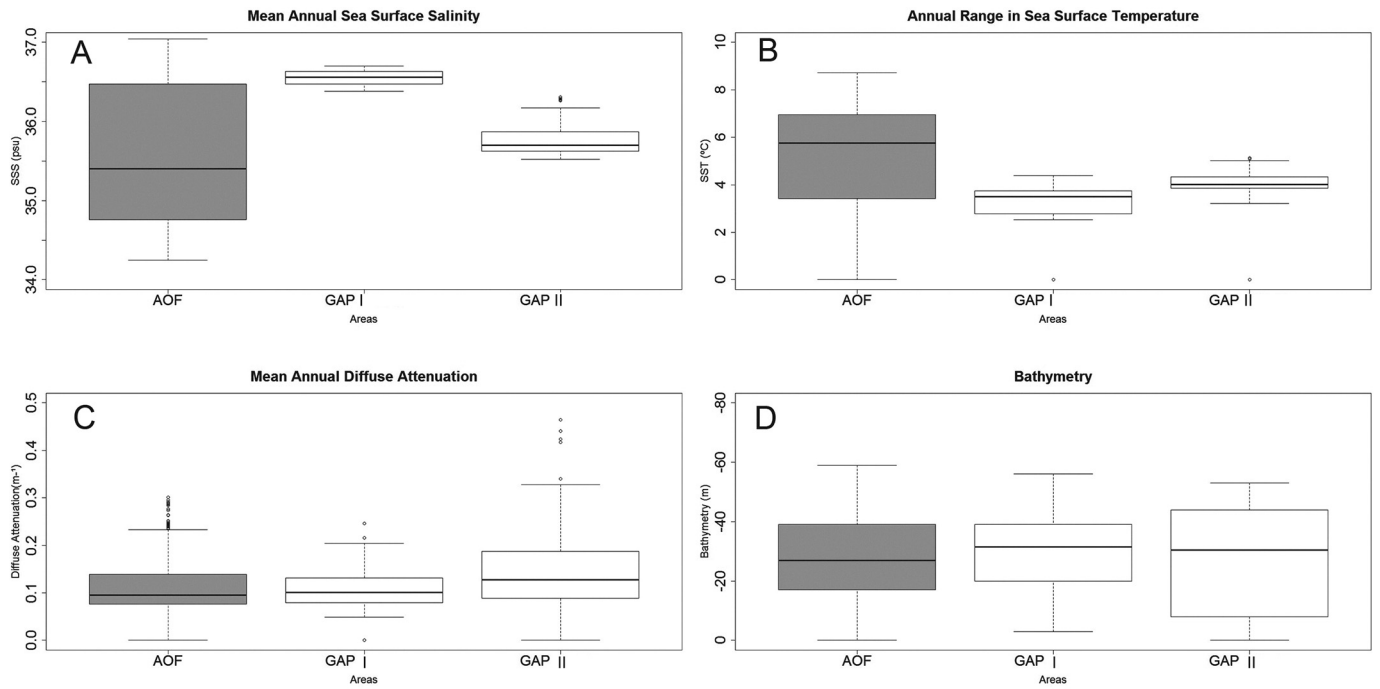
Fig. 6. Boxplot of environmental values extracted from random points grouped according area occupied by franciscana (AOF) and those not occupied. Boxplot represents median, 25th and 75th percentiles, and 5th and 95th are represented by the errors bars. In A) Mean Annual Sea Surface Salinity; B) Annual Range in Sea Surface Temperature; C) Mean Annual of Diffuse Attenuation; and D) Bathymetry.
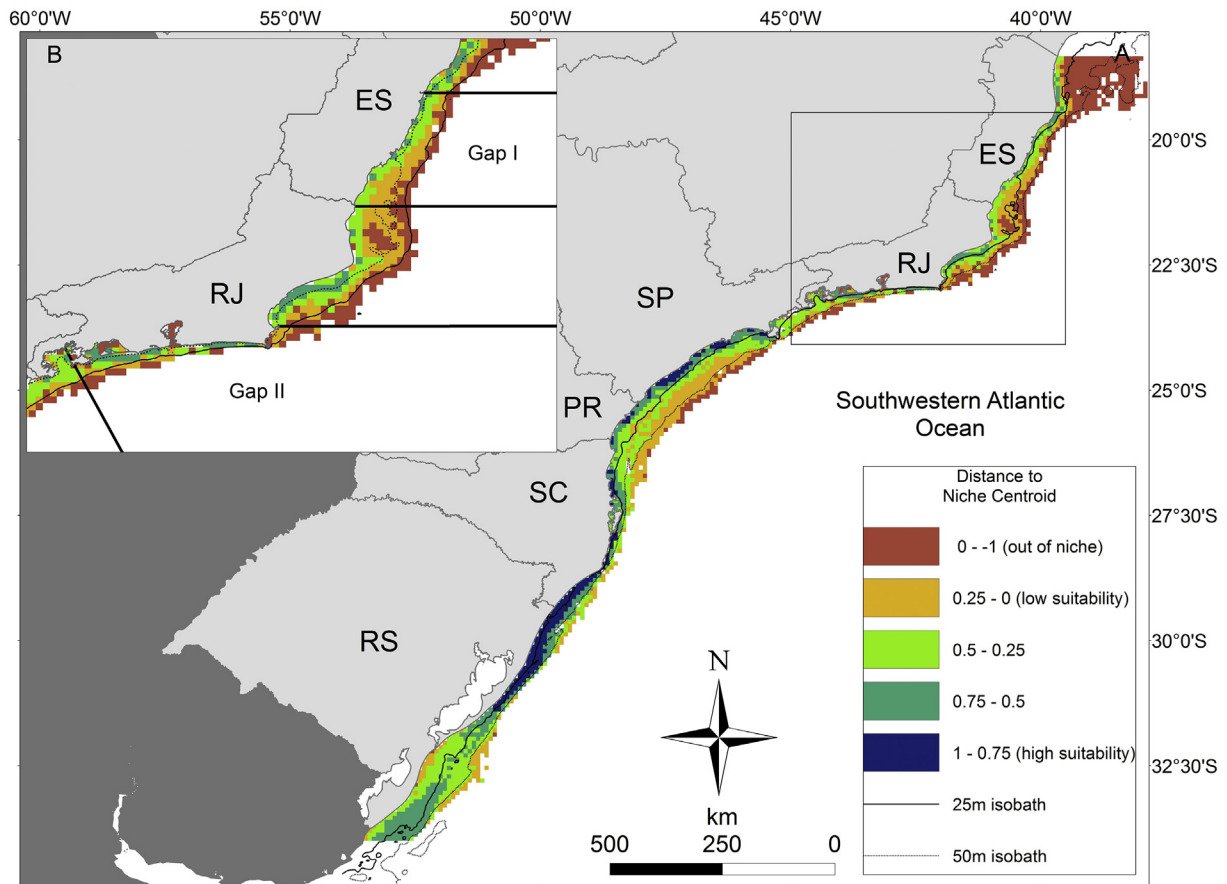

Fig. 7. A) Habitat suitability model with continuous values of the distance to the niche centroid representing the franciscana's fundamental niche. B) A map zoom is provided to visualize the environmental suitability of northern and southern distributional gaps. Values between 0 and 1 represent the relative distance between the points and the centre of the ellipsoid; −1 represent areas out of the ellipsoid or unsuitable; 0 means areas on the edge of the ellipsoid or with low suitability; and 1 means areas on the center of the ellipsoid or with high environmental suitability (H. Qiao 2017, p.c.).
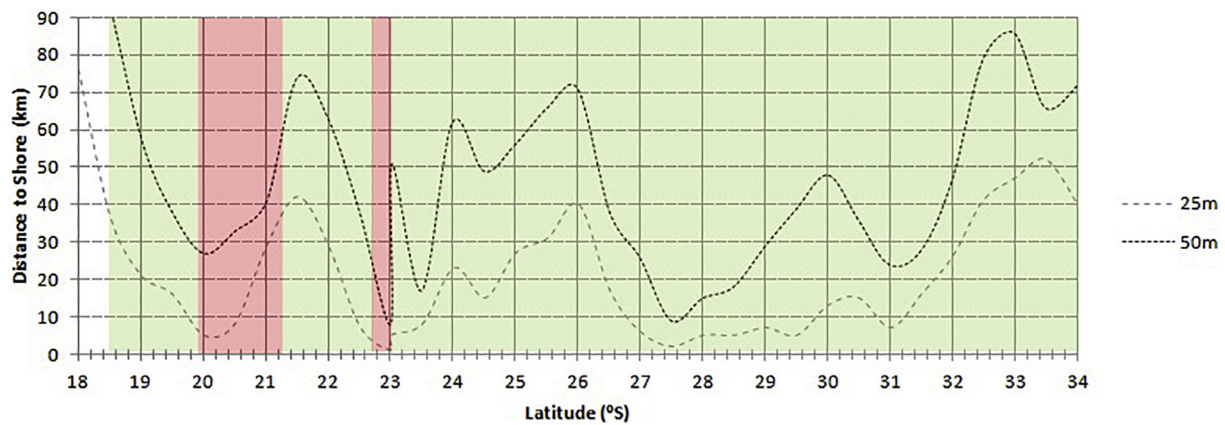
Fig. 8. Distance to shore of the 25 m and 50 m isobaths in relation to latitude. Area occupied by franciscana are represented in green; distributional gaps are represented in red (left, Gap I; right, Gap II). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

play a key role to explain for the absence of franciscana in these areas.

5. Conclusions

The shelf habitat is very important to franciscana. However, a wide shelf does not necessarily result in an increased presence of franciscana if other conditions such as salinity are not suitable. For example, the Brazilian continental shelf is very large at the north portion of the Espírito Santo, in the region of the Abrolhos Bank, however franciscana range seems to be limited longitudinally, possible due to higher and salt levels recorded there.

The new limits of FMAs and the habitat suitability model presented here could be used as a guide to planning studies and actions that aim the conservation of the franciscana in Brazil. Further studies should investigate franciscanas' prey availability in those areas considered distributional gaps as well the possible relevance of other biotic interactions. Finally, changes in the coastal environment and habitat loss caused by human activities, such as industrial port development, should also be considered in conservation plans for the species.

Acknowledgements

References

Alonso, M.B., Eljarrat, E., Gorga, M., Secchi, E.R., Bassoi, M., Barbosa, L., Bertozzi, C.P., Marigo, J., Cremer, M., Domit, C., 2012. Natural and anthropogenically-produced brominated compounds in endemic dolphins from western South Atlantic: another risk to a vulnerable species. Environ. Pollut. 170, 152–160.

Anonymous, 2015. Report of the VIII Workshop for the Research and Conservation of the Franciscana Dolphin (*Pontoporia blainvillei*) in the Soutwestern Atlantic. UNEP/CMS, São Francisco do Sul, Brazil. http://www.pontoporia.org

Azevedo, A.F., Fragoso, A.B.L., Lailson-Brito Jr., J., Cunha, H.A., 2002. Records of the franciscana (*Pontoporia blainvillei*) in the southwestern Rio de Janeiro and north-ernmost São Paulo state coasts - Brazil. LAJAM 1 (1), 191–192.

Barbato, B.H.A., Secchi, E.R., Di Beneditto, A.P.M., Ramos, R.M.A., Bertozzi, C., Marigo, J., Bordino, P., Kinas, P.G., 2012. Geographical variation in franciscana (*Pontoporia blainvillei*) external morphology. J. Mar. Biol. Assoc. UK 92 (8), 1645–1656.

Bassoi, M., 2005. Feeding Ecology of Franciscana Dolphin, *Pontoporia blainvillei* (Cetacea: Pontoporiidae), and Oceanographic Processes on the Southern Brazilian Coast. University of Southamptom, Southamptom, England (PhD Dissertation).

Baumgartner, M.F., Mullin, K.D., May, L.N., Leming, T.D., 2001. Cetacean habitats in the northern Gulf of Mexico. Fish. Bull. 99 (2), 219–239.

Bittar, V.T., Di Beneditto, A.P.M., 2009. Diet and potential feeding overlap between *Trichiurus lepturus* (Osteichthyes: Perciformes) and *Pontoporia blainvillei* (Mammalia: Cetacea) in northern Rio de Janeiro, Brazil. Zoologia 26 (2), 374–378.

Castro, B.M., Miranda, L.B., 1998. Physical oceanography of the western Atlantic continental shelf located between 4° N and 34° S - Coastal segment. In: Robinson, A., Brink, K.H. (Eds.), The Sea. The Global Coastal Ocean - Regional Studies and Synthesis. Wiley, New York, pp. 209–251.

Costa-Urrutia, P., Abud, C., Secchi, E.R., Lessa, E.P., 2012. Population genetic structure and social kin associations of franciscana dolphin, *Pontoporia blainvillei*. J. Hered. 103, 92–102.

Cremer, M.J., Simões-Lopes, P.C., 2005. The occurrence of *Pontoporia blainvillei* (Gervais & d'Orbigny) (Cetacea, Pontoporiidae) in an estuarine area in southern Brazil. Rev. Bras. Zool. 22 (3), 717–723.

Cremer, M.J., Simões-Lopes, P.C., 2008. Distribution, abundance and density estimates of franciscanas, *Pontoporia blainvillei* (Cetacea: Pontoporiidae), in Babitonga Bay, southern Brazil. Rev. Bras. Zool. 25 (3), 397–402.

Crespo, E.A., Harris, G., González, R., 1998. Group size and distributional range of the franciscana, *Pontoporia blainvillei*. Mar. Mamm. Sci. 14 (4), 845–849.

Cunha, H.A., Medeiros, B.V., Barbosa, L.A., Cremer, M.J., Marigo, J., Lailson-Brito Jr., J., Azevedo, A., Solé-Cava, A., 2014. Population structure of the endangered franciscana dolphin (*Pontoporia blainvillei*): reassessing management units. PLoS One 9 (1), e85633.

Da Silva, V.M.F., Fettuccia, D., Rodrigues, E.S., Edwards, H., Moreno, I.B., de Moura, J.F., Wedekin, L., Bazzalo, M., Emin-Lima, R., Carmo, N.A.S., Siciliano, S., Utreras, V., 2010. Report of the working group on distribution, habitat characteristics and preferences, and group size. LAJAM 8 (1–2), 31–38.

Danilewicz, D., 2003. Reproduction of female franciscana (*Pontoporia blainvillei*) in Rio Grande do Sul, southern Brazil. LAJAM 2 (2), 67–78.

Danilewicz, D., 2007. A toninha, *Pontoporia blainvillei* (Mammalia: Cetacea), no litoral norte do Rio Grande do Sul: mortalidade acidental em redes de pesca, abundância populacional e perspectivas para a conservação da espécie. Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil (PhD dissertation).

Danilewicz, D., Rosas, F., Bastida, R., Marigo, J., Muellbert, M., Rodriguez, D., Lailson-Brito Jr., J., Ruoppolo, V., Ramos, R., Bassoi, M., Ott, P.H., Caon, G., Rocha, A.M., Catão-Dias, J.L., Secchi, E., 2002. Report of the working group on biology and ecology. LAJAM 1, 25–42.

Danilewicz, D., Secchi, E.R., Ott, P.H., Moreno, I.B., Bassoi, M., Borges-Martins, M., 2009. Habitat use patterns of franciscana dolphins (*Pontoporia blainvillei*) off southern Brazil

in relation to water depth. J. Mar. Biol. Assoc. UK 89, 943–949.

Danilewicz, D., Moreno, I.B., Ott, P.H., Tavares, M., Azevedo, A.F., Secchi, E.R., Andriolo, A., 2010. Abundance estimate for a threatened population of franciscana dolphins in southern coastal Brazil: uncertainties and management implications. J. Mar. Biol. Assoc. UK 90, 1649–1657.

Danilewicz, D., Zerbini, A., Andriolo, A., Secchi, E.R., Sucunza, F., Ferreira, E., Denuncio, P., Flores, P.A.C., 2012. Abundance and distribution of an isolated population of franciscana dolphins (*Pontoporia blainvillei*) in southeastern Brazil: red alert for FMA I? In: Int. Whaling Comm. Working Paper SC/64/SM17.

Danilewicz, D., Ott, P.H., Secchi, E., Andriolo, A., Zerbini, A., 2013. Occurrence of the Atlantic spotted dolphin, *Stenella frontalis*, in southern Abrolhos Bank. Braz. Mar. Biodivers. Rec. 6, 1–3.

Di Beneditto, A.P.M., 2003. Interactions between gillnet fisheries and small cetaceans in northern Rio de Janeiro, Brazil: 2001-2002. LAJAM 2 (2), 79–86.

Di Beneditto, A.P.M., Ramos, R.M.A., Lima, N.R.W., 2001. Sightings of *Pontoporia blainvillei* (Gervais & D'Orbigny, 1844) and *Sotalia fluviatilis* (Gervais, 1853) (Cetacea) in South-Eastern Brazil. Braz. Arch. Biol. Techn. 44 (3), 291–296.

Di Beneditto, A.P.M., Rezende, C.E., Camargo, P.B., Kehrig, H.A., 2013. Trophic niche comparison between two predators in northern Rio de Janeiro state, Brazil: a stable isotopes approach. Biota Neotrop. 13 (3), 29–33.

Dinno, A., 2017. Dunn.test: Dunn's Test of Multiple Comparisons Using Rank Sums. R package version 1.3.5. https://CRAN.R-project.org/package=dunn.test.

do Amaral, K.B., Alvares, D.J., Heinzelmann, L., Borges-Martins, M., Siciliano, S., Moreno, I.B., 2015. Ecological niche modeling of *Stenella* dolphins (Cetartiodactyla: Delphinidae) in the southwestern Atlantic Ocean. J. Exp. Mar. Biol. Ecol. 472, 166–179.

Dormann, C.F., Elith, J., Bacher, S., Buchmann, C., Carl, G., Carré, G., Marquéz, J.R.G., Gruber, B., Lafourcade, B., Leitão, P.J., Münkemüller, T., McClean, C., Osborne, P.E., Reineking, B., Schröder, B., Skidmore, A.K., Zurell, D., Lautenbach, S., 2013. Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecography 36, 27–46.

ESRI, 2013. ArcGIS Desktop: Release 10.2.2. Environmental Systems Research Institute, Redlands, California, USA.

Flores, P.A.C., 2009. Occurrence of franciscana (*Pontoporia blainvillei*) in Baía Norte, southern Brazil. LAJAM 7 (1–2), 93–95.

Gariboldi, M.C., Túnez, J.I., Dejean, C.B., Failla, M., Vitullo, A.D., Negri, M.F., Cappozzo, H.L., 2015. Population genetics of Franciscana dolphins (*Pontoporia blainvillei*): introducing a new population from the southern edge of their distribution. PLoS One 10 (7), e0132854.

Gariboldi, M.C., Túnez, J.I., Failla, M., Hevia, M., Panebianco, M.V., Viola, M.N.P., Vitullo, A.D., Cappozzo, H.L., 2016. Patterns of population structure at microsatellite and mitochondrial DNA markers in the franciscana dolphin (*Pontoporia blainvillei*). Ecol. Evol. 1, 13.

Gomez, J.J., Cassini, M.H., 2015. Environmental predictors of habitat suitability and biogeographical range of Franciscana dolphins (*Pontoporia blainvillei*). Glob. Ecol. Conserv. 3, 90–99.

Gross, J., Ligges, U., 2015. Nortest: Tests for Normality. R package version 1.0-4. https://CRAN.R-project.org/package=nortest.

Higa, A., Hingst-Zaher, E., De Vivo, M., 2002. Size and shape variability in the skull of *Pontoporia blainvillei* (Cetacea: Pontoporiidae) from Brazillian coast. LAJAM 1 (1), 145–152.

Hoss, D., Valente, A.L., Ott, P.H., 2017. Gastrointestinal helminths of the franciscana *Pontoporia blainvillei* (Mammalia: Cetartiodactyla) in the northern coast of Rio Grande do Sul state, southern Brazil. Oecologia Aust. 21 (1), 72–82.

Jeffree, E.P., Jeffree, C.E., 1994. Temperature and the biogeographical distributions of species. Funct. Ecol. 8 (5), 640–650.

Kinas, P.G., 2002. The impact of incidental kills by gill nets on the franciscana dolphin (*Pontoporia blainvillei*) in southern Brazil. B. Mar. Sci. 70, 409–421.

de la Torre, A., Alonso, M.B., Martínez, M.A., Sanz, P., Shen, L., Reiner, E.J., Lailson-Brito Jr., J., Torres, J.P.M., Bertozzi, C., Marigo, J., Barbosa, L., Cremer, M., Secchi, E., Malm, O., Eljarrat, E., Barceló, D., 2012. Dechlorane-related compounds in franciscana dolphin (*Pontoporia blainvillei*) from southeastern and southern coast of Brazil. Environ. Sci. Technol. 46, 12364–12372.

Mahiques, M., Sousa, S., Furtado, V., 2010. The southern Brazilian shelf: general characteristics, quaternary evolution and sediment distribution. Braz. J. Oceanogr. 58, 25–34.

Martins, A.S., Haimovici, M., 2016. Seasonal mesoscale shifts of demersal nekton assemblages in the subtropical South-Western Atlantic. Mar. Biol. Res. 13 (1), 88–97.

Mendez, M., Rosenbaum, H.C., Subramaniam, A., Yackulic, C., Bordino, P., 2010. Isolation by environmental distance in mobile marine species: molecular ecology of franciscana dolphins at their southern range. Mol. Ecol. 19, 2212–2228.

MMA, 2014. Lista Nacional Oficial de Espécies da Fauna Ameaçadas de Extinção – Mamíferos, Aves, Répteis, Anfíbios e Invertebrados Terrestres. Portaria MMA nº. 444, de 17 de dezembro de 2014. Brazil. http://www.icmbio.gov.br/cepsul/images/stories/legislacao/Portaria/2014/p_mma_444_2014_lista_esp%C3%A9cies_ame%C3%A7adas_extin%C3%A7%C3%A3o.pdf, Accessed date: 15 May 2017.

Moreno, I.B., Martins, C.C.A., Andriolo, A., Engel, M.H., 2003. Sightings of franciscana dolphins (*Pontoporia blainvillei*) off Espírito Santo, Brazil. LAJAM 2 (2), 131–132.

Moreno, I.B., Zerbini, A.N., Danilewicz, D., Santos, M.C.O., Simões-Lopes, P.C., Laílson-Brito Jr., J., Azevedo, A.F., 2005. Distribution and habitat characteristics of dolphins of the genus *Stenella* (Cetacea: Delphinidae) in the Southwest Atlantic Ocean. Mar. Ecol. Prog. Ser. 300, 229–240.

de Moura, J.F., da Silva, É.R., Sholl, T.G.C., Siciliano, S., 2009. Franciscana dolphin (*Pontoporia blainvillei*) on the north-east coast of Rio de Janeiro state, Brazil, recorded during a long term monitoring programme. Mar. Biodivers. Rec. 2, 1–4.

Netto, R.F., Siciliano, S., 2007. Contribuição ao conhecimento da distribuição da toninha *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) no estado do Espírito Santo, sudeste do Brasil. Bol. Mus. Biol. Mello Leitão 21, 35–45.

Ott, P.H., 1998. Análise das capturas acidentais da toninha, *Pontoporia blainvillei*, no litoral norte do Rio Grande do Sul, sul do Brasil. Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil (MSc Thesis).

Ott, P.H., 2002. Diversidade genética e estrutura populacional de duas espécies de cetáceos do Atlântico Sul Ocidental: *Pontoporia blainvillei* e *Eubalaena australis*. Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil (PhD Dissertation).

Ott, P.H., Danilewicz, D., 1998. Presence of franciscana dolphins (*Pontoporia blainvillei*) in the stomach of a killer whale (*Orcinus orca*) stranded in southern Brazil. Mammalia 62 (4), 605–609.

Ott, P.H., Secchi, E.R., Moreno, I.B., Danilewicz, D., Crespo, E.A., Bordino, P., Ramos, R., Di Beneditto, A.P., Bertozzi, C., Bastida, R., Zanelato, R., Perez, J., Kinas, P.G., 2002. Report of the working group on fishery interactions. LAJAM 1, 55–64.

Ott, P.H., Domit, C., Siciliano, S., Flores, P.A.C. 2015. Memórias do VII Workshop para a coordenação de pesquisa e conservação de *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844). http:// www.pontoporia.org

Palacios, D., Baumgartner, M., Laidre, K., Gregr, E., 2013. Beyond correlation: integrating environmentally and behaviourally mediated processes in models of marine mammal distributions. Endanger. Species Res. 22, 191–203.

Phillips, S.J., Anderson, R.P., Schapire, R.E., 2006. Maximum entropy modeling of species geographic distributions. Ecol. Model. 190, 231–259.

Pinedo, M.C., 1995. Development and variation in external morphology of the franciscana, *Pontoporia blainvillei*. Rev. Bras. Biol. 55, 85–96.

Prado, J.H.F., Secchi, E.R., Kinas, P.G., 2013. Mark-recapture of the endangered franciscana dolphin (*Pontoporia blainvillei*) killed in gillnet fisheries to estimate past bycatch from time series of stranded carcasses in southern Brazil. Ecol. Indic. 32, 35–41.

Prado, J.H.F., Mattos, P.H., Silva, K.G., Secchi, E.R., 2016. Long-term seasonal and interannual patterns of marine mammal strandings in subtropical western South Atlantic. PLoS One 11 (1), e0146339.

Qiao, H., Peterson, A.T., Campbell, L., Soberón, J., Ji, L., Escobar, L.E., 2016. NicheA: creating virtual species and ecological niches in multivariate environmental scenarios. Ecography 39, 1–9.

R Development Core Team, 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna. www.r-project.org Retrieved 23 July 2016.

Ramos, R.M.A., Di Beneditto, A.P., Siciliano, S., Santos, M.C.O., Zerbini, A., Bertozzi, C., Vicente, A.F.C., Zampirolli, E., Alvarenga, F.S., Lima, N.R.W., 2002. Morphology of the franciscana (*Pontoporia blainvillei*) off southeastern Brazil: sexual dimorphism, growth and geographic variation. LAJAM 1 (1), 129–144.

Redfern, J.V., Ferguson, M.C., Becker, E.A., Hyrenbach, K.D., Good, C.P., Barlow, J., Kaschner, K., Baumgartner, M., Forney, K., Balance, L., Fauchland, P., Halpin, P., Hamazaki, T., Pershing, A.J., Qian, S.S., Read, A., Reilly, S.B., Torres, L., Werner, F., 2006. Techniques for cetacean–habitat modeling. Mar. Ecol. Prog. Ser. 310, 271–295.

Rocha-Campo, C.C., Danilewicz, D., Siciliano, S., 2010. Plano de ação nacional para a conservação do pequeno cetáceo toninha: *Pontoporia blainvillei*. In: Série Espécies Ameaçadas nº 10. Instituto Chico Mendes de Conservação da Biodiversidade, Brasília, Brazil.

Rossi-Santos, M.R., Oliveira, G., 2016. Combining cetacean soundscape ecology and niche modeling to contribute in the mapping of the Brazilian Continental Shelf. In: Finkl, C.W., Makowski, C. (Eds.), Seafloor Mapping along Continental Shelves. Research and techniques for visualizing benthic environments Springer International Publishing, Switzerland, pp. 146–165.

Santos, M.C.O., Netto, D., 2005. Killer whale (*Orcinus orca*) predation on a franciscana dolphin (*Pontoporia blainvillei*) in Brazilian waters. LAJAM 4, 62–72.

Santos, M.C.O., Vicente, A.F.C., Zampirolli, E., Alvarenga, F.S., Souza, S., 2002. Records of franciscana (*Pontoporia blainvillei*) from the coastal waters of São Paulo state, southeastern Brazil. LAJAM 1 (1), 169–174.

Santos, M.C.O., Oshima, J.E.F., da Silva, E., 2009. Sightings of franciscana dolphins (*Pontoporia blainvillei*): the discovery of a population in the Paranaguá Bay estuarine complex, southern Brazil. Braz. J. Oceanogr. 57 (1), 57–63.

São Pedro, S.L., Alves, J.M.P., Barreto, A.S., Lima, A.O.S., 2015. Evidence of positive selection of aquaporins genes from Pontoporia blainvillei during the evolutionary process of cetaceans. PLoS One 10 (7), e0134516.

Sbrocco, E.J., Barber, P.H., 2013. MARSPEC: ocean climate layers for marine spatial ecology. Ecology 94, 979.

Secchi, E.R., 2006. Modelling the Population Dynamics and Viability Analysis of Franciscana (*Pontoporia blainvillei*) and Hector's Dolphins (*Cephalorhynchus hectori*) under the Effects of Bycatch in Fisheries Parameter Uncertainty and Stochasticity. University of Otago, Dunedin, New Zealand (PhD Dissertation).

Secchi, E.R., Ott, P.H., 1999. A profundidade Como um fator determinante da distribuição de toninhas, *Pontoporia blainvillei*, conforme indicado pelos índices CPUE. In: Report of the Third Workshop for Coordinated Research and Conservation of the Franciscana Dolphin (*Pontoporia blainvillei*) in the Soutwestern Atlantic, . http://www.pontoporia.org/.

Secchi, E.R., Zerbini, A.N., Bassoi, M., Dalla-Rosa, L., Möller, L.M., Rocha-Campos, C.C., 1997. Mortality of franciscanas, *Pontoporia blainvillei*, in coastal gillnetting in southern Brazil: 1994-1995. Rep. Int. Whaling Comm. 47, 653–658.

Secchi, E.R., Wang, J.Y., Murray, B.W., Rocha-Campos, C.C., White, B.N., 1998. Population differentiation in the franciscana (*Pontoporia blainvillei*) from two geographic locations in Brazil as determined from mitochondrial DNA control region sequences. Can. J. Zool. 76, 1622–1627.

Secchi, E.R., Danilewicz, D., Ott, P.H., 2003a. Applying the phylogeographic concept to identify franciscana dolphin stocks: implications to meet management objectives. J. Cetacean Res. Manag. 5 (1), 61–68.

Secchi, E.R., Ott, P.H., Danilewicz, D., 2003b. Effects of fishing by-catch and conservation status of the franciscana dolphin, *Pontoporia blainvillei*. In: Gales, N., Hindell, M., Kirkwood, K. (Eds.), Marine Mammals: Fisheries. Tourism and Management Issues CSIRO Publishing, Collinwoop, Australia, pp. 174–191.

Secchi, E.R., Kinas, P.G., Muelbert, M., 2004. Incidental catches of franciscana in coastal gillnets fisheries in the Franciscana management area III: period 1999-2000. LAJAM 3 (1), 61–68.

Siciliano, S., Di Beneditto, A.P., Ramos, R., 2002. A toninha, *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) (Mammalia, Cetacea, Pontoporidae), nos estados do Rio de Janeiro e Espírito Santo, costa sudeste do Brasil: Caracterização dos hábitats e fatores de isolamento das populações. Boletim do Museu Nacional 476, 1–15.

Siciliano, S., de Moura, J.F., Secco, H.K.C., 2015. Considerações sobre a distribuição da toninha (*Pontoporia blainvillei*, Gervais & d'Orbigny, 1844) na costa centro-norte do estado do Rio de Janeiro, Brasil. In: Ott, P.H., Domit, C., Siciliano, S., Flores, P.A.C. (Eds.), Memórias do VII Workshop para a coordenação de pesquisa e conservação de *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844).

Tybergheim, L., Verbruggen, H., Pauly, K., Troupin, C., Mineur, F., De Clerck, O., 2012. Bio-ORACLE: a global environmental dataset for marine species distribution modelling. Glob. Ecol. Biogeogr. 21, 272–281.

Wei, T., Simko, V., 2016. R package version 0.77. In: Corrplot: Visualization of a Correlation Matrix, . http://CRAN.R-project.org/package=corrplot.

Xu, S., Yang, Y., Zhou, X., Xu, J., Zhou, K., Yang, G., 2013. Adaptive evolution of the osmoregulation-related genes in cetaceans during secondary aquatic adaptation. BMC Evol. Biol. 13, 189.

Zerbini, A., Secchi, E., Danilewicz, D., Andriolo, A., Laake, J., Azevedo, A., 2011. Abundance and distribution of the franciscana (*Pontoporia blainvillei*) in the Franciscana management area II (southeastern and southern Brazil). In: Int. Whaling Comm. Working Paper SC/62/SM7.

Zerbini, A.N., Secchi, E., Crespo, E., Danilewicz, D., Reeves, R., 2017. *Pontoporia blainvillei*. The IUCN Red List of Threatened Species 2017: e.T17978A123792204. http://www.iucnredlist.org/details/17978/0/, Accessed date: 15 May 2018.