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Variabilidade espaço-temporal dos impactos da pesca de arrasto sobre as assembleias do macrozoobentos e estrutura do substrato em áreas rasas do estuário da Lagoa dos Patos, RS, Brasil

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A amizade o tesouro mais valioso que
podemos ter

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RESUMO

A distribuição de sedimentos e macrofauna em ambientes estuarinos é modulada por variações nos parâmetros ambientais, como ventos e descargas fluviais, gerando mudanças nas condições hidrodinâmicas. A pesca de arrasto é uma das perturbações mais prejudiciais que afetam a fauna bentônica e seu habitat, sendo melhor entendido seu impacto quando avaliado junto com a variabilidade natural. Foram analisadas as variações espaço-temporais da granulometria (Capítulo 1) e da macrofauna (Capítulo 2) em duas áreas rasas com diferentes condições hidrodinâmicas do estuário da Lagoa dos Patos e a sua relação com parâmetros ambientais entre os anos 2015-2017. Analisou-se desde o enfoque ecossistêmico (EAF) o impacto da pesca de arrasto nas densidades, estrutura das assembleias e estratificação vertical do macrobentos (Capítulo 2), e na estrutura populacional do tanaidáceo *Monokalliapseudes schubarti* (Capítulo 4), por ser esta uma espécie dominante do estuário e um importante elo trófico desse ecossistema. Para isto, foram realizadas amostragens sazonais de sedimento e macrofauna nas duas áreas e aplicados arrastos experimentais de diferente intensidade em zonas marcadas dentro dessas áreas durante a época da safra do camarão nos três anos. As amostragens avaliaram o estrato superficial (0-5 cm) e um mais profundo (5-10 cm). Foi observada uma variação sazonal na composição granulométrica e no teor de matéria orgânica, com maiores percentuais de sedimentos finos (areia muito fina, silte e argila) e matéria orgânica nas zonas mais profundas, principalmente no inverno e primavera. Em zonas protegidas a direção e velocidade do vento foram os principais fatores geradores da variabilidade granulométrica, enquanto que nas zonas expostas as diferenças na batimetria tiveram um efeito mais significativo. A macrofauna respondeu espacialmente às variações na granulometria e temporalmente às variações na salinidade e temperatura. A composição das assembleias foi diferente entre as duas áreas amostradas, apresentando uma maior riqueza e densidade de espécies na área com maior hidrodinâmica por circulação local. O efeito do arrasto sobre o fundo foi maior nas áreas rasas com maiores porcentagens de sedimentos finos. Igualmente, nestas áreas foram detectados maiores sinais de impacto no macrobentos, principalmente observando-se diminuição de densidades totais no estrato mais superficial. Não houve perda da estratificação vertical das assembleias macrobentônicas após o arrasto, mantendo-se maiores densidades e riqueza de espécies no estrato superficial. Foram observadas alterações nas densidades em algumas espécies de importância para as teias tróficas estuarinas como *Erodona mactroides*, *Heleobia australis, H. charruana, Heteromastus similis, Laeonereis acuta* e *Monokalliapseudes schubarti.* As respostas destas espécies frente ao arrasto variaram temporalmente, e foram mais marcadas as variações de densidade após o arrasto nas zonas onde foi simulada uma alta intensidade de pesca de arrasto. Foram observados decréscimos nas densidades de machos e juvenis de *M. schubarti* no estrato superficial, e houve desaparecimento de fêmeas ovigeras em áreas com maiores percentuais de areias médias. Conclui-se, portanto, que o impacto do arrasto sobre espécies-chave estuarinas pode comprometer o funcionamento do ecossistema, pois a macrofauna bentônica serve como importante fonte de alimento para muitas espécies estuarinas, incluindo recursos pesqueiros.

Palavras chave: Lagoa dos Patos, pesca de arrasto, impacto, macrofauna, parâmetros ambientais

ABSTRACT

The distribution of sediments and macrofauna in estuarine environments is modulated by variations in environmental parameters, such as winds and river discharges, resulting in hydrodynamic shifts. Trawling fisheries are one of the most harmful anthropogenic impacts on benthic fauna and its habitat, and to properly understand this effect, natural variability of assemblages must be assessed. The spatio-temporal variations in granulometry and macrofauna of the Patos Lagoon Estuary were analyzed in two shallow areas; with different hydrodynamic conditions as well as their relationship with environmental parameters between the years 2015 and 2017. The trawling impact with an ecossistemic approach (EAF) on the densities, assemblages structure and vertical stratification of the macrofauna was analyzed, as well as on the population structure of the tanaid *Monokalliapseudes schubarti*, one of the dominant species of the estuary. Seasonal sampling of sediment and macrofauna were carried out in both areas and experimental trawls of different intensities were performed in zones within these areas during three shrimp fishing seasons. The sampling evaluated the superficial stratum (0-5 cm) and a deeper stratum (5-10 cm). A seasonal variation was observed in the grain size composition and the organic matter content, with higher percentages of fine sediments (very fine sand, silt and clay) and organic matter in deeper zones, mainly in winter and spring. In zones protected from the main estuarine channel currents, the direction and wind speed were the main factor causing granulometric variability, whereas in the exposed zones to those currents, the bathymetry played a greater role. The macrofauna spatially responded to variations in sediment texture and temporarily to variations in salinity and temperature. The composition of the assemblages was different between the two sampled areas, presenting a greater species richness and density in the area with higher hydrodynamics caused by currents. The effect of trawling over the bottom was higher in the area with higher percentages of fine sediments. Also, in these areas were detected greater signs of impact in the macrobentos, indicated by a decrease of total densities in most of the superficial stratum. However, there was no loss of vertical stratification of the fauna after trawling, remaining higher densities and especies richness in the superficial stratum. Changes in densities have been observed in some species of importance for the estuarine trophic webs such as *Erodona mactroides, Heleobia australis, H. charruana, Heteromastus similis, Laeonereis acuta* and *Monokalliapseudes schubarti*. The response of these species to trawling varied temporarily and with more marked variations in densities after trawl impact in areas where high trawling intensity was simulated. Decreases in the densities of males and juveniles of *M. schubarti* were observed in the superficial stratum, and there was disappearance of ovigerous females in areas with higher percentages of medium sands. We highlight that the trawling impact on estuarine key species may compromise ecosystem function, since benthic macrofauna provides food sources for many estuarine species, including some fishery resources.

Key words: Patos Lagoon, trawling, impact, macrofauna, environmental parameters

INTRODUÇÃO

Eventos discretos que removem organismos ou desestruturam a comunidade influenciando a disponibilidade de alimento, espaço ou modificando o ambiente físico são considerados como perturbações (Begon et al. 2005). Estas podem ocorrer de forma natural, por eventos climáticos, ou ser ocasionadas pelo homem. A pesca de arrasto é uma das perturbações antropogênicas mais prejudiciais sobre a fauna bentônica e o seu hábitat (Kaiser et al. 2002, Collie et al. 2005, 2016, Brown et al. 2005, Hiddink et al. 2017). Está destinada a capturar organismos na interface água-sedimento, causando perturbações crônicas no fundo (Jennings et al. 2002, Kaiser et al. 2002). As redes de arrasto deslizam na superfície do substrato com o uso de tralhas e/ou portas, que removem o sedimento e capturam os organismos bentônicos (Eleftheriou & McIntyre 2005). Estes petrechos causam a raspagem ou aragem do substrato, ressuspendem os sedimentos e destroem feições do fundo, ocasionando alteração da estrutura e complexidade do habitat (Kaiser et al. 2002, Brown et al. 2005, Foden et al. 2010, Sciberras et al. 2016), mortalidade incidental de espécies não alvo, alterações na biomassa, produtividade e estrutura de comunidades bentônicas, alterações na estrutura trófica e mudanças na função e estrutura de ecossistemas (Jennings et al. 2002, Hermsen et al. 2003, Mangano et al. 2013, 2014, 2015, Collie et al. 2016). Além disso, menos perceptíveis são as alterações nos fluxos de nutrientes e contaminantes, exposição das camadas anóxicas do sedimento e eventuais aumentos da demanda biológica de oxigênio devido à ressuspenção do sedimento (Olsgard et al. 2008, Collie et al. 2016). Também tem sido observado, que espécies infaunais de menor tamanho podem apresentar aumentos de abundância como resposta às perturbações por pesca, devido à redução da biomassa de grandes predadores bentônicos, assim como também outros organismos que podem ser beneficiados pela redução da competição (Jennings et al. 2002, Romano et al. 2016). Além dos efeitos antes expostos, redes de arrasto podem gerar marcas de 1,5 a 12 m de largura e 0,01-0,6 m de profundidade na superfície do substrato (Foden et al., 2010; Mangano et al., 2013, 2014). Estas diferenças na profundidade das marcas repercutem no grau de perturbação dentro do sedimento que pode ocasionar o arrasto. Scisberras et al. (2016) demostraram que o arrasto pode incrementar a quantidade de clorofila *a* e amônio superficial em camadas inferiores aos 5 cm de profundidade. Isto implica que o arrasto pode misturar a estratificação vertical dos sedimentos misturando camadas superficiais e profundas, isto com repercussão direta nos organismos infaunais os quais também poderiam perder sua estratificação vertical com soterramento de espécies epifaunais semelhanteas ao soterramento observado da chlorofila *a*. No entanto, o grau de perturbação causado pelo arrasto está associado ao peso da rede, à velocidade usada, à natureza do fundo e à frequência com a qual é usada, assim como aos níveis de perturbações naturais que ocorrem no local (Jennings et al. 2002, Costa & Netto 2014).

Apesar de serem reconhecidas as consequências negativas das redes de arrasto, esta arte de pesca é amplamente utilizada nas pescarias de camarão e sustentam uma porção substancial das aproximadamente 3,5 milhões de toneladas capturadas anualmente deste crustáceo (FAO 2016). A maioria dos estudos sobre o impacto da pesca nos fundos costeiros e oceânicos têm sido realizados sobre a plataforma continental em sedimentos arenosos, embora os fundos lamosos cubram mais de 50% da superfície da Terra. Estes são habitats típicos de muitas espécies de importância comercial e são frequentemente sujeitos à pesca de arrasto intensiva (Mangano et al. 2013). Nas plataformas continentais geralmente atua a pesca industrial, enquanto que em zonas marinhas mais perto à costa e estuarios a pesca é principalmente artesanal. Apesar de 50% da captura de peixes a nível mundial ter sua origem em pescarias artesanais (FAO 2017), seu impacto tem sido pouco investigado, especialmente aquelas relacionadas a ambientes estuarinos (Costa & Netto 2014). O impacto da pesca de arrasto geralmente é avaliado baseado na comparação entre áreas com pouca ou nenhuma atividade pesqueira. No entanto, é natural a sobreposição de resultados com os originados pela própria variabilidade natural dos parâmetros e processos que atuam no substrato, que por sua vez podem estar associados à atividade da pesca (Blyth et al. 2004). Assim, torna-se essencial o desenvolvimento de experimentos com replicação, em diferentes pontos da área de pesca a ser avaliada, e áreas controle sem a influência desta atividade.

Uma das estratégias atuais de avaliação e manejo de recursos pesqueiros é a implementação do enfoque ecossistêmico para as pescarias (EAF, Ecosystem Approach to Fisheries). Mais do que considerar a espécie alvo, essa abordagem propõe estratégias mais holísticas de análises ecossistêmicas, incluindo seu funcionamento e variabilidade natural (Garcia & Cochrane 2005, Bolam et al. 2014). A recente ênfase na gestão ambiental com base nesta abordagem aumentou a necessidade da compreensão dos efeitos das perturbações antrópicas sobre os ecossistemas que estão associados a atividades de pesca. Isto é fundamental para a utilização de medidas de gestão com o objetivo de manter a estrutura e função das comunidades (Mangano et al. 2014). Neste sentido resulta indispensável o conhecimento da variabilidade natural do ecossistema para um melhor entendimento da interferência antrópica.

Os invertebrados bentônicos desempenham importantes funções ecológicas como decomposição, ciclagem de nutrientes, e como elementos das teias tróficas atuando como consumidores e presas, ligando desse modo a produção primária aos níveis tróficos superiores (Ysebaert & Herman 2002, Tagliapietra & Sigovini 2010, Mola & Abdel Gawad 2014, Piló et al. 2015). Os ecossistemas bentônicos na plataforma continental provêm bens e serviços ecossistêmicos importantes, como a sustentação de importante produção pesqueira, assim como proporcionando alimento para espécies de peixes demersais (Rijnsdorp et al. 2016). O zoobentos também desempenha um papel importante no funcionamento dos ecossistemas marinhos, suportando uma ampla diversidade de espécies (Rijnsdorp et al. 2016). Nas regiões estuarinas o macrobentos está constituído principalmente por espécies suspensívoras, detritívoras e carnívoras, as quais compreendem um importante elo trófico entre a produtividade primária e detritos com os níveis superiores das tramas tróficas (Brown & McLachlan 2006). Adicionalmente, têm um papel importante mediando os processos físicos e químicos dos sedimentos, incluindo a bioturbação, degradação de matéria orgânica e metabolismo de contaminantes (Barros et al. 2008).

A distribuição espacial destes organismos não é homogênea, geralmente sendo encontrados em agrupamentos ou manchas (Dole-Olivier & Marmonier 1992, Beseres & Feller 2007, Barros et al. 2008). A relação entre os organismos e o substrato representa o principal fator para seu estabelecimento e sobrevivência (Espinoza & Morales 2008). Em ambientes costeiros, as flutuações de variáveis físicas como o grau de exposição às ondas, o tamanho de grão dos sedimentos e a turbidez possuem uma determinante influência sobre as espécies e populações, e podem alterar a composição e riqueza das assembleias e comunidades (Clark et al. 1996). Características do sedimento como tamanho de grão, proporções de lama, presença de cascalhos, além da disponibilidade de alimento, são determinantes importantes das comunidades bentônicas. Além disso, a topografia do fundo influencia o zoobentos em diferentes escalas espaciais (Rijnsdorp et al. 2016). Em fundos moles, a distribuição espacial do macrobentos posue uma variabilidade tridimensional, a qual resulta em variações nas interações entre espécies e na actividade dos organismos (Rosa & Bemvenutti 2006). A maioria dos organismos bentônicos nestes tipos de fundos ocupam os primeiros 5 cm de profundidade, respondendo a caracterisiticas fisicoquimicas do sedimento como distribuição de oxigênio e temperatura (Netto & Lana 1995). Estas variações na posição vertical dos organismos implicam também na disponibilidade principalmente da epifauna para predadores assim como a sua suceptibilidade as perturbações do sedimento (Posey et al. 1995, Rosa & Benvenuti 2006).

Desta forma, para entender a variabilidade da abundância e estrutura das assembleias do macrobentos estuarino, é necessária a avaliação das respostas aos diferentes parâmetros ambientais (Giménez et al. 2014).

Os processos de sedimentação são muito importantes na variabilidade da macrofauna estuarina. Estes estão controlados pela interação entre descargas fluviais (que controlam a salinidade, aporte de nutrientes e contaminantes), intrusão de água marinha e distribuição da biota (Pino et al. 1999). Além disso, a dinâmica dos sedimentos (erosão, ressuspensão, deposição) é determinada pela sinergia entre as condições hidrodinâmicas (por exemplo: velocidade das correntes) e propriedades do sedimento na interface coluna d'água/sedimento (por exemplo: tamanho do grão) que apresentam marcadas flutuações temporais (Ha & Park 2012). Adicionalmente, a dinâmica sedimentar é influenciada por variáveis biológicas, como a presença de organismos bioestabilizadores como o microfitobentos e os poliquetas, e dos bioturbadores como crustáceos e bivalves (Dyer et al. 2000, Widdows & Brinsley 2002). A profundidade também afeta a hidrodinâmica gerada por ondas induzidas pelo vento, a qual diminui conforme aumenta a profundidade e, portanto, menores tamanhos de grão tendem a ser encontrados (Sedano et al. 2014).

Estudos avaliando o efeito das pescarias de arrasto sobre o macrobentos são escassos no Brasil. Angonesi (2005) avaliou o efeito de dois tipos de redes de arrasto manual artesanal (berimbau e coca) sobre o macrobentos de enseadas rasas estuarinas, não identificando efeitos diretos destas redes sobre a fauna. Pelaes & Borzone (2007) analisaram a perturbação causada pelo arrasto de camarão na plataforma continental interna do Paraná, comparando áreas arrastadas com áreas onde houve o cessamento da atividade em anos anteriores. Nesta região de plataforma rasa (12-14 m de profundidade), não foram evidenciadas diferenças para nenhum descritor ecológico entre áreas com ou sem arrasto, apenas variações temporais de origem natural. Prantoni et al. (2013), avaliando uma área adjacente da plataforma continental interna do Paraná, não encontraram efeitos consistentes sobre as assembleias macrobentônicas antes e após a pesca de arrasto com redes de portas. A única espécie que mostrou alguma evidência de impacto foi o tanaidáceo *Monokalliapseudes schubarti*, que aumentou suas densidades após o arrasto. Costa & Netto (2014) analisaram o impacto de uma rede de arrasto manual tipo berimbau no estuário de Santo Antônio (Laguna, Santa Catarina). Eles analisaram o efeito do arrasto em áreas estuarinas com e sem vegetação submersa, concluindo que a pesca de arrasto não produz uma perda de diversidade bentônica e causa danos mínimos ao macrobentos estuarino. No entanto, evidenciaram impactos significativos sobre a meiofauna, com diminuições das densidades de nematodas e ostracodas.

No Estuário da Lagoa dos Patos (ELP), assim como ao longo do litoral e zonas estuarinas do Atlântico Sudoeste, a espécie de camarão *Penaeus paulensis* (camarão rosa) é um dos principais recursos pesqueiros (Costa & Netto 2014). A pescaria desta espécie se desenvolve no ELP sobre a população juvenil, a qual utiliza o estuário como uma zona berçário, e ocorre de maneira intensiva entre fevereiro e maio quando é liberada a pesca com redes passivas (D'Incao 1991, Benedet et al. 2010). No ELP a pesca de arrasto encontra-se proibida, mas os pescadores artesanais ainda utilizam esta arte de pesca para a captura do camarão rosa em profundidades que variam entre 1,5 a 14 m (Benedet et al. 2010). Entre as redes de arrasto utilizadas neste estuário encontram-se a rede de coca e berimbau (redes manuais) e a rede de arrasto de portas (com uma ampla variação de modelos), sendo as portas arrastadas por uma embarcação motorizada para garantir a abertura horizontal da rede. Segundo Benedet et al. (2010), os arrastos com as redes de porta são realizados tanto de dia quanto de noite com duração de 35 a 60 minutos cada, podendo ser feitos pequenos arrastos de 15 minutos para a avaliação da abundância do local. Estes autores também caracterizam a rede típica utilizada, a qual posue "tralhas de cabo tipo polipropileno (PP) 8 a 10 mm. Na tralha superior são colocados flutuadores de isopor para que a rede tenha abertura vertical e, na inferior, são usadas correntes (pesos) para que ela trabalhe removendo a parte superior do substrato". As portas utilizadas "são construídas de acordo com as características da embarcação e da rede. A parte central é de madeira, parafusada em uma armação de ferro (sapata) e medem de 60 x 35 a 130 x 55 cm pesando de 8 a 40 kg, sendo as de 12 a 18 kg as mais utilizadas pela frota artesanal".

No ELP a intensificação dos eventos reprodutivos e de recrutamento da maioria dos macroinvertebrados bentônicos ocorre do final da primavera ao verão (Bemvenuti & Netto 1998, Rosa & Bemvenuti 2006). Estes períodos podem se extender até o outono em épocas muito chuvosas (Colling et al. 2007), coincidindo então as épocas de maior recrutamento da macrofauna bentônica com a época de intensificação da pesca de arrasto. Desta forma, essa simultaneidade entre época da safra do camarão e a época de intensificação de recrutamento do macrobentos pode potencializar um eventual impacto da pesca de arrasto sobre estes organismos.

No ELP aproximadamente 40 taxa de macroinvertebrados bentônicos têm sido reportados, dos quais o gastrópode *Heleobia australis*, o bivalve *Erodona mactroides,* o tanaidáceo *Monokalliapseudes schubarti* e os poliquetas *Nephtys fluviatilis* e *Heteromastus similis* têm sido evidenciados como numericamente dominantes (Bemvenuti & Netto 1998, Colling et al. 2007, Colling 2011). *Monokalliapseudes schubarti* é uma espécie endêmica do Atlântico Sul, ocorrendo desde Cabo Frio (RJ) até a Península Valdés (Argentina) (Rosa-Filho & Bemvenuti 1998, D'Amico & Bala 2004, Elías et al. 2005). A espécie apresenta um papel ecológico importante na transferência de energia dos detritos para níveis tróficos superiores, uma vez que atua como comedor de depósito e suspensívoro (Fonseca & D'Incao 2003, Montagnolli et al. 2004, Freitas-Júnior et al. 2013). *Monokalliapseudes schubarti* é um dos principais itens alimentares de peixes (Bemvenuti 1990) e crustáceos, incluindo o siri azul *Callinectes sapidus* (Oliveira et al. 2006) e o camarão rosa *Penaeus paulensis* (Soares et al. 2004), sendo também reportada na dieta de aves (D'Amico & Bala 2004). A espécie apresenta uma distribuição estratificada da sua população, com juvenis ocupando profundidades menores em relação aos adultos (Rosa-Filho & Bemvenuti 1998). Este tanaidáceo constrói tubos em forma de *u* de até 15 cm de profundidade; devido à fertilização ocorrer dentro do tubo, os machos desta espécie fazem incursões dos seus tubos aos tubos das fêmeas (Freitas-Júnior et al. 2013), podendo assim ficar mais suscetíveis à predação, dispersão pelas correntes próximas à interface sedimento-coluna d'água e impactos do arrasto.

A macrofauna bentônica da Lagoa dos Patos apresenta marcadas flutuações sazonais e/ou interanuais na abundância dos organismos e das variáveis físico-químicas (Bemvenuti, 1987; Bemvenuti & Netto, 1998; Rosa, 2003; Colling et al., 2007; Colling, 2011). Existem duas situações ambientais e bióticas: a primeira no verão e outono com um aumento da salinidade as densidades do macrobentos aumentam, principalmente pelo recrutamento de espécies dominantes (Bemvenuti, 1987; Bemvenuti & Netto, 1998) durante anos típicos sem influencia de eventos atmosféricos extremos; e a segunda correspondente aos anos muito chuvos em que nos meses de primavera e verão ocorrem baixas densidades do macrozoobentos, baixos valores de salinidade e pouca variação no nível do substrato (Colling et al., 2007). Desta forma pode ser deduzido que flutuações na salinidade media do estuário afetam a abundancia das comunidades do macrobentos. Estabelecendo-se que moderada/elevada salinidade é propícia tanto à ocorrência de elevadas densidades da macrofauna, quanto à atividade pesqueira sobre a megafauna disponível; enquanto áreas de baixa salinidade ou condições límnicas, condicionam cenários com baixas densidades da macrofauna, e pouco produtivos quanto aos recursos pesqueiros da megafauna (Colling et al., 2007).

Na região de Rio Grande, o inverno e primavera, geralmente é uma época chuvosa, sendo que o verão e o outono são períodos mais secos. Contudo, variações interanuais observadas nos trabalhos de Bemvenuti (1987), Bemvenuti & Netto (1998) e de Colling et al. (2007), reforçam a necessidade de contemplar vários anos de amostragem nas interpretações de mudanças nas comunidades bentônicas e nas conclusões derivadas das investigações. Amostragens em apenas um ano podem levar a uma interpretação específica para as condições ambientais do período amostrado.

Desta forma, buscando seguir o enfoque ecossistêmico para a avaliação do impacto da pesca, surge a necessidade de avaliar a variabilidade natural do macrozoobentos e o substrato para o melhor entendimento dos impactos causados pelo arrasto. Assim, as perguntas que deram origem a esta tese foram: Qual a variação natural sazonal e interanual da macrofauna bentônica e do sedimento? Qual a resposta da macrofauna frente às perturbações da pesca de arrasto de portas? Esta resposta muda com o tempo ou é sempre igual? As diminuições na abundância, diversidade e riqueza de espécies reportadas para ecossistemas marinhos ocorrem em igual intensidade num estuário? Além de alterações nas assembleias, podem ocorrer mudanças a nível populacional?

Desta forma levantaram-se as seguintes hipóteses:

1. Espera-se que ocorra uma variabilidade espacial e temporal na composição granulométrica e percentuais de matéria orgânica relacionado com variações nos parâmetros ambientais.
2. Espera-se que a macrofauna bentônica responda à estrutura espacial e temporal da composição granulométrica do substrato, e à variabilidade temporal dos parâmetros da coluna d'água.
3. Espera-se maior abundância e riqueza de espécies do macrobentos em zonas sem influência da pesca de arrasto em relação às zonas impactadas.
4. A pesca de arrasto modifica a estratificação vertical da macrofauna nos primeiros 10 cm de profundidade do sedimento.
5. Maior frequência de perturbação de arrasto implicará em modificações mais acentuadas na densidade e riqueza do macrobentos, quando comparadas a perturbações de menor frequência.

6. A resposta da macrofauna frente à mesma frequência de perturbação da pesca será semelhante nas escalas temporais e espaciais avaliadas.

7. A pesca de arrasto altera a estrutura vertical do tanaidáceo *Monokalliapseudes schubarti*, afetando principalmente as densidades de juvenis e machos que habitam o estrato mais superficial do sedimento.

Assim o objetivo da presente tese foi: Avaliar o impacto da pesca de arrasto na estrutura das assembleias do macrobentos no estuário da Lagoa dos Patos, desde o enfoque ecossistémico.

Os objetivos específicos levantados foram:

- Determinar a variação espaço-temporal do sedimento em duas áreas do estuário da Lagoa dos Patos e a sua relação com parâmetros ambientais.
- Determinar a variação espaço-temporal do macrobentos em diferentes estratos do substrato, nas mesmas duas áreas e a sua relação com parâmetros ambientais.
- Avaliar a resposta do macrobentos quanto à variação na estrutura das assembleias macrobentônicas frente a altas e baixas intensidades de pesca.
- Avaliar a resposta na estrutura da população de *Monokalliapseudes schubarti,* por ser uma espécie dominante, frente à perturbação da pesca.

METODOLOGIA

Área de Estudo

A Lagoa dos Patos está localizada no segmento mais meridional do litoral brasileiro, e compreende a maior laguna do tipo estrangulada do planeta (Kjerfve 1986). Recebe aportes de água doce principalmente dos rios Jacuí, Taquari, Sinos, Caí, Gravataí e Camaquã, além do Canal São Gonçalo (Fernandes et al. 2005), os quais representam o principal aporte de nutrientes e material em suspensão para o estuário (Wallner-Kersanach et al. 2016). Apesar de apresentar uma elevada hidrodinâmica, principalmente regida pela velocidade e direção do vento, o efeito da maré é baixo (0,4 m) (Möller et al. 2001, Fernandes et al. 2005, Wallner-Kersanach et al. 2016). Na margem oeste encontra-se o saco do Arraial, que é uma zona protegida das correntes principais do estuário, mas por ser bastante raso, resulta sujeita a ação de ondas provocadas por vento e que promovem ação de erosão, deposição e transporte, resultando assim em um ambiente muito dinâmico (Colling et al. 2007, Souza & Hartmann 2008). Neste saco, comumente ocorre a pesca artesanal de arrasto na época de safra do camarão (fevereiro a maio). Por ser uma enseada rasa protegida da circulação do canal principal do estuário, o sedimento está composto principalmente por areia fina. Estas zonas são consideradas áreas de criação, alimentação e proteção para variadas espécies de peixes e invertebrados de importância ecológica e econômica (Angonesi 2005).

Foram selecionadas duas áreas dentro do saco do Arraial: uma na margem oeste da ilha da Pólvora (Área 1) e outra na margem leste da ilha dos Cavalos (Área 2) (Fig. 1). Em cada área foram marcadas três zonas de 20 x 20 m com a utilização de calões de madeira e adicionalmente foram monitoradas 3 zonas, sem marcação visível entre as duas áreas.

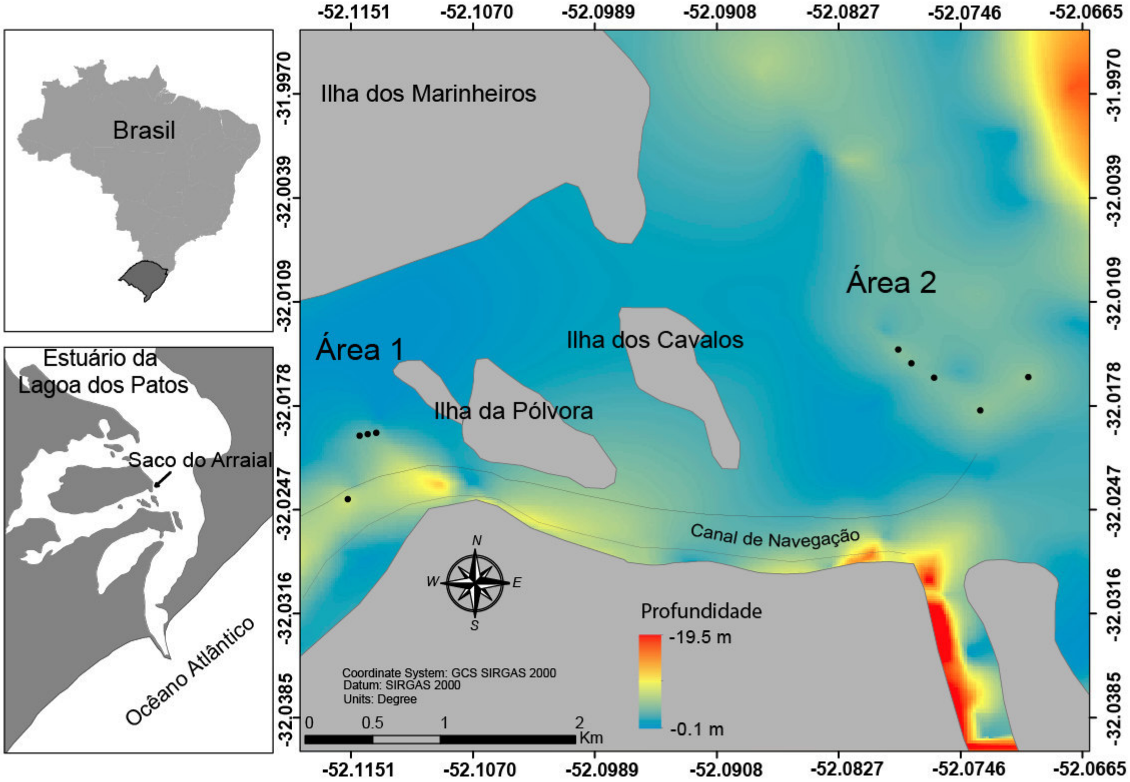

Figura 1. Mapa batimétrico da região de estudo mostrando as duas áreas amostradas dentro do Saco do Arraial.

Coleção de Amostras

Amostras de macrozoobentos e de sedimentos foram coletadas entre fevereiro de 2015 e maio de 2017, cobrindo três safras de pesca e a variabilidade sazonal ao longo de dois anos e meio. Resultando em 9 coletas em 2015 (fevereiro, março, abril, maio, duas coletas em junho, duas em julho e uma em novembro), 10 coletas em 2016 (janeiro, fevereiro, março, abril, maio, duas em junho, julho, agosto e novembro) e 3 coletas em 2017 (janeiro, março, maio). As coletas foram realizadas a bordo da lancha *Larus*-FURG e de um barco de pesca artesanal. As amostras foram coletadas com uma draga *van Veen* e subamostradas com um tubo de PVC de 10 cm de diâmetro para a separação do sedimento em dois estratos, o mais superficial constituindo os primeiros 5 cm (Estrato 0-5) e o mais profundo constituído pelos 5 cm seguintes (Estrato 5-10). Em cada zona amostral foram coletadas 3 réplicas para análise de macrofauna e duas para análise de sedimentos. As amostras para análise do macrobentos foram peneiradas a bordo com uma malha de 300 µm e preservadas em formol a 10%. Aquelas amostras para análise de sedimento foram congeladas para o seu posterior processamento no laboratório.

Foram gravadas imagens do fundo para a observação de possíveis marcas deixadas por redes de arrasto, através de um sonar de varredura lateral ou *side scan sonar* Lowrance HDS 9-Gen 2 com um tradutor programado para tomar imagens entre 450-800 kHz. Com este mesmo equipamento e oo ecobatimetro da lancha *Larus* foram feitas medidas da profundidade local. Temperatura da àgua e salinidade foram medidas *in situ* com um termômetro de mercúrio e um refratômetro manual, respectivamente.

Metodologia específica Capítulo 1: Variações espaço-temporais de sedimentos em um estuário subtropical de micro maré (*Spatial and temporal variations of sediments in a subtropical micro tidal estuary*)

Neste capítulo foram avaliadas as nove zonas amostradas, nomeadas da seguinte forma: pontos 1P, 2P, 3P e 4P aqueles próximos à ilha da Pólvora, e pontos 1C, 2C, 3C, 4C e 5C aqueles próximos a ilha dos Cavalos (Apêndice 1, Fig. 1). Utilizaram-se as amostras de sedimento coletadas antes do arrasto nas zonas onde este foi simulado, para evitar possíveis alterações ocasionadas pelo petrecho. Foi analisado somente o estrato superficial (0-5 cm). Durante o período estudado, as amostragens foram compostas da seguinte forma: duas amostragens no verão, três no outono, três no inverno e uma na primavera de 2015; três no verão, três no outono, três no inverno e uma na primavera de 2016; duas no verão e uma no outono de 2017. Foram classificadas como zonas rasas aquelas com profundidade média menor a 2,0m (1P, 2P, 3P; e 1C, 2C, 3C) e como profundas aquelas com profundidade média igual ou superior aos 2,0m, encontrando-se estas mais perto ao canal de navegação (4P, 4C, 5C) (Tabela I).

Tabela I. Profundidade (m) de cada ponto de coleta (mínima, média e máxima)

Área	Zona	Ponto	Profundidade		
			Mín	Média	Máx
Área 1: Pólvora	Rasa	1P	0,9	1,3	2,0
		2P	0,9	1,3	2,0
		3P	0,9	1,3	2,0
	Profunda	4P	1,5	2,7	4,0
Área 2: Cavalos	Rasa	1C	1,5	1,7	2,5
		2C	1,5	1,7	2,1
		3C	1,5	1,9	2,1
	Profunda	4C	1,9	2,1	2,6
		5C	1,9	2,0	2,6

As análises de granulometria foram realizadas pelo método de peneiramento sequencial através de peneiras com intervalo entre 4 mm e 0,063 mm a intervalo de um phi, e por pipetagem para os tamanhos de grão menores que 0,063 mm (Suguio 1973). Como as frações maiores a 1 mm estiveram compostas por fragmentos de conchas de bivalves e gastrópodes mortos, estas foram desconsideradas da análise. Foram utilizados aproximadamente 30 g de sedimento para as análises de granulometria. O teor de matéria orgânica foi calculado pela diferença de peso depois de calcinação por 2h a 375°C, utilizando aproximadamente 5 g de sedimento.

Foram obtidos dados da precipitação acumulada mensal estimada por imagens satelitais para a bacia de drenagem da Lagoa dos Patos utilizando um quadrado de coordenadas -54.8437,-32.2998,-50.0537,-28.96 no período entre 1997 e 2017. Estas estimações proveem do satélite MERRA-2 Model M2TMNXFLX v5.12.4 do sistema de dados Giovanni desenvolvido e mantido pela NASA GES DISC (*National Aeronautics and Space Administration - Goddard Earth Sciences Data and Information Services Center*) e seus resultados podem ser reproduzidos no link: https://giovanni.gsfc.nasa.gov/giovanni/#service=ArAvTs&starttime=1997-01-01T00:00:00Z&endtime=2017-09-30T23:59:59Z&shape=gpmLandMask/shp_0&bbox=-54.8437,-32.2998,-50.0537,-28.96&data=M2TMNXFLX_5_12_4_PRECTOT(units%3Dmm%2Fmonth)&variableFacets=dataFieldMeasurement%3APrecipitation%3B&portal=GIOVANNI&format=json. Foram calculadas a média da velocidade e a moda da direção do vento para as 24 horas antes das 9 am do dia da coleta, e para as 72 horas antes (3 dias) a partir dos dados horários fornecidos pelo instituto nacional de metereologia (IMET- 8º Distrito de Meteorologia - Porto Alegre – Brasil, Estação Rio Grande). Foi utilizado este critério uma vez que Möller et al. (2001) estimaram que frentes frias que condicionam a velocidade e direção dos ventos podem durar entre 3 e 16 dias. Os dados diários do nível de água da laguna foram obtidos junto à Agencia Nacional das Águas (ANA), estação Rio Grande – Regatas, com os quais foram calculadas médias mensais para o período entre 1997 e 2017. A profundidade de fechamento das ondas foi calculada segundo a metodologia proposta por Hallermeier (1981), utilizando os dados para a Lagoa dos Patos desde janeiro até julho de 2015, fornecidos pelo programa Rede Ondas, da Universidade Federal de Rio Grande (http://www.redeondas.furg.br/index.php/pt/).

A porcentagem média sazonal de areia, silte e argila por ponto foram plotadas e classificadas através de diagramas de Shepard (1954). Os parâmetros estatísticos foram calculados segundo as equações de Folk & Ward (1957), considerando na análise estatística a média, mediana, seleção (desvio padrão), assimetria e curtose, parâmetros calculados a partir da utilização do programa SYSGRAN (Camargo 1999).

Para caracterizar a variação espacial e temporal das zonas de estudo baseado na distribuição dos tamanhos de grão e nos parâmetros estatísticos, foram realizadas Análises de Componentes Principais (ACP). Para avaliar a significância das variações espaço-temporais se realizou uma análise de variância multivariada (Permanova, Anderson et al. 2008) com permutações dos resíduos sob um modelo reduzido, a partir de uma matriz de distância Euclidiana. O desenho incluiu os fatores Área (2 níveis: Área1 e Área 2 - fixo), Ponto (9 níveis: 1P a 4P, 1C a 5C - aleatórios, aninhados à Área), Ano (3 níveis: 2015, 2016 e 2017 - Aleatório) e Estação (4 níveis: verão, outono, inverno e primavera - Aleatório, aninhado ao Ano). A análise foi aplicada para a distribuição dos tamanhos de grão, e para os parâmetros estatísticos. Estas análises foram realizadas com os programas PRIMER V6 + PERMANOVA *add-on* (Anderson et al. 2008) e R (R core team 2017).

Metodologia específica Capítulo 2: Variações espaço-temporais de assembleias macrobentônicas em áreas estuarinas rasas com diferentes condições hidrodinâmicas (***Temporal and spatial distribution of macrobenthic assemblages in areas of an estuarine shallow mudflat with different hydrodynamic conditions***)

Foram utilizadas as zonas controle e sem marcas visíveis ao longo das 4 estações do ano. Estas zonas foram nomeadas consecutivamente como os pontos 1, 2, 3, 4 e 5 (Apêndice 2, Fig.

1). Como parâmetros ambientais foram utilizadas a temperatura, salinidade, profundidade, teor de matéria orgânica e granulometria. A metodologia das duas últimas foi a mesma utilizada no Capítulo 1. Medidas diárias de salinidade foram obtidas através do projeto PELD-FURG (Programa de Estudos de Longa Duração), provenientes da Ilha dos Marinheiros.

Para caracterizar os pontos e avaliar as diferenças sazonais considerando os parâmetros ambientais, foi realizada uma Análise de Componentes Principais (ACP). Para avaliar possíveis relações entre as espécies e as variáveis ambientais foram realizadas correlações de Spearman.

As seguintes medidas univariadas foram calculadas: densidade total (N), número de espécies (S), índice de diversidade de Shannon-Wiener (H') e índice de equitatividade de Pielou (J'). A frequência de ocorrência foi calculada pela presença de indivíduos no número total de amostras coletadas em cada local.

Variações espaço-temporais da fauna foram analisadas através da Permanova com seguinte modelo: Área (2 níveis: Área 1 e Área 2 - fixo), Ponto (5 níveis: Ponto 1 ao Ponto 5 - aleatório, aninhado em Área), Ano (3 níveis: 2015 a 2017 – aleatório), Estação (4 níveis: verão, outono, inverno, primavera - aleatório, aninhado no Ano), Estrato (2 níveis: 0-5 cm e 5-10 cm - fixo). Para os dados univariados da macrofauna (densidade total, riqueza de espécies, diversidade e equitatividade), foi utilizada uma Permanova com 4999 permutações de unidades de dados brutos para analisar a possível variabilidade entre fatores (Anderson 2005). A Permanova foi realizada usando uma matriz de distância euclidiana. Testes par a par também usaram 4999 permutações aleatórias para obter p-valores através da correção de Monte Carlo. Para os dados multivariados (estrutura da assembleia), a Permanova (com 999 permutações de resíduos sob um modelo reduzido) foi baseada numa matriz de similaridade de Bray-Curtis construída a partir dos dados de densidade transformados pela raiz quadrada (ind.m^{-2}). A transformação foi usada para diminuir a importância das espécies dominantes. Foram somados valores de um (+1) para reduzir o efeito da ausência de indivíduos em algumas amostras (Clarke et al. 2006). Testes para a par foram realizados para os fatores que resultaram significativos (p-valor <0,05), usando a correção de Monte Carlo.

Metodologia específica Capítulo 3: Resposta de assembleias macrobentônicas de fundos inconsolidados à pesca de arrasto artesanal em um estuário subtropical (*Response of soft-bottom macrobenthic assemblages to artisanal trawling fisheries in a subtropical estuary*)

Foram utilizadas as amostras coletadas nas zonas controle (C1, C2) e aquelas onde foi simulado o impacto da pesca de arrasto (HI1, HI2, LI1, LI2) (Apêndice 3, Fig. 1) entre os meses de verão e outono de 2015 a 2017, resultando em 5 avaliações em 2015 (fevereiro, março, abril, maio e junho), 6 em 2016 (janeiro a junho) e 3 em 2017 (janeiro, março e maio). Nas zonas onde foram realizadas simulações de pesca, foram coletadas 3 réplicas para macrofauna e duas para sedimento antes e após o arrasto. Para as zonas controle, foram coletadas 3 réplicas para macrobentos e duas para sedimento. Utilizaram-se as amostras de sedimento e macrofauna dos dois estratos (0-5 cm e 5-10 cm) antes e após a simulação da pesca. O processamento das amostras de sedimento foi o mesmo utilizado no Capítulo 1, e o das amostras de macrofauna o mesmo utilizado na Metodologia Geral.

Nestes experimentos, a pesca de arrasto intensiva foi simulada com 5 arrastos consecutivos nas zonas HI1 (Simulado 1 Alto) e HI2 (Simulado 2 Alto), enquanto que a baixa intensidade de pesca foi simulada com 2 arrastos consecutivos nas zonas LI1 (Simulado 1 Baixo) e LI2 (Simulado 2 Baixo) (Apêndice 3; Fig. 1). Os arrastos foram realizados a bordo de um barco de pesca artesanal de 8 m a uma velocidade aproximada de 2 nós. A rede utilizada era uma rede de portas típica de pesca no local (Benedet et al. 2010), a qual posuia uma malha de 12 mm de abertura entre nós opostos e 5 m de largura, com portas de madera parafusada em uma armação de ferro de 80 x 50 cm, pesando ao todo15 kg cada porta.

Foram gravadas imagens do fundo com o sonar de varredura lateral tanto nas zonas controle como nas impactadas antes do arrasto para verificar a ausência de marcas. O sonar seguiu gravando durante os arrastos para observar a perturbação causada pela rede de pesca no fundo e foi escaneada mais uma vez todas as zonas após o arrasto para a observação da possível permanência das marcas deixadas pelas redes. As informações deste equipamento estão detalhadas na Metodologia Geral.

Para caracterizar as duas áreas e avaliar as diferenças sazonais baseando-se nas variáveis ambientais, foi realizada uma Análise de Componentes Principais (ACP) utilizando como variáveis a salinidade, temperatura, profundidade, teor de matéria orgânica e a granulometria. A contribuição de cada espécie nas possíveis diferenças entre as duas áreas foi avaliada com uma análise de contribuição de espécies SIMPER (*Similarity Percentages*). Diferenças entre as espécies do macrobentos e as variáveis ambientais foram avaliadas através de uma Análise de Correspondência Canônica (CCA).

O impacto da pesca foi analisado usando-se um delineamento aninhado para avaliar diferenças nas densidades totais, riqueza, diversidade, equitatividade e estrutura da assembleia entre os períodos de arrasto (antes e depois do arrasto) e a estratificação vertical da macrofauna para cada área, ao longo dos três anos amostrados. Os fatores testados foram: Tratamento (5 níveis: Controle, Antes do alto impacto, Após o alto impacto, Antes do baixo impacto e Após o baixo impacto - fixo), Mês (6 níveis: janeiro a junho - aleatório, aninhado em Estação), Estação (2 níveis: Verão e Outono - aleatório, aninhado em Ano), Ano (3 níveis: 2015, 2016 e 2017 - aleatório), e Estrato (2 níveis: 0-5 cm e 5-10 cm - fixo). Todos os fatores fixos foram ortogonais entre si.

Para os dados univariados da macrofauna (densidade total, riqueza de espécies, diversidade e equitatividade), foi utilizada uma Permanova com 4999 permutações de unidades de dados brutos para analisar a possível variabilidade entre fatores (Anderson 2005). Foi utilizado o teste de permutação de Monte Carlo para obter os p-valores ($P < 0,05$) (Anderson & Robinson 2003, Anderson 2005). A Permanova foi realizada usando uma matriz de distância euclidiana. Os resultados significativos foram avaliados com comparações *a posteriori*, que também utilizaram 4999 permutações aleatórias para obter p-valores através da correção de Monte Carlo.

Para os dados multivariados (estrutura da assembleia), a Permanova foi baseada na matriz de similaridade de Bray-Curtis construída a partir dos dados de abundância. Estes valores foram transformados pela raiz quadrada ($ind.m^{-2}$) para diminuir a importância das espécies dominantes. Valores de um (+1) foram somados para reduzir o efeito da ausência de indivíduos em algumas amostras (Clarke et al. 2006). A significância estatística das relações foi avaliada através do teste de permutação de Monte Carlo. Testes par a par foram aplicados aos fatores que resultaram significativos, usando a correção de Monte Carlo. A mesma abordagem estatística foi aplicada aos dados de tamanho de grão, mas ao invés de uma matriz de similaridade de Bray-Curtis, foi empregada a matriz de distância euclidiana. Adicionalmente foram feitas análises de agrupamento (Cluster) a partir da matriz de similaridade de Bray-Curtis para observar possíveis diferenças entre os tramatentos.

Metodologia específica Capítulo 4: Resposta do peracarido infaunal *Monokalliapseudes schubarti* (Tanaidacea: Kalliapseudidade) à perturbação do arrasto de fundo e à sazonalidade num estuário do sul do Brasil (*Responses of the infaunal peracarid* Monokalliapseudes schubarti *(Tanaidacea: Kalliapseudidae) to bottom trawling disturbance and seasonality in an estuary of Southern Brazil*)

Para avaliar as variações espaciais e temporais de *Monokalliapseudes schubarti* em zonas sem impacto do arrasto, foram utilizadas as amostras das quatro estações de 2015 das zonas controle e aquelas sem marcação visível nomeadas como Zona 1 ao 5, obedecendo assim um critério de proximidade geográfica (Apêndice 4, Fig. 1). Para avaliar o impacto da pesca do arrasto sobre a população desta espécie, foram utilizadas as amostras das zonas onde foi simulada a pesca de arrasto, antes e após o arrasto entre fevereiro e junho de 2015. Durante este ano *M. schubarti* representou 70% da macrofauna encontrada, ressaltando assim a sua importância como espécie dominante na área de estudo. Os indivíduos com tamanhos menores a 2,5 mm foram definidos como juvenis, a não ser que apresentassem características secundárias de machos. Os machos foram diferenciados das fêmeas baseado na morfologia do propódio (Apêndice 4, Fig. 2; Mañé-Garzón 1949). As fêmeas ovadas foram identificadas pela presença do marsúpio (Fonseca & D'Incao 2003).

Como parâmetros ambientais foram utilizados a temperatura, salinidade, granulometria e velocidade do vento. Com os dados horários de velocidade do vento foram calculadas as médias diárias para cada dia de coleta. A granulometria foi tratada de acordo com o Capítulo 1.

Para as zonas não impactadas, o delineamento para as análises estatísticas foi constituído por cinco fatores: Área (2 níveis: Área 1 e Área 2 - fixo), Zona (5 níveis: Zona 1 a Zona 5 - aleatório, aninhado na Área), Estrato (2 níveis: 0-5 cm a 5-10 cm - fixo), Estação (4 níveis: verão, outono, inverno e primavera - fixo) e data de amostragem (9 níveis: coletas realizadas entre fevereiro e novembro - aleatório, aninhado na Estação).

A Permanova foi usada para analisar o conjunto de dados univariados (densidades totais e densidades de cada categoria morfológica: machos, fêmeas, fêmeas ovigeras e juvenis) em resposta ao modelo estatístico completo (incluindo interações). Para cada fator, 4999 permutações de unidades de dados brutos foram aplicadas para obter os p-valores (Anderson 2005). Como para alguns fatores, houve poucos números de permutações possíveis, usamos o p-valor de Monte Carlo (Anderson & Robinson 2003, Anderson 2005). A Permanova foi realizada utilizando uma matriz de distância euclidiana das densidades totais / categorias morfológicas. A significância dos p-valores foi fixada em 0,05. Os resultados significativos foram avaliados com comparações *a posteriori*, que também utilizaram 4999 permutações aleatórias para obter p-valores através da correção de Monte Carlo. Análises de correlação de Spearman foram realizadas para avaliar as relações entre as densidades totais / categorias morfológicas de *M. schubarti* e os parâmetros ambientais.

Os efeitos do impacto nas densidades totais / categorias morfológicas foram testadas com uma Permanova univariada através do seguinte modelo: Zona (2 níveis: Baixo impacto, Alto impacto - fixo), Tempo (2 níveis: antes, depois - fixo), Estrato (2 níveis: 0-5 cm e 5-10 cm - fixo) e Mês (5 níveis: fevereiro a junho - aleatório). Todos os testes foram realizados usando um procedimento de permutação (com 4999 permutações), conforme descrito para a Permanova acima. As análises foram realizadas usando o pacote de software Primer V6 + PERMANOVA *add on* (Anderson et al. 2008) e o software R V3.4.1 (R Core Team 2017).

SÍNTESE DOS RESULTADOS E DISCUSSÃO

Capítulo 1. Variações espaço-temporais de sedimentos em um estuário subtropical de micro maré (*Spatial and temporal variations of sediments in a subtropical micro tidal estuary,* artigo em preparação para ser submetido na revista Brazilian Journal of Oceanography, apresentado no Apêndice 1)

O substrato de zonas rasas geralmente é influenciado de forma direta pela ação hidrodinâmica em suas diferentes intensidades, a qual pode ser gerada pela própria circulação local ou pela atuação de ondas moduladas por ventos. Nos resultados do presente trabalho observa-se que nas zonas rasas de ilha da Pólvora o as ondas geradas por ventos são o principal agente causante das variações temporais dos sedimentos, enquanto que nas zonas rasas da ilha dos Cavalos assim como nas mais profundas as variações batimétricas são o principal fator correlacionado com as variações nos tamanhos de grão. A ação de agentes hidrodinâmicos no substrato pode implicar na remobilização dos sedimentos mais leves, impedindo a deposição do material fino e favorecendo o predomínio das frações mais grossas, ou na deposição de sedimentos em situações e locais menos dinâmicos (Dyer et al. 2000, Santos et al. 2004, Souza & Hartmann 2008). O estresse ocasionado pelas ondas geradas pelo vento é uma função complexa de altura da onda e profundidade da coluna d’água. A altura da onda depende da velocidade e direção do vento, do tempo de duração de tempestades, da profundidade de fechamento das ondas e da profundidade da água no local de formação da onda (West & West 1991).

A precipitação da bacia de drenagem da Lagoa dos Patos apresentou uma oscilação bastante grande no período de estudo, variando entre 20-336 mm por mês. Os meses mais secos foram de maio a julho de 2016 e os mais chuvosos aqueles do inverno e primavera de 2015 e 2016, destacando outubro de 2015 como o mês mais chuvoso do período de estudo (Apêndice 1, Fig. 2).

O nível da laguna perto da sua desembocadura apresentou uma grande variação mensal, sendo os meses com maior altura os correspondentes ao inverno e primavera 2015 e outono 2016. Os meses com o nível mais baixo foram maio de 2015, fevereiro e outubro de 2016, e abril e maio de 2017 (Apêndice 1, Fig. 3). Após 2013, as oscilações do nível da laguna estiveram acima da média histórica com exceção de janeiro de 2016 quando o nível baixou a 53 cm. Os anos de 2014 a 2016 foram muito chuvosos, tendo sido influenciados por dois eventos de *El Niño*, um fraco (2014-2015) e um forte (2015-2016). No entanto, quando comparada à serie histórica de pluviosidade, pôde ser observado que em outros eventos de *El Niño* houve maiores taxas de precipitação, mas por períodos de tempo menores. O aumento do nível também pôde ter sido favorecido por uma maior retenção de água no estuário a consequência da ampliação dos molhes da barra de Rio Grande. Como foi modelado por Fernandes et al. (2005) analisando possíveis cenários de alterações na circulação do estuário como consequência da obra, a ampliação poderia ocasionar aumento dos fluxos de vazante e diminuição dos fluxos de enchente, diminuição da distância atingida pela água salgada no interior do estuário e aumento do nível de água no interior do estuário. Esta obra terminou em 2011, mas foi seguido por um ano de baixa pluviosidade como consequência do fenômeno *La Niña*.

O conteúdo de matéria orgânica (MO) variou de 0-10,59%. Os maiores valores foram observados no inverno e primavera de 2015 para todos os pontos. Particularmente, durante inverno e primavera de 2015 e 2016 os pontos mais profundos perto da ilha dos Cavalos (4C e 5C) foram os que apresentaram maior valor de MO. Os ventos NE formam um gradiente de pressão que favorece a vazão da laguna, os quais são predominantes ao longo do ano, enquanto que os ventos SW são predominantes no inverno e primavera e favorecem o predomínio de condições polihalinas no estuário (Möller et al. 2001). Simultaneamente, existe uma alta variabilidade na descarga dos principais afluentes da Lagoa dos Patos, ocorrendo uma alta descarga no inverno e primavera, seguida por uma baixa descarga no verão e outono (Möller et al. 1996). Isto pode explicar o maior teor de matéria orgânica no inverno e a primavera, já que os rios aportam grandes quantidades de sedimentos em suspensão que depositarão nas zonas mais calmas do estuário. Adicionalmente, o alto nível da laguna, outro indicativo de maior descarga fluvial, poderia explicar o alto teor de matéria orgânica no substrato entre 2015 e 2017, quando comparado com os valores reportados para o Saco do Arraial por Rosa & Bemvenuti (2004) entre 0,2-0,8%, e por Rosa & Bemvenuti (2006) entre 0,09-3,15%.

Com relação à textura do sedimento, os resultados apontam um predomínio das fácies arenosas em ambas as áreas estudadas do Saco do Arraial, apesar das diferenças entre os pontos principalmente quanto ao teor de silte e argila. Isto coincide com o reportado por Antiqueira & Calliari (2005) e por Tomazelli & Villwock (2000), que reportam que nas zonas rasas inferiores a 4 m de profundidade da Lagoa dos Patos o sedimento é predominantemente arenoso. Apesar dos sedimentos de ambas as áreas serem constituidos predominantemente por areias, pôde-se observar que as áreas do lado exposto da ilha dos Cavalos apresentaram maior quantidade de frações grossas, enquanto que as áreas na margem protegida da ilha da Pólvora apresentaram maiores quantidades de sedimentos finos. A maior porcentagem de areias medias a grossas nos pontos próximos à ilha dos Cavalos reflete a hidrodinâmica do local, pois estão mais expostos à maior energia da água e da circulação local associada ao canal principal da laguna.

Na área próxima à Ilha da Pólvora (exposta para os ventos S-SE) a direção do vento nas 24 horas antes da coleta foi o fator que mais influenciou na variação da granulometria, afetando negativamente a porcentagem de silte, e positivamente de areia fina a média, isto é, ventos de quadrante S-SW favorecem a formação de ondas que ressuspendem os sedimentos ocasionando a remoção do silte e, portanto, aumenta a porcentagem de areias. Adicionalmente, a velocidade do vento também influenciou negativamente na porcentagem do silte, sendo que ventos muito fortes podem ressuspender estes tamanhos de grão mais leves (Apêndice 1, Tabela II). Na área próxima à Ilha dos Cavalos, o vento não foi tão importante para a variabilidade dos sedimentos como foi a profundidade, a qual se correlacionou positivamente com a porcentagem de silte e argila e negativamente com areias finas a muito grossas. Desta forma, quanto maior a profundidade, maior a porcentagem de silte e argila e menor a de areias mais grossas (Apêndice 1, Tabela II). A temperatura foi um fator correlacionado negativamente com a porcentagem de silte nas duas áreas, indicando que nos meses mais frios houve um aumento da quantidade de silte na região. Contudo, acredita-se que seja uma correlação indireta coincidindo com a sazonalidade dos ventos e descarga fluvial. Na Ilha da Pólvora, a batimetria entre os pontos das zonas rasas estudadas foi semelhante, e menos profunda que na Ilha dos Cavalos. Isto sugere que o vento pode ter, na Ilha da Pólvora, uma maior influência na resuspenssão, erosão e transporte de sedimentos, sendo por isto um dos fatores com maior correlação nas variações da granulometria. Na Ilha dos Cavalos, os pontos são mais profundos e apresentam maiores diferenças batimétricas entre sí. Isto implica na provável menor influência dos ventos nas variações da granulometria, sendo por isto a profundidade um dos fatores mais correlacionados com os diferentes tamanhos de grão.

Adicional ao efeito direto dos agentes hidrodinâmicos, a erosão dos sedimentos finos também responde à atuação dos organismos bentônicos. Organismos como poliquetas que segregam muco, e diatomáceas que compõem o microfitobentos, podem atuar para uma maior resistência à erosão, aumentando a coesão dos grãos de sedimento. Por outro lado, alguns bivalves e anfipodas podem desestabilizar o sedimento durante a construção das suas galerias (Kornman & De Deckere 1998, Dyer et al. 2000). Desta forma, o estabelecimento das comunidades bentônicas pode ter uma influência importante nos processos de estabilização e erosão e de sedimentos (Kornman & De Deckere 1998, Widdows et al. 2000, Widdows & Brinsley 2002). O declínio da produtividade biológica simultaneamente às fortes tempestades do inverno pode facilitar a erosão, criando um ciclo de erosão e deposição como observado por Dyer et al. (2000). Durante o inverno, há um reconhecido declínio da abundância da macrofauna em áreas rasas da Lagoa dos Patos (Bemvenuti & Netto 1998, Rosa & Bemvenuti 2006), que pode se estender até a primavera em períodos de elevadas precipitações e menores salinidades (Colling et al. 2007). Neste contexto, em períodos com menores abundâncias do zoobentos, este pode ser um fator adicional para o aumento dos processos erosivos que ocorram nas enseadas rasas estuarinas.

Além da variação sazonal na composição granulométrica, foi encontrada uma variação sazonal nos parâmetros estatísticos dos sedimentos, principalmente nas zonas mais profundas (4P e 5C) no inverno e primavera. Nestas estações houveram maiores valores na mediana, indicando sedimentos mais finos e menores em assimetria, indicando um ambiente de menor energia. Isto complementa o já observado através da textura de sedimentos, observando-se menores tamanhos de grão como consequência do maior aporte de sedimentos finos nestas estações. Além deste componente temporal condicionando o aporte destes sedimentos, também é importante reconhecer que nestas sazões ocorrem os ventos de maior intensidade que condicionam processos erosivos, atuando principalmente nas áreas rasas e marginais de ilhas, devido á ação das ondas.

Capítulo 2. Variações espaço-temporais de assembleias macrobentônicas em áreas estuarinas rasas com diferentes condições hidrodinâmicas (*Temporal and spatial distribution of macrobenthic assemblages in areas of an estuarine shallow mudflat with different hydrodynamic conditions,* artigo em preparação para ser submetido na revista Journal of the Marine Biological Association, apresentado no Apêndice 2)

A temperatura variou entre os 9-28 °C com valores médios entre 20-23 °C na primavera e verão e entre 14-17 °C no outono e inverno. A salinidade medida nos dias de coleta variou entre 0-36, sendo os maiores valores no outono, com média de 21 ± 10 e os menores em todas as primaveras (2015 e 2016) com média de 0 ± 0. Através dos dados diários foi observada uma diferença no padrão de salinidades entre os três anos, com valores muito baixos (<5) em 2015 e um único pico de salinidade no outono; uma maior oscilação durante 2016 com valores próximos a 5 e um único pico de aumento de salinidade entre final de verão e começo do outono; e os maiores valores de salinidade foram observados em 2017 com um pico no final do verão e outro maior no final do outono. Os baixos valores de salinidade medidos durante o período de estudo se devem a épocas de alta precipitação devido a eventos *El Niño*, que ocasionam fortes chuvas na região, incrementando a descarga dos rios (Fernandes et al. 2002, Barros et al. 2014). A Análise de Componentes Principais mostrou uma separação espacial entre as zonas amostradas, principalmente com relação aos pontos com presença de sedimentos grossos (biodetritos) simultâneo à elevado teor de finos no Ponto 2 (ponto mais profundo da Ilha da Pólvora- Área 1; 2,7 m). Além disso, o Ponto 3 (ponto mais raso da Ilha dos Cavalos- Área 2; 1,7 m) apresentou uma maior proporção de areias finas e médias, ao contrário do Ponto 5 (Área 2; 2,0 m) que apresentou as maiores proporções de silte e argila (Apêndice 2, Fig. 3; Fig. 4). Quanto às tendências temporais, os pontos mais rasos tanto da Área 1 quanto da Área 2 apresentam uma maior estabilidade temporal dos tamanhos de grão, ao contrário dos pontos mais profundos das duas áresas, que apresentam uma maior variação temporal.

As densidades da macrofauna variaram sazonalmente, com incrementos somente no outono seguindo os aumentos da salinidade, e diminuições no inverno e primavera seguindo as tendências de decréscimo da salinidade e temperatura. Este padrão temporal, com baixas densidades nas primaveras e altas nos outonos, coincide com o reportado para anos anteriores influenciados por eventos de *El Niño* (Colling et al. 2007). Ao contrário, os resultados contrastam com os reportados para anos sem a influência destes eventos climáticos, quando a salinidade aumenta simultaneamente com a temperatura durante o verão, que induzem os eventos reprodutivos da macrofauna e recrutamentos das principais espécies do macrozoobentos (Bemvenuti & Netto 1998, Rosa & Bemvenuti 2004, 2006).

Como o esperado, a fauna foi mais abundante e com maior riqueza no Estrato 0-5. Resultados semelhantes com maior abundância e número de espécies nas camadas mais superficiais do sedimento foram observados na Baia de Chesapeake (Schaffner 1990) e no estuário de Douro (Mucha et al. 2004). Estas preferencias da fauna no estrato mais superficial têm sido associadas a uma maior disponibilidade de matéria orgânica recien assentada ou a material em suspensão (Dauwe et al. 1998), assim como a maiores concentrações de nitrogênio (Yang et al. 2017) e oxigênio (Baillie 1986).

Foram identificados 34086 indivíduos pertencentes a 9 grandes grupos: Amphipoda, Tanaidacea, Isopoda, Cumacea, Mysidacea, Decapoda, Bivalvia, Gastropoda, Polychaeta e Nemertea. Destes grupos, foram identificados 12 gêneros e 10 espécies, sendo a mais frequente e abundante *Monokalliapseudes schubarti*. Esta riqueza de espécies resulta ligeiramente menor que o previamente reportado para a ilha das Pombas (oeste da ilha da Pólvora no Saco do Arraial), onde foram encontrados 14 gêneros e 12 espécies (Colling et al. 2007). Essa menor riqueza pode se dever à baixa salinidade que predominou durante o período de estudo que pôde ter afetado algumas espécies e a ter amostrado em uma área espacialmente diferente. A maior riqueza foi encontrada no Ponto 3 (ponto mais raso na Área 2), que é justamenteo ponto com maior hidrodinâmica causada pelas ondas induzidas pelo vento assim como pelas correntes de circulação do canal principal, o que pode ser inferido pela maior porcentagem de areias medias quando comparado aos outros pontos. Em regiões sublitorais do oeste da Finlândia, Valanko et al. (2010) também encontraram maiores riquezas de espécies em zonas com maior hidrodinâmica, quando comparada a distribuição de pós-larvas ao longo de um gradiente de exposição a hidrodinâmica imposta pela ação dos ventos. Da mesma forma, no mar de Wadden um maior número de espécies foi encontrado em sedimentos com tamanhos de grão intermediários, quando comparados aos sedimentos muito finos ou muito grossos (Beukema & Dekker 2012). Tanto a diversidade quanto a equitatividade, no presente estudo, não apresentaram maiores diferenças temporais nem espaciais, semelhante ao reportado no ELP por Colling et al. (2007).

As áreas apresentaram uma dissimilaridade de 57,94% entre elas, principalmente pela abundância de *Monokalliapseudes schubarti* (contribuição de 29,16%) a qual foi maior na Área 2, e do poliqueta *Heteromastus similis* (contribuição de 19,50%) que foi maior na Área 1. O tanaidáceo apresentou altos valores de correlação com o incremento da salinidade e da profundidade, o que explica sua maior abundância no outono (período de maior salinidade) e na Área 2 (área com maior profundidade) (Apêndice 2, Tabela II). O poliqueta, por sua vez, apresentou correlação negativa com os sedimentos finos e positiva com as areias finas. Mesmo sendo significativas, o valor das correlações diminuiu conforme aumentou o tamanho de grão (Apêndice 2, Tabela II). Isto é esperado já que a espécie predominou no Ponto 2, o qual apresentou maiores proporções de fragmentos de conchas de moluscos mortos e o principal tamanho de grão foi o de areias finas. Adicionalmente foi a única espécie com correlações significativas com o teor de matéria orgânica (Rho= -0,164). Em lagunas costeiras do Uruguai, esta espécie também se correlacionou positivamente às porcentagens de areias finas e cascalhos (Giménez et al. 2014). Entretanto, ao contrário do encontrado no presente trabalho, os autores reportam uma correlação positiva com as porcentagens de lama. Provavelmente devido às baixas salinidades encontradas nas épocas de maior porcentagem de lama, as densidades da espécie tenham diminuído resultando numa correlação negativa. Os poliquetas são reconhecidos por serem estabilizadores de sedimento devido à produção de mucos que facilitam a retenção de sedimentos finos (Dyer et al. 2000). Como capitelideo, *H. similis* segrega mucopolissacarídeos para aglutinar grãos de areia e possivelmente selecionar matéria orgânica (Fauchald & Jumars 1979). Desta forma, regiões com baixa hidrodinâmica favorecem a deposição de matéria orgânica e sedimentos finos, favorecendo condições propicias para a espécie. *Nephtys fluviatilis* é uma espécie bastante móvel e preda *H. similis* (Bemvenuti 1994), portanto é de esperar sua presença nas mesmas condições ambientais que a presa.

Quando analisadas as áreas separadamente, foi observado que a dissimilaridade entre os pontos da Área 1 foi de 55,96%, sendo as principais espécies responsáveis *M. schubarti* (29,87% de contribuição), *H. similis* (25,51%) e *Nephtys fluviatilis* (16,93%). Na Área 2, a semelhança entre os Pontos 3 e 4 é maior (dissimilaridade de 48,21%) que entre os Pontos 3 e 5 (dissimilaridade de 50.33%). As principais espécies responsáveis por estas dissimilaridades entre pontos são *Erodona mactroides* (~26,76%), *M. schubarti* (~26,36%) e *Heleobia australis* (~13%).

Estas espécies responsáveis pela dissimilaridade entre os pontos apresentaram uma marcada sazonalidade, com as maiores abundâncias no outono. Os poliquetas particularmente aumentaram em grandes quantidades no outono de 2015 no Ponto 2, alcançando densidades de 6260,09 ± 4025,21 ind.m^{-2} (media ± desvio padrão). Em ambas as áreas, *M. schubarti* apresentou altas densidades no final do verão, outono e inverno em todos os pontos, exceto o Ponto 1. A espécie apresentou uma clara sazonalidade com picos de abundância no outono e inverno, seguido por densidades muito baixas na primavera. Os maiores valores foram encontrados no outono de 2016 nos Pontos 2 (6238,87 ± 3055,54 ind.m^{-2}) e 4 (6603,87 ± 5570,93 ind.m^{-2}), e no outono de 2017 no Ponto 3 (6387,41 ± 2820,07 ind.m^{-2}). Na Área 2, também houve uma maior abundância de *E. mactroides*, que foi encontrada mais frequentemente em altas densidades no Ponto 3, o qual está localizado em uma área mais exposta às correntes locais e indica uma maior influência da hidrodinâmica. Em um estudo prévio considerando os recrutamentos deste bivalve, foi observado que as maiores abundâncias ocorreram, de fato, em uma área mais exposta à circulação local da mesma enseada rasa, quando comparada a uma área protegida desta circulação onde os recrutamentos foram menos significativos (Colling et al. 2010). Por sua vez, o Ponto 5 que também está localizado na Área 2 (mais exposta à circulação do canal principal do estuário) apresentou maiores recrutamentos de *E. mactroides* no outono de 2016 (3955,53 ± 6206,39 ind.m^{-2}). A espécie também apresentou uma clara sazonalidade com aumentos no outono, e um segundo pico de abundância no verão de 2016, o que espacialmente ocorreu de forma recorrente no Ponto 3. A espécie *E. mactroides* apresenta larvas planctônicas cujos adultos encontram-se em maior abundância em regiões do alto estuário, as quais são levadas pelas vazantes ao baixo estuário durante a primavera e o verão, recrutando em enseadas abrigadas (Colling et al. 2010). Nesse sentido, a ocorrência das maiores abundâncias do bivalve nos Pontos da Área 2, além de sua correlação positiva e significativa com tamanhos maiores de grão (Apêndice 2, Tabela II), indica que seus recrutamentos ocorrem preferencialmente em locais com um mínimo de hidrodinâmica que garanta o aporte de suas larvas. Desta forma, a disposição espacial da Área 2 parece ser a mais propícia ao recebimento de suas larvas meroplanctônicas, quando comparada à Área 1, que se localiza em uma região menos próxima ao canal principal do estuário, e apresenta maiores percentuais de sedimentos finos, indicando menor influência de correntes locais. No Golfo da Finlândia, foi observado que em áreas com ausência de marés regulares, a dispersão do meroplâncton de espécies infaunais é favorecida pelas ondas e correntes induzidas pelo vento e pode ser modificada pelo regime hidrodinâmico local (Valanko et al. 2010). Em enseadas rasas da Nova Zelândia, foi observado que bivalves juvenis se dispersam em escalas de metros dentro de um ciclo de maré (Norkko et al. 2001), o que destaca a importância da influência das forçantes hidrodinâmicas locais sobre a dispersão de recrutas. Da mesma forma que o bivalve *E. mactroides*, o gastrópodo *H. australis* também apresentou maiores densidades na Área 2. As maiores densidades foram encontradas no Ponto 3, principalmente na primavera de 2015 (2716,24 ± 3196,65 ind.m^{-2}). Esta espécie é encontrada em elevadas abundâncias (> 30.000

ind.m^{-2}) nos canais profundos do ELP (Pinotti et al. 2011). Considerando a disposição da Área 2 no presente trabalho, é possível que as maiores abundâncias deste gastrópodo tenham relação com os locais onde ocorre de forma mais abundante, como a região do canal portuário mais próximo à Área 2, quando comparado à Área 1, com picos esporádicos de abundância.

Capítulo 3. Resposta de assembleias macrobentônicas de fundos inconsolidados à pesca de arrasto artesanal em um estuário subtropical (*Response of soft-bottom macrobenthic assemblages to artisanal trawling fisheries in a subtropical estuary,* artigo publicado no jornal Estuaries, Coastal and Shelf Science, apresentado no Apêndice 3)

A média das densidades totais do macrozoobentos da Área 1 foi de 909,0 ± 1305,8 ind.m^{-2} (média ± desvio padrão). Diferenças significativas na estratificação por mês (Pseudo-F = 4,1255, P = 0,0002) foram encontradas, onde maiores densidades ocorreram no Estrato 0-5 em quase todos os meses (Apêndice 3, Fig. 4). A Permanova também detectou diferenças significativas nas densidades totais por mês (Pseudo-F = 28,383, P = 0,0002), com maiores incrementos no final do verão e início do outono (Apêndice 3, Fig. 4). A resposta das densidades totais da fauna ao tratamento do arrasto variou entre os estratos (Pseudo-F = 4,4103, P = 0,0308). Foram detectados mais sinais de impacto no Estrato 0-5, com decréscimo nas densidades após o arrasto em alguns meses (marcado como setas descendentes na Fig. 4 do Apêndice 3). Por outro lado, as densidades aumentaram em alguns meses após a pesca de arrasto no Estrato 5-10 cm, evidenciando o enterramento de organismos (marcados como setas ascendentes na Fig. 4 do Apêndice 3). As maiores diferenças nas densidades entre os tratamentos foram observadas entre os anos de 2016 e 2017, quando as abundâncias da macrofauna foram maiores. Foi detectado impacto principalmente na Área 1, uma vez que diminuiram as densidades e houve alteração na estrutura da comunidade. Adicionalmente, o distúrbio físico causado pelo arrasto variou acentuadamente entre as duas áreas, e parece estar relacionado principalmente à estrutura do sedimento: a Área 1 apresentou maiores teores de silte, argila e areias muito finas, enquanto a Área 2 foi caracterizada por uma granulometria mais grossa. Tal como pôde se ver através de imagens do sonar de varredura lateral (Apêndice 3, Fig. A.1 e Apêndice 4, Fig. 4), as marcas no fundo deixadas pelas redes de arrasto permaneceram visíveis e contínuas na maior parte dos casos na Área 1. Por outro lado, as marcas deixadas pelas redes de arrasto foram imediatamente recheadas por sedimentos vizinhos na Área 2. Padrões semelhantes, com impactos de arrasto sendo mais evidentes em zonas caracterizadas por níveis mais baixos de perturbação natural também foram observados por

Brylinsky et al. (1994), Jennings et al. (2001), Hiddink et al. (2006), e Prantoni et al. (2013). Na Baía de Fundy (Canadá), Brylinsky et al. (1994) detectaram que os impactos diretos na fauna bentônica foram causados principalmente pelas portas e não pelas correntes. As redes utilizadas pelos autores possuíam portas de 180 kg e deixaram marcas de até 5 cm de profundidade. Não conseguimos medir a profundidade das marcas de arrasto neste estudo, no entanto, se uma porta de 180 kg deixou marcas de 5 cm de profundidade, podemos supor que as nossas portas de 15 kg terão impacto sobre uma camada de sedimentos mais superficial. Essas observações poderiam implicar na ressuspensão do sedimento, o que foi percebido visualmente durante a ação do arrasto, mas possivelmente não sendo suficiente para resultar em alterações granulométricas nos estratos analisados. Isso também pode explicar os maiores sinais de impacto sobre os macrobentos do Estrato 0-5 e a manutenção da estratificação vertical.

A riqueza de espécies variou de 0 a 7 espécies por amostra na Área 1. A distribuição das espécies entre os estratos variou de forma diferente em cada mês (Pseudo-F = 1,9402, P = 0,0472) (Apêndice 3, Fig. 6). Além disso, a riqueza variou de maneira diferente para cada tratamento em cada mês (Pseudo-F = 1,7806, P = 0,0088). O Estrato 0-5 apresentou maior riqueza e foi mais variável entre os meses do que entre os tratamentos. Em aproximadamente 36% das amostragens ao longo dos três anos, a riqueza diminuiu após o arrasto de alto impacto no Estrato 0-5, perdendo-se entre 1-3 espécies, e em 29% das réplicas do experimento no Estrato 5-10.

A riqueza de espécies foi maior na Área 2, variando entre 0 a 10 espécies por amostra. A maior riqueza ocorreu geralmente no Estrato 0-5, com menor riqueza em 2015 (Pseudo-F = 885,39, P = 0,0002). Diferenças significativas foram detectadas na interação entre tratamento, estrato e estação (Pseudo-F = 2,612, P = 0,0126). Foram observados decréscimos na riqueza de espécies após o arrasto de alto impacto em poucas ocasiões: quatro no Estrato 0-5 (maio de 2015, janeiro de 2016, abril de 2016, junho de 2016) e cinco no Estrato 5-10 (abril de 2015, maio de 2015, janeiro de 2016, abril de 2016, março de 2017) (ver os números acima das colunas da Fig. 7 do Apêndice 3).

Não foram observadas diferenças na diversidade e equitatividade entre os anos. O índice de diversidade de Shannon não apresentou diferenças significativas entre os tratamentos para nenhuma área (Área 1 média H '= 0,85 ± 0,45; Área 2 média H' = 0,86 ± 0,41). A diversidade foi sempre maior no Estrato 0-5 na Área 1 (Pseudo-F = 21,174, P = 0,0398; H’= 1,08 ± 0,32) quando comparado ao Estrato 5-10 (H’ = 0,62 ± 0,45). A mesma tendência, com maiores valores no Estrato 0-5 (Pseudo-F = 57,69, P = 0,0118; H '= 1,09 ± 0,30) e menores no Estrato 5-10 (H' = 0,63 ± 0,37) foi observada para a Área 2.

A estrutura da macrofauna apresentou uma variação mensal na resposta frente ao tratamento de pesca (Pseudo-F= 1,2338, P=0,028) e a estratificação (Pseudo-F= 3,6269, P=0,001) na Área 1. Somente 4 das 10 espécies encontradas nesta área apresentaram diferenças significativas frente ao impacto da pesca de arrasto em algum nível: *Erodona mactroides* apresentou diferenças nas abundâncias na interação entre tratamento, mês e estrato (Pseudo-F = 2,5043, P = 0,0002), com diferenças restritas ao Estrato 0-5. *Heleobia australis* apresentou diferentes respostas ao tratamento por estação (Pseudo-F = 5,9964, P = 0,0002), com decréscimos significativos de densidade no outono de 2017, após alto (t = 2,6737, P = 0,0268) e baixo (t = 9,9924, P = 0,0002) impacto de arrasto. Também foram detectadas respostas na interação de estação, tratamento e estrato (Pseudo-F = 5,4449, P = 0,0002), com diferenças ocorrendo apenas no Estrato 0-5. Da mesma forma, diferenças significativas nas densidades de *Heleobia charruana* foram detectadas para a interação de tratamento e estação (Pseudo-F = 2,1867, P = 0,0388). A espécie foi registrada apenas no verão de 2015 e diminuiu suas densidades após o arrasto de baixo impacto. *Laeonereis acuta* também mostrou respostas diferentes ao tratamento de arrasto em cada estrato por estação (Pseudo-F = 3,0158, P = 0,0078). Foram observados decréscimos nas densidades tanto no verão quanto no outono de 2015 e 2017, e no verão de 2016 no Estrato 0-5 após o arrasto de alto impacto. Observamos decréscimos marcados das densidades de *Monokalliapseudes schubarti* no outono de 2015 (15%) e 2017 (60%) após o arrasto de alto impacto, contudo não foram detectados resultados significativos.

Na Área 2 foram observadas maiores densidades e riqueza no Estrato 0-5, com exceção de abril e maio de 2015 (Apêndice 3, Fig. 7). A estrutura de assembleias e densidades das espécies dominantes apresentaram variabilidade entre meses e uma marcada variabilidade sazonal. Ao analisar as espécies separadamente, foram detectadas diferenças significativas frente ao tratamento do arrasto apenas nas densidades de *Heteromastus similis* (Pseudo-F = 4,9073, P = 0,0126), com decréscimos após arrasto de alto impacto, permanecendo significativamente diferentes da zona controle (t = 4,6444, P = 0,015).

A resposta da macrofauna bentônica à perturbação do arrasto depende da biologia e do comportamento de cada espécie. As atividades de arrasto podem ressuspender ou deslocar a macrofauna como resultado do distúrbio físico criado pelas redes (Dell et al. 2013, Johnson et al. 2014, Collie et al. 2016). Assim, as espécies mais leves da epifauna, como os crustáceos peracáridos, provavelmente são mais suscetíveis à ressuspensão e ao deslocamento pelas forçantes hidrodinâmicas e sedimentares naturais, do que os organismos mais pesados, como os moluscos. Construtores de tubos como *Laeonereis acuta* e *Monokalliapseudes schubarti*, que constroem tubos com profundidade superior a 5 cm, poderiam sentir a aproximação das portas e descer dentro do tubo, evitando o impacto, como sugerido por Brylinsky et al. (1994) para poliquetas tubícolas. Este comportamento evasivo pode explicar aumentos nas densidades no Estrato 5-10 após o arrasto. Considerando a importância do detritívoro *L. acuta* e do detritívoro/suspensívoro *M. schubarti* como principais presas de peixes e crustáceos no ELP (Soares et al. 2004, Oliveira et al. 2006, Bemvenuti & Colling 2010), qualquer impacto antrópico sobre as suas populações e, portanto, da disponibilidade de alimentos para as teias tróficas precisam ser cuidadosamente avaliadas.

O gastrópode *H. australis* e o bivalve *E. mactroides* apresentaram respostas semelhantes frente à pesca de arrasto, com evidências de impacto relacionadas aos períodos de sua maior abundância. A sobreposição temporal da atividade pesqueira e as maiores abundâncias de *H. australis*, por exemplo, que apresenta recrutamento e crescimento mais rápido durante os meses mais quentes (Carcedo & Fiori 2012), representam um ponto chave para a sustentabilidade das redes tróficas estuarinas. Tem sido reportada a importância de *H. australis* (Carcedo & Fiori 2012) e *E. mactroides* (Oliveira et al. 2006, Mendoza-Carranza & Vieira 2008) como alimento para crustáceos e peixes, sendo estas espécies as principais presas do caranguejo *Callinectes sapidus* (Oliveira et al. 2006).

Apesar da falta de um padrão linear e contínuo na resposta das espécies macrobentônicas à pesca de arrasto, registramos algumas evidências do impacto do arrasto no macrozoobentos e no substrato. Estes impactos foram evidenciados através da aplicação de arrastos experimentais mensais com um único barco. Extrapolando estas observações para todo o cenário pesqueiro do ELP, onde Kalikoski & Vasconcellos (2012) sugerem que pelo menos 370 embarcações estão equipadas para utilizar redes de arrasto, os impactos ambientais desta pescaria seriam exponencialmente maiores.

Capítulo 4. Resposta do peracarido infaunal *Monokalliapseudes schubarti* (Tanaidacea: Kalliapseudidade) à perturbação do arrasto de fundo e à sazonalidade num estuário do sul do Brasil (*Responses of the infaunal peracarid* Monokalliapseudes schubarti *(Tanaidacea: Kalliapseudidae) to bottom trawling disturbance and seasonality in an estuary of Southern Brazil*, artigo submetido no jornal Marine Biology Research, apresentado no Apêndice 4)

Foram identificados 12916 indivíduos do tanaidáceo *Monokalliapseudes schubarti* (densidade média ± desvio padrão: 2383,36 ± 3120,19 ind.m^{-2}) nas cinco zonas não impactadas. Dentre eles, 54,5% foram fêmeas, 31,7% juvenis e 10,4% machos e 0,8% fêmeas ovigeras. As densidades totais entre as duas áreas foram significativamente diferentes (Pseudo-F= 8,9662, P=0,026), sendo maiores na Área 2 (Apêndice 4, Fig. 5 e 6). Houve uma clara sazonalidade com baixas densidades no verão, aumentos no outono e diminuições no inverno e primavera (Apêndice 4, Fig. 5 e 6). Adicionalmente, as densidades exibiram interações significativas entre zonas, estratos e data de amostragem (Pseudo-F= 2,3368, P=0,0052). Os resultados da estrutura da população foram semelhantes aos das densidades totais com interações significativas entre zonas, estratos e data de amostragem (P<0,025) para todas as categorias morfológicas. Quando analisada a estratificação vertical, não houve diferenças significativas nas densidades das fêmeas (ovigeras e não ovigeras). No entanto, houve diferenças para os juvenis (Pseudo-F= 7,1221, P=0,014) e machos (Pseudo-F= 6,3392, P=0,023). As fêmeas ovigeras foram encontradas apenas no verão e outono, com maiores densidades no outono, e as maiores abundâncias estiveram no Estrato 5-10. Os machos geralmente estiveram mais associados ao Estrato 5-10, ocorrendo durante o ano todo, com maiores densidades no outono e inverno. Os juvenis foram encontrados em ambos os estratos, mas com maior abundância no Estrato 0-5, apresentando picos de densidades no outono e inverno (Apêndice 4, Fig. 5 e 6).

A variação sazonal das densidades totais de *M. schubarti* coincidiram com as flutuações já reportadas para a macrofauna em períodos com predominância de cenários límnicos na região estuarina do ELP (Colling et al. 2007). No ano de 2015, houve uma maior taxa de precipitação devido à ocorrência do fenômeno de *El Niño* (NOAA 2017), que aumenta a descarga dos rios da bacia de drenagem da Lagoa dos Patos (Fernandes et al. 2002, Barros et al. 2014). Variações temporais semelhantes nas variações do macrobentos também foram observadas no estuário de Lavaca - Colorado (Pollack et al. 2011) e no golfo de Riga - Mar Báltico (Kotta et al. 2009).

Entre os parâmetros ambientais medidos, a salinidade apresentou uma forte correlação com a densidade de todas as categorias morfológicas, com grandes decréscimos nas densidades em relação a salinidades inferiores a 5. Houve fortes correlações positivas entre as densidades de fêmeas ovígeras e a temperatura (Apêndice 4, Tabela I). As variações em salinidade e temperatura têm efeitos importantes no crescimento, reprodução e sobrevivência de macroinvertebrados, devido a mudanças nas taxas metabólicas pelo estresse térmico e osmótico (Uwadiae 2009). Em outros peracaridos, tem sido demonstrada a incapacidade de se reproduzir a baixas salinidades, mesmo que o adulto possa tolerar amplas variações (Steele & Steele 1991), assim como que certas temperaturas são indispensáveis para o início e término do período reprodutivo (Panov & McQueen 1998).

Dentre as caraterísticas do sedimento, a porcentagem de areia média teve maiores influências na densidade, apresentando correlações positivas com todas as categorias da população (Apêndice 4, Tabela I). As porcentagens de sedimentos finos (areia muito fina, silte e argila) apresentaram correlações negativas com a densidade total da espécie. As maiores densidades foram observadas com porcentagens de areia média superior a 7% e porcentagens de sedimentos finos menores a 15%. Como sugerido por Rosa-Filho & Bemvenuti (1998), as caraterísticas do sedimento são um fator limitante na distribuição de *M. schubarti*, o qual precisa de uma certa porcentagem de silte e argila. Nossos resultados foram semelhantes aos de Leite et al. (2003) e Freitas-Júnior et al. (2013), que também encontraram maiores densidades de *M. schubarti* relacionadas a porcentagens de silte e argila entre 4-10%, e mais baixas quando as porcentagens destes tamanhos de grão são maiores do que 13%.

A resposta de *M. schubarti* frente ao arrasto foi diferente entre as áreas estudadas, sendo sugerido que em áreas estuarinas rasas muito dinâmicas o efeito do arrasto muda com o tempo e interatua com os efeitos das perturbações naturais. Não foi identificado um claro padrão de impacto do arrasto. A resposta do *M. schubarti* foi diferente entre os meses, as zonas e os estratos (Pseudo-F= 2,8531, P=0,0322) na Área 1, enquanto que na Área 2 foram entre os meses, estratos e a sua interação (P=0,0002). Nossos resultados sugerem que um alto impacto de arrasto pode afetar a distribuição dos juvenis que estão no estrato mais superficial (0-5 cm), assim como os machos que deixam os seus tubos incursionando na busca das fêmeas para os processos reprodutivos (Apêndice 4, Fig. 7 e 8). As perturbações do arrasto podem ressuspender ou deslocar a macrofauna como resultado da perturbação física ocasionada pela rede (Dell et al. 2013, Johnson et al. 2014, Collie et al. 2016). Assim, a redução das densidades de *M. schubarti* nas camadas superficiais podem ser o resultado de mortalidade nas zonas arrastadas ou ressuspensão induzida pelo arrasto junto com a ressuspensão do sedimento. Fica a dúvida se os juvenis que ainda precisam do cuidado parental podem sobreviver após serem ressuspendidos e deslocados a relativas distâncias de sua progenitora.

As fêmeas ovígeras foram impactadas principalmente na Área 2, aquela composta por sedimentos mais grossos e, por consequência, uma possível maior hidrodinâmica, registrando-se desaparecimentos desta categoria após o arrasto. Os machos e juvenis, por sua vez, foram impactados em ambas as áreas. Isto sugere que a espécie sofre um maior impacto em áreas com maior hidrodinâmica por impactar a quase todos os componentes da população.

CONSIDERAÇÕES FINAIS

Nos baseando no enfoque ecossitémico da pesca, foram levantados dados sobre a variação sazonal e espacial do macrobentos e o sedimento, a modo de após compreender a dinâmica natural destes componentes do ecossistema entender de uma forma mais clara o impacto que a pesca de arrasto pode induzir. Desta forma, ao analizar as variações granulométricas ao longo de duas áreas com diferentes graus de exposição a hidrodinâmica causada pela circulação de correntes do canal principal estuarino, divididas por sua vez em duas zonas cada uma de acordo com as profundidades médias e expostas a diferentes graus de perturbação de ondas ocasionadas pelos ventos foi observado que: a perturbação ocasionada pelas ondas induzidas pelo vento teve maior influencia nas zonas mais rasas e com menor exposição á hidrodinâmica imposta pelas correntes do canal principal. Por outra parte nas zonas mais profundas, as variações batimétricas foram as principais responsáveis pela variação espacial dos sedimentos. A elevada taxa de precipitação ocasionada pelos eventos de *El Niño* cujo efeito predominou no período de estudo, repercutiu em maiores teores de matéria orgânica e sedimentos finos na região de estudo quando comparado com os valores obtidos em anos anteriores. Esta variabilidade nos sedimentos e nas taxas de precipitação, que induzem quedas na salinidade, influenciaram a variabilidade espacial e temporal da fauna macrobentônica, a qual apresentou diferenças significativas entre as áreas amostradas. Desta forma na área mais exposta à circulação do canal principal do estuário apresentou uma maior densidade e riqueza de espécies. Provavelmente essa maior condição hidrodinâmica induzida pelas correntes tenha favorecido o aporte de larvas para certas espécies com desenvolvimento meroplanctônico. Adicionalmente estas condições favorecem a oxigenação do substrato o que pode favorecer o desenvolvimento de espécies infaunais. Tem que ser ressaltado que as condições hidrodinâmicas impostas tanto pelas correntes como pelo vento nesta área mesmo sendo superiores que na Área 1 são de nível moderado, caracterizando por tanto o Saco do Arraial como zona protegida das correntes do canal principal do estuário e favorecendo por tanto sua utilização por recrutas de peixes e crustáceos como área berçário. Conhecendo então a variabilidade do sedimento e da fauna entre as áreas amostradas, foram aplicadas duas intensidades de pesca simulada em cada uma para avaliar a resposta da macrofauna e seu habitat à perturbação física imposta por redes de arrasto. Foi observado que na Área 1, aquela mais protegida das correntes do canal principal, o impacto do arrasto foi mais evidente, tanto nas marcas deixadas pelas redes no fundo como pela frequência de vezes em que foram detectados sinais de impacto tanto na densidade total como na estrutura das assembleias. Adicinalmente nesta área foi determinada que quase a metade das espécies identificadas apresentaram alterações nas suas densidades como consequência dos arrastos. Adicionalmente, foi observado em ambas as áreas as redes de arrasto exercieram perturbações principalmente no estrato superficial, não conseguindo atingir a distribuição estratificada da macrofauna. Finalmente, quando focado o impacto do arrasto sobre uma população, no caso a do tanaidáceo *Monokalliapseudes schubarti*, foram observados decréscimos nas densidades de machos e juvenis nas duas áreas, mas mais frequente na área com maiores porcentagens de sedimentos finos. Adicionalmente, na área mais exposta à circulação das correntes do canal principal do estuário observaram-se perdas das fêmeas ovígeras após o arrasto, implicando em maior impacto sobre a espécie nesta área. Tanto nas assembleias como na população do tanaidáceo, os sinais de impacto foram mais evidentes nas épocas com maiores densidades, as quais se desplazam entre verão e outono seguindo os padrões de variação da salinidade. Estas épocas de maior densidade de organismos, refletindo maior recrutamento e recuperação da macrofauna, coincidem com a época da safra da pesca, a qual pode influenciar estes processos de recrutamento, afetando principalmente a recuperação da macrofauna após perturbações naturais como elevadas precipitações e consequentes quedas na salinidade. Evidenciando assim que o arrasto apresenta consequências negativas tanto para assembleias como para populações de invertebrados macrobentônicos. Isto numa área berçário, onde adicionalmente o arrasto ocorre de maneira muito mais intensiva que o aqui simulado, pode repercutir nas tramas tróficas e no funcionamento do ecossistema.

CONCLUSÕES

- Poucos centímetros de diferenças na profundidade podem ocasionar mudanças importantes na granulometria de áreas rasas estuarinas.

- Existe uma variabilidade sazonal na granulometria e teor de matéria orgânica, a qual se viu intensificada em maiores cotas batimétricas, mesmo com pequenas amplitudes de profundidade.
- A variabilidade das taxas de precipitação e as consequentes variações nas descargas fluviais, como principal fonte de sedimentos finos, assim como a ação dos ventos, são os principais fatores temporais determinantes das tendências das propriedades granulométricas no estuário da Lagoa dos Patos. Os resultados desses forçantes implicam em uma maior variação das principais porcentagens de sedimentos a maiores profundidades.
- As caraterísticas do sedimento, relacionadas à hidrodinâmica local, estão associadas à variabilidade espacial da macrofauna, enquanto que as variações dos parâmetros da coluna de água (temperatura e salinidade) atuam na variabilidade sazonal destas assembleias.
- As mudanças nas assembleias macrobentônicas se relacionam mais consistentemente com a variabilidade natural que com a perturbação da pesca, sendo altamente influenciadas pelas caraterísticas do sedimento e a salinidade.
- O impacto do arrasto sobre as assembleias macrobentônicas foi detectado principalmente em zonas com maior predominância de sedimentos mais finos, com respostas diferentes entre os meses e os estratos.
- O arrasto ocasionou decréscimos nas densidades totais da macrofauna, detectados principalmente no estrato superior, e sinais de enterramento. Estas consequências ficaram mais evidentes em épocas com maiores densidades de macrofauna e após o alto impacto de arrasto.
- As maiores densidades do macrobentos apresentaram sobreposição temporal com a safra de camarão, amplificando o impacto ecossistêmico.
- As espécies que apresentaram variações significativas nas suas densidades como consequência do arrasto foram *Monokalliapseudes schubarti, Heteromastus similis, Laeonereis acuta, Erodona mactroides*, *Heleobia australis* e *H. charruana.*
- As densidades de *M. schubarti* diminuíram em até 60% em algumas estações após o arrasto. Foram principalmente afetadas as fêmeas ovigeras, os juvenis e machos, sendo maior o impacto sobre a população desta espécie na área com maior hidrodinâmica.

- A configuração da rede de arrasto e portas utilizadas no presente trabalho diminuiu as densidades de algumas das espécies dominantes da macrofauna estuarina, mas não desestruturou a distribuição vertical das associações analisadas.
- O impacto imposto pelas atividades de arrasto sobre componentes específicos de tramas tróficas estuarinas podem comprometer a função ecológica deste ecossistema.
- Esta tese evidencia alguns impactos ocasionados pela pesca de arrasto sobre assembleias e espécies macrobentônicas em uma área reconhecida como berçário da região estuarina da Lagoa dos Patos. Tais impactos devem ser reconhecidos e extrapolados com cuidado para todo o cenário pesqueiro da Lagoa dos Patos, reforçando que variações nas caraterísticas batimétricas e granulomêtricas.

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APÊNDICE

Apêndice 1.

Esse apêndice refere-se ao artigo intitulado “**Spatial and temporal variations of sediments in a subtropical micro tidal estuary”** em preparação para submissão na revista: Braziliam Journal of Oceanography

Spatial and temporal variations of sediments in a subtropical micro tidal estuary

Running title: Variability of estuarine sediments

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ABSTRACT

Sedimentation processes in transitional coastal zones, such as estuaries, are controlled by the interaction of river, marine and biota processes. This study analyzed the spatial-temporal variations in sediment texture in shallow mudflats of different hydrodynamic conditions of the

Patos Lagoon Estuary (Southern Brazil) and the influence of environmental parameters in these variations between the years 2015-2017. The organic matter content presented a spatial and temporal variation. There were higher values at deeper zones near the navigation channel along winter and spring, mainly of 2015, due to higher precipitation rates. The more protected areas presented smaller grain sizes and their variability was more correlated with wind intensity and direction. In more exposed areas the water depth was more influent in the granulometric variations. The greater seasonality in the granulometric variation was observed in the deeper zones, which presented the smaller grain sizes and lower values of asymmetry, decreasing these values in winter and spring. We concluded that a few centimeters of difference in depth could cause large variations in the sedimentology of shallow estuarine areas. Within these areas, the proximity to the coast favors the occurrence of essentially sandy sediments. From the environmental variables measured, wind, temperature, rainfall and bathymetry are the most influential in the temporal variation of the sediments, which also correlates with seasonal macrofauna variations.

Descriptors: Patos Lagoon, sedimentology, environmental factors, grain size

RESUMO

Os processos de sedimentação em ambientes costeiros transicionais como os estuários estão controlados pela interação de processos fluviais, marinhos e a biota. Este estudo analisou as variações espaço-temporais na textura sedimentar em zonas com diferentes condições hidrodinâmica do estuário da Lagoa dos Patos (Sul do Brasil) e a influência de parâmetros ambientais nestas variações entre os anos 2015-2017. O teor de matéria orgânica apresentou uma variação espacial e temporal. Valores mais elevados nas zonas mais profundas próximas ao canal de navegação ocorreram no inverno e primavera de 2015 devido as maiores precipitações pluviométricas. As zonas mais abrigadas apresentaram menores tamanhos de grão e a sua variabilidade esteve mais correlacionada com a intensidade e direção do vento. Em zonas menos abrigadas a profundidade da lâmina d´agua exerce maior influência nas variações granulométricas. A maior sazonalidade na variação granulométrica foi observada nas zonas mais profundas, as quais apresentaram os menores tamanhos de grão e menores valores de assimetria, diminuindo estes valores no inverno e primavera. Conclui-se que poucos centímetros de diferença na profundidade podem ocasionar grandes variações na sedimentologia de zonas rasas estuarinas. Nestas zonas, a proximidade da costa propicia a ocorrência de sedimentos essencialmente arenosos. Dos parâmetros ambientais medidos, o vento, temperatura, pluviosidade e batimetria são as mais importantes na variação temporal dos sedimentos, a qual também se correlaciona com variações sazonais da macrofauna.

Descritores: Lagoa dos Patos, sedimentologia, fatores ambientais, tamanho de grão

INTRODUCTION

Estuaries occupy a unique position among coastal systems allowing a relatively simplistic view of water inflows and outflows, although they are subject to complex variations in time and space. The degree of fluvial influence (as a result of precipitation in the drainage basin) affects, not only sediment distribution but also controls salinity, nutrients, pollutants and biota distribution (Pino et al., 1999). The estuaries act as filters for the particulate and dissolved materials that originate in the drainage basin and are transported to the sea (Siegle et al., 2009). In saline or fresh waters of an estuary, deposition and erosion are the main mechanisms that control fluctuations in suspended sediment concentration (Wolanski et al., 1996). In this way, estuarine hydrodynamic is a key factor, and it is the result of the interaction of processes that control the inlet morphodynamics (waves, tides, fluvial discharge and sediment supply) and those that act in the mixing of the water in the estuary (Siegle et al., 2009). Sediment dynamics (erosion, resuspension, deposition) are determined by the synergy between the hydrodynamic condition (e.g. stream velocity) and sediment properties (e.g. concentration and grain size) at the water/sediment interface, which present a temporal variability in their measurements (Ha and Park, 2012). The water depth also affects the hydrodynamics, decreasing the hydrodynamic of the bottom with increasing depths and therefore smaller sediment grain sizes may be found (Sedano et al., 2014).

Estuaries on micro tidal coasts have several common characteristics as a result of low tidal currents in relation to wave action and the formation of small inlets (Cooper, 2002). In shallow tidal estuaries, wind is an important factor that influences the resuspension of sediments that can affect the water column in large proportions, thus affecting biological and ecological processes of both the water column and the bottom (Ha and Park, 2012).

Field measurements in intertidal zones also show that the erodibility of cohesive sediments depends on the physical properties of the sediment, such as sediment grain size and apparent density, and on biological variables, such as microphytobenthos' sediment stabilization. Thus, the seasonal cycle of erosion and deposition is reinforced by the biological cycle (van der Wal et al., 2010). The asymmetry of waves in estuaries results in the transport and deposition of muddy sediments (fine sand, silt, clay and organic matter) in mudflats and saltmarshes. The coexistence of mud and sand in these environments is usually explained as the result of their different suspension behaviors and the variability of currents (Pino et al., 1999).

Sedimentological analyzes are useful tools to explain the complex mechanism of estuaries (Kumar Maity and Maiti, 2018). Sediments are typically described by particle size class distribution based on grain size logarithm (Avramidis et al., 2008). In addition to the mean size, the use of measures such as mean, variance and asymmetry highlight important features that may help in the interpretation of depositional environments and sediment transport

(Avramidis et al., 2008; Kumar Maity and Maiti, 2018). The distribution of sediment grain sizes is affected by several factors such as source area, climate type, length and energy of sediment transport in the depositional environments (Kumar Maity and Maiti, 2018). Generally, fine sediments are well selected while coarse ones are poorly selected; negative asymmetries are associated with high energy environments and positive asymmetries with low energy environments (Friedman, 1961).

Sediments of muddy bottoms may vary in space and time as a consequence of bottom erodibility. This erodibility depends on interactions between physical processes, sediment properties and biological processes, particularly the balance between bio-stabilizers and bioturbators (Widdows and Brinsley, 2002). These variations in erodibility and sediment transport processes may play an important role in the seasonal variations of sedimentation and erosion (Andersen et al., 2005). In the Patos Lagoon, the winter and early spring seasons are characterized by strong winds that induce waves that cause erosion in the muddy bottoms (Möller et al., 2001), but also by low temperatures that decrease the abundance of organisms (Rosa and Bemvenuti, 2006). Quantitative changes in the organic matter content and chlorophyll *a* (chl a) in the eroded material suspended in the water column are determined by the velocity of the currents (Widdows et al., 2000).

The aims of the present study are to evaluate the spatial and temporal variations in sediment texture in zones of different hydrodynamic conditions of the Patos Lagoon microtidal estuary and the influence of environmental parameters in these variations.

MATERIAL AND METHODS

Study area

The Patos Lagoon is a chocked lagoon located at southern Brazil (Kjerfve 1986), between Porto Alegre (30º 30'S, 51° 13'W) and Rio Grande cities (32º 12' S, 52° 5'W). The estuarine region had a single communication with sea through a channel of maximum depth of 18m. It is a micro-tidal estuary with mean tidal amplitude of 0.47 m (Fernandes et al., 2005). The main inflow of fresh water at the north region comes from the hydrographic basin of the Guaíba system (the Jacuí, Taquarí, Caí, Sinos and Gravataí Rivers), supplying 86% of its average total freshwater input. Water discharge of this system varies from de 41-22.000 m^3s^{-1}. At central region the lagoon receives the inflow of the Camaquã river, which contribution may reach 5.000 m^3s^{-1} on flood periods (Möller and Castaing, 1999). Finally, in the estuarine region, receives inflow of the São Gonçalo channel which contribution came from the Mirim lagoon (Wallner-Kersanach et al., 2016). This hydrographic basin contributes to the sedimentary dynamics inputting higher values of suspended sediments converting the estuary in a depositional environment. At estuarine region the suspended sediments vary between 4-32mg/ml near bottom, conditioned by flocculation process, with a sedimentation rate of 5-8 mm per year (Tomazelli and Villwock, 2000).

In addition to river discharge, the lagoon hydrodynamic is influenced by wind regime, which regulates the estuarine subtidal circulation in intervals from 3 to 16 days, coinciding with the passage of frontal systems over the area (Fernandes et al., 2002). Northeast winds favor flushing of the lagoon water, generating sediment transport to the ocean. While when the wind comes from the southwest the Ekman transport piles up water close to the mouth, generating inflow currents which transport sediment to inside the estuary (Möller et al., 2001; Antiqueira and Calliari, 2005). The estuarine region has a variable hydrodynamic in function of the meteorological parameters and its morphometry, having in most of the area depth less than 1m, being the deeper zones in relationship with the navigation channels (Souza and Hartmann, 2008).

The study was carryout in the inlet Saco do Arraial, at west margin of the Patos Lagoon Estuary, which is partially protected for events of high energy and currents for the lagoon main channel (Figure 1). This inlet is shallow (1-4m depth), so wind incidence converts it in a highly dynamic environment with respect to the process of erosion, transport and deposition of sediments (Souza and Hartmann, 2008).

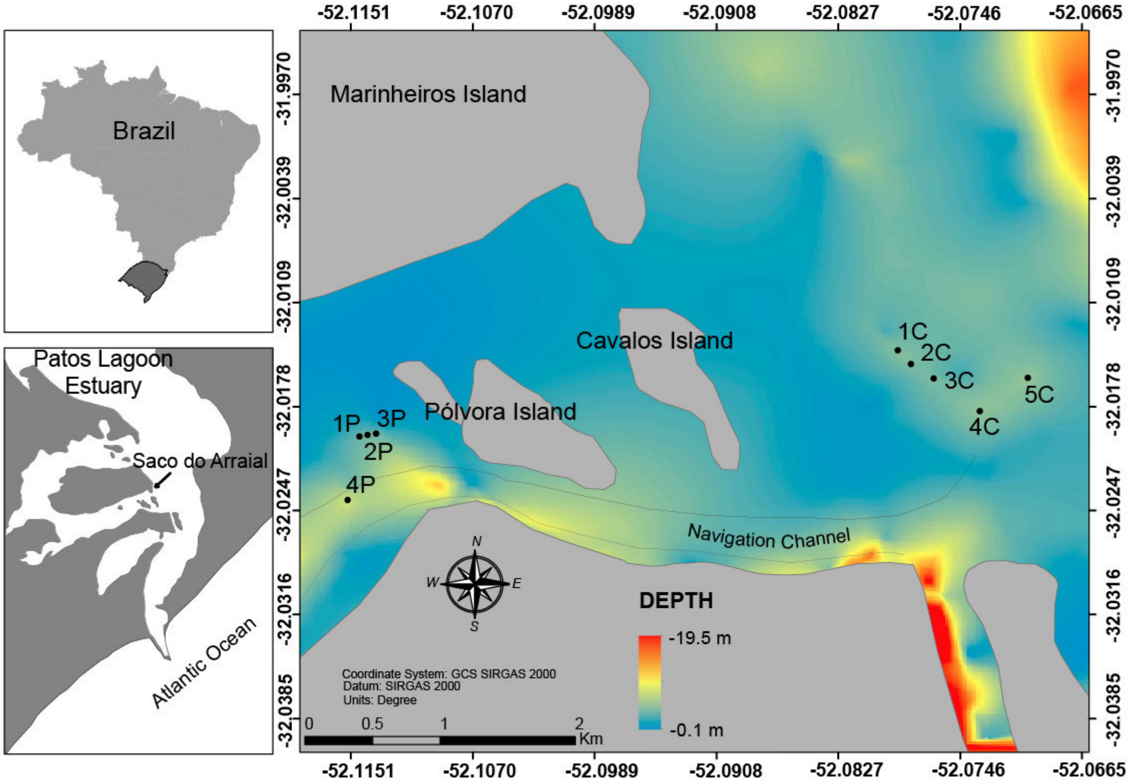

Figure 1. Bathymetric map of the study area with localization of the sampling points. Modified from Ortega et al. (2018)

Sampling

There were selected two areas: the protected zone of the Pólvora island (island west side), and the exposed zone of the Cavalos island (island east side) (Figure 1). In the first area we selected 4 sites (1P to 4P), and five in the second one (1C to 5C) (Figure 1). Depth was measured in each sampling, showing a variation between 0.9 - 4m, considering differences between sites and tidal variations. Even the low difference in depth within sampling sites, due to the study being developed in shallow areas, there was classified as shallow and deeper zones respectively those with depth less than 2,0 m (1P, 2P and 3P, 1C, 2C and 3C) and depth equal or superior to 2,0 m which were near to the navigation channel (4P, 4C and 5C) (Table 1).

Table 1. Characteristics of each sampling site with relationship to depth (minimum, mean and maximum)

Area	**Zone**	**Site**	**Depth**		
			Min	Mean	Max
Pólvora island	Shallow	1P	0.9	1.3	2.0
		2P	0.9	1.3	2.0
		3P	0.9	1.3	2.0
	Deep	4P	1.5	2.7	4.0
Cavalos island	Shallow	1C	1.5	1.7	2.5
		2C	1.5	1.7	2.1
		3C	1.5	1.9	2.1
	Deep	4C	1.9	2.1	2.6
		5C	1.9	2.0	2.6

Samplings were developed between February 2015 and May 2017, collecting between one and three times per season, totalizing 343 sedimentary samples. At each sampled point, two sediment samples were collected with a Van Veen grab and sub-sampled with a core of 10 cm diameter to separate a portion of 10 cm in diameter x 5 cm in depth. In this way two replicates of each stratum were obtained for each site. Samples were refrigerated until processing on laboratory. Granulometric analyses were performed by dry mechanical sieving through a column of sieves of different mesh sizes from 4 mm to 0.063 mm and by pipetting analysis for grain sizes <0.063 mm, using approximately 30 g of sediment. The granulometric procedures followed the Wentworth classification system in intervals of one phi (Suguio, 1973). The grain size composition was expressed as the percentage of the total sample weight. Organic matter content was calculated by differences on weight before and after calcination for 2 hours at 375 °C, utilizing approximately 5 g of sediment.

Is important to highlight that in the whole study period dredging activities associated to channels and port basins which involves high remobilization of sediments to the water column with probable effects at superficial sedimentation were inexistent.

We also measured *in situ* water temperature with a mercury thermometer and salinity with a refractometer. Monthly precipitation data for the Patos Lagoon drainage basin were obtained from the satellite MERRA-2 Model M2TMNXFLX v5.12.4 provided by the Giovanni online data system, developed by the NASA Goddard Earth Sciences Data and Information Services Center (NASA GES DISC). To this we selected a square area limited by the coordinates -54.8437, -32.2998, -50.0537, -28.96 and download the data of total surface precipitation from January/1997 to August/2017, results that can be reproduced following the link https://giovanni.gsfc.nasa.gov/giovanni/#service=ArAvTs&starttime=1997-01-01T00:00:00Z&endtime=2017-09-30T23:59:59Z&shape=gpmLandMask/shp_0&bbox=-54.8437,-32.2998,-50.0537,-28.96&data=M2TMNXFLX_5_12_4_PRECTOT(units%3Dmm%2Fmonth)&variableFacets=dataFieldMeasurement%3APrecipitation%3B&portal=GIOVANNI&format=json. Hourly data of wind speed and direction were obtained from the National Institute of Meteorology (Instituto Nacional de Meteorologia - IMET- 8º Distrito de Meteorologia - Porto Alegre – Brasil, Estação Rio Grande). It was calculated the wind mean speed and the wind direction mode for the 24 and 72 h previous to collect day, since Möller et al. (2001) estimates that cold fronts which drives condition wind speed and direction may last between 3 and 16 days. Daily values of lagoon water level were obtained from the national agency of water (Agencia Nacional das Aguas - ANA -, station Rio Grande - Regatas). We calculated monthly mean of water level for the period 1997-2017. Depth of waves fetch was calculated with the methodology proposed by Hallermeier (1981), using data for the Patos Lagoon from January to July 2015 provided by the program Rede Ondas of the Federal University of Rio Grande (http://www.redeondas.furg.br/index.php/pt/).

Data Analysis

The mean percentage of sand, silt and clay by season were plotted and classified by Shepard diagrams (Shepard, 1954). The statistical parameters were calculated following the equations of Folk and Ward (1957), considering in the statistical analysis the mean, median, selection (standard deviation), asymmetry and kurtosis with the software SYSGRAN (Camargo, 1999).

To characterize the spatial and temporal variation of the areas based on the distribution of grain sizes and the statistical parameters, it was performed a Principal Component Analyses (PCA). To evaluate the significance of the spatial and temporal variations we performed a multivariate analysis of variance (Permanova) with residuals permutations under a reduced model, for an Euclid distance matrix. The model design included the factors: Area (Pólvora island, Cavalos island-fixed), Site (the 9 sampled sites-random, nested in Area), Year (random) and Seasons (random, nested in Year). The analysis was performed to de distribution of grain sizes and for the statistical parameters. All analyses were performed with the software PRIMER V6 + PERMANOVA add-on (Anderson et al. 2008) and R V3.4.1 (R Core Team 2017).

RESULTS

Precipitation

Precipitation presented a fairly large oscillation, varying between 0-300 mm by month. Dryer month were from May to July 2016 and the rainiest those from winter and spring of 2015 and 2016, highlighting October of 2015 as the rainiest month of the studied period (Figure 2). The historical data shows that there were long periods of high precipitation, some of the related to *El Niño* events.

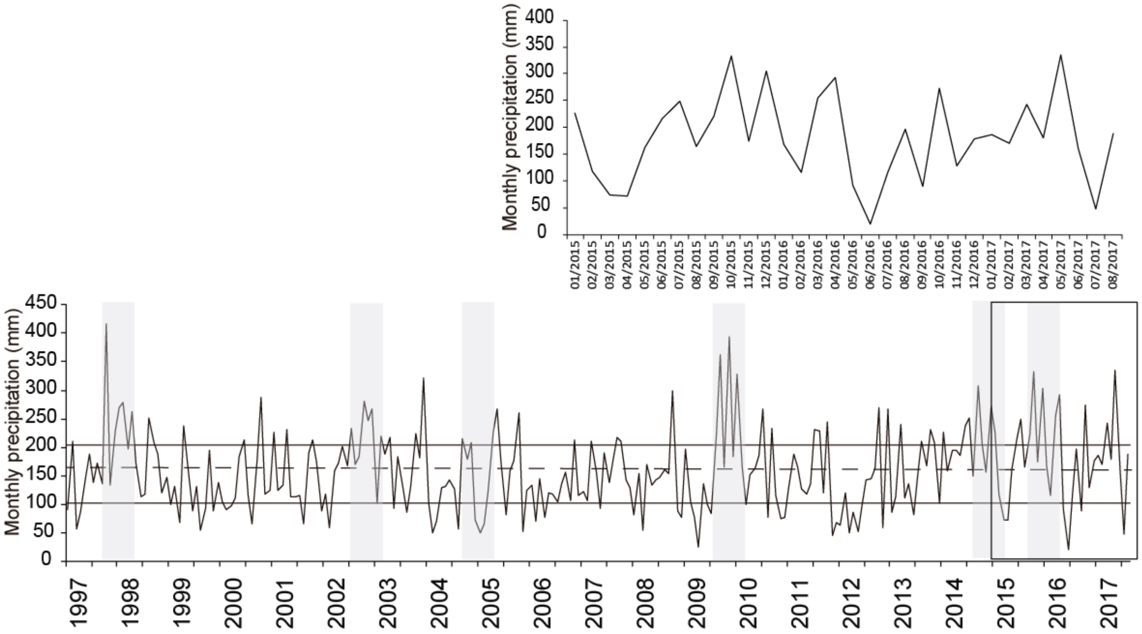

Figure 2. Monthly accumulated precipitation (mm) for the Patos Lagoon dreinage basin in last 20 years with zoom to the studied period. Horizontal continuous lines indicate the first

and third historic quantile. The dashed lines indicate the historic mean value. Gray columns indicate years when *El Niño* events occurred.

Lagoon level

The lagoon level near the mouth had a fairly monthly variation, being the months with higher level those corresponding to winter and spring of 2015 and autumn of 2016 (Figure 3). The periods with higher lagoon levels correspond to those of higher precipitation rates (Figure 2). Observing the complete temporal series, there was a high temporal variation on the lagoon levels with values over the third historic quantile in the years 1997-1998, 2002-2003, 2010 and after 2014. Values under the first historic quantile were observed on the years 1997, 1999-2000, 2004-2007, 2009, 2011, 2012. After 2013 oscillations on lagoon level were over the historic mean with exception of January 2016 when the level decreased to 53 cm (Figure 3).

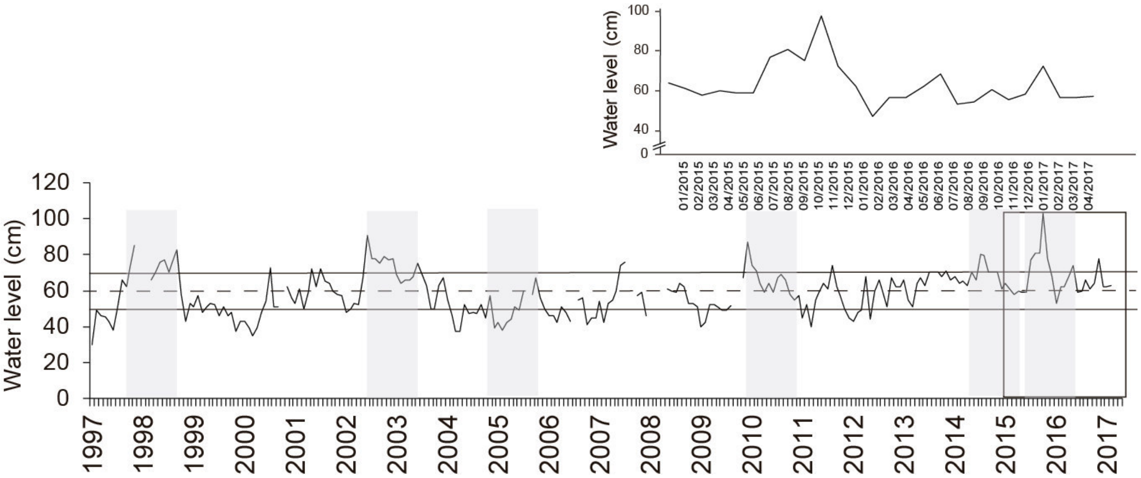

Figure 3. Monthly mean of Patos lagoon water level measured as cotas by the National Agency of water (ANA) at the station Rio Grande-Regatas for the last 20 years with zoom to the studied period. Horizontal continuous lines indicate the first and third historic quantile.

The dashed lines indicate the historic mean value. Gray columns indicate years when *El Niño* events occurred.

Closure depth

The maximum water depth for nearshore erosion by extreme wave condition (dl) was calculated based on the mean wave height (Hs = 0.37 m) and its standard deviation (σ = 0.25 m), giving as result a depth of 3.48 m. The maximum water depth for motion initiation by median wave condition (di) was calculated separately for the shallow zones of Pólvora island and Cavalos island obtaining the same result for both (di = 8.82 m). For the deeper zones of both areas we obtained the same result (di = 12.43 m), which was nearly 50% deeper than for shallow zones.

Organic matter

The organic matter percentage varied between 0 - 10.59%. The higher values were observed in winter and spring of 2015. The highest values were determined in the deeper sites of Cavalos island (4C and 5C) along 2015-2016. In some occasions (winter and spring of 2015, autumn and winter of 2016 and autumn of 2017) the deepest site of Pólvora island (4P) had higher organic matter percentage than the other sites of the area (Figure 4). The variability of organic matter percentage was fairly higher in spring than in the other seasons.

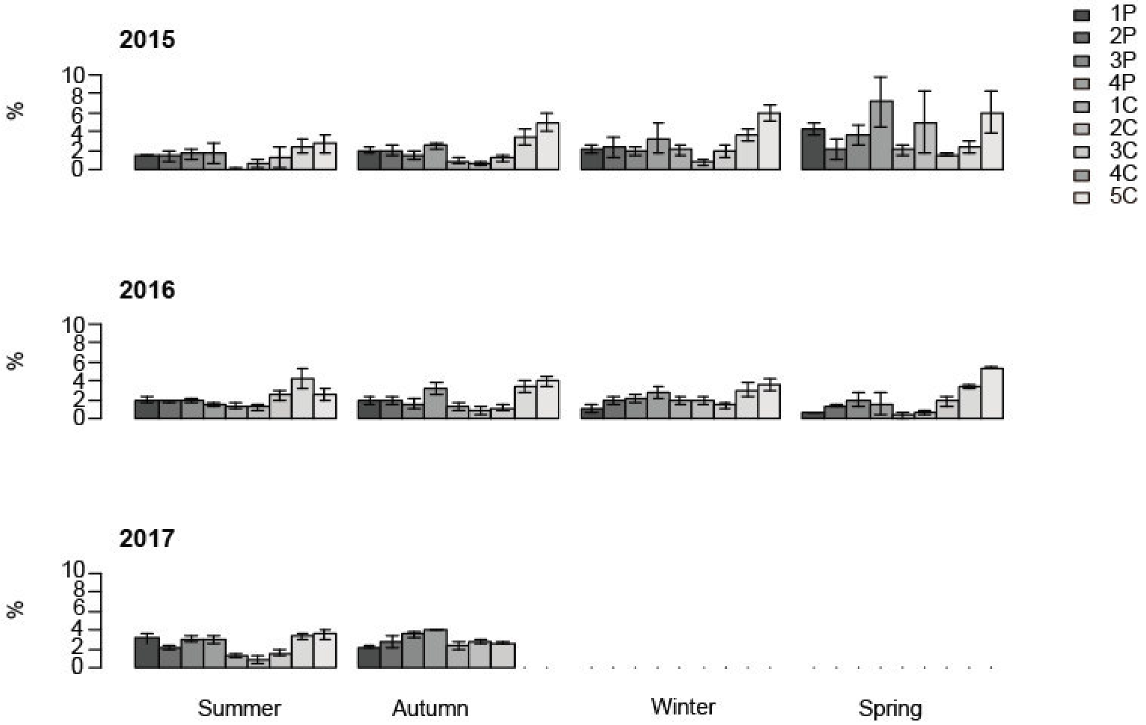

Figure 4. Seasonal mean percentages of sediment organic matter at the nine sampled sites. Vertical bars indicate standard error.

Relationship between grain size and environmental parameters

At the area of Pólvora island (exposed to S-SE winds) the wind direction in the 24 h prior to sampling was the most influent factor in the granulometric variation, correlated negatively with silt percentage and positively with medium and fine sands. This means that winds in the S-SE direction favored silt resuspension and thus increases the relative percentage of sand. In addition, wind speed also has a negative correlation with silt percentage, being that high-speed winds may also favor the resuspension of silt (Table 2). At the area near Cavalos island, wind was not correlated to sediment variability as was water depth. This last factor was positively correlated with silt and clay percentages and negatively with fine to very coarse sands

(Table 2). Temperature was negatively correlated with silt percentage in both areas, indicating that during cold months there is an increase in the relative percentage of silt.

Table 2: Combination of parameters that resulted with significant correlations with sediment grain sizes (p<0.05)

Area	Parameters	Rho	p-value
Pólvora	Silt: Wind direction 24h	-0.230	0.004
	Silt: Wind speed 24h	-0.196	0.015
	Silt: Salinity	0.194	0.016
	Silt: Temperature	-0.238	0.003
	Fine sand: Wind direction 24h	0.213	0.008
	Medium sand: Wind direction 24h	0.268	0.001
	Medium sand: Salinity	0.238	0.003
	Medium sand: Depth	0.362	0.004
	Very coarse sand: Depth	0.344	0.013
	Mean: Wind direction 24h	-0.256	0.001
	Median: Wind direction 24h	-0.248	0.001
Cavalos	Silt: Temperature	-0.208	0.004
	Silt: Depth	0.349	0.001
	Clay: Depth	0.375	<0.001
	Very fine sand: Depth	0.436	<0.001
	Fine sand: Depth	-0.398	<0.001
	Medium sand: Depth	-0.333	0.003
	Coarse sand: Wind direction 24h	0.201	0.006
	Coarse sand: Depth	-0.162	0.026
	Very coarse sand: Wind direction 24h	-0.145	0.046
	Very coarse sand: Wind speed 24h	-0.150	0.040
	Very coarse sand: Temperature	-0.216	0.003
	Mean: Depth	0.370	<0.001
	Median: Depth	0.370	<0.001

Spatial and Temporal distribution of sediment texture

Shallow zones of both areas (Pólvora island and Cavalos island) are composed mainly by sand, independently of seasonal or interannual variation (Figure 5). At site 4P in winter of 2015 the sediment can be classified as silty sand, changing in spring of this same year to clayey sand; on 2016 and 2017 it changed to sandy sediments. At site 4C the samples were classified as sand, but with a higher percentage of silt and clay than in shallower zones, and only in winter of 2015 were classified as silty sand. The site 5C was the most variable and with higher percentages of fine fractions. Over summer and autumn of 2015 sediments of this site were classified as silty sand, on winter changed to silty clayey sand and in spring changed again to silty sand but with a higher percentage of silt and clay than on summer and autumn. On summer and autumn of 2016 site 5C sediments were classified as sand and in winter and spring as silty sand, with higher percentage of silt and clay on spring. This classification remained until summer 2017 (Figure 5).

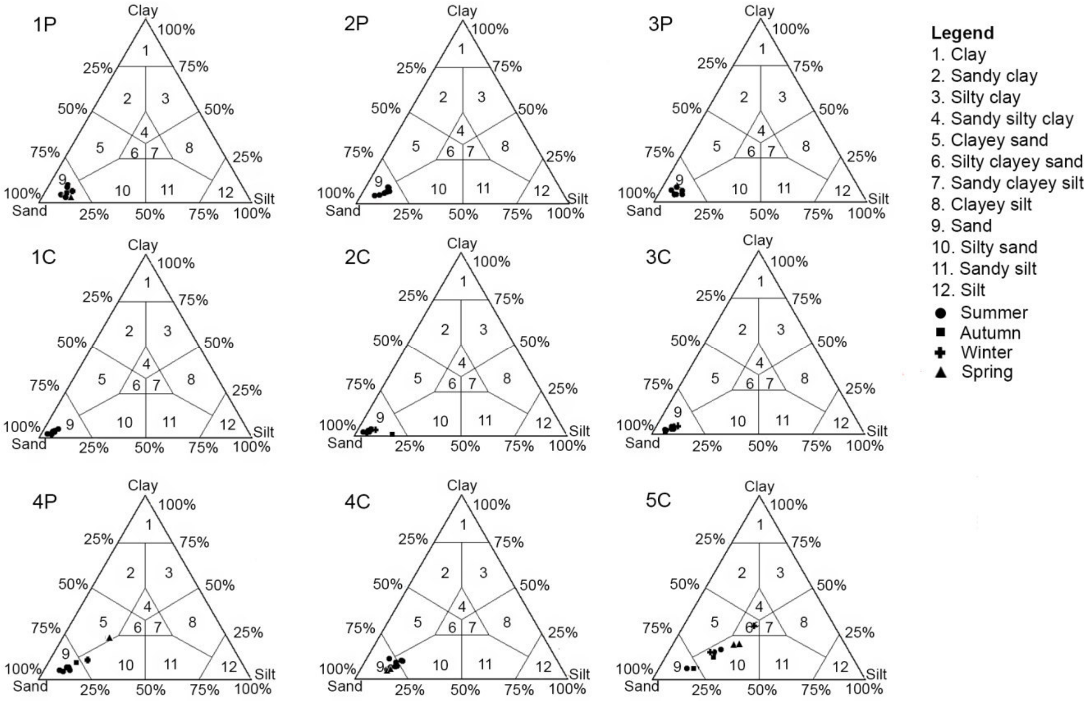

Figure 5. Shepard ternary diagram with seasonal mean values of sampled sediments over the three studied years on the Patos Lagoon Estuary. Sites 1P-3P correspond to shallow zones of Pólvora island, 1C-3C shallow zones of Cavalos island, 4P deeper zone of Pólvora island and 4C-5C deeper zones of Cavalos island.

The main grain sizes in both studied areas were those less than 0,250 mm, thus we analyzed in details the seasonal variations of these sizes for each zone. For the area near Pólvora island there was a decrease of the very fine sediments (very fine sand, silt and clay) with the proximity to the island independently of the season. In this way, it was determined that sediments in Site 1P were composed by approximately 45% of these very fine sediments, the site 2P by 36% and 3P by 33%. The site 4P (deeper and longest from coast) presented a seasonal variation being summer the season with less very fine sediments (~33%) and were increasing up to ~54% at spring (Figure 6A). Similarly, in the area near the Cavalos island there was a decrease of very fine sediments with proximity to the island. The site 1C presented approximately 14% of very fine sediments, site 2C presented 16%, site 3C a 22%, site 4C a 55% and 5C a 71%. In the site 5C was observed a high seasonal variation with a gradual increase of the fine sediments from summer to spring, arising to 83% of the granulometric composition (Figure 6B). Comparing both areas, it was observed that shallow zones presented a higher stability of granulometric composition, being finer in the Pólvora island. In contrast, deeper zones presented a broad seasonal variation (Figure 6).

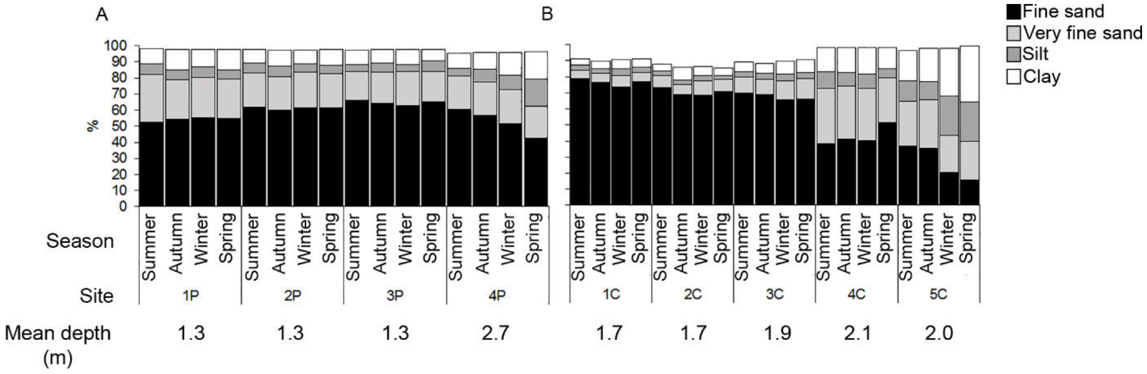

Figure 6. Seasonal variation of the main grain sizes of each site. A) Pólvora island area, B) Cavalos island area.

The principal component analysis (PCA) explained the 70.4% of variation between zones, based on the grain size (Figure 7A). The analysis showed a clear separation between the four studied zones (shallow zones of Pólvora island, shallow zones of Cavalos island, deeper zones of both areas). Samples of each site remained inside its group independently of seasonal variations in almost all cases. Depth is an important factor separating groups in both axis, so as the grain size.

The first axis clearly separates the shallow and deep zones from the Cavalos island area, showing that shallow zones presented a higher content of medium and fine sand, while the deeper zones presented higher values of very fine sediments. The second axis separates the shallow and deep zones of the Pólvora island area, where the deep zones presented high percentages of very coarse and coarse sediments, which were not present in the shallow zones (Figure 7A). This imply that different processes are acting in the Polvora and Cavalos islands areas, so different drivers explain the separation between shallow and deeper zones of each area.

No significant differences were found between the areas of Pólvora island and Cavalos island. However, sites within them were different (Pseudo-F=54.58, P=0.001). Temporarily, there were significant differences between years (Pseudo-F=2.54, P=0.021) and sites within each area varied differently in the between seasons of each year (Pseudo-F=1.62, P=0.005) (Table 3). Pairwise tests showed that differences between sites on the Pólvora island were mainly by the distance from coast and disappear on spring. In the Cavalos island almost all period presented significant differences between all sites, being sites 4 and 5 similar on winter and spring. On spring of 2016 there were not differences in grain sizes between any site of Cavalos island.

Table 3. Permanova results of the variation in grain size structure between areas, sites and temporal scales. * significant values

			Grain size structure		Mean		Median		Sorting		Skewness		Kurtose	
Source	Component	df	Pseudo-F	P(perm)	Pseudo-F	P(perm)	Pseudo-F	P(perm)	Pseudo-F	P(perm)	Pseudo-F	P(perm)	Pseudo-F	P(perm)
Area	Fixed	1	0,318	0,992	0,047	1,000	0,275	0,971	0,050	1,000	3,452	**0,043***	0,090	1,000
Year	Random	2	2,545	**0,021***	9,652	**0,001***	1,770	0,125	10,322	**0,001***	0,631	0,766	2,730	**0,035***
Site(Area)	Random	7	54,587	**0,001***	34,964	**0,001***	9,827	**0,002***	43,940	**0,001***	11,064	**0,001***	13,903	**0,001***
Season(Year)	Random	7	1,369	0,218	0,809	0,600	1,193	0,313	1,871	0,106	1,184	0,323	0,201	0,975
AreaxYear	Random	2	1,144	0,343	2,923	**0,03***	0,340	0,978	6,345	**0,001***	2,528	0,053	1,124	0,368
AreaxSeason(Year)	Random	7	0,689	0,744	0,927	0,475	0,519	0,798	2,015	0,093	0,591	0,745	0,476	0,831
Site(Area)xYear	Random	14	1,332	0,178	1,215	0,313	2,161	0,056	1,021	0,449	1,939	0,056	1,146	0,352
Site(Area)xSeason(Year)	Random	47	1,622	**0,005***	1,611	**0,032***	1,388	0,140	1,445	**0,045***	1,567	**0,048***	1,283	0,141
Res		254												

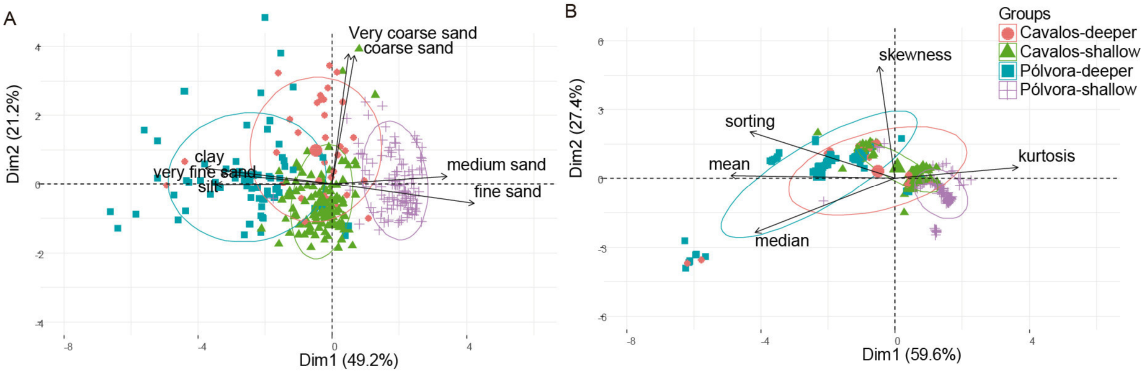

Figure 7. Principal component analysis showing the spatial and temporal variation in sediments between sampled zones. A) based on grain size distribution, B) based on grain-size statistical parameters. Ellipses grouped 70% of points from each centroid.

The principal component analysis based on grain-size statistical parameters (Figure 7B) explained the 87% of variation between zones, being the coefficient with higher value the mean. Basically, the first axis explained the variability (59.6%), clearly separating the shallow zones of the cavalos island area from the deeper zones of this area. This axis does not explain much of the variation of the zones from the Pólvora island area, which remained in the centerof the axis. The axis 2 mainly explain the seasonal variability in the deeper zones of both areas, which is reflected in the more variable distribution of points (Figure 7B). Points out from the ellipses from the deeper zones, located to the left, with higher values of median represent the samples from winter and spring.

There were significant differences in the median parameter between the interaction of seasons per site (Pseudo-F= 2.6618, P=0.001) (Table 3). At Pólvora island area, the deeper point (4P) presented differences between seasons. The summer had significantly lower median values than spring (Figure 8A). At the Cavalos island, sites 1C and the deeper ones (4C and 5C) showed significant differences on winter and spring, with lower values in the second one (Figure 8A). Sorting presented significant differences between sites (Pseudo-F= 6.1752, P=0.002) (Table 3), with less sorting values in the shallower zones of Cavalos island (Figure 8B). The skewness also presented different seasonal variations for each site (Pseudo-F= 2.3924, P=0.006) (Table 3), with significant differences only in the deeper sites of both areas (Figure 8C). The kurtoses did not present significant seasonal differences, only between sites (Pseudo-F= 27.224, P=0.001) (Table 3). In both areas the deeper zones presented the lower kurtoses values (Figure 8D).

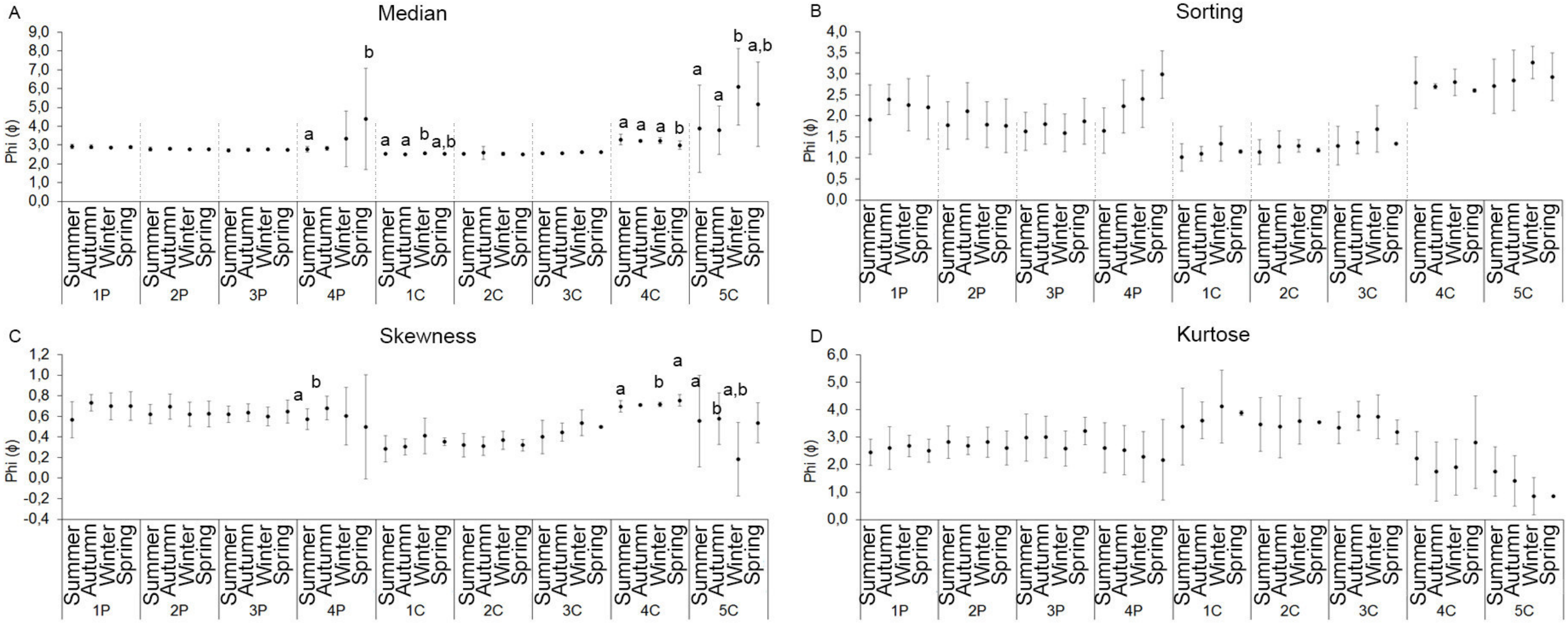

Figure 8. Seasonal variation of grain-size statistical parameters for each site. A) Median, B) Sorting, C) Skewness, D) Kurtose. Vertical bars indicate the standard deviation. Low-case letters indicate significant variations among seasons for each site (P<0.05).

The relationship between grain-size statistical parameters through scatter plots allow the identification of sedimentary environment in relation to its energy level. In this sense, figure 9 shows the relationship between mean and skewness evidencing four different populations. The shallow zones near Cavalos island with larger grain-sizes (lower mean value in the phi scale) and a skewness between 0.3-0.4. The zones near Pólvora island showed a higher value of skewness, which tilts data to the fine fractions, corresponding also to a smaller mean size. The site 4C showed a skewness even higher, being the data more tilt to the fine fractions, with a mean size even smaller. Site 5C and the spring values of site 4P as the fourth group with the lowest mean values and a skewness tilt to fine fractions (Figure 9).

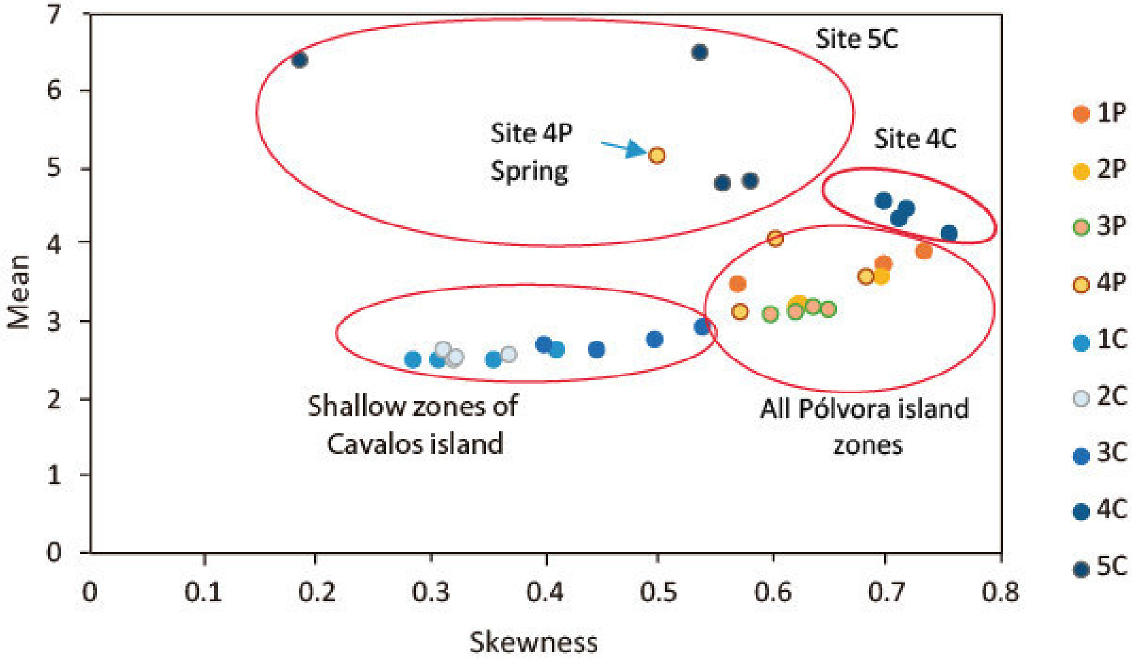

Figure 9. Scatter plot between mean grain size and asymmetry

DISCUSSION

Mudflats are less understood than saltmarshes and sandy beaches because of the cohesive nature of its sediments, which are often very soft (Dyer et al., 2000). Sediments deposited into estuaries are influenced in varying degrees by river discharge, tidal height, wind speed, the concentration of suspended sediments and flocculation, as demonstrated in the Severn (Allen and Duffy, 1998; Manning et al., 2010), Scheldt (Temmerman et al., 2003) and Swan River (Kurup et al., 1998) estuaries.

Strong currents also affect shallow subtidal flats and the action of wind induced waves. This causes sediment remobilization and difficult the deposition of fine material, favoring the predominance of coarser grain sizes (Dyer et al., 2000; Souza and Hartmann, 2008; Santos et al., 2004). The stress caused by wind-induced waves is a function of waves height and water depth. The wave height depends on wind speed and direction, storm duration and water depth in the wave generation area (West and West, 1991).

Möller et al. (2001) stated that wind is the main factor driving the circulation patterns of the Patos Lagoon in periods of low river discharge. The NE winds, predominant along year, form a pressure gradient that favors the seaward flows, while SW winds predominant in winter and spring favors the landward flows (Möller et al., 2001; Fernandes et al., 2002). Simultaneously, exist a seasonal pattern of the fresh water discharge with higher flows in winter and spring and low flows at summer and autumn (Möller et al., 1996). This may explain the higher abundance of organic matter percentages on winter and spring, as the rivers provide large amounts of suspended sediments that will deposit in the calmer zones of the estuary. Coinciding with this, Pino et al. (1999) reported high seasonality in the organic matter percentages and muddy sediments with peaks in winter for the Queule estuary (Chile). Also, in the Arasalar estuary (India) (Bragadeeswaran et al., 2007) and Kemaman estuary (Malasya) (Kamaruzzaman et al., 2002) higher clay content was recorded during the post monsoon (rainy period). Even when water temperature showed high correlation values with the very fine sediments, we believed to be an indirect correlation coinciding with seasonality of winds and river discharge.

The deeper zones, which have a higher percentage of organic matter due to the lower influence of winds and waves causing resuspension of sediments, were the ones that presented the highest seasonal variation in grain size. It was determined that wind induced waves generated in the lagoon may erode until 3.48 m depth. This limit is close to the maximum depth sampled, indicating that in deeper zones just in maximum conditions of winds/waves would be bottom remobilization. In shallower zones, the short period and low amplitude of the wind induced waves are able to cause greater resuspension and to hinder the deposition of the finer sediments.

Considering the historic series of the Patos Lagoon estuary water level, for the last 20 years, it can be observed that after 2013 the depth remained high. Also, from 2014 to 2016 there were long periods of high precipitation rates in all the drainage basin, including two *El Niño* events (one weak and one very strong). *El Niño* and *La Niña* events of the *El Niño* Southern Oscillation (ENSO) in the Pacific cause abundant rainfall and drought, respectively, in southern Brazil, Uruguay and northeastern Argentina (Philander, 1990). However, those high precipitation rates estimated for 2014 to 2016 were lower than in previous strong *El Niño* events but for longer periods. These events in the Patos Lagoon increase the freshwater discharges, decreasing salinity and increasing the fine sediment loads into the estuary

(Fernandes et al. 2002, Barros et al. 2014). This abnormal long period of high lagoon water level may be also favored by the elongation of jetties in the Barra of Rio Grande in 2011, ended in drought period due to *La Niña* event. When extreme drought condition ended, and precipitacion levels rised water levels increased, as modeled by Fernandes et al. (2005) analyzing possible scenarios of changes in the circulation of the estuary as a consequence of the jetties elongation. The model showed that this magnification could lead to an increase in seaward flow and a decrease in landward flows, a decrease in the distance reached by salt water and an increase in the water level inside the estuary.

These scenarios main explain the higher percentage of organic matter when compared with previous studies in nearby areas like Pombas island (Saco do Arraial), where was observed organic matter percentages between 0.2 - 0.8% (Rosa and Bemvenuti, 2004), 0.09 - 3.15% (Rosa and Bemvenuti, 2006) during years without the influence of *El Niño* and 0.74 - 1.22% (Colling et al., 2010) during years with influence of *El Niño*. Also, in the Pombas island was observed a different pattern of increments of organic matter were, instead of increase the fine sediments and organic matter percentages simultaneously with increases of water discharge, these increases were recorded just in the months after higher freshwater discharge and was related to the benthic recovery of post-*El Niño* (Colling et al, 2010). Our results were similar to the patterns observed in the Queule estuary, where in years with high precipitation rates, like those with influence of the *El Niño* events, the fine sediments and organic matter reach maximum peaks (Pino et al., 1999).

In periods of increased water level in the estuarine islands of the Patos Lagoon with a relief less than 0.8 m above the mean water level, become vulnerable to erosion. The wind induced waves from NW-SW and NE-SE directions, which may reach 0.2 m height and a frequency of 5 s stacked on the islands causing erosion (Marangoni and Costa, 2009). In this sense, when NE-SE winds are strong resuspend sediments in areas at exposed side of Cavalos island, erode this island and sediments will deposit in the most protected areas and with less hydrodynamics as are for example the zones of greater depth in the channels and the areas to the west side of the Saco do Arraial islands (Souza and Hartman, 2008). In the Yangtze delta, has being demonstrated that wind (by changes in hydrodynamic and inducing waves) may induce daily to seasonal changes in surface sediment grain size on unvegetated flats which contributes to process of accretion and erosion (Yang et al., 2008).

Sediment texture in both studied areas of the Saco do Arraial indicate a predominance of sandy facies, in spite of the differences between the sites mainly regarding the content of silt and clay. This coincide with reports of Antiqueira and Calliari (2005) and Tomazelli and Villwock (2000), who found that in the shallow areas below the 4 m depth of the Patos Lagoon the sediment is predominantly sandy. Souza et al. (2008) surveyed the granulometry for the Saco do Arraial, determining that the region adjacent to the western margin of the Pólvora island was composed of silty sand, but with sand regions to the north of the island. In our results the sites on the western margin of the Pólvora island are composed mainly of sand in the 4 seasons of the year, indicating a greater resuspension of fine sediments in the studied years than previously reported. The sites in the exposed part of the Cavalos island coincide with that determined by Souza et al. (2008) being sand bottoms. Although both areas are predominantly sandy, it was observed that the exposed areas on the eastern side of the Cavalos island presented larger amounts of coarse fractions while the protected areas on the western margin of the Pólvora island had larger amounts of fine sediments. It should be noted that the coarsest sand fractions found were calcareous remains of bivalve and gastropods shells, abundant in the adjacent region of Cavalos island.

The higher percentage of medium to coarse sand at points close to Cavalos island reflects the hydrodynamics generated by local circulation on the area, where the sites to the east of the island are more exposed to the greater energy of the water associated with the main channel of the lagoon. Souza and Hartmann (2008) recorded that the distribution of the sediments in this area is interspersed with abrupt contacts between fine sand and medium silt and also with the occurrence of biodetritus of the mollusk *Erodona mactroides*.

Within the shallow zone of Pólvora island the bathymetry is more similar and shallower than within the zones of Cavalos island. This opens a gap for the wind to have a greater influence on resuspension, erosion and sediment transport, being therefore one of the factors with higher correlation in granulometry variations. While in the sites of Cavalos island there are more variations on bathymetry, reflecting in less direct influence of wind in the variations of granulometry and a higher correlation with depth. The deeper zones of both areas (Sites 4P, 4C and 5C) were nearby the navigation channels. It has been recognized that these channels are deposition areas (Souza et al., 2008), which explains higher percentages of fine sediments. Antiqueira and Calliari (2005) also reported that exist a direct correlation between depth and fine sediments, coinciding with observations in the present study. In both areas it was observed a decrease of fine fractions according to the proximity of the islands. Similar results were found for the Yang Tse estuary, where sediments tend to become coarser towards the land in wave-dominated environments of intertidal flats (Yang et al. 2008).

In addition to the direct effect of environmental agents, the influence of benthic organisms on the erosion of fine sediments must be considered. Organisms comprising the microphytobenthos like diatoms, and macrofaunal species like polychaetes secrete mucus that can perform a resistance force against erosion processes by increasing the cohesion among grains. Bivalves and amphipods, in contrast, tend to destabilize the sediment in the construction of its galleries (Kornman and De Deckere, 1998; Dyer et al., 2000). In this sense, the establishment of benthic communities may have an important influence on the erosion and sediment transport processes (Kornman and De Deckere, 1998; Widdows et al., 2000; Widdows and Brinsley, 2002). Declining biological productivity along with strong winter storms facilitates erosion, creating a cycle of erosion and deposition (Dyer et al., 2000). It has been reported that on shallow areas of the Patos Lagoon estuary occurs a decrease of macrofauna on winter (Bemvenuti and Netto, 1998; Rosa and Bemvenuti, 2006), which can be extended to spring on high precipitation periods with consequent decrease on salinities (Colling et al., 2007). The years 2015-2017 were years of high rainfall mainly in the spring, causing decline of the macrofauna, which could facilitate together with the strong winds the erosion of the sediment of the shallow sites and their deposition in deeper zones.

We concluded that a few centimeters of difference in depth may cause large variability in the sedimentology of shallow estuarine areas. There is a temporal variation in sediment texture and organic matter content in the shallow areas of the Patos Lagoon, intensified with increasing depths, even in small bathymetric differences. Within estuarine shallow areas, the proximity to the island margins imply in the predominance of coarser sediments. The estuarine shallow areas, which are protected from the higher hydrodynamics of deeper channels, have higher content of very fine sand. The variability of rainfall rates as a main source of fine sediments through freshwater discharge, and the action of winds, are the main temporal factors determining trends of the granulometric properties in the Patos

Lagoon estuary. The results of these main forcing factors imply in a more variety of granulometric profiles in deeper depths, following atmospheric and hydrographic trends.

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Apêndice 2.

Esse apêndice refere-se ao artigo intitulado “**Temporal and spatial distribution of macrobenthic assemblages in areas of an estuarine shallow mudflat with different hydrodynamic conditions”** em preparação para submissão na revista: Journal of the Marine Biological Association

Temporal and spatial distribution of macrobenthic assemblages in areas of an estuarine shallow mudflat with different hydrodynamic conditions

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Abstract

The sediment and macrofauna distribution in estuarine environments are modulated by variations in environmental parameters as winds, river discharges, and suspended sediment inputs, driving changes on hydrodynamic effects. The aim of this study was to evaluate the responses of macrofauna (abundance, richness, diversity, and assemblage structure) to variations on environmental parameters in two areas of the same shallow mudflat with different hydrodynamic conditions within small-scale distances on the Patos Lagoon Estuary. The assemblages were different between the two areas but presented a similar temporal trend with higher densities at autumn related with increases of salinity. The macrofauna presented higher densities, species richness and diversity in the superficial stratum (0-5 cm). The site with presumed higher hydrodynamic condition due to the proximity to main estuarine channel circulation was constituted mainly by medium sands and had a higher diversity and abundance of organisms. The lowest abundances were found in the most protected site from the main channel circulation, which have a high percentage of fine sediments without relevant temporal variations. The tanaid *Monokalliapseudes schubarti* was the most abundant species in the studied mudflat, with higher abundances on deeper zones. There was a clear zonation in the scale of areas for the polychaetes and the bivalve *Erodona mactroides*. The polychaetes densities were higher in the protected area relative to the main channel circulation while the bivalve presented higher abundances in the exposed area. Spatial variations in the macrofauna were related to sediment characteristics while temporal trends were related to water properties. This study highlights the importance of local hydrodynamic and small-scale variations on sediment properties and depth in the composition and abundance of the estuarine macrobenthos.

Key words: Benthos, Estuaries, Biodiversity, Patos Lagoon, macrofauna environmental relationships

INTRODUCTION

Estuaries are among the most productive ecosystems, mainly due to the presence of nutrients and multiple food resources for the trophic web, as a result of autoctonus, riverine or marine inputs, which are produced by the planktonic and benthic compartments (Malet et al. 2008, Ubertini et al. 2012). These ecosystems support important ecological functions and services being highly explored by humans, which compromise its integrity (Alves et al. 2015). Estuarine communities have to cope with extremely variable physical and chemical parameters that define these ecosystems as highly dynamic environments. The understanding of the relationships between species distribution and environmental characteristics is a major goal in the search of forces driving species distribution and biological parameters (Mola & Abdel Gawad 2014, Alves et al. 2015). The hydrodynamic of an estuary is a result of complex processes related to tidal features, estuarine morphology, freshwater discharge, wind velocity and direction, waves and depth (Siegle et al. 2009, Xia et al. 2010). The sediment dynamics depends on water circulation, salinity, biological interaction and granulometric size (Wang & Adutta 2013). The water currents usually transport silt and clay while sands are usually transported along the bottom by the processes of saltation, rolling, and sliding (Wang & Adutta 2013). Although worldwide estuaries have similar driving forces, they differ in their patterns of river runoff, tides, wind and coastal geology, which affect the physicochemical properties of the sediments and the water column (Giménez et al. 2014). Therefore, it is important to identify particular spatial and temporal patterns of estuarine benthic assemblages, which remains unaddressed in some estuaries, because functional and taxonomic diversity are prime candidates for scale dependence (Barros et al. 2012, Barnes & Hamylton 2015).

Macrobenthic organisms contribute in many important ecological functions, such as decomposition, nutrient recycling, and as food web components, acting both as consumers and prey (Mola & Abdel Gawad 2014, Ysebaert & Herman 2002, Piló et al. 2015). They also have an important role in mediating physical and chemical processes in the sediment-water interface and in interstitial water, including the degradation of organic matter (Barros et al. 2008).

The interaction between macrobenthic assemblages and sedimentary properties are essential for the description of seabed systems. Studies of benthic assemblages are of great interest for analyzing the structure and evolution of coastal systems (Dauvin et al. 2017), due to the lack of homogeneous distribution in space, driven by many different ecological processes (Barros et al. 2008). The distribution of macrofauna is highly dependent to the nature of sediment, grain size distribution (Giménez et al. 2014, Vanaverbeke et al. 2011), morphodynamic (Carcedo et al. 2015), and bottom topography (Colling et al. 2007). However, organic matter content, food availability, biological and trophic relations may also influence the macrofauna distribution (Dauvin et al. 2017).

The identification of the relevant scales of variability in space and time is a prerequisite to understanding factors and processes generating patterns in biotic and abiotic components of ecosystems, especially in estuaries, which are characterized by a high degree of spatial and temporal variability (Morrisey et al., 1992; Ysebaert & Herman 2002). In this sense, it has been observed spatial distribution of estuarine macrobenthic species through upstream-downstream areas, responding mainly to the salinity influence (Zajac & Whitlatch, 1982; Chainho et al. 2006; Blanchet et al., 2014). The zonation of benthic species from the supralittoral to deeper zones of estuaries are well recognized as well, where assemblages vary according to the features of substrate and depth. The sediment in shallower zones is composed mainly by sand and the mud percentage increase with depth, switching the composition from the dominance of superficial epifauna as some bivalves in shallower zones to the dominance of infaunal organisms in deeper zones (Capítoli et al. 1978; Logan et al. 1984).

Benthic assemblages play an essential role of the bottom stability, reworking the sediments through the bioturbation (e.g. amphipods and bivalves) or bio-stabilization (e.g. polychaetas) (Dyer et al. 2000, Widdows & Brinsley 2002). Both sediment properties and faunal distribution may also vary according to hydrodynamic forces (circulation, tides, waves), input of suspended sediments through drainage basin (Dauvin et al. 2017).

There are increasing worldwide impacts on marine and estuarine environments, which arises a need for monitoring, assessment and management of ecological integrity, to promote an ecological sustainability of this ecosystems (Veríssimo et al. 2012). Macrobenthic assemblages are frequently mentioned as good indicators of environmental quality and are included in impact analyses, but the establishment of causal relationships between anthropogenic stressors and its effects on the structure of benthic assemblages is difficult (Veríssimo et al. 2012, Barros et al. 2012). The habitat preferences of macrobenthic organisms, their life history traits, their limited mobility and relatively long life-spans, make them highly suitable to assess environmental conditions (Veríssimo et al. 2012, Piló et al. 2015). Moreover, due to their high taxonomic diversity, macrobenthic assemblages comprise species that show different tolerances to stress, being therefore sensitive indicators of change (Veríssimo et al. 2012, Piló et al. 2015). Nevertheless, estuaries present high complexity with strong natural gradients that may confound the evaluation of anthropogenic stressors (Barros et al. 2012).

The macrobenthic ecology through the salinity and depth gradient on the Patos Lagoon Estuary was investigated by Capitoli *et al.* (1978), and the distribution and seasonal patterns of macrofauna by Bemvenuti *et al.* (1978), Bemvenuti & Netto (1998), Capítoli & Bemvenuti (2006), Rosa & Bemvenuti (2006, 2007) and Colling et al. (2007, 2010). However, no research has been carried out regarding the seasonal and interannual responses of benthic macrofauna concerning possible differences according to hydrodynamic conditions in small-scale (few kilometers) distances. In this manner, we evaluated the responses of macrofauna (abundance, richness, diversity, and assemblage structure) to variations on environmental parameters in two areas of a same shallow mudflat with different hydrodynamic conditions within small-scale distances on this subtropical estuary. We hypothesize that the dominant macrobenthic species in a same estuarine shallow mudflat may vary as a response of different levels of exposure to hydrodynamic effects on bottom granulometry. In addition, we hypothesize that even different assemblage estructures will present similar temporal responses and trends in its abundances, evidencing that seasonal and interannual variations on macrobenthos mainly depends of the seasonality of estuarine salinity. Independently of the spatial and temporal variations, we expect a maintenance of the vertical stratification of macrobenthic properties like higher densities, species richness, diversity and equitatibility on the superficial strata.

MATERIALS AND METHODS

Description of study area

The Patos Lagoon is a choked lagoon located in southern Brazil (Kjerfve 1986) (Fig. 1). Its estuarine region presents a unique communication to the sea through an 18 m depth channel, receiving freshwater inputs from the Guaiba system (Jacuí, Taquarí, Caí, Sinos and Gravataí rivers) and the Camaquã river with a discharge between 41 m^3s^{-1} to 22,000 m^3s^{-1} and the São Gonçalo channel which contribution came from the Mirim lagoon (Wallner-Kersanach et al. 2016). Those tributaries have a great contribution on suspended sediments, varying near bottom between 4-32 $mg.l^{-1}$ with a sedimentation rate of 5-8 mm per year, making the estuary a deposition environment (Tomazelli & Villwock 2000). In addition to the rivers input, the lagoon hydrodynamic is influenced by wind regime, regulating the subtidal circulation (Möller et al. 2001, Fernandes et al. 2002). Estuarine discharge is influenced by the NE-E winds predominant among year and the S-SW which increase in frequency and intensity on winter and spring creating currents that transport sediments to estuary interior (Möller et al. 2001).

The study was carried out in a shallow inlet called “Saco do Arraial” with depth ranging from 1 to 4 m, this the maximum characterizing a navigation channel (Fig. 1). Even that this inlet is relatively protected from events of high energy from the main lagoon, the incidence of strong winds and the low depth cause a highly dynamic environment with respect to the erosion, transport and deposition of sediments (Colling et al. 2007; Souza & Hartmann 2008).

Experimental design

Two areas were chosen: the west side of the Pólvora island (more protected area concerning the hydrodynamic of the lagoon main channel circulation) -Area 1; and the east side of Cavalos island (exposed to the hydrodynamic of the lagoon main channel circulation)- Area 2 (Fig. 1). We defined five sites distributed on shallow mudflats between both areas (Fig. 1).

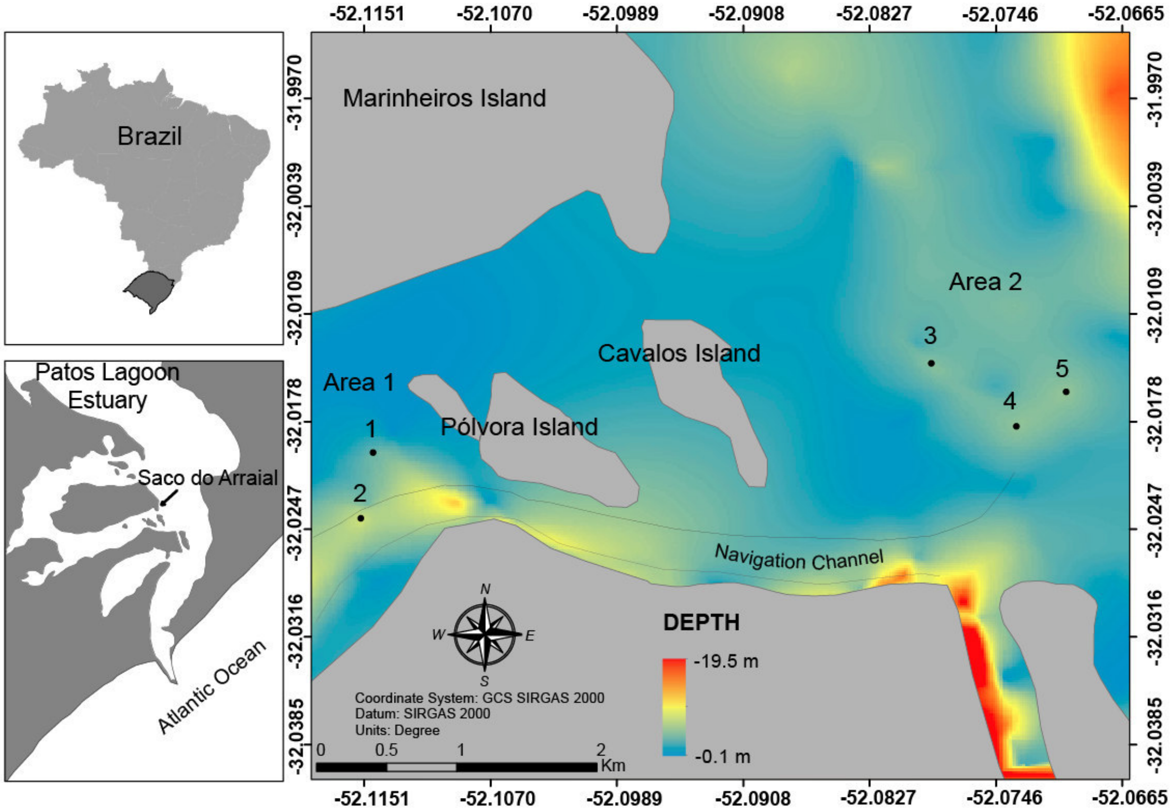

Figure 1. Map of the studied area with bathymetry and location of sampling sites. Adapted from Ortega et al. (2018)

Sampling

Sampling were performed between February 2015 and May 2017 concerning a seasonal approach through three years. There were two sampling in summer, three in autumn, three in winter and one in spring of 2015; three sampling in summer, autumn, winter and one in spring of 2016; two in summer and one in autumn of 2017. In each sampling, three replicates of sediment for macrofauna analysis and two for granulometric and organic matter analyses were collected with a *van Veen* grab. Each grab sample was subsampled with a core of 10 cm diameter (0.0078 m^2) and sectioned into 0-5 cm and 5-10 cm depth strata. The benthic macrofauna was sieved with a 300 µm mesh net and preserved in 4% formaldehyde and identified to the lower taxonomic level possible. The water temperature was measured with a mercury thermometer (°C) and salinity with a refractometer on each sampling date. Daily salinity measures of the three sampled years were obtained from the Brazilian Long Term Ecological Research (PELD) project in the Pombas island (Saco do Arraial) a few meters west to the sampling points. This data was averaged by 15 days. Depth was measured in each sampling event with an ecobathymeter. Granulometric analyses were performed by dry mechanical sieving through a column of sieves of different mesh sizes from 4 mm to 0.063 mm. The granulometric procedures followed the Wentworth classification system in intervals of one phi (Suguio 1973). The grain size composition was expressed as the percentage of the total weight. Grain sizes finer than 0.125 mm (very fine sand, silt and clay) were named as fine sediments, and those coarser than 0.500 mm (very coarse sand, granule and pebble) as coarser sediments. Organic matter content was calculated by differences on weight between before and after calcination for 2 hours at 375 °C.

Statistical analysis

In order to characterize the sampling sites based on environmental variables, a Principal Component Analysis (PCA) was performed using the variables: depth, organic matter percentage and all grain sizes percentages.

Univariated measures were calculated: total abundance (N), number of species (S). Shannon-Wiener's diversity index (H') and Pielou evenness index (J'). The frequency of occurrence was calculated by the presence of individuals in the total number of samples collected in each site.

Spatiotemporal variations in the fauna were analyzed with a Permutational Multivariate Analysis of Variance (Permanova, Anderson et al. 2008) through the following model: Area (fixed, 2 levels), Site (random, nested in Area, 5 levels), Year (random, three levels), Season (random, nested in Year, four levels), Stratum (fixed, two levels). For the univariate macrofaunal data (total densities, species richness, diversity and evenness), a Permanova with 4999 permutations of raw data units was used to analyze the possible variability among factors (Anderson 2005). Permanova was performed using a Euclidian distance matrix. Pairwise tests also used 4999 random permutations to obtain P-values through Monte Carlo correction. For multivariate data (assemblage structure), the Permanova (with 999 permutations of residuals under a reduced model) was based on the Bray-Curtis similarity matrix constructed from the square-root-transformed abundance data (ind.m^{-2}). Transformation was used to down-weigh the importance of the dominant species. Dummy values of one were added to reduce the effect of the absence of individuals in some samples (Clarke et al. 2006). Pairwise tests were run for those factors that resulted significant (p-value <0.05), using p-values under the Monte Carlo correction.

To visualize the patterns in the spatial distributions of macrobenthic assemblages, Principal Coordinate Analysis (PCoA) were performed from the same Bray-Curtis similarity matrix used for the PERMANOVA. The relative contribution of each taxon to the dissimilarity between areas and sites was assessed with the Similarity Percentages (SIMPER) analysis. To evaluate possible relations between the most frequent macrofaunal species (FO >10% in all Sites) and environmental variables Spearman correlations were performed.

RESULTS

Environmental variables

Temperature ranged between 9 and 28°C, with mean values of 20-23°C in spring and summer, and mean values of 14-17 °C in autumn and winter (Fig. 2A). Field measured salinity ranged between 0-36, being the higher values in autumn (21±10) (mean ± standard deviation) and the lower values at spring (0±0). Through daily salinity values was observed that during entire 2015 the salinity was under 5, except for autumn when higher values were recorded (Fig. 2B). In 2016, there was a marked salinity oscillation, with lower salinities recorded during all year and rising values above 5 recorded from late summer to early autumn (Fig. 2C). In 2017, salinities were higher when compared to previous years, with a fortnightly mean above 15 during autumn (Fig. 2D).

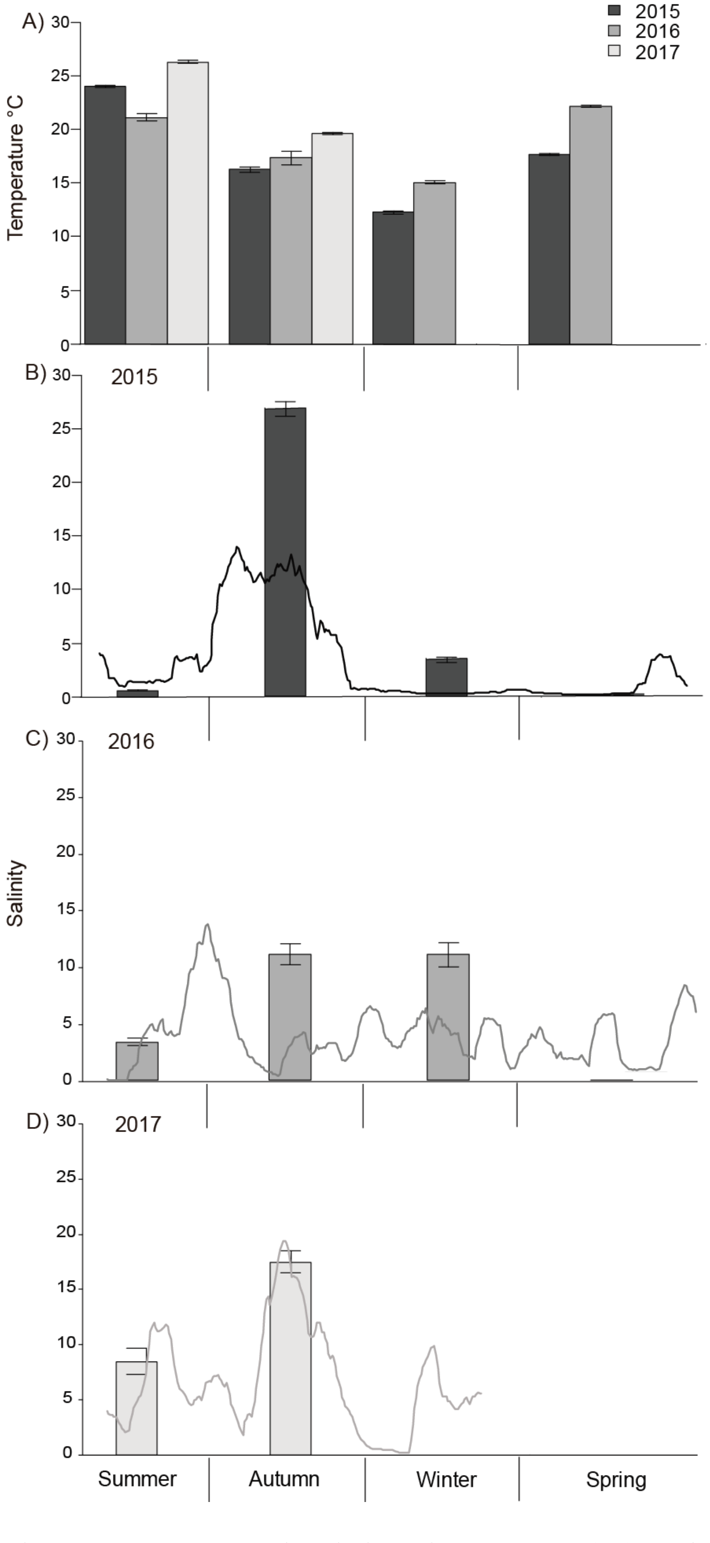

Figure 2. Mean seasonal variation of temperature (°C) on the three sampled years (A), and salinity in the 2015 (B), 2016 (C) and 2017 (D) years. Vertical bars on temperature and salinity columns indicate standard error. Lines in the salinity charts indicate mean fortnightly values, while the columns indicate the mean seasonal value of the *in situ* measures

Mean depth was of 1.3±0.3 m on Site 1 and 2.5±0.9 m on Site 2 from Area 1; 1.7±0.3 m on Site 3; 2.0±0.1 on Site 4 and 1.9±0.4 m on Site 5 from Area 2. There were not significant differences in granulometric composition among strata. The most abundant granulometric fraction in almost all zones was the fine sediments. The Area 1 was represented by 39.8±7.7% of fine sediments on Site 1 and 39.7±9.6% on Site 2; while in the Area 2 there were 13.9±2.9% on Site 3; 59.0±5.7% on Site 4 and 61.4±14.7% on Site 5. The highest values of this grain sizes in Area 1 were found in spring of 2015 in Site 2, while in Area 2 were in winter and spring of 2015, and spring of 2016 in Site 5, reaching more than 70% of sediment composition. The highest variability on fine sediments were observed on Sites 2 and 5 (Fig. 3). Is important to note that higher percentages of coarse sediments on Site 2 were due to biodetritic fragments of shells, mainly of dead *Erodona mactroides* and *Heleobia* spp. Organic matter percentage varied between 0.25-7.81%, with a seasonal variation where highest values were determined on winter and spring. No significant differences of the organic matter percentages were found between strata.

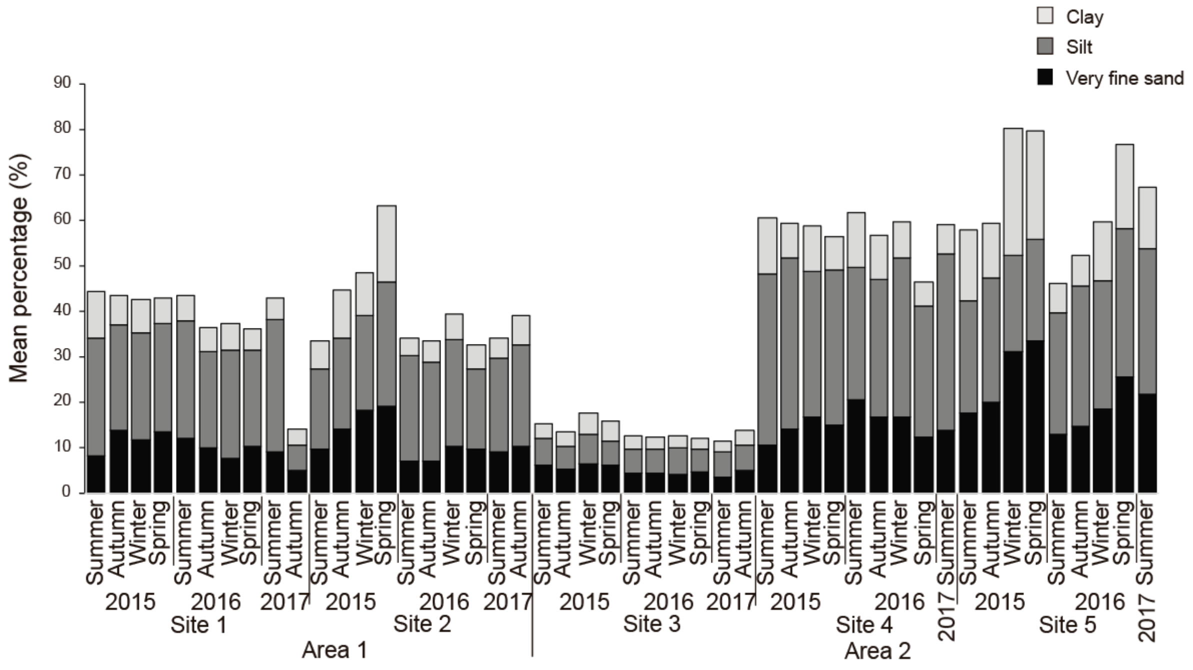

Figure 3. Seasonal variation of fine sediments mean percentages in all sampled sites

The Principal Component Analysis (PCA) represented 72.6% of sites variation, showing that Site 3 differs from the others due a higher percentage of medium and fine sands (Fig. 4). The variation coefficient that influenced more on the first axis were the percentage of fine sediments (-0.54) and the percentage of fine sands (0.53), while in the second axis was the coarse sediment (-0.64). Site 1 had the lower percentage of coarse sediments and lower depths. The sites nearest to the navigation channel (Site 2 and 5) are those with higher variability, where Site 2 had higher percentages of coarse sediments and Site 5 the higher percentages of fine sediments and organic matter content. Deeper depths were recorded at Site 2 (Fig. 4). Larger distribution of points in the PCA was observed on Sites 2 and 5, indicating a higher variability on grain size composition among samples, while in Sites 1, 3 and 4 the grouping of points represents a more stable composition of sediments. The clear separation by the first axis among sites from the Area 2, and the separation of sites from Area 1 given by the second axis indicate that different processes are acting in both areas causing variability between shallower and deeper sites.

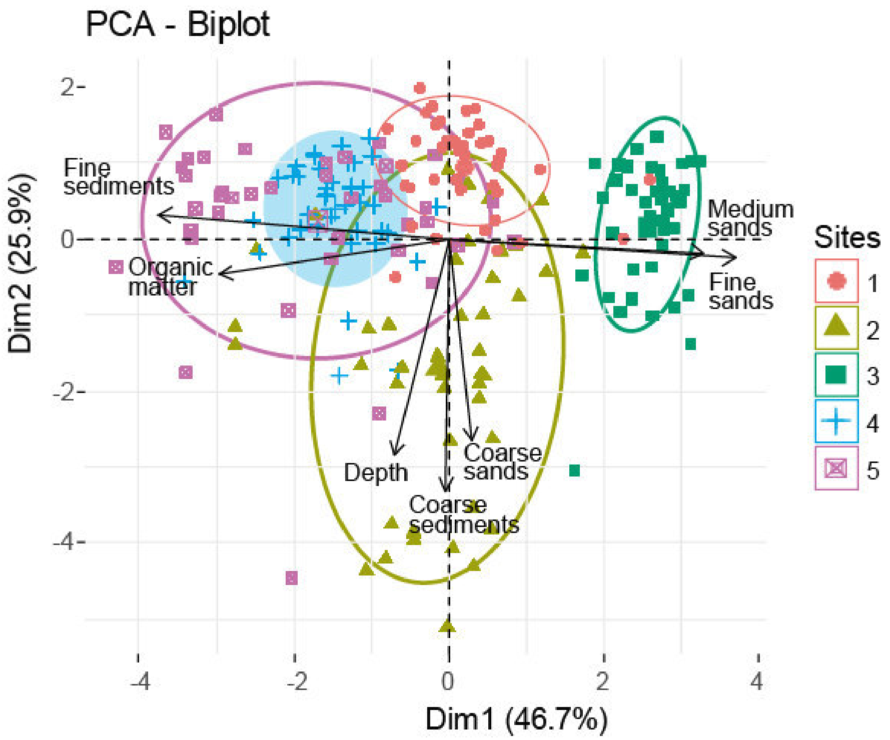

Figure 4 Principal component analysis(PCA) showing the variation of sites according to environmental variables

Macrobenthos composition and structure

We quantified 34086 individuals, belonging to nine taxonomic groups: Amphipoda, Cumacea, Isopoda, Mysidacea, Tanaidacea, Decapoda, Bivalvia, Gastropoda, Polychaeta and Nemertea. Within this groups, 12 genera and 10 species were identified, being the most abundant and frequent the tanaid *Monokalliapseudes schubarti* (Table I).

Table I. Species found per site with their mean density (ind.m^{-2}), standard deviation and occurrence frequency for both studied areas

		Area	**1**				**2**					
		Site	**1**		**2**		**3**		**4**		**5**	
Group	**Specie**	**Epifaunal/ Infaunal**	**Mean± Standard deviation**	**FO (%)**	**Mean± Standard deviation**	**FO (%)**	**Mean± Standard deviation**	**FO (%)**	**Mean± Standard deviation**	**FO (%)**	**Mean± Standard deviation**	**FO (%)**
Amphipoda	**Gammaridea**	Epifaunal	-	0.0	127.3±0	1.5	-	0.0	-	0.0	-	0
Bivalvia	***Erodona mactroides***	Infaunal	502.2±749.4	14.0	789.9±1364	40.8	1743.5±4668	65.1	1727.3±2194	43.1	2316.4±3858	43.1
Cumacea	***Diastylis sympterygiae***	Infaunal	-	0.0	127.3±0	0.8	233.4±253	9.3	-	0.0	127.3±0	0.8
Decapoda	**Panopeidae**	Epifaunal	-	0.0	-	0.0	127.3±0	0.8	-	0.0	-	0
	***Pinnixa* sp.**	Epifaunal	-	0.0	-	0.0	-	0.0	-	0.0	127.3±0	2.4
Gastropoda	***Heleobia australis***	Epifaunal	382.0±507.3	13.2	780.7±1148	28.5	1374.7±1851	47.3	1400.6±1595	28.5	749.8±738	22.0
	Heleobia charruana	Epifaunal	127.3±0	0.8	356.5±188	3.8	2437.3±2528	5.4	1364.2±1972	5.7	551.7±374	4.9
	***Heleobia* sp.**	Epifaunal	891.3±720.2	1.6	-	0.0	148.5±52	4.7	594.2±482	2.4	-	0.0
Isopoda	***Kupellonura* sp.**	Epifaunal	254.6±0	0.8	226.4±199	6.9	264.8±164	58.1	316.7±225	61.0	232.2±188	41.5
	Uromunna peterseni	Epifaunal	201.6±220.3	9.3	165.5±61	7.7	145.5±46	10.9	127.3±0	4.1	127.3±0	0.8
Mysidacea	***Mysidopsis* sp.**	Infaunal	127.3±0	0.8			127.3±0	0.8				
Polychaeta	***Heteromastus similis***	Infaunal	1264.2±1513.6	85.3	2065.0±2312	90.8	575.4±466	80.6	370.4±393	69.1	422.6±405	74.0
	Laeonereis acuta	Infaunal	327.7±325.9	41.9	254.6±245	17.7	159.2±63	3.1	-	0.0	127.3±0	0.8
	Nephtys fluviatilis	Infaunal	732.5±613.4	65.1	866.3±760	73.1	514.0±687	62.0	560.2±657	59.3	376.6±307	56.1
	Polychaeta n.i	Infaunal	127.3±0	1.6	-	0.0	-	0.0	-	0.0	-	0.0
Tanaidacea	***Monokalliapseudes schubarti***	Infaunal	1122.6±1537.6	78.3	2220.5±2638	90.0	2846.7±3313	97.7	3271.2±3684	99.2	2511.5±3531	98.4
	Sinelobus stanfordi	Epifaunal	127.3±0	2.3	169.8±73	2.3	152.8±56	3.9	127.3±0	2.4	254.6±254	3.3
Nemertea			127.3±0	3.9	127.3±0	3.8	-	0.0	-	0.0	-	0
N.I.			-	0.0	127.3±0	0.8	127.3±0	0.8	-	0.0	-	0

The mean density of macrofauna was 1474.2±2538 ind.m^{-2}, being lower at Area 1 (1219.4±1798 ind.m^{-2}) than at Area 2 (1618.8±2851 ind.m^{-2}). The vertical distribution was significantly different (Pseudo-F=13.673, P=0.001) independently of the area or season, with higher densities on the Stratum 0-5. There was a clear seasonality (Pseudo-F= 8.1922, P=0.0008) with higher densities in autumn in both areas. Total densities varied differently between sites and seasons (Pseudo-F= 2.2243, P=0.0062), with a more marked seasonality at deepest sites (Site 2, Area 1; Site 4 and 5, Area 2) than at shallowest ones (Site 1, Area 1; Site 3, Area 2) (Fig. 5).

Species richness was similar between areas, but differ among sites (Pseudo-F= 8.3451, P= 0.0014). Site 1 showed significantly less richness than Site 2 (P=0.0154), while within Area 2 just the Site 3 was different from Site 5 (P=0.0308). The vertical stratification was significantly different (Pseudo-F= 27.773, P= 0.0004), with higher values recorded on Stratum 0-5. There was a different seasonal variation in the strata (Pseudo-F= 2.9032, P=0.0458), with no detected differences in the Stratum 0-5 among seasons. In contrast, in the Stratum 5-10, the richness in autum was significantly higher than in summer (P=0.023) and spring (P=0.006), plus the richness in spring was significantly lower than in winter (P=0.013). There was also a marked seasonal variability in some zones (Pseudo-F= 1.7508, P= 0.0248). In Site 1 the species richness in the summer of the three years was lower than other seasons, and in Site 2 the lowest richness was found in spring of 2016. The highest richness was found in Site 3, mainly in summer, autumn and spring of 2015 (Fig. 5)

The diversity just presented significant differences among zones (Pseudo-F= 4.8217, P= 0.015) and strata (Pseudo-F= 38.299, P=0.0002), but no differences were detected between Areas. This index showed higher values at Stratum 0-5. Site 3 was significantly more diverse than Site 4 (P=0.0002) and 5 (P=0.0004). No differences were observed between other sites diversity. Species evenness was similar between areas, sites and seasons (Fig. 5), being significantly higher (Pseudo-F=11.43, P=0.0028) at Stratum 0-5.

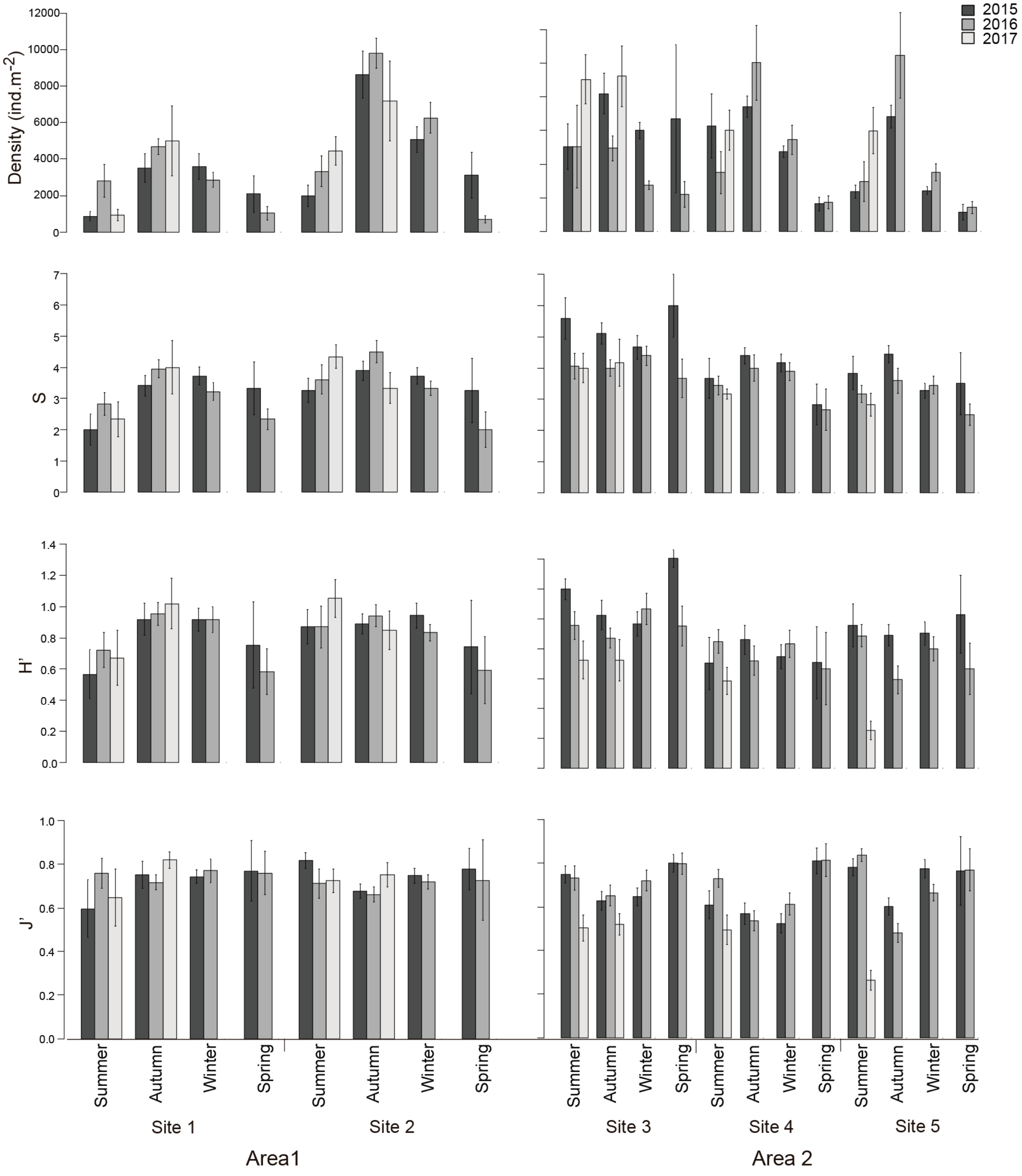

Figure 5 Seasonal variation on total densities, species richness (S), diversity (H') and equitability (J') at each sampled site of both Areas

The assemblage structure varied significantly between areas (Pseudo-F= 5.2523, P=0.003), the strata (Pseudo-F= 13.57, P= 0.001) and sites (Pseudo-F= 4.4293, P=0.001). In addition, the assemblage varied in a different way within the seasons and strata for each area (Pseudo-F= 1.9995, P=0.012). In Area 1 the assemblage of the Stratum 0-5 was different between summer and autumn in 2015 (P=0.0253), and between autumn and winter in 2016 (P=0.0297). For the Stratum 5-10, the differences were detected only between summer and autumn in 2016 (P=0.022). In Area 2 the macrobenthic assemblage of Stratum 0-5 of summer differ from autumn (P=0.021) and winter (P=0.018), being the latter different from spring (P=0.0291) in 2015. While in the next year, the assemblage of summer differs from autumn (P= 0.013), winter (P=0.044) and spring (P=0.022), being also different from winter and spring (P=0.0471). Concerning the Stratum 5-10, the assemblage of autumn was different from spring (P=0.032) and summer (P=0.008) on 2015, and autumn was different from summer on 2016.

The PCoA explained the 62.6% of variation between the Areas. The graph showed a clear separation of the assemblage between sites of Area 1 and those of Area 2 (black ellipses). In addition, it could be observed that first axis separate autumn and winter assemblages from Area 2 at left side and the assemblages of summer and spring of the Area 1 at the right side; the second axis separate at up side the assemblages from summer and spring of the Area 2 and down side the assemblages of autumn and winter from Area 1 (Fig. 6).

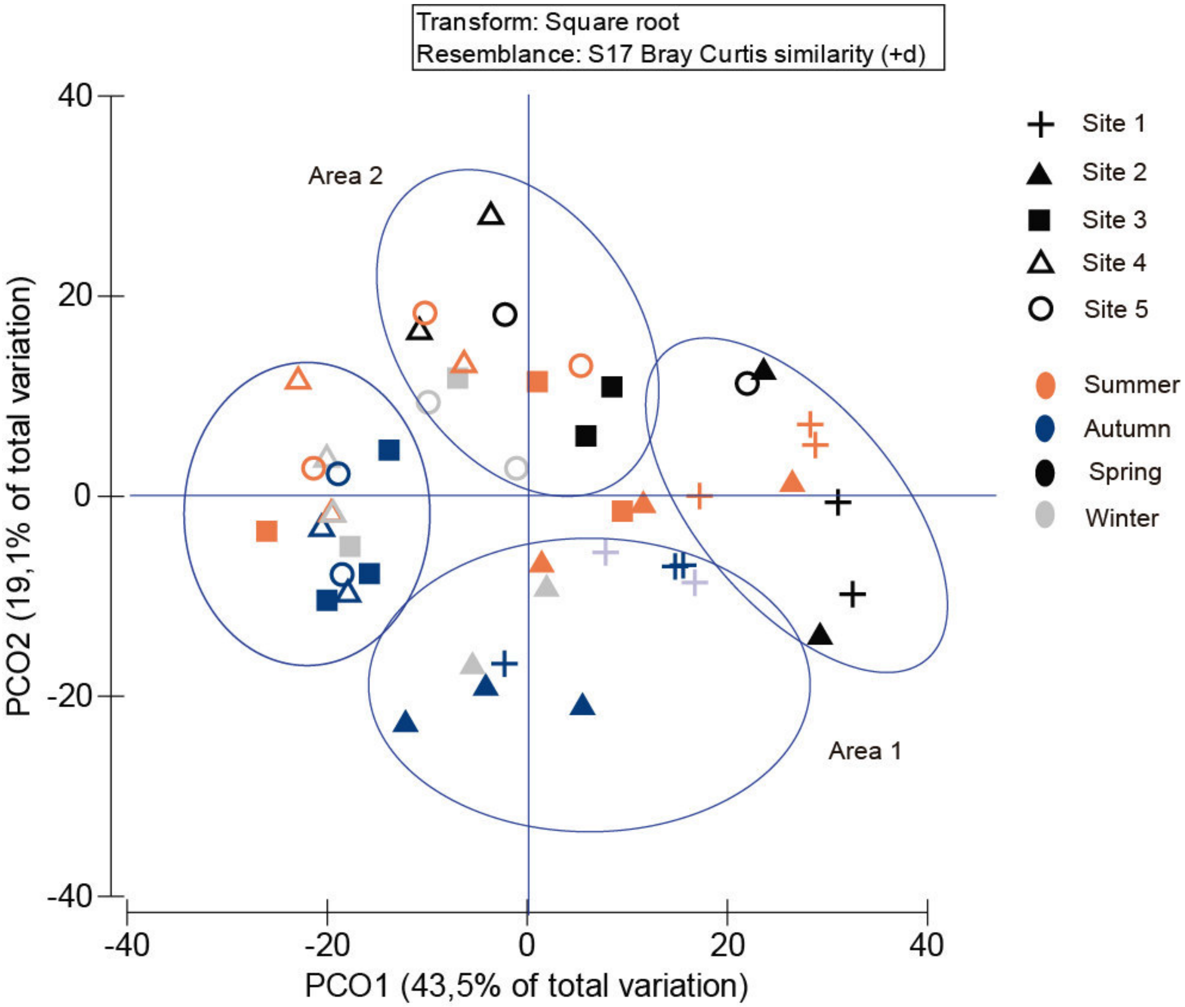

Figure 6 Principal coordinates analysis (PCoA) showing the seasonal separation of macrobenthic assemblages at different sites.

Both areas had a dissimilarity of 57.94% mainly due to the higher abundance of *M. schubarti* (29.16% contribution) at Area 2 (Table I), and of the polychaeta *Heteromastus similis* (19.50% contribution) at Area 1 (Table I). When analyzed the areas separately, it was observed that dissimilarity between sites of Area 1 was 55.96%, being *M. schubarti* (29.87%), *H. similis* (25.51%) and the polychaeta *Nephtys fluviatilis* (16.93%) the main responsible for these differences, which presented higher abundances on Site 2. Within Area 2, Sites 3 and 4 were more similar (dissimilarity: 48.21%) than when compared Sites 3 and 5 (dissimilarity: 50.33%). The main species responsible for the differences between sites were *M. schubarti* (~26.76%), the bivalve *Erodona mactroides* (~26.36%) and the gastropod *Heleobia australis* (~13%).

There was a neat and clear seasonality at most frequent and abundant species densities. The polychaetes presented their highest increments in autumn, particularly in autumn of 2015 in Site 2, when reached densities of 6260.09 ± 4025.21 ind.m^{-2} (mean ± standard deviation). In both areas *M. schubarti* was highly abundant in summer, autumn and winter, with exception of Site 1. It presented a marked seasonality with higher abundances in autumn and winter, followed by very low densities in spring. The highest values were found in autumn of 2016 in Sites 2 (6238.87 ± 3055.54 ind.m^{-2}) and 4 (6603.87 ± 5570.93 ind.m^{-2}), and in autumn of 2017 on Site 3 (6387.41 ± 2820.07 ind.m^{-2}). On Area 2 there was also a higher abundance of *E. mactroides*, which was frequently and highly abundanton Site 3. However, their highest densities were found in autumn of 2016 in Site 5 (3955.53 ± 6206.39 ind.m^{-2}). This species also increased its abundances mainly in autumn, but in 2016 there were also increments in summer for Site 3. *Heleobia australis* presented sporadic peaks of abundances, with higher densities on Site 3, mainly in spring of 2015 (2716.24 ± 3196.65 ind.m^{-2}). (Figure 7).

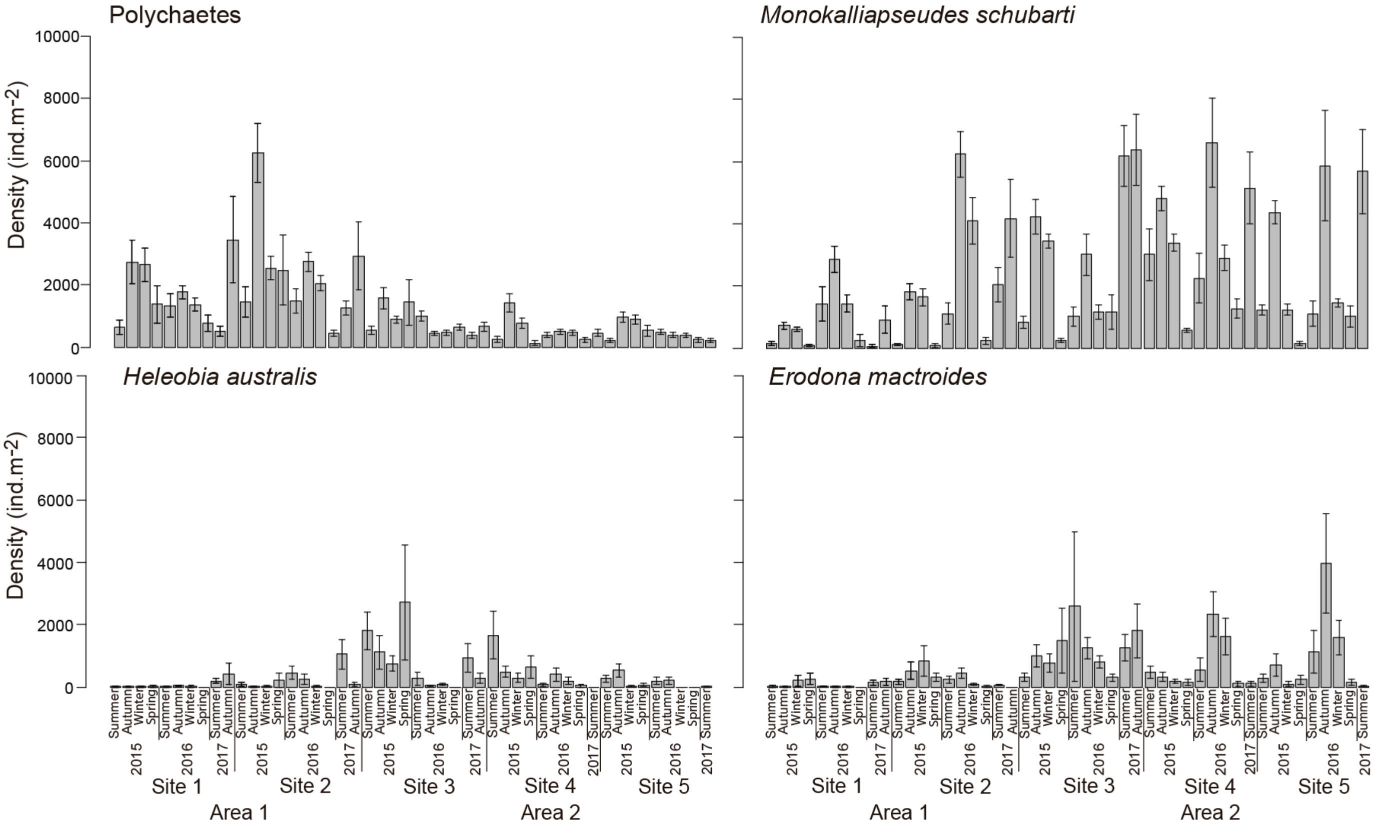

Figure 7. Mean seasonal density of polychaetes, *Monokalliapseudes schubarti, Heleobia australis* and *Erodona mactroides* for each sampled year and site.

Almost all species were positively correlated with salinity. *Monokalliapseudes schubarti* was strongly positive correlated with salinity and depth, but negatively with temperature (Table II). The polychaete

Heteromastus similis was positively correlated with fine sand percentages and the correlation values decreases with the grain sizes increments (Table II). *Nephtys fluviatilis* had strong positive correlations with coarse sands and coarser sediments (Table II). Both species had negative correlations with temperature. They had higher densities when the percentages of fine sediments between 20-40 and fine sands between 50-70%, with coarser sediments and organic matter values under 5%. *Erodona mactroides* was correlated positively with medium and coarse sands (Table II). This species was abundant in zones with sediments composed by less than 5% of coarser sediments, 2-4% of coarse sands, 4-10% medium sands, 30-60% of fine sands. *Heleobia australis* correlated negatively with fine sediments and positively with coarser grain sizes (Table II). This gastropod species was abundant in zones composed by less than 0.6% of organic matter, less than 6% of coarses sands, 10-20% of fine sediments, and its densities increased proportionally with increments of fine sands up to 30%. Higher densities of *M. schubarti* were observed with percentages of fine sands over 30%. The species is positively correlated with salinity and depth. (Table II)

Table II. Spearman correlation value (Rho value) of the environmental variables with the most frequent species in the studied areas. * indicates significant correlations

	Monokalliapseudes schubarti	*Heleobia australis*	*Erodona mactroides*	*Heteromastus similis*	*Nephtys fluviatilis*
Temperature	-0.141*	0.063	-0.075	-0.203*	-0.207*
Salinity	0.409*	-0.035	0.098*	0.235	0.207*
Depth	0.260*	0.064	0.05	0.029	0.047
Organic matter percentage	0.03	-0.077	-0.018	-0.164*	-0.044
Fine sediments (<125um)	0.034	-0.152*	-0.075	-0.187*	-0.019
Fine sand	-0.051	0.170*	0.078*	0.187*	0.029
Medium sand	0.026	0.127*	0.148*	0.180*	0.006
Coarse sand	0.057	0.132*	0.272*	0.107*	0.353*
Coarse sediments (>500um)	0.095*	0.242*	0.259*	0.164*	0.331*

DISCUSSION

In shallow areas, sediment dynamics are influenced by the currents and the wind-formed waves that resuspend sediments and difficult fine material deposition (Dyer et al. 2000, Santos et al. 2004, Souza

& Hartmann 2008). In the Patos Lagoon, N-NE winds favor the lagoon discharge and enhance the erosion of the islands (Möller et al. 2001), resuspending material at shallower zones from Area 2- directly exposed to these winds- and favoring deposition at Area 1- protected by the Pólvora Island-, and in the deeper zones. Conversely, the SW winds are predominant during winter and spring and allow polyhaline scenarios at the estuarine region. At the same time, these southerly winds act in the estuarine hydrodynamic as a barrier to the seaward flows (Möller et al. 2001, Costa et al. 1988), thus allowing the deposition of fine sediments in the shallow estuarine areas. Increases of organic matter and fine sediments during winter were also found in the Queule river (Chile), being the variations in sedimentological parameters related to rainfall and tidal range (Pino et al. 1999). Also, this higher hydrodynamic caused by wind and currents in the PLE may explain the lower percentages of fine sediments recorded on Site 3 (the shallower zone of Area 2) and thus its separation from the other sites on the PCA.

Macrobenthic densities varied seasonally, with peaks on autumn following the increases of salinity values. The sampled years were very rainy, with higher precipitation rates on 2015-2016 (www.inmet.gov.br), as a consequence of two consecutive *El Niño* events: *El Niño* 2014-2015 event was characterized as weak, and *El Niño* 2015-2016 as strong (NOAA, 2017). These events in the Patos Lagoon are the main forcing factors inducing higher freshwater discharges, implying in limnetic scenarios, thus decreasing salinity and increasing the fine sediment inputs into the estuary (Fernandes et al. 2002, Barros et al. 2014). This implies in a scenario where macrofaunal densities are very low during spring, with later recruitment events occurring mainly at the end of summer and autumn (Colling et al. 2007). These observations contrast to that reported for years without the influence of *El Niño* events*,* when the simultaneity between higher salinities and temperature for spring and summer, induces the recruitments, increasing the macrobenthos densities (Bemvenuti & Netto 1998, Rosa & Bemvenuti 2004, 2006). Salinity and temperature have significant effects on species growth, reproduction and survival, inducing changes in the metabolic rates caused by both osmotic and thermic stresses (Uwadiae 2009). There are reports of reduction of reproductive activities on estuarine species when salinities are very low (Steele & Steele 1991), and also that certain temperatures are important for the induction and termination of reproductive resting stages (Panov & McQueen 1998).

As expected, we found higher abundances, richness and diversity of the macrozoobenthos on the Stratum 0-5. Analyzing the small-scale vertical distribution of the macrobenthos in subtidal flats of Chesapeake Bay (USA), was found that organism densities were highest near the surface (81% of total fauna in the 0-2 cm layer), decreasing to deeper layers (Schaffner 1990). They also demonstrate a high degree of association among some species within estuarine assemblages, especially epifauna wich may enhace species richness and diversity. In the Douro estuary (Portugal), higher number of species and higher densities were found in the first 5 cm depth sediment in four of five analyzed sites, when compare with a vertical stratification up to 15cm (Mucha et al. 2004). This preference of macroinfaunal organisms for the upper most superficial layers were related in the North Sea to the availability of freshly deposited or (re-)suspended material (Dauwe et al. 1998). In addition, total nitrogen content decreased with sediment depth between 0-15 cm in estuaries of Dianchi Lake (China) (Yang et al. 2017) and has been proved that there is a decrease in sediment oxygenation with depth (Baillie 1986). Theses conditions may favor higher faunal preferences for upper layers.

In the Patos Lagoon estuary, aproximatelly 40 taxa has being identified (Bemvenuti & Netto 1998, Colling et al. 2007, Colling 2011). In the Saco do Arraial mudflats 14 genera and 12 species were previously observed near Pombas island (Colling et al. 2007), while in the present study we found a lower species richness. (12 genera and 10 species). This lower richness may have occurred due to the spatial location of sampling, but mainly due to a more prolonged period of lower salinities, when compared to that recorded for *El Niño* 2002-2003 (Colling et al. 2007). The higher richness at Site 3 (Area 2) may be due to the higher hydrodynamic, conditioning the predominance of higher percentage of medium sands. In sublittoral sites of western Finland, for example, higher pós-larvae richness was related to sites with higher hydrodinamic when compared to the distribution among a gradient fo hidrodynamic exposition induzed by wind action (Valanko et al. 2010). At Wadeen Sea, the species number of benthic organisms was higher at intermediate sediments size classes than sites with very fine or very coarse sediments (Beukema & Dekker 2012), which coincides with our results. Diversity and evenness index did not show significant variations between sampling sites, as well as between months, coinciding with the reports of Colling et al. (2007). However, slightly higher diversity values where found in sites with coarser sediments, similar to was found by of Vanaverbeke et al. (2011) in the North Sea.

Different abiotic parameters may affect macrozoobenthic assemblages' variations in spatial and temporal scales. It is well known that the distribution of macrobenthos is related to abiotic factors (e.g., salinity, water depth, bottom water temperature, hydrodynamic conditions, sediment grain size and dissolved oxygen) (Ysebaert & Herman 2002, Como & Magni 2009, Nishijima et al. 2015, Veiga et al. 2017). Temperature and salinity may be the main responsible for seasonal variations, however, these factors may not drive spatial variations in small-scale distances. In the coastal lagoon of Cabras (Sardinia, western Mediterranean) some critical areas for macrofauna development could be identified based on the hydrodynamic (e.g. inner water circulation) and sedimentary features of the basin (e.g. sediment transport) (Magni et al. 2004). It has been reported that water circulation and wind speed indirectly influence recruitment, resuspension and dispersal of individuals (Vanaverbeke et al. 2011), which may explain spatial patterns of macrobenthos dispersion and occurence. The current speed, bathymetry and salinity are the most important environmental factors explaining the distribution pattern of benthic assemblages in the eastern part of the Bay of Seine (Dauvin et al. 2017). However, macrofaunal distribution patterns on the Belgian coast have been related mainly to water column processes: suspended particulate matter, food availability and hydrodynamic stress (Vanaverbeke et al. 2011).

Monokalliapseudes schubarti was the most abundant and frequent species in all sites, with exception to Site 1 where the polychaete *Heteromastus similis* presented higher abundances and frequency of occurrence. The highest densities of *M. schubarti* were recorded on the higher depths of Area 2 and the Site 2 of Area 1. The species presented significant correlations with temperature and salinity, as well as depth. In small coastal plain estuaries of Southern Brazil, however, this tanaid presented the strongest relationships with characteristics of sediments and the lowest relationships with water properties (Pagliosa & Barbosa 2006). Also, Leite et al. (2003) and Freitas-Júnior et al. (2013) found that densities of the species where highest at low to intermediate percentages of silt and clay (4-10%), with lower abundances related to percentages higher than 13%.

Heteromastus similis was the species, which better correlates with sediment properties and organic matter percentages. This coincides with the spatial distribution of sediments, while in Area 1 were found higher percentages of fine sands and coarse sediments, and the accumulation of dead seashells. In coastal lagoons and estuaries from Uruguay, this species was positively correlated with percentages of gravel, fine sands and mud (Giménez et al. 2014). The absence of positive correlation *between H. similis* and fine sediments found here was probably due the low salinities in the seasons with higher percentages of this grain sizes. Polychaetes are recognized as sediment stabilizers due to the production of mucus, which acts in retention of fine sediments (Dyer et al. 2000). As a capitellid this species produce a mucopolysaccharide to agglutinate grains of sand and organic particles (Fauchald & Jumars 1979). In this sense, protected sites that favor the presence of higher percentages of fine sediments and organic matter enhance proper conditions to the presence of the species. Consistently, all polychaetas found in this study had higher abundances on Area 1. *Nephtys fluviatilis* had also their highest significant correlation coefficients related to sediment properties. The specie is a quite mobile organism and seems to exert influence on the young forms of *H. similis* trough predation (Bemvenuti 1994), which justify its higher abundances on the same conditions.

The recruits of the mollusk species *Heleobia australis* and *Erodona mactroides* were positively correlated with coarser sediments. Variations in *E. mactroides* abundances has been related with sediment properties, with higher abundances observed in areas dominated by medium and coarse sands in uruguaian estuaries (Giménez et al. 2014), which coincide with our findings for the Area 2. In the PLE, the adults of *E. mactroides* inhabit the upper estuarine zones (Bemvenuti et al. 1978, Capítoli et al. 1978, Colling et al. 2010). Their larvae are carried by ebb tides to the lower estuary during spring and summer, recruiting in sheltered inlets (Colling et al. 2010). These recruitments present an intrinsic relation to local hydrodynamic and currents, which are responsible for a high spatial and temporal variability over the PLE shallow flats (Colling et al 2010), which explain the marked differences between Areas and sites in our study. Similar trends were found in New Zealand tidal flats, where juvenile bivalves dispersed over a scale of meters within tidal cycles (Norkko et al. 2001). *Heleobia australis* is an opportunistic deposit feeder, which benefits on the absence of other sedentary epifaunal species (Bemvenuti et al. 2003). High abundances of this gastropod (>30,00 ind.m^{-2}) were found in the deep channels of the Patos lagoon Estuary (Pinotti et al. 2011). Considering the location of Area 2 in the present study, it is possible that higher abundances of this gastropod were related to regions where it occurs in their greater densities, as the harbour channel near Area 2, when compared with Area 1.

The present study evidenced that the spatial distribution of macrobenthic species are related to the variability of the sediment properties, depth and to local hydrodynamic conditions. The temporal variability of water properties was directly related to seasonal differences in the benthic macrofaunal assemblages. Concerning the vertical distribution of benthic organisms, our findings evidenced that the superficial layers present higher densities and species richness, which independs of any spatial variability of granulometric features or temporal trends of salinity scenarios of the esturine region. In this sense, we evidenced that even when depth and seawards zonation of estuarine macrobenthic fauna has been well studied, small-scale variations in local hydrodynamic conditions should be understood as little variations may imply in the dominance of different species.

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Apêndice 3

Esse apêndice refere-se ao artigo intitulado “Response of soft-bottom macrobenthic assemblages to artisanal trawling fisheries in a subtropical estuary” publicado na revista Estuarine Coastal and Shelf Science

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Response of soft-bottom macrobenthic assemblages to artisanal trawling fisheries in a subtropical estuary

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ABSTRACT

Bottom trawling is one of the most harmful and widespread activities affecting benthic habitats and fauna. In this study, we analyzed the impact of shrimp trawling fisheries on the densities, assemblage structure and vertical stratification of the benthic macrofauna in shallow mudflats of the Patos Lagoon Estuary (Brazil). Experimental trawls of different intensities were performed during three shrimp fishing seasons (2015–2017), comparing macrofauna among before and after trawling, and a control zone. The changes in macrofaunal assemblages were more consistently related to natural variability than to trawling impact, being mostly influenced by sediment structure and salinity variation. The trawling impact was mainly detected in the area with higher percentages of fine sediments, with different macrofaunal responses in each month and stratum. Some non-significant decreases on total densities after trawling were observed, mainly on the superficial stratum, and signs of burial activities. The response to trawling disturbance of each species was different and it highly depended on their natural variability. Only five species showed significant variation to the trawling treatment (*Erodona mactroides*, *Heleobia australis, H. charruana, Heteromastus similis and Laeonereis acuta*), with temporally different responses. Decreasing densities were more related to the high impact treatment. In some seasons, decreases on the abundance of *Monokalliapseudes schubarti* after high impact trawls were up to 60% of the seasonal mean densities. Trawling fisheries may reduce macrobenthic densities but not their vertical stratification. The impact of trawling on key species may compromise the ecosystem function, as benthic macrofauna provide food sources for many aquatic resources. This study highlights trawling impacts in an estuarine nursery area, which should be fully considered since they may be reflected in the trophic webs.

1. Introduction

Estuaries are very productive ecosystems, providing important ecological services but different human activities have compromised their ecological integrity (Alves et al., 2015; Rehitha et al., 2017). Trawling fisheries are one of the most harmful anthropogenic impacts on seabed (Kaiser et al., 2002; Collie et al., 2016; Hiddink et al., 2017). The impact of bottom fisheries on the seafloor result in harmful effects on benthic flora and fauna, but also on the physical environment (Kaiser et al., 2006, 2016; Collie et al., 2016; Hiddink et al., 2017). These gears can promote sediment suspension; decreasing macrofaunal bioturbation processes; remove, injure, or kill a wide range of benthic organisms, and induce evolutionary changes on population demography and on the ecosystems structures and functions, among other consequences (Engel and Kvitek, 1998; Brown et al., 2005; Foden et al., 2010; Mangano et al., 2013, 2014; Sciberras et al., 2016).

The widespread use of bottom gears is controversial, leading to calls for bans of trawling in order to minimize ecosystem deterioration (Cinner et al., 2005; Watling, 2013; European Parliament, 2016). The studies regarding fishing impact on bottom communities have focused on large-scale or industrial fisheries, giving little attention to small-scale or artisanal fisheries, especially in estuaries (Costa and Netto, 2014). Despite of its apparently small-scale catches, artisanal fisheries contribute 50% of the world fish gross capture and two-thirds of world catches destined for direct human consumption (FAO, 2017).

A method to evaluate the impact of fisheries is the Ecosystem

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Approach to Fisheries (EAF). This method requires knowledge of how target species may be affected, but also takes into consideration the impact of the fisheries in the whole ecosystem, including trophic web disruption and the effects of multiple and potentially interacting pressures, as natural variability and climate change (Garcia and Cochrane, 2005; Bolam et al., 2014). The EAF aims to safeguard both function and biodiversity, therefore, trawling impacts on benthic community functioning needs to be understood prior to the enforcement of a given management action (Bolam et al., 2014). The natural variability generally is known for decades, but in practice it is not taken into account when evaluating impact, mainly because it is difficult to predict. When this variability is considered, it generally improves management performance (Garcia and Cochrane, 2005).

Coastal lagoons are one of the most productive ecosystems in the world, providing services to humans and acting as nursery grounds to marine and estuarine species, some of which has commercial interest (Miththapala, 2013; Sheaves et al., 2015). Artisanal trawling is widely performed in the Patos Lagoon Estuary (southern Brazil) targeting the pink shrimp *Farfantepenaeus paulensis* (Benedet et al., 2010; Kalikoski and Vasconcellos, 2012). Legal regulations for the estuarine fisheries ban the use of bottom trawling in the area (MMA, 2004). However, due to the lack of effective control, it is widely used during shrimp season. This fishery starts in the austral summer and ends in autumn (February to May), allowing only the use of passive fyke-nets (D'Incao, 1991; Benedet et al., 2010). The period of intense trawling matches a critical recruitment period for the macrobenthos at the Patos Lagoon Estuary. There is an increase of macrofauna reproduction and recruitment in late spring and summer (Bemvenuti and Netto, 1998; Rosa and Bemvenuti, 2006), which can be extended to autumn, influenced by large-scale climate events, such as ENSO (Colling et al., 2007). Therefore, it may magnify the impact due to the synchronicity of trawling and macrobenthos reproduction and recruitment seasons.

This potential impact on the macrobenthos may affect the whole estuarine ecosystem, due to the important role of these assemblages in the nutrient cycling, sediment dynamics and food webs, since act as both consumers and prey (Mola and Abdel Gawad, 2014; Piló et al., 2015; Dauvin et al., 2017). Due to the limited mobility and short life span, benthic macrofauna is considered as a good biological indicator of impact, being capable of responding to disturbances in the water column and sediment (Dauvin et al., 2017).

This study evaluates the potential impact of trawling fisheries on the grain size composition and macrofaunal densities, species richness, diversity, evenness, assemblage structure and vertical stratification in a subtidal shallow mudflat of the Patos Lagoon estuarine region. We hypothesized that trawls should reduce the percentage of very fine sediments (very fine sand, silt and clay), reduce the macrofaunal densities, richness, diversity, alter the assemblage structure and modify the previous vertical stratification of the macrobenthos.

2. Materials and method

2.1. Study area

The Patos Lagoon Estuary (PLE) is located in the southern Brazilian coastal plain (Kjerfve, 1986) (Fig. 1). Five major tributary rivers constitute significant freshwater sources to the PLE from an extensive drainage basin (201,626 km^2) (Fernandes et al., 2005). The freshwater discharge and the action of predominant winds (from NE during the warm seasons and from S during the cold seasons) tightly control the hydrodynamics of the PLE (Fernandes et al., 2002). In the western margin, it has a shallow (1–4 m depth) inlet called 'Saco do Arraial', characterized as a dynamic environment concerning the erosion processes, sediment transportation, and deposition (Colling et al., 2007; Souza and Hartmann, 2008), which is our study region.

2.2. Experimental design

Two areas commonly used by artisanal fishermen in the Saco do Arraial were selected: 1) the sheltered area near Pólvora Island-Area 1; and 2) the more exposed area near Cavalos Island -Area 2 (Fig. 1). Depth was measured in every sampling, ranging between 0.9 and 2.5 m. Three zones of 20 × 20 m were marked in each area with wood sticks: the first one without any impact (Control zone), the second where two consecutive experimental tows were performed (Low-impact zone), and the third where five consecutive experimental tows were conducted (High-impact zone) (Fig. 1). In each sampling, trawling followed an imaginary line randomly selected into the marked square. Trawls were performed using a typical artisanal fishing boat of 8 m in length at a speed of approximately 2 knots (3.7 km/h), equipped with an otter trawl net for shrimp capture, with 12 mm between opposite knots and 15 kg boards.

2.3. Sampling

Sampling was carried out monthly between January and June 2015–2016, and in January, March and May 2017, resulting in five sampling campaigns in 2015, six in 2016 and three in 2017. In each sampling, three replicates of sediment samples were collected for macrofauna analysis and two for granulometric and organic matter analyses in the control zones. In the impacted zones, three samples were collected before and after trawling for fauna analysis and two for granulometric and organic matter analyses. The post-trawling samplings were immediately collected after the induced impact. Samples were collected with a *van Veen* grab, and from each grab, one subsample was taken with a 10 cm diameter core (0.0078 m^2). This subsample was sectioned into 0–5 cm and 5–10 cm strata. The benthic macrofauna was sieved with a 300 μm mesh net and preserved in 4% formaldehyde. Water temperature was measured with a mercury thermometer (°C) and salinity with a refractometer. Granulometric analyses were performed by dry mechanical sieving through a column of sieves of different mesh sizes from 4 mm to 0.063 mm and by pipetting analysis for grain sizes < 0.063 mm. The granulometric procedures followed the Wentworth classification system in intervals of one phi (Suguio, 1973). The grain size composition was expressed as the percentage of the total sample weight. Organic matter content was calculated by differences on weight before and after calcination for 2 h at 375 °C.

2.4. Statistical analysis

In order to characterize and evaluate seasonal differences on the studied area, a Principal Component Analysis (PCA) was performed using all measured parameters (salinity, temperature, depth, organic matter percentage and all granulometric percentages). Similarity Percentages (SIMPER) of the species contributions were calculated to identify the main species responsible for the differences on faunal composition between the studied areas. A canonical correspondence analysis (CCA) was performed to evaluate possible relations between the macrofaunal species and environmental parameters.

The fishing impact was analyzed using a nested design in order to evaluate differences in total densities, species richness, diversity, evenness and assemblage structure between trawling periods (before and after trawling) and vertical macrofaunal stratification for each area along the three sampled years. The tested factors were Treatment (fixed- Control, Before high impact, After high impact, Before low impact and After low impact), Month (random, nested in Season), Season (random, nested in Year), Year (random), and Stratum (fixed- 0–5 cm, 5–10 cm). All fixed factors were orthogonal among themselves. The possible variability among factors was tested through a permutational multivariate analysis of variance (Permanova, Anderson et al., 2008).

For the univariate macrofaunal data (total densities, species

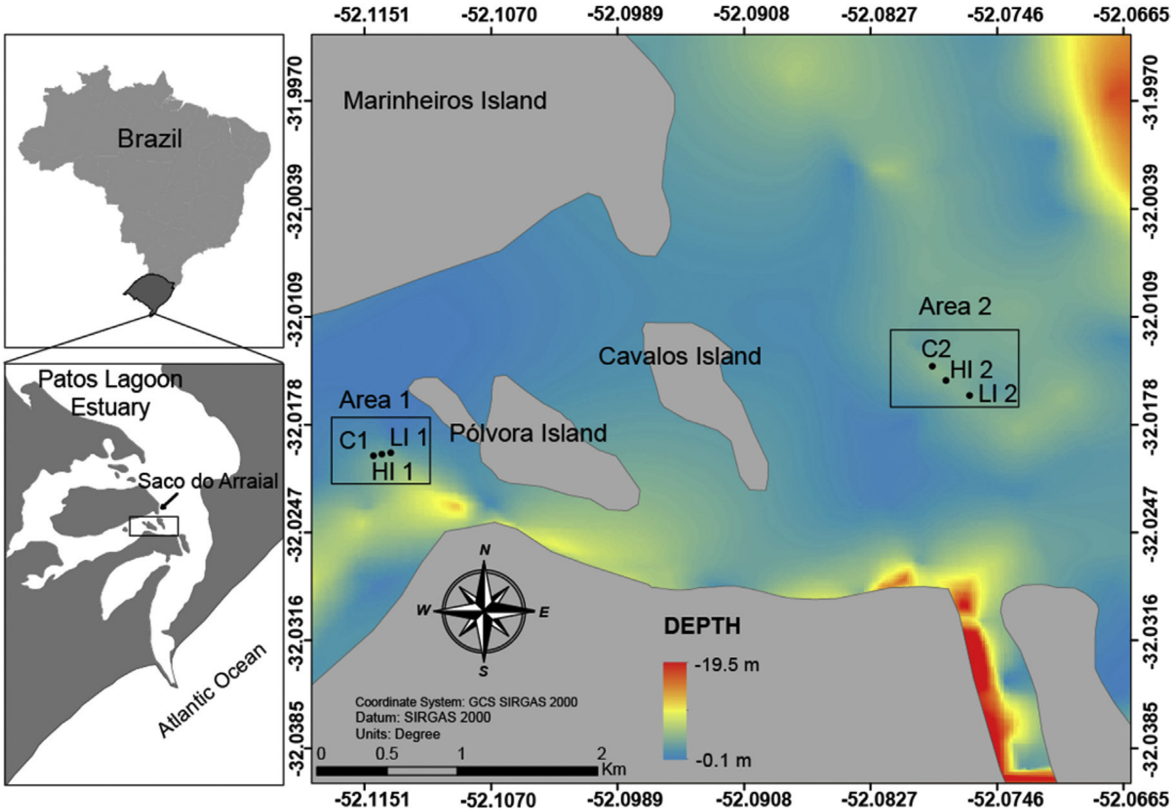

Fig. 1. Map of the study area: Saco do Arraial, Patos Lagoon Estuary, Brazil. C1 and C2: control zones, HI1 and HI2: high-impacted zones, LI1 and LI2: low-impacted zones.

richness, diversity and evenness), a Permanova with 4999 permutations of raw data units was used to analyze the possible variability among factors (Anderson, 2005). We used the Monte Carlo permutation test to obtain P-values ($P < 0.05$) (Anderson and Robinson, 2003; Anderson, 2005). This test assumes not only exchangeability, but also a linear model on dissimilarities that is appropriate for choosing reasonable statistical tests analogous to those used in univariate Anova (Anderson and Millar, 2004). Permanova was performed using a Euclidian distance matrix. The significant results were evaluated with *a posteriori* pairwise comparisons, which also used 4999 random permutations to obtain P-values through Monte Carlo correction.

For multivariate macrofaunal data (assemblage structure), the Permanova was based on the Bray-Curtis similarity matrix constructed from the square-root-transformed abundance data (ind.m^{-2}). Transformation was used to down-weight the importance of the dominant species. Dummy values of one were added to reduce the effect of the absence of individuals in some samples (Clarke et al., 2006). The statistical significance of the relationships was evaluated using Monte Carlo permutation test. Pairwise tests were run for those factors that resulted significant, using the Monte Carlo correction. The same statistical approach was applied to grain size data, but instead of a Bray-Curtis similarity matrix, the Euclidean distance matrix was employed.

3. Results

3.1. Environmental variables

Salinity varied between 0 and 34, presenting higher values at late autumn (May and June) and lower at early summer (January–February). Temperature varied between 10 and 27 °C, with the early summer presenting the highest values and at late autumn the lowest values. Organic matter varied between 0 and 8.24% per sample (minimum and maximum determined values), with higher mean values recorded in Area 1 (approx. 2% in Area 1 and 1.5–1.16% in Area 2). Fine sands constitute the main sediment fraction for both areas. Zones from Area 1 were characterized by more than 14% of silt and clay, and less than 3% of medium sand, while the zones from Area 2 presented less than 9% of silt and clay and more than 10% of medium sands (Table 1).

The Principal Component Analysis (PCA) explained approx. 51% of variance: the axis 1 explained 37% of variation while axis 2 the 14.5% of the variation. The first axis separated spatially both areas, and the second one was related to the temporal variability (Fig. 2). The Area 1 exhibited higher percentages of silt, clay and very fine sands (fine sediments), and organic matter, while Area 2 was slightly deeper and composed by higher percentage of fine sands, medium sands, coarse sands, very coarse sands, granules and pebbles (coarser sediments) (Fig. 2; Table 1). Temperature and salinity correlated negatively and their relationship was higher than those related to sediment characteristics, characterizing a temporal trend.

3.2. Trawl impact on grain size composition

The sediment texture varied within the interaction of the factors month, stratum and treatment in both areas (Pseudo-F Area 1 = 2630.5, Pseudo-F Area 2 = 1.23E+05, P = 0.001), indicating a high variability on sediment response. Pairwise tests showed that differences on treatment occurred among zones (Control, High and Low-impact zones) and not between before and after trawling ($p < 0.001$). For Area 1, the Control zone exhibited approximately 39 ± 9% (mean ± standard deviation) of very fine sediments, while High-impact zone had 35 ± 6% and Low-impact zone 31 ± 6%. For the Area 2, the Control zone had approximately 13 ± 2% of very fine sediments, High-impact zone had 15 ± 6% and Low-impact zone 21 ± 6%. Differences between strata were subtle, with higher percentages of very fine sediments in Stratum 0–5 for both areas (differences of ± 2% between strata).

Table 1
Mean and standard deviation (SD) of the environmental variables in both studied areas.

Area	1				2			
Season	Summer		Autumn		Summer		Autumn	
Variables	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Temperature (°C)	23	3	17	4	24	2	17	4
Salinity	4	4	19	8	3	6	21	11
Depth (m)	1.30	0.17	1.34	0.20	1.70	0.24	1.84	0.27
Organic matter (%)	2.07	0.82	1.85	0.74	1.51	1.00	1.16	0.66
Pebbles (%)	0.003	0.015	0.005	0.022	0.008	0.024	0.017	0.046
Granules (%)	0.018	0.033	0.015	0.028	0.088	0.095	0.072	0.053
Very coarse sand (%)	0.040	0.041	0.035	0.038	0.092	0.058	0.069	0.041
Corse sand (%)	0.224	0.124	0.215	0.097	0.293	0.426	0.200	0.090
Medium sand (%)	2.839	4.205	2.530	1.840	10.432	3.166	11.790	2.604
Fine sand (%)	61.963	7.088	62.995	6.008	71.925	6.136	71.078	4.606
Very fine sand (%)	20.369	5.214	19.039	4.048	8.702	3.926	7.406	2.462
Silt (%)	5.266	2.138	5.682	1.496	3.169	1.299	3.417	1.268
Clay (%)	9.176	2.973	9.400	2.977	5.239	2.138	5.900	3.287

3.3. Macrobenthic structure

A total of 45529 specimens were quantified, comprising 9 major groups (Amphipoda, Isopoda, Tanaidacea, Cumacea, Decapoda, Bivalvia, Gastropoda, Polychaeta and Nemertea). The most abundant and frequent species in both zones was the tanaid *Monokalliapseudes schubarti* (Table 2). Juveniles from the gastropod genus *Heleobia* were grouped as *Heleobia* sp. due to the lack of identification structures.

The assemblage structure was significantly different between both areas (Permanova, Pseudo-F = 17.842, P = 0.001). The dissimilarity percentage between both, Area 1 and Area 2, was of 62%. Seven species were the responsible for 94% of differences: the peracarids *Monokalliapseudes schubarti* (30.63%) and *Kupellonura* sp. (7.34%), the mollusks *Erodona mactroides* (14.68%) and *Heleobia australis* (9.52%), with higher abundances on Area 2; and the polychaetes *Heteromastus similis* (15.07%), *Nephtys fluviatilis* (11.36%) and *Laeonereis acuta* (5.68%) with higher abundances on Area 1.

The CCA explained 80.32% of the variance (54.47% at CCA1 and 25.85% at CCA2). Polychaetes were more correlated with the fine sediments, which were present in higher percentages on Area 1. The gastropods were predominant in Area 2 and more correlated with higher abundances of coarser sands. The tanaid *M. schubarti* correlated positively with salinity and higher percentages of medium sands, which were predominant on Area 2 (Fig. 3).

3.4. Trawl impact on macrofauna

3.4.1. Macrobenthic total densities

The mean total density at Area 1 was 909.0 ± 1305.8 ind.m^{-2} (Mean ± Standard deviation). The response of faunal total densities to treatment varied between strata (Pseudo-F = 4.4103, P = 0.0308). The pairwise test detected no statistically significant differences between strata for any treatment, nor differences between treatments for each stratum, although we detected more signals of impact on Stratum 0–5, with decrease on densities after trawling in some months (marked as descending arrows on Fig. 4). In addition, densities increased in some months after trawling on Stratum 5–10 cm, indicating the burial of organisms (marked as ascending arrows on Fig. 4). The greatest differences in densities between treatments were observed between years 2016 and 2017 when faunal abundances were higher. Significant differences in the stratification by month (Pseudo-F = 4.1255, P = 0.0002) were found, where higher densities occurred in Stratum 0–5 in almost all months (Fig. 4). The Permanova also detected significant differences in total densities by month (Pseudo-F = 28.383, P = 0.0002), with higher increments at late summer and early autumn (Fig. 4).

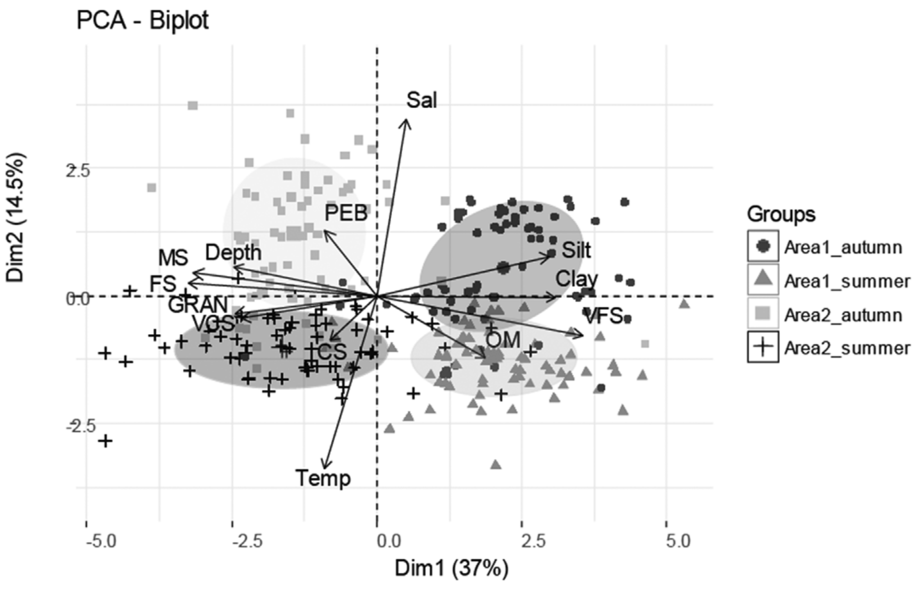

Fig. 2. Principal Component Analysis Plot (Dim1/Dim2) used to separate areas and seasons by the environmental variables. PEB: pebbles, GRAN: granules, VCS: very coarse sand, CS: coarse sand, MS: medium sand, FS: fine sand, VFS: very fine sand, Temp: water temperature, Sal: water salinity. Ellipses grouped 50% of points from each centroid.

Table 2
List of species found on the Saco do Arraial with their mean density and frequency of occurrence by treatment. C: Control zone, LI: Low-impact zone, HI: High-impact zone. FO: frequency of occurrence.

Group	Specie	Epifauna/ Infauna	C1	% FO	LI1 Be	% FO	LI1 Af	% FO	HI1 Be	% FO	HI1 Af
			Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)
Amphipoda	**Gammaridea**	Epifauna	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cumacea	***Diastylis sympterygiae***	Infauna	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Isopoda	***Kupellonura* sp.**	Infauna	254.6	0.3	0.0	0.0	0.0	0.0	254.6	0.3	0.0
	***Sphaeromopsis* sp.**	Epifauna	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Uromunna peterseni	Epifauna	127.3	1.6	127.3	1.5	233.4	1.8	200.1	2.1	141.5
Tanaidacea	***Monokalliapseudes schubarti***	Infauna	1294.3	35.2	992.7	34.3	1212.3	35.3	1328.9	34.3	1254.3
	Sinelobus stanfordi	Epifauna	127.3	0.3	127.3	0.3	0.0	0.0	0.0	0.0	254.6
Decapoda	**Panopeidae**	Epifauna	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Bivalvia	***Erodona mactroides***	Infauna	297.1	3.9	497.7	6.4	583.0	5.6	369.2	6.1	245.6
Gastropoda	***Heleobia australis***	Epifauna	418.4	4.5	516.8	4.9	617.0	3.9	1077.4	4.0	262.1
	Heleobia charruana	Epifauna	127.3	0.3	159.2	1.2	127.3	0.3	0.0	0.0	127.3
	***Heleobia* sp.**	Epifauna	0.0	0.0	127.3	0.3	254.6	0.6	0.0	0.0	0.0
Polychaeta	***Heteromastus similis***	Infauna	1232.0	21.9	1396.9	20.3	1230.2	22.8	1247.0	20.8	1114.5
	Laeonereis acuta	Infauna	405.1	10.6	437.9	11.9	474.3	11.9	445.6	13.5	364.4
	Nephtys fluviatilis	Infauna	714.4	17.4	606.4	17.2	731.5	16.3	712.5	15.9	675.1
	Poliqueta n. i	Infauna	127.3	0.6	127.3	0.6	0.0	0.0	0.0	0.0	0.0
Nemertea	**Nemertea**	Infauna	127.3	0.6	127.3	0.6	127.3	0.3	127.3	0.6	127.3

Group	% FO	C2	% FO	LI2 Be	% FO	LI2 Af	% FO	HI2 Be	% FO	HI2 Af	% FO
		Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)		Mean density (ind.m^2)	
Amphipoda	0.0	0.0	0.0	0.0	0.0	127.3	0.5	0.0	0.0	0.0	0.0
Cumacea	0.0	254.6	1.7	191.0	0.5	127.3	1.4	191.0	1.7	145.5	1.6
Isopoda	0.0	288.4	10.6	221.5	11.5	203.7	12.4	275.5	11.7	309.9	12.0
	0.0	0.0	0.0	0.0	0.0	127.3	0.2	0.0	0.0	0.0	0.0
	2.8	152.8	2.2	178.3	2.3	178.3	1.1	175.1	1.7	169.8	1.4
Tanaidacea	37.5	3033.6	34.8	3231.1	40.2	3392.5	38.2	3349.7	37.2	3269.3	37.0
	0.3	169.8	0.6	350.1	1.8	350.1	1.8	254.6	1.3	534.8	1.1
Decapoda	0.0	127.3	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Bivalvia	4.3	2136.7	11.9	4168.3	9.7	4526.4	9.0	4391.3	9.6	3169.3	10.4
Gastropoda	5.3	1612.8	8.4	1199.3	7.1	2432.8	6.3	1733.6	8.3	1571.4	8.6
	0.3	2546.5	1.3	1491.5	1.6	1344.0	2.0	1909.9	1.7	1888.6	1.4
	0.0	169.8	0.6	254.6	0.7	509.3	0.5	286.5	0.8	1527.9	1.1
Polychaeta	21.4	588.4	16.0	534.3	14.0	484.6	15.2	596.6	14.9	487.7	13.3
	9.0	159.2	0.9	127.3	0.5	191.0	0.5	152.8	1.1	200.1	1.6
	16.4	532.7	10.6	494.8	10.1	390.3	10.4	543.8	10.2	368.1	10.4
	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Nemertea	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

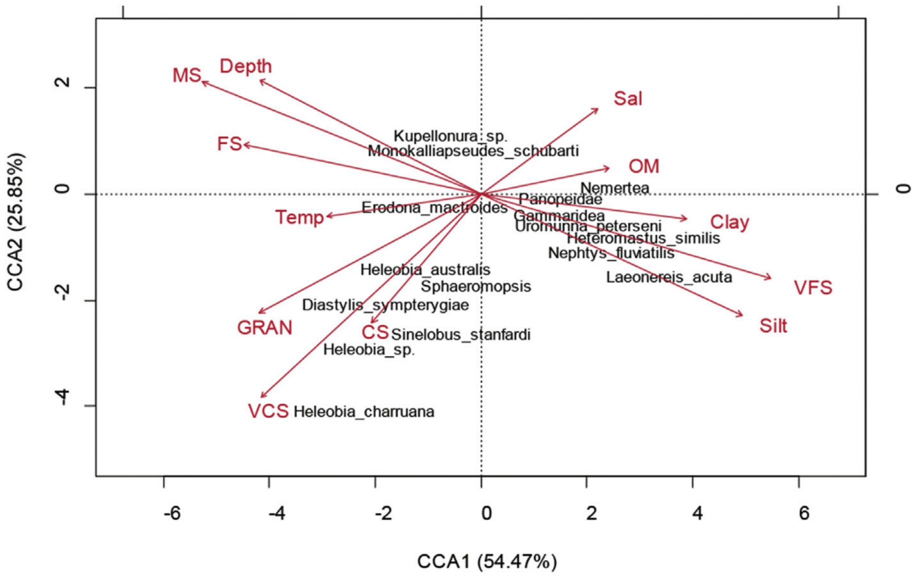

Fig. 3. Canonic Correspondence Analysis of fauna sampled on the Saco do Arraial in correlation with environmental variables. GRAN: granule, VCS: very coarse sand, CS: coarse sand, MS: medium sand, FS: fine sand, VFS: very fine sand, OM: organic matter, Sal: water salinity, Temp: water temperature.

At Area 2, the mean total density was 1910.9 ± 3709.4 ind.m^{-2}. The response of total densities to treatment varied monthly (Pseudo-F = 1.611, P = 0.0236). Pairwise tests evidenced some months with differences between zones, and only in March (2017) there were differences between before and after trawling in the Low-impact zone (t = 14.775, P = 0.002). Although there were no-significant differences, a decrease in densities was observed in some months in both strata (marked as descending arrows on Fig. 5), as well as increases in densities on Stratum 5–10 (marked as ascending arrows on Fig. 5). Stratification also differed monthly (Pseudo-F = 9.2454, P = 0.0002). In almost all months, the total densities were significantly higher at Stratum 0–5, with exception of January of 2015. There were also significant differences within months (Pseudo-F = 18.117, P = 0.0002). The highest faunal densities were observed at the end of summer and autumn.

3.4.2. Species richness

Species richness ranged from 0 to 7 by sample at Area 1. The

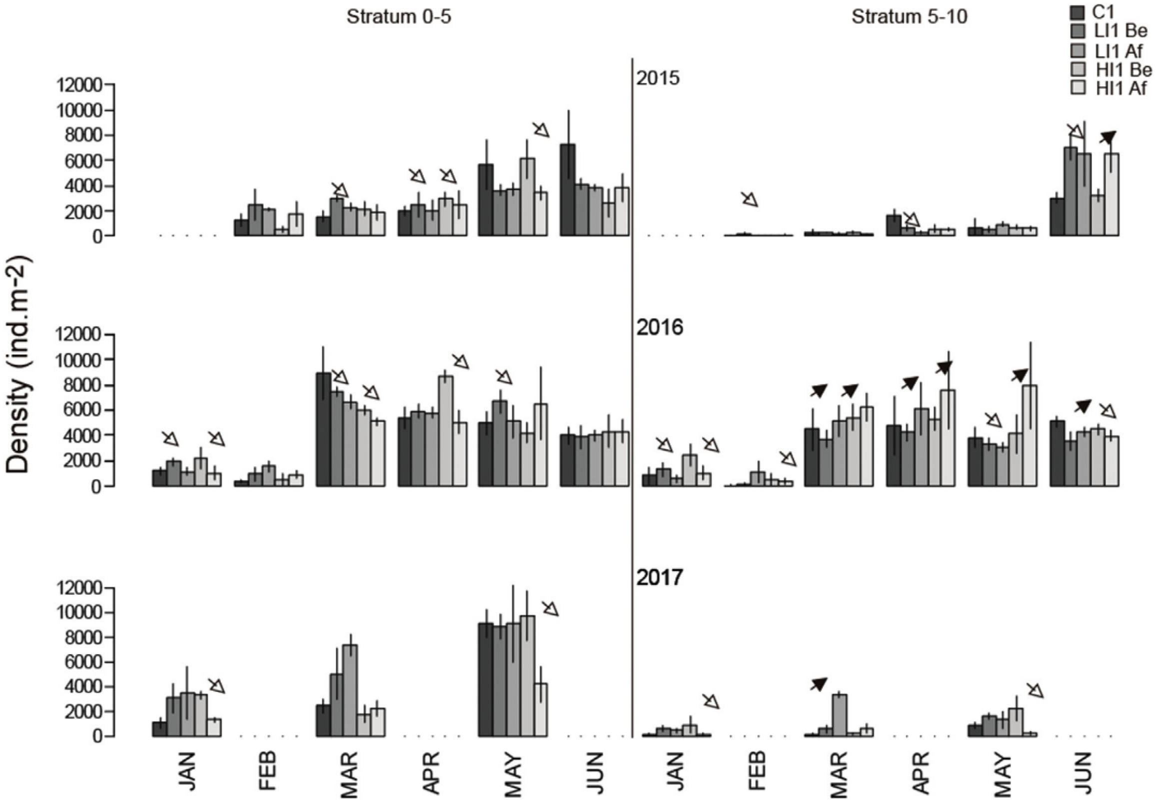

Fig. 4. Monthly variation on mean density (ind.m^{-2}) of macrobenthic organisms in each treatment for Area 1. Vertical bars indicate standard error. C1: Control zone, HI1 Be: High-impact zone before trawling, HI1 Af: High-impact zone after trawling, LI1 Be: Low-impact zone before trawling, LI1 Af: Low-impact zone after trawling.

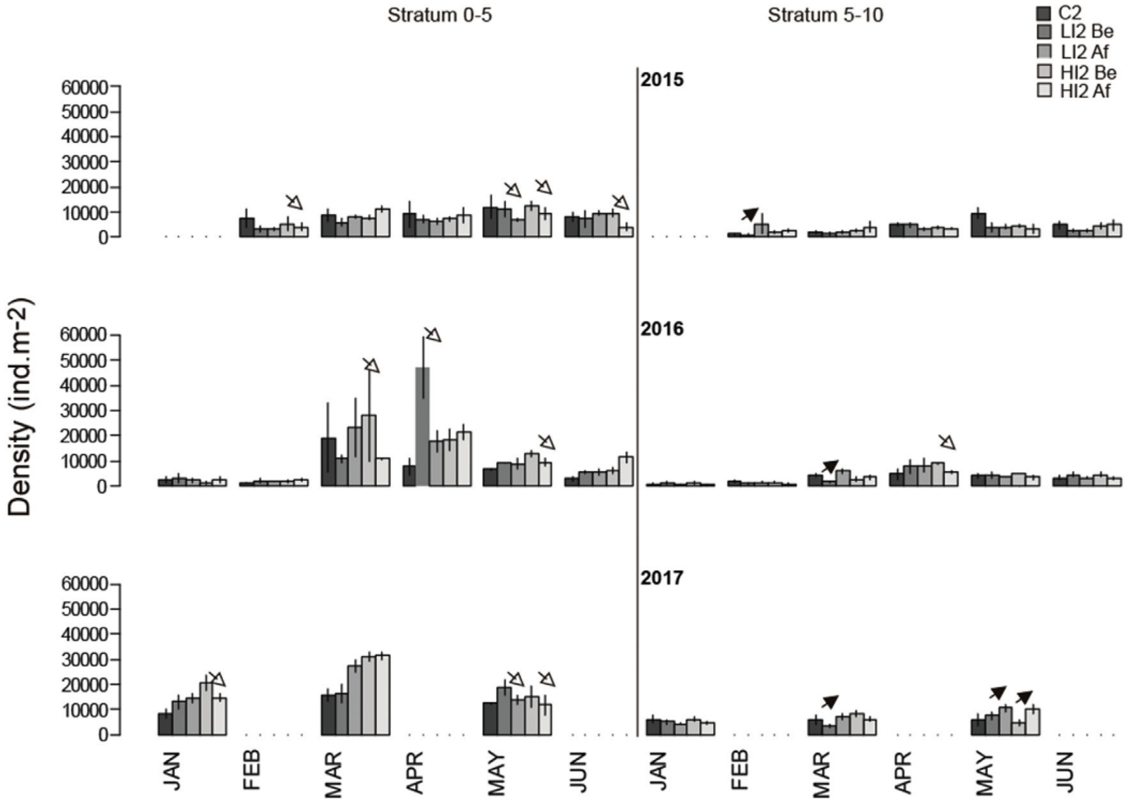

Fig. 5. Monthly variation on mean density (ind.m^{-2}) of macrobenthic organisms in each treatment for Area 2. Vertical bars indicate standard error. C2: Control zone, HI2 Be: High-impact zone before trawling, HI2 Af: High-impact zone after trawling, LI2 Be: Low-impact zone before trawling, LI2 Af: Low-impact zone after trawling.

distribution of species between strata varied differently in each month (Pseudo-F = 1.9402, P = 0.0472) (Fig. 6). In addition, the richness varied differently for each treatment at each month (Pseudo-F = 1.7806, P = 0.0088). The Stratum 0–5 presented higher richness and was more variable within months than within treatments. The Control zone tended to exhibit less or equal richness than the impacted ones. In 36% of sampling events along the three years, richness decreased after high impact trawls on the Stratum 0–5, loosing between 1 and 3 species, and at 29% of experiment replicates on Stratum 5–10 (Fig. 6).

Species richness was higher at Area 2 and ranged from 0 to 10 species per sample, although it was less variable than Area 1. Higher richness occurred generally in the Stratum 0–5, with lower richness in 2015 (Pseudo-F = 885.39, P = 0.0002). Significant differences were detected regarding the interaction of treatment, stratum and season (Pseudo-F = 2.612, P = 0.0126). Differences after trawling occurred mainly in autumn, with decreases on species richness after a high impact trawling in few occasions: four in Stratum 0–5 (May 2015, January 2016, April 2016, June 2016) and five on Stratum 5–10 (April 2015, May 2015, January 2016, April 2016, March 2017) (see numbers above columns on Fig. 7).

3.4.3. *Species diversity and evenness*

No differences among years for both diversity and evenness were observed. Shannon diversity index presented no significant differences between treatments for any area (Area 1 mean H' = 0.85 ± 0.45; Area 2 mean H' = 0.86 ± 0.41). The diversity was always higher in Stratum 0–5 in the Area 1 (Pseudo-F = 21.174, P = 0.0398; H' = 1.08 ± 0.32) when compared to the Stratum 5–10 (H' = 0.62 ± 0.45). The same trend was observed for the Area 2, with higher values in the Stratum 0–5 (Pseudo-F = 57.694, P = 0.0118; H' = 1.09 ± 0.30) and lower in the Stratum 5–10 (H' = 0.63 ± 0.37). No significant differences were recorded between seasons in Area 1 (Summer H' = 0.71 ± 0.5; Autumn H' = 0.99 ± 0.34). In Area 2, significant differences within seasons (Pseudo-F = 8.69, P = 0.0078) were observed (Summer H' = 0.93 ± 0.42; Autumn H' = 0.79 ± 0.39).

The evenness presented no significant differences between treatments for any area (Area 1 mean J' = 0.68 ± 0.31; Area 2 mean J' = 0.60 ± 0.23). There were no differences in the stratification at the Area 1 (Stratum 0–5 J′ 0.79 ± 0.31; Stratum 5–10 J′ 0.56 ± 0.31) and Area 2 (Stratum 0–5 J′ 0.67 ± 0.23; Stratum 5–10 J′ 0.53 ± 0.23). No differences were observed for seasons at the Area 1 (Summer J′ 0.62 ± 0.31; Autumn J′ 0.74 ± 0.31), and Area 2 (Summer J′ 0.64 ± 0.23; Autumn J' 0.56 ± 0.23).

3.4.4. *Macrobenthic assemblage structure*

At Area 1, the Cluster analysis highlighted similarities between the High -impact and Low-impact zones before trawling, grouping those treatments with the Low-impact zone after trawling. A second group was formed by the samples of the Control zone and High-impact zone after trawling. At Area 2, the Control zone separated from the Low-impact and High-impact zones, which formed a cluster with high similarity between before and after trawling (Fig. 8).

A marked seasonal variability was observed for Area 1 (Pseudo-F = 2.2681, P = 0.027), with the highest abundances occurring in autumn. The treatment response of the assemblage structure (Pseudo-F = 1.2338, P = 0.028) and the stratification (Pseudo-F = 3.6269, P = 0.001) exhibited a monthly variation. The temporal variability of the dominant species presented different responses considering the treatments (Fig. 6). Only 4 of the 10 species in this area presented significant differences within treatment at some level: *Erodona mactroides* showed differences in abundances between treatment, month and strata (Pseudo-F = 2.5043, P = 0.0002), with differences restricted to the Stratum 0–5. In June 2015 *E. mactroides* densities increased in both impacted zones after trawling, but this increment in the Low-impact zone implied in significant differences with the Control zone

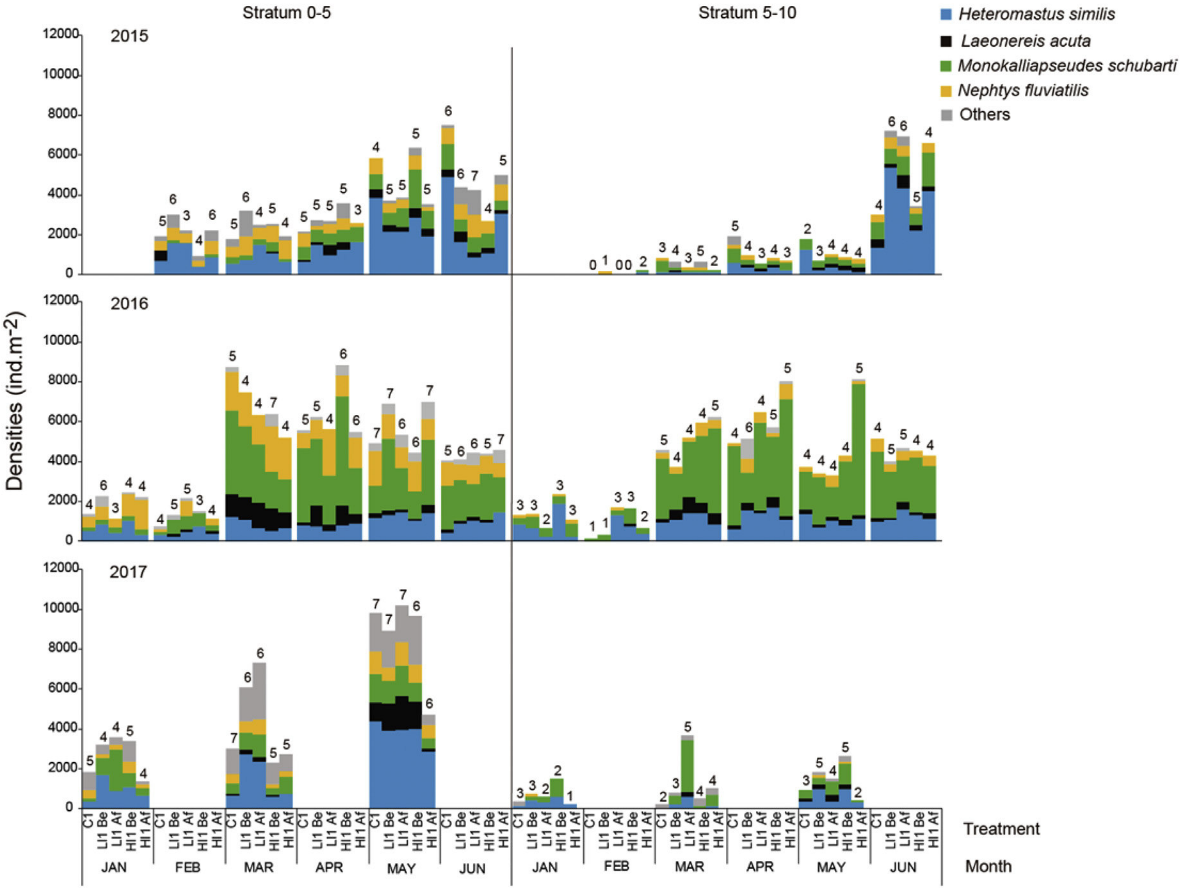

Fig. 6. Monthly mean density of species found in each treatment along the three sampled years in the Area 1. Numbers above columns indicate species richness. C1: Control zone, HI1 Be: High-impact zone before trawling, HI1 Af: High-impact zone after trawling, LI1 Be: Low-impact zone before trawling, LI1 Af: Low-impact zone after trawling.

(t = 3.7262, P = 0.021). In January 2017, densities decreased significantly after high-impact trawls (t = 4.0508, P = 0.0154). *Heleobia australis* showed different responses to treatment by season (Pseudo-F = 5.9964, P = 0.0002), with significant decreases of densities in autumn of 2017 after both high-impact (t = 2.6737, P = 0.0268) and low-impact (t = 9.1924, P = 0.0002) trawling. Also, responses in the interaction of season, treatment and stratum were detected (Pseudo-F = 5.4449, P = 0.0002), with differences occurring only in the Stratum 0–5. Similarly, significant differences in *Heleobia charruana* densities for the interaction of treatment and season were estimated (Pseudo-F = 2.1867, P = 0.0388). The species was recorded only on summer of 2015 and decreased its densities after low-impact trawling. *Laeonereis acuta* also showed different responses to trawling treatment in each stratum by season (Pseudo-F = 3.0158, P = 0.0078). Even when pairwise tests did not detect significant differences, decreases on densities were observed on both seasons of 2015 and 2017, and in summer of 2016 on Stratum 0–5 after high-impact trawls. We observed marked decreases of *Monokalliapseudes schubarti* densities on autumn of 2015 (15%) and 2017 (60%) after high-impact trawling, but non-significant effects of this treatment were detected.

Monthly variation in each stratum was detected at Area 2 (Pseudo-F = 1.9864, P = 0.001). Higher densities and richness were observed on the Stratum 0–5, excepting April and May of 2015 (Fig. 7). The assemblage structure and densities of the dominant species presented variability between months and a marked seasonal variability. When analyzing species separately, significant differences on densities were detected of *Heteromastus similis* within treatment (Pseudo-F = 4.9073, P = 0.0126), with decreases after high-impact trawling, remaining significantly different from Control zone (t = 4.6444, P = 0.015).

4. Discussion

Estuarine assemblages are adapted to deal with a very dynamic ecosystem in which physicochemical characteristics vary in time and space, often presenting unpredictable environmental scenarios (Elliott and Quintino, 2007; Alves et al., 2015; Rehitha et al., 2017). The present study highlights the role of environmental conditions on the macrofauna of a subtropical estuary, which can promote variations in benthic assemblages through time and space. The responses of the macrofauna to different intensities of trawling impact, when detected, were related to the temporal trends of the species composition, abundance, and granulometric characteristics, but also on their vertical distribution.

In the shallow estuarine zones assessed in this study, the benthic macrofauna is subjected to a dynamic habitat, where the sedimentation process related to winds and currents may change in scale of days. The variability of topographic profiles of this shallow mudflat is sometimes up to 10 cm, associated to erosion and deposition processes of mud and sand sediments, with autumn being the more dynamic season (Colling et al., 2007). These erosional and depositional processes can be intensified depending on the wind speed and direction (Souza, 2002), with higher dynamic in the Area 2. Moreover, the occurrence of ENSO-El Niño events in 2015 and 2016 (NOAA, 2017) resulted in a long rainy period in southern Brazil. In consequence, a greater discharge of freshwater drains from de Patos Lagoon drainage basin to the estuarine region. These events of higher freshwater discharge imply limnetic scenarios and a greater input of silt and clay to the estuary (Tomazelli and Villwock, 2000), increasing the mud content in the sediments of shallow soft bottoms. Considering the important role of these natural

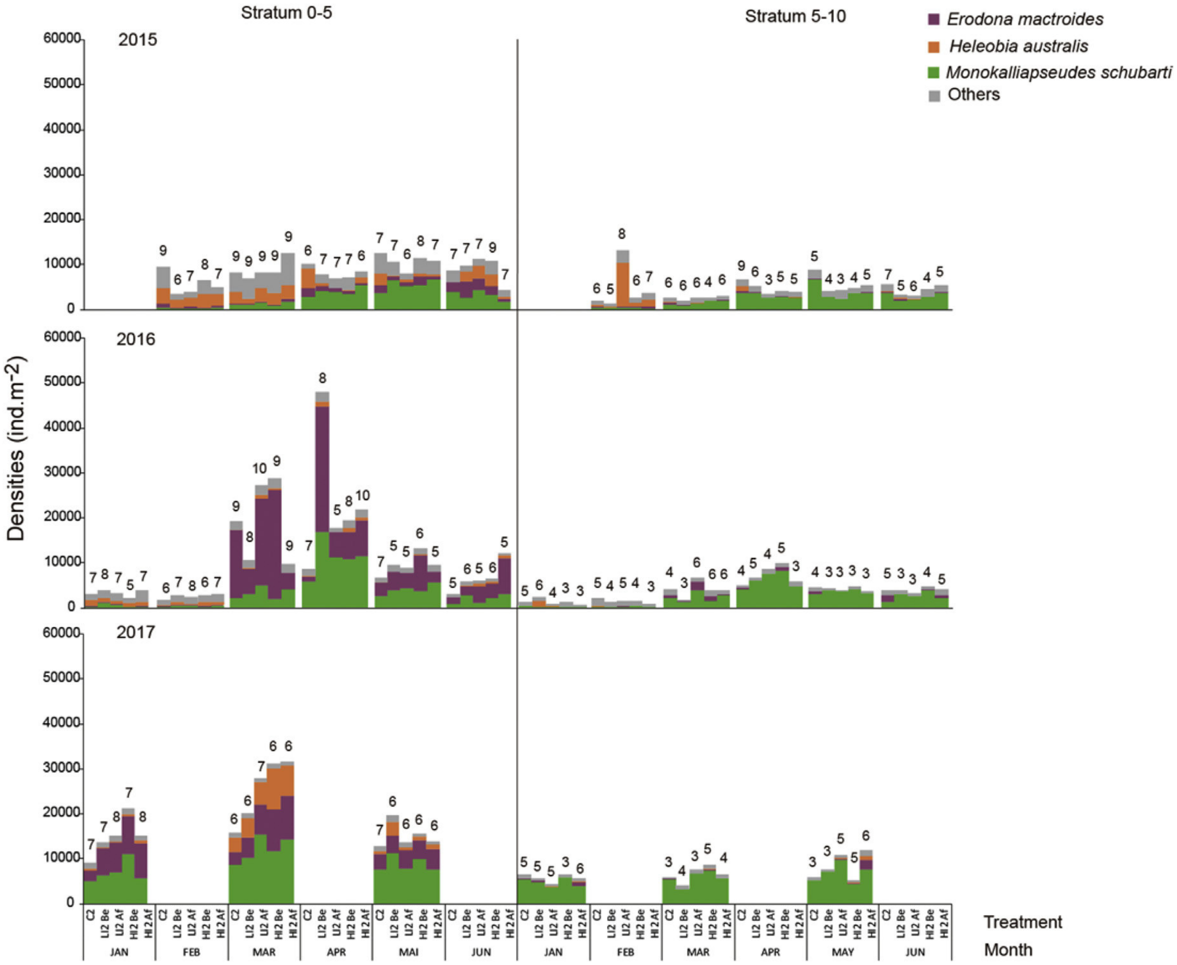

Fig. 7. Monthly mean density of species found in each treatment along the three sampled years on Area 2. Numbers above columns indicate species richness. C2: Control zone, HI2 Be: High-impact zone before trawling, HI2 Af: High-impact zone after trawling, LI2 Be: Low-impact zone before trawling, LI2 Af: Low-impact zone after trawling.

disturbances, it is necessary to recognize a possible overlap between the effects of trawling impact and the natural physic disturbances, both contributing to the temporal variation of benthic richness and diversity.

Trawling activities impacted the total densities and assemblages structure mainly in the sheltered area near the Pólvora Island (Area 1). Furthermore, the physical disturbance caused by trawling varied markedly between both areas and seems to be primarily related to the sediment structure: Area 1 presented higher contents of silt, clay and very fine sands, while Area 2 was characterized by a coarser granulometry. As seen by using a side-scan sonar (Fig. A1), marks in the soft

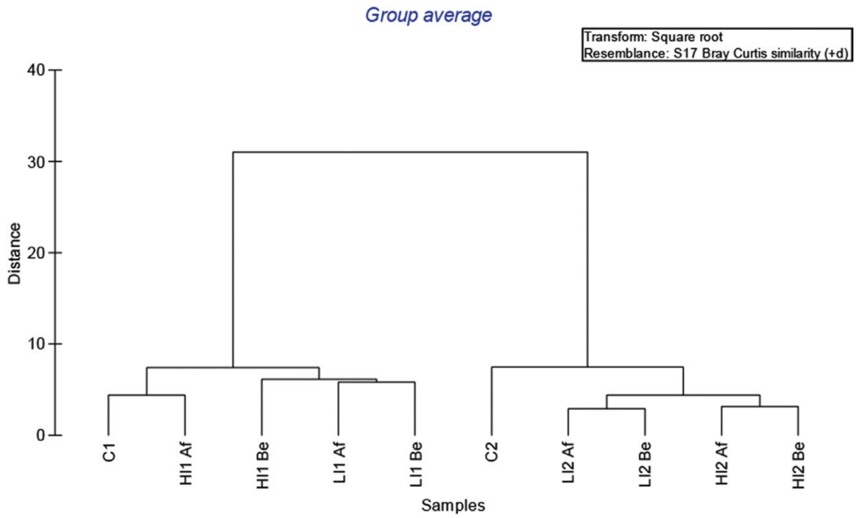

Fig. 8. Dendrogram showing differences between treatments for both areas.

bottom left by otter trawls remained visible in the majority of cases at Area 1. Conversely, the tracks left by otter trawls were immediately filled by neighboring sediments at Area 2. Similar patterns of higher impacts of trawling on zones characterized by lower levels of natural disturbance were also observed by Brylinsky et al. (1994), Jennings et al. (2001), Hiddink et al. (2006) and Prantoni et al. (2013). For instance, in the Bay of Fundy, Brylinsky et al. (1994) detected that direct impacts on benthic fauna were caused mainly by doors rather than rollers, with marks up to 5 cm depth made by trawl doors of 180 kg. We could not measure the depth of trawl marks, however, if a 180 kg door left marks of 5 cm depth, we suppose that our 15 kg doors will impact a more superficial sediment layer. These observations could imply that the sediment resuspension visually observed during trawling action could not be enough to result in granulometric changes in the analyzed strata. This may also explain the higher signals of impact over the macrobenthos of the Stratum 0–5 and the maintenance of vertical stratification.

Evaluating the effects of dredging in soft bottoms of the PLE has highlighted the efficient strategies of resilience performed by the dominant estuarine species. Macrobenthic invertebrates performed rapid recolonizations after dredging impacts in a scale of months (Bemvenuti et al., 2005). The same pattern was observed after the impact of a sulphuric acid spill on macrobenthic fauna of PLE, which presented clear recovering after six months (Bemvenuti et al., 2003). This evidence of fast macrofaunal recovering suggest an absence of cumulative effects in our experiments through subsequent years, as well as on intra-annual scales.

The faunal response to the trawling disturbance depends on the biology and behavior of each species. Trawling activities can carry or displace macrofauna as a result of the physical disturbance created by the trawl gear (Dell et al., 2013; Johnson et al., 2014; Collie et al., 2016). Thus, epifaunal weightless species, such as peracarid crustaceans probably are more susceptible to resuspension and displacement by currents than heavier organisms such as mollusks. Tube-builders such as *Laeonereis acuta* and *Monokalliapseudes schubarti*, that build tubes deeper than 5 cm may sense the approach of doors and move down into their tubes avoiding the impact, as suggested by Brylinsky et al. (1994) for tube-dwelling polychaetes. This evasive behavior may explain increasing densities on the Stratum 5–10 after trawling. Considering the importance of the deposit feeder *L. acuta* and deposit/suspension feeder *M schubarti* as a main prey for fishes, crustaceans and birds in the PLE (Bemvenuti and Colling, 2010), any anthropogenic impact over their populations and therefore, food availability to food webs need to be carefully evaluated.

The gastropod *H. australis* and the bivalve *E. mactroides* presented similar responses to trawl fisheries, with impact related to periods of their higher abundances. The gastropod showed decreasing densities after the low and high impact trawling of 2017, while the bivalve presented increasing and decreasing densities after both trawling intensities during some months of 2015 and 2017. The temporal overlapping of fisheries activity and higher abundances of *H. australis*, for example, which presents recruitments and faster growth during warming months (Carcedo and Fiori, 2012), represents a key point for the sustainability of the estuarine food webs. The importance of this fauna as food items for crustaceans and fishes was recognized for *H. australis* (Carcedo and Fiori, 2012) and *E. mactroides* (Oliveira et al., 2006), with these species being the main prey among all macrobenthic species for the crab *Callinectes sapidus* (Oliveira et al., 2006).

Despite the lack of a consistent pattern on the response of macrobenthic species to trawling fisheries, we showed some evidence of trawling impact in the macrozoobenthos and bottom. These impacts were shown through the application of experimental tows with a single boat once in a month. Extrapolating these observations to the whole fisheries scenario of the PLE, where Kalikoski and Vasconcellos (2012) suggest that at last 370 boats (30% of the total number of motorized boats in the estuary) are equipped to conduct trawls, the environmental impacts will be exponentially higher.

Trawling on shallow estuarine areas that are recognized nursery grounds for fishes (Costa et al., 2016) and shrimps (Noleto-Filho et al., 2017) may not affect only the target species, but the whole estuarine communities. This potentially affects the food availability for this delicate food web. A better understanding of the impacts caused by trawling fisheries and the ecosystem variability is an important tool for the improvement of fisheries management. Therefore, this assessment of ecological impacts and adequate dimensioning of anthropogenic activities is vital information for addressing effective governmental policies for the protection and conservation of estuarine nursery areas. For instance, our results may be used to determine priority areas for conservation, establishing closed areas based on their susceptibility to fisheries activities.

The different responses of macrobenthic assemblages highlight the relation between the detection of anthropogenic pressure and the natural variability of the ecosystem. The detection was closely related to the granulometric properties of the sediment which suggest a demand of studies evaluating trawling impact in different estuarine habitats, taking into account the hydrodynamics and the spatio-temporal variability of fauna and environment.

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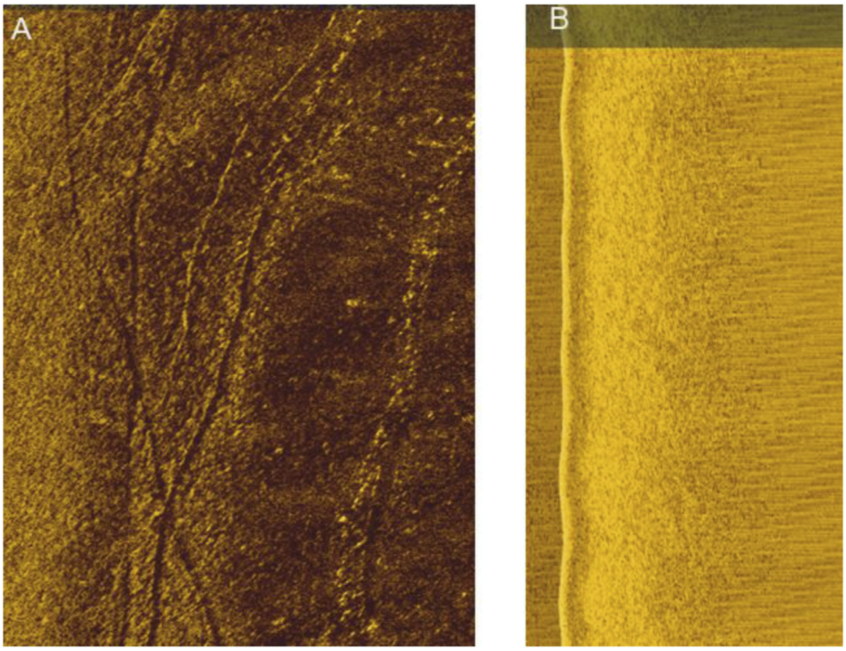
Appendix Figure A1



Imagens of the side scan sonar over the fishing experimental zones of the Patos Lagoon estuary. A) Tracks left by trawl gears on Area 1 and B) absence of tracks on Area 2.

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Apêndice 4.

Esse apêndice refere-se ao artigo intitulado “Responses of the infaunal peracarid *Monokalliapseudes schubarti* (Tanaidacea: Kalliapseudidae) to bottom trawling disturbance and seasonality in an estuary of Southern Brazil” submetido na revista Marine Biology Research

Responses of the infaunal peracarid *Monokalliapseudes schubarti* (Tanaidacea: Kalliapseudidae) to a bottom trawling disturbance and seasonality in an estuary of Southern Brazil

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Responses of the infaunal peracarid *Monokalliapseudes schubarti* (Tanaidacea: Kalliapseudidae) to a bottom trawling disturbance and seasonality in an estuary of Southern Brazil

Trawl fisheries are known to have negative effects on benthic assemblages. The infaunal tanaid *Monokalliapseudes schubarti* is one of the dominant macrobenthic species in the shallow mudflats of southern Brazil and is thus susceptible to the impact of trawling. Furthermore, *M. schubarti* plays an important role in local trophic webs as a primary consumer and as a prey. This work investigated 1) the temporal variations in the population densities and structure of *M. schubarti* in the Patos Lagoon Estuary and 2) the potential impacts of shrimp trawling on the population densities, population structure, and vertical stratification of the species in two shallow areas of the estuary. In the years sampled, the species exhibited significant temporal variability in its total densities and population structure, with a reproductive period observed only during late summer and autumn, following an increase in salinity within the estuary. There were significant differences in *M. schubarti* densities, which appeared to be associated with variations in sediment texture and salinity. No well-defined pattern of trawling impact was identified; however, after high-impact trawling, decreases in the abundance of males and juveniles in the 0-5 cm sediment surface layer were recorded, and ovigerous females disappeared from the sampled area with relatively higher percentages of medium sand. Changes in the densities of different morphological categories observed after trawling were likely due to mortality, resuspension, or rapid movement deeper into their tubes.

Keywords: macrobenthos; tanaid; population structure; disturbance; experimental fishery

Introduction

The use of towed bottom-fishing gears is among the most harmful anthropic activities to the assemblages of benthic fauna and their habitat (Kaiser et al. 2002; Brown et al. 2005; Hiddink et al. 2007; Lambert et al. 2014; Mangano et al. 2015; Sciberras et al. 2016). The use of such gears can result in the suspension of sediments and contribute to changes in macrofaunal bioturbation, benthic organisms mortality, and changes in population demography (Engel & Kvitek 1998; Brown et al. 2005; Mangano et al. 2013; Mangano et al. 2014; Sciberras et al. 2016).

Trawls are the most common gear used in shrimp fisheries and support a substantial portion of the ~3.5 million tonnes of shrimp harvested each year (FAO 2016). Shrimp fisheries exploit resources at the industrial scale in oceans zones and at small scales in estuaries and coastal zones. Although small-scale fisheries provide approximately 50% of the global fish capture and two-thirds of the catches for direct human consumption (FAO 2017), the ecological impacts of these fisheries have received little attention (Costa & Netto 2014).

Studies of the impact of trawl fisheries on macrobenthic communities are scarce in Brazil. Pelaes & Borzone (2007) analysed the disturbances caused by shrimp trawling on the inner continental shelf of Paraná State by comparing areas currently trawled with areas where trawling has ceased for some years. They found no significant differences in any ecological descriptor between trawled and non-trawled areas, differences were just in the temporal patterns. Prantoni et al. (2013) evaluated the inner continental shelf off Paraná State and did not observe consistent effects on macrobenthic assemblages before and after trawling. The only species that exhibited some evidence of being affected was the tanaid *Monokalliapseudes schubarti*, which displayed a density increase after trawling. Costa & Netto (2014) analysed the impact of a mini trawl in an estuary on vegetated and non-vegetated bottoms and concluded that trawling did not produce a loss of benthic diversity and caused minimal damage to the estuarine benthos. They found a significant decrease in the total meiofauna after trawling and lower densities of nematodes and ostracods in non-vegetated areas.

Bottom trawling is prohibited within the Patos Lagoon Estuary (PLE) in southern Brazil but are widely used in pink shrimp *Penaeus paulensis* fisheries. Only passive nets (fyke nets) are allowed in the PLE, but artisanal bottom trawls are widely used (Benedet et al. 2010) due to a lack of effective control by fishery management authorities. This estuarine-dependent species uses the estuary as a nursery ground and constitutes the most important resource for artisanal fishing in the region (Moller et al., 2009). The fishery season encompasses the austral warmer months and early autumn, ranging from February to May (D'Incao 1991; Benedet et al. 2010), and regulation is based on the monitoring of shrimp size (Kalikoski and Vasconcellos, 2012). In the PLE, the recruitment of macrobenthic species intensifies during late spring and summer (approx. November to April)

(Bemvenuti & Netto 1998; Rosa & Bemvenuti 2006). The simultaneity between the fishing season and the recruitment season highlights the potential impacts of trawling fisheries on macrobenthic assemblages.

The tanaid *Monokalliapseudes schubarti* (Mañé-Garzón 1949) is a peracarid crustacean endemic to the southern Atlantic (Rosa-Filho & Bemvenuti 1998). The species is one of the dominant components of the macrobenthic assemblages in the shallow coastal regions of southern and southeastern Brazil (Leite et al. 2003; Colling et al. 2007; Neves & Valentin 2011; Freitas-Júnior et al. 2013). Moreover, *M. schubarti* plays an important ecological role in the transfer of energy from detritus to higher trophic levels. This tanaid is the main prey of many fish and crustaceans species, including important fisheries resources such as the blue crab *Callinectes sapidus* Rathbun, 1896 and the pink-shrimp *Penaeus paulensis* (Pérez Farfante, 1967) (Bemvenuti & Colling 2010), and feeds as a deposit and filter feeder (Montagnolli et al. 2004; Fonseca & D'Incao 2006; Freitas-Júnior et al. 2013). The peracarid *M. schubarti* continuously reproduces throughout the year, with peaks in the spring and summer (Fonseca & D'Incao 2003; Freitas-Júnior et al. 2013). Therefore, the impact of trawl fisheries occurs in critical periods of reproduction for the macrobenthic community. This tanaid exhibits a vertically stratified size structure, with juveniles burrowing less than adults (Rosa-Filho & Bemvenuti 1998). Because fertilization occurs inside the tubes individuals inhabit, males must leave their tubes to find females (Freitas-Júnior et al. 2013), which makes them more susceptible to predation, dispersal by water turbulence, currents and trawling impacts.

This study investigated the population variability of *M. schubarti* and the potential impacts of trawling fisheries on the species densities, population structure, and vertical stratification within the sediment in the shallow areas of the PLE. Specifically, we evaluated 1) the relationships among seasonal variations in population parameters and variation in both water physico-chemical properties and sediment properties and 2) the effects of trawling on the total densities, population structure and vertical stratification of the species. We hypothesize that the population densities and reproductive period would exhibit similar trends and present positive correlations with both temperature and salinity, as these parameters display a marked seasonality, with increasing values during summer months. In addition, we hypothesize that trawling activities would reduce the overall population densities, and alter the vertical stratification of the species, with the strongest effects on juveniles and males due to their preference for strata near the sediment surface.

Materials and methods

Study area

The Patos Lagoon is a choked lagoon located in the southern Brazilian coastal plain (Kjerfve 1986), and it extends in a NE-SW direction from the city of Porto Alegre (30º30'S, 51°13'W) to Rio Grande (32º12'S, 52°5'W). Saco do Arraial is an inlet in the western margin of the PLE (Figure 1). Although this inlet is sheltered from high-energy events in the lagoon, the high incidence of winds and its shallow depth result in dynamic erosion, sediment transport, and sediment deposition processes (Souza & Hartmann 2008). Two areas in the Saco do Arraial with depths of 1.3–2.5 m were selected for the current study: 1) a more sheltered area near Pólvora Island (Area 1) and 2) a more exposed area near Cavalos Island (Area 2) (Figure 1). Both areas are commonly used by artisanal fishermen who employ a variety of fishing gear, including towed bottom trawls.

Sampling methodology

The tanaid M. schubarti was sampled across five zones of Area 1 (Zones 1 and 2) and Area 2 (Zones 3, 4, and 5) (Figure 1). In each zone, three sediment samples were collected with a *van Veen* grab. From each grab, one subsample was collected with a 10 cm-diameter core (0.0078 m^2) and sectioned into 0-5 cm and 5-10 cm strata. Sampling was performed in 2015 from the oceanographic vessel *Larus* (Universidade Federal do Rio Grande) three times per season, except in the spring, when only one sampling campaign was conducted. To assess the impacts of trawling, additional 20 x 20 m zones were marked in each area with wooden sticks, one to simulate a high impact trawling and the other to simulate a less intensive impact. Simulations were performed based on five continuous tows in each of the high-impact zones (High-Impact 1 (HI1; Area 1) and High-Impact 2 (HI2; Area 2)) and two continuous tows in the low-impacted zones (Low-Impact 1 (LI1; Area 1) and Low-Impact 2 (LI2; Area 2)) (Figure 1). Simulations were conducted with an otter trawl net (mesh size of 12 mm between opposite knots and 15 kg boards) deployed from an 8 m artisanal fishing vessel, with the net towed at a speed of approximately 2 knots. Three biological samples were collected before and after trawling using the methodology described above, and sampling was repeated monthly between February and June (2015).

In some field trips the vessel was outfitted with Lowrance HDS 9-Gen 2 side-scan sonar with a transducer programmed to take images at 600 kHz. The bottom images were captured prior to the trawl impact in all zones of each area, followed by data collection during and after trawling.

Samples were sieved with a 0.3 mm mesh net on the vessel (according to the juvenile tanaid size) and fixed in 4% formaldehyde. Approximately 70% of macrofauna were constituted by *M. schubarti*, highlighting its importance as a dominant species in the study area. Four morphological categories were considered: males, females, ovigerous females and juveniles. The males were distinguished from females based on propodus morphology (Mañé-Garzón 1949; Figure 2). Individuals with a total length of less than 2.5 mm were defined as juveniles unless they exhibited secondary male characteristics. Ovigerous females were identified by the presence of a marsupium (Fonseca & D'Incao 2003).

During each sampling event, the parameter temperature was measured using a mercury thermometer (°C), and salinity was recorded using an optic refractometer. In addition, two sediment samples were collected in each zone for the granulometric analysis. The analyses of grain sizes were performed by dry mechanical sieving through a column of sieves from 4 mm to 0.063 mm. This process followed the Wentworth classification system at intervals of one phi and with pipetting for grain sizes < 0.063 mm (Suguio 1973). Grain size composition was expressed as a percentage of the total sample weight. Wind speed data were provided by the National Meteorology Institute (Instituto Nacional de Meteorologia). Wind speed was measured hourly and averaged over each field day.

Statistical analysis

In the non-impacted zones, the experimental design consisted of five factors: Area (two levels, fixed), Zone (five levels, random, nested in Area), Stratum (two levels: 0-5 and 5-10 cm, fixed), Season (four levels, fixed) and Sampling date (random; 9 field trips between February and November, nested in Season). The *M. schubarti* densities exhibited considerable variability among samples and a predominance of zero values, making traditional analyses (which assume normality of errors) unsuitable. Thus, non-parametric approaches were used (Anderson & Millar 2004). Permanova was used to analyse the univariate data set (total densities and densities of each category) according to the complete experimental design (including interactions). For each factor, 4999 permutations of raw data units were applied to obtain P-values (Anderson 2005). As for some factors there was a low number of possible permutations, we used the Monte Carlo P-value (Anderson & Robinson 2003, Anderson 2005). These permutation tests assume not only exchangeability but also that a linear model on dissimilarities which is appropriate for choosing reasonable statistical tests analogous to those used in univariate ANOVA (Anderson & Millar 2004). Permanova was conducted using a Euclidian distance matrix of the total and morphological categories densities. The significance of P-values was set at 0.05. The significant results were evaluated via *a posteriori* pairwise comparisons, which also used 4999 random permutations to obtain P-values through Monte Carlo correction. Spearman correlation analyses were performed to evaluate the relationships between the total densities and morphological categories of *M. schubarti* and the environmental parameters.

The effects of trawling impact on total densities/morphological categories was tested with a univariate Permanova through the following model: Zone (fixed; Low-impact, High-impact), Time (fixed; before, after) Stratum (fixed; 0-5 and 5-10 cm) and Month (random; February to June). All tests were performed using a permutation procedure (with 4999 permutations of appropriate units), as described for the Permanova above. The analyses were performed using the Primer V6 software package with the PERMANOVA add-on (Anderson et al. 2008) and R V3.4.1 software (R Core Team 2017).

Results

Environmental variables

Water temperature ranged from 10°C to 25.7°C over the study period, with an average of 24.09 ± 0.64°C in the summer, 16.35 ± 2.52°C in the autumn, 12.31 ± 1.19°C in the winter, and 17.70 ± 0.25°C in the spring. Although the samples were collected in an estuary inlet in the middle estuary, the mean salinity was very low throughout most of the year, with high values recorded only in autumn (0.48 ± 0.35 in the summer, 26.36 ± 6.27 in the autumn, 3.16 ± 2.09 in the winter, and 0.18 ± 0.17 in the spring).

The non-impacted zones in Area 1 (Zones 1 and 2) were composed of more than 70% of fine and very fine sands, with Zone 2 having a 1-10% higher percentage of very fine sediments (silt + clay) (Figure 3). Within Area 2 (Zones 3, 4, and 5), the grain size distribution was more variable among zones. Zone 3 was composed of more than 80% medium and fine sands, whereas Zones 4 and 5 exhibited percentages of medium and fine sands of 18-48% (Figure 3). Higher percentages of silt and clay were observed in the winter and spring (Figure 3). In both areas, there were significant effects of zone, sampling date, and their interaction on grain size composition ($P<0.005$). There were no significant differences within strata. Zones with fishing simulations in Area 1 (High-

Impact and Low-Impact zones) displayed grain size distributions similar to that of Zone 1, whereas in Area 2, the High-Impact and Low-Impact zones exhibited characteristics similar to those of Zone 3.

Side-scan sonar images

The side-scan sonar images illustrated that the ground marks left by the trawl net were deeper and remained continuous throughout almost the entire trawl track in Area 1(Figure 4). After trawling these marks were still visible on the bottom. However, in Area 2 the remaining marks were inconsistent and only visible along a few parts of the track (Figure 4) and they quickly filled with sediment.

Population structure in non-impacted zones

A total of 12916 individuals of *M. schubarti* (mean density ± standard deviation: 2383.36 ± 3120.19 ind.m^{-2}) were identified within the five zones. Among them, 7041 (54.5%) individuals were females, 105 (0.8%) were ovigerous females, 4095 (31.7%) were juveniles, and 1343 (10.4%) were males. Some adult individuals (2.6%) could not be sexed because of a lack of diagnostic characteristics. The ratio of females to males was 5:1.

The total densities in both areas were significantly different (Pseudo-F= 8,9662, P=0.026), with higher densities in Area 2 (Figures 5 and 6). The data exhibited clear and significant seasonality (Pseudo-F= 8.5785, P=0.002), with low densities in the summer and increases in autumn followed by decreases in the winter and spring. The densities were lower in the spring than in the summer (Figures 5 and 6). The total densities differed significantly among the different zones of each area and for different strata and sampling dates (Pseudo-F= 2.3368, P=0.0052). The results of the analysis of the population structure of *M. schubarti* were very similar to those of the total density, with significant variable effects in the interaction of the factors zone, stratum and sampling date (P<0.025) for all morphological categories. When separately analysing the females densities (ovigerous and not ovigerous) based on the stratum depth, no significant differences were observed. However, there were significant differences in juveniles (Pseudo-F= 7.1221, P=0.014) and males (Pseudo-F= 6.3392, P=0.023) abundance by stratum. Ovigerous females were recorded only in the summer and autumn, with higher densities in the latter season, and they were more abundant in the 5-10 cm stratum. Males occurred throughout the year, with the highest densities observed in the autumn and winter, and they generally were more abundant in the 5-10 cm stratum. Juveniles were found in both strata, with a higher abundance in the 0-5 cm stratum, and they exhibited density peaks in the autumn and winter (Figures 5 and 6).

Among the environmental variables, salinity displayed a significant and strong correlation with density in all population categories (Table I). Salinities below 5 were associated with low species densities. Wind speed was negatively correlated with density in almost all population categories (Table I). Temperature was significantly and positively correlated with the density of ovigerous females. For the grain size distribution, the percentage of medium sand appeared to have the strongest influence on density, with high positive correlations observed in all population categories. The percentages of the finest sediments (very fine sand, silt and clay) had negative correlations with the density of *M. schubarti* (Table I). High species densities were observed in zones with more than 7% medium sand and less than 15% very fine sediments (very fine sand, silt and clay combined).

Impacts of trawl fisheries

A total of 12937 individuals (1882.50 ± 2818.80 ind.m^{-2}) were identified from the four zones (High and Low-Impact zones of both areas): 7265 females, 116 ovigerous females, 3885 juveniles, and 1513 males.

In Area 1, the response of the total density of *M. schubarti* to trawling was different among months in each zone and stratum (Pseudo-F= 2.8531, P=0.0322). Pairwise tests showed that the densities significantly varied with time (before *vs.* after) in Stratum 0-5 of the Low- impact zone only in May (P=0.0366). In Area 2, the total densities were significantly affected by month, stratum and their interaction (P=0.0002). The densities were generally higher in Stratum 0-5 and in autumn months.

The population structure in Area 1 significantly varied within strata and months. The ovigerous females densities varied significantly within months, zones, time and stratum (Pseudo-F= 3.6266, P= 0.0084), but pairwise tests did not detect significant differences between before and after trawling for any combination of these factors. The juveniles densities varied significantly based on the zone, time and stratum interaction (Pseudo-F= 6.3046, P=0.016) and the response to trawling was different within months (Pseudo-F= 3.3889, P=0.011). Pairwise tests did not detect significant differences for any combination of significant factors. The males densities in response to trawling was different in each zone and stratum (Pseudo-F= 4.6218, P= 0.0356), without detectable differences in the pairwise tests. In Area 2, the females densities exhibited a different response to trawling in each month (Pseudo-F= 2.5576, P=0.0432), with a significant decrease only in May (P=0.043). The response of ovigerous females differed in each zone and month (Pseudo-F= 3.8516, P=0.0062), but pairwise tests did not detect differences between before and after trawling for any combination of zone and month. Additionally, juveniles did not exhibit a significant response to trawling activities. The males response to trawling was different in each zone (Pseudo-F= 4.7245, P=0.029), but the pairwise test did not reveal differences between before and after trawling in any zone.

Considering only the autumn months when *M. schubarti* densities were higher, we observed some variable differences in total densities between before and after trawling, with increasing values in some cases and decreasing values in others (Figures 7 and 8). In the high-impact zones of both areas, the densities of males in the 0-5 cm stratum decreased after trawling in April, May and June. In the same stratum, juveniles occurred at lower densities after trawling in May and June in Area 1 and April and June in Area 2. Moreover, in Area 2 a total loss of ovigerous females was recorded after high-impact trawling during April and May, and no ovigerous females were observed in June.

Discussion

Population structure

The seasonal variations in the total density of *M. schubarti* observed in the present study coincide with the predicted fluctuations in macrobenthic species during high-rainfall periods. The pattern of salinity differed from the expected pattern due to the high rates of precipitation caused by an *El Niño* event (NOAA 2017), which increased the freshwater discharge from the Patos Lagoon drainage basin (Fernandes et al. 2002; Barros et al. 2014). The low densities of *M. schubarti* observed in the spring and summer coincided with those reported by Colling et al. (2007), who sampled during an *El Niño* event (2002-2003) and observed a similar seasonal pattern of macrobenthic assemblages. Similar temporal trends were observed in the Lavaca-Colorado estuary (Pollack et al. 2011) and the Gulf of Riga (Kotta et al. 2009). The pattern observed here is in contrast to that reported for years without the influence of *El Niño*, when macrobenthos densities were higher in the spring and summer in the PLE (Bemvenuti & Netto 1998; Rosa & Bemvenuti 2006) and other subtropical/temperate estuaries (Drake et al. 2002; Magni et al. 2015).

Salinity and temperature variations have significant effects on the growth, reproduction, and survival of macroinvertebrates due to changes in the metabolic rates caused by both osmotic and thermic stresses (Uwadiae 2009). In peracarids such as the amphipod *Gammarus lawrencianus* Bousfield, 1956, although adults can tolerate a wide range of salinities, they are unable to reproduce at low salinities (Steele & Steele 1991). Certain temperatures are important for the induction and termination of reproductive resting stages in the amphipod *Hyalella azteca* (Saussure) (Panov & McQueen 1998). Despite previous reports that *M. schubarti* can continuously reproduce over the entire year, with peaks in the spring and summer (Fonseca & D'Incao 2003; Freitas-Júnior et al. 2013), the extended periods of low salinity observed in the present study suggest that this behaviour is negatively influenced by salinity. Thus, it is likely that the low salinities and total species densities reflected in the absence of ovigerous females during spring. The ability of females to support different developmental stages in the marsupium (eggs, embryos, and mancas) (Fonseca & D'Incao 2003) might explain the occurrence of juveniles throughout the year.

The *M. schubarti* densities were negatively correlated with high percentages (>15%) of fine sediments (<125 mm), and higher densities were observed in sediments with a percentage of medium sand higher than 7%. As suggested by Rosa-Filho & Bemvenuti (1998), the sediment characteristics are a limiting factor for the occurrence of the species*,* that require a minimum percentage of silt and clay. Even if very fine sediments are required for tube stability, the presence of coarser sediments might increase the oxygenation of the substrate. Our results are similar to the findings of Leite et al. (2003) and Freitas-Júnior et al. (2013), who showed that *M. schubarti* densities were higher at low to intermediate percentages of silt and clay (4-10%) and lower at higher percentages (>13%).

Impacts of trawl fisheries

The response of *M. schubarti* was different among the sampled areas, suggesting that in shallow, highly dynamic estuarine areas, the effects of trawling vary with time and have interactive effects with natural disturbances. According to Mangano et al. (2013), the effects of trawling vary with bottom geology and the natural disturbance regime.

No well-defined pattern of trawling impact was identified, although some changes in both the total density and morphological categories of *M. schubarti* were observed before and after trawling. Our results suggest that high-impact trawling might affect the distributions of juveniles, which are more abundant in the surface sediment layers (0-5 cm) than in deeper layers (5-10 cm), and males, which leave their tubes in search of available females for reproduction. Trawl perturbations can carry or displace macrofauna as a result of the physical disturbance created by the trawl gear (Dell et al. 2013; Johnson et al. 2014; Collie et al. 2016). Thus, the reduction in *M. schubarti* densities in the surface layer might be the result of mortality at trawling sites or trawling-induced resuspension and displacement to other areas, together with resuspended sediment. The potential re-suspension of juveniles may hamper their survival capability in the first life stages, because they require parental care and are highly susceptible to disturbances. It is unknown whether these individuals can survive without the female progenitor care.

There were also differences between the two areas, with ovigerous females primarily impacted in the area composed of coarser sediments and more exposed to the hydrodynamics of the main channel of the lagoon (Area 2). However, males and juveniles were the categories more frequently affected in both areas. This finding suggests that trawl fisheries have a stronger impact on *M. schubarti* in sediments with relatively higher percentages of medium sand and affect nearly all components of the population. Side-scan sonar images showed that on bottoms with higher percentages of medium sand (impacted zones in Area 2), as the net skimmed the substrate, neighbouring sand filled the trawl track. Thus, we assume that organisms from the neighbouring substrate might be transported along with the sand filling the track. In areas with higher percentages of very fine sand, silt and clay (Area 1), tracks left by the net lasted longer than those in other areas. A similar observation was reported by Brylinsky et al. (1994), who observed that in the macrotidal estuary of the Minas Basin (Canada), the extent of the initial physical disturbance caused by trawling was greater in siltier sediments than in sediments characterized by relatively coarse sand. They also suggested that tube-dwelling polychaetes, which inhabit tubes that extend deeper than 5 cm, might move down in their tubes when they detect an otter trawl approaching. We assume that the lower densities of *M. schubarti* adults observed after trawling might reflect evasive action similar to that of tube-dwelling polychaetes.

One of the greatest challenges in understanding the impacts of trawling on *M. schubarti* is their patchy distribution (Freitas-Júnior et al. 2013). This type of distribution, which is very common in benthic species (Dole-Olivier & Marmonier 1992; Barnes and Ellwood 2012; Beseres & Feller 2007), might have influenced our results regarding the effects of trawling because some patches of organisms might be sampled while others are not, regardless of the disturbance. In addition, particularly for a species such as *M. schubarti* that requires parental care and has a marsupium that can hold up to 87 eggs or 47 mancas at one time (Fonseca & D'Incao 2003), each reproductive event of a female will produce a patch of organisms. This feature might also be associated with random increases in the number of individuals collected per sampling unit, as also detected by Prantoni et al. (2013) on the inner shelf of Paraná state (Brazil).

In conclusion, the main environmental factors that influenced *M. schubarti* were sediment characteristics and water salinity. The grain size composition might determine the species abundance, with individuals preferring muddy sand bottoms with relatively high content of medium sand. Furthermore, salinity might influence the reproductive cycle and abundance of *M. schubarti*. Bottom trawling did not have a clear impact on the total density or population structure of the studied tanaids. However, lower densities of males and juveniles in the 0-5 cm stratum were recorded after high-impact trawling, and ovigerous females disappeared from bottoms with coarser sands. These varied responses of tanaids total densities and in the morphological categories to bottom trawl disturbances highlight the importance of future studies concerning the impacts of fisheries on benthic populations, in order to identify consequences for key species and possible implications for assemblages, communities and ecosystems.

Benthic macrofauna provide a necessary food source for many aquatic resources such as crabs (Branco et al. 2002; Oliveira et al. 2006) and shrimps (Soares et al., 2005); therefore, their abundance can influence the trophic structure in coastal ecosystems (Posey et al. 2002; Harmelin–Vivien 2009; Possamai et al. 2018). This study evaluated the trawling impact in a recognized nursery area of living resources including some of commercial interest. Thus, any evidence of impacts should be carefully evaluated because it might cascade down trophic webs.

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Table I. Spearman correlation coefficients of environmental variables and the densities of different population categories of *M. schubarti*. * Significant correlation.

	Females		**Ovigerous females**		**Juveniles**		**Males**		**Total**	
	Rho	**p-value**	**Rho**	**p-value**	**Rho**	**p-value**	**Rho**	**p-value**	**Rho**	**p-value**
Temperature	-0.305*	2.16E-14	0.289*	5.62E-13	-0.102*	0.01	-0.157*	1.13E-04	-0.246*	9.38E-10
Salinity	0.413*	2.20E-16	0.142*	4.60E-04	0.435*	2.20E-16	0.312*	5.04E-15	0.452*	2.20E-16
Wind speed	-0.248*	5.11E-09	0.116*	6.95E-03	-0.250*	3.90E-09	-0.150*	4.84E-04	-0.292*	4.57E-12
%Pebble	0.082*	0.04	-0.119*	3.39E-03	0.005	0.89	-0.003	0.93	0.063	0.12
%Granule	0.286*	9.49E-13	-0.045	0.27	0.218*	7.01E-08	0.152*	1.81E-04	0.288*	6.51E-13
%Very coarse sand	0.267*	3.12E-11	-0.098*	0.02	0.266*	3.31E-11	0.074	0.07	0.280*	2.79E-12
%Coarse sand	0.099*	0.01	-0.105*	9.55E-03	0.147*	2.83E-04	-0.045	0.27	0.099*	0.02
%Medium sand	0.330*	2.20E-16	0.062	0.12	0.302*	4.36E-14	0.260*	1.05E-10	0.329*	2.20E-16
%Fine sand	0.134*	9.81E-04	0.033	0.41	0.155*	1.33E-04	0.136*	8.47E-04	0.130*	1.33E-03
%Very fine sand	-0.239*	2.99E-09	-0.009	0.82	-0.198*	9.84E-07	-0.180*	8.61E-06	-0.219*	5.70E-08
%Silt	-0.199*	8.31E-07	-0.082*	0.04	-0.180*	8.59E-06	-0.206*	3.72E-07	-0.197*	1.12E-06
%Clay	-0.109*	7.53E-03	-0.026	0.51	-0.146*	3.14E-04	-0.122*	2.69E-03	-0.119*	3.35E-03

Figure 1. Study area map showing the two areas (Areas 1 and 2) and the zones (1-5; HI1, LI1, HI2, and LI2). (■) 1-5: sampled zones for population structure analysis without trawling impacts. (●) HI1 and HI2: High-Impact Zones. LI1 and LI2: Low-Impact Zones.

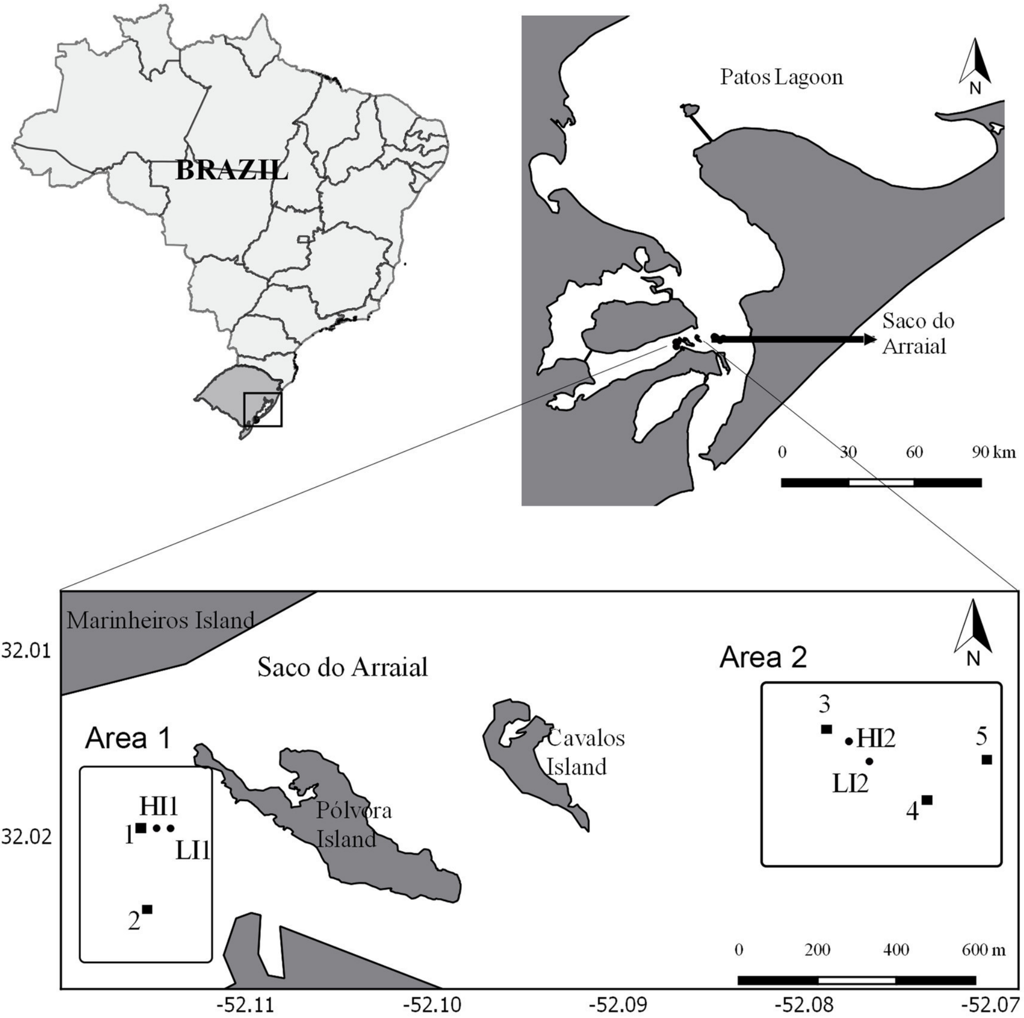

Figure 2. Electronic microscopy photograph of *M. schubarti* with details of first propodus A) Male and B) Female

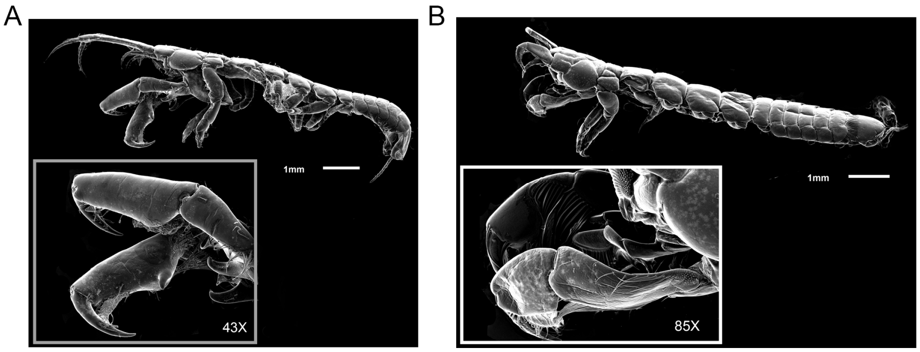

Figure 3. Main grain size distribution of each stratum in the sampled zones of both areas in 2015. Zones 1 and 2 are in Area 1, and Zones 3, 4, and 5 are in Area 2.

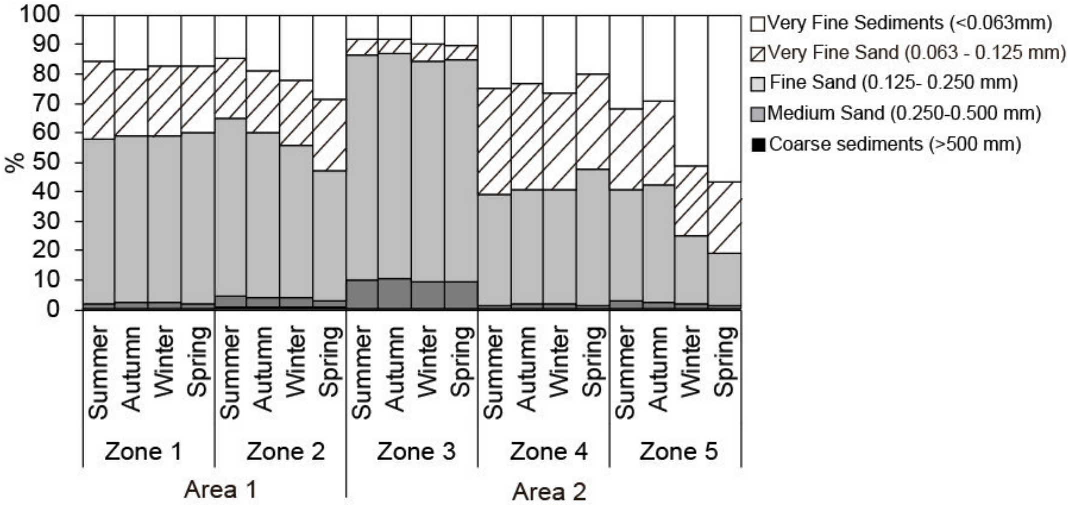

Figure 4. Side-scan sonar images of the control and high-impacted zones of both studied areas: Area 1-Pólvora Island and Area 2- Cavalos Island

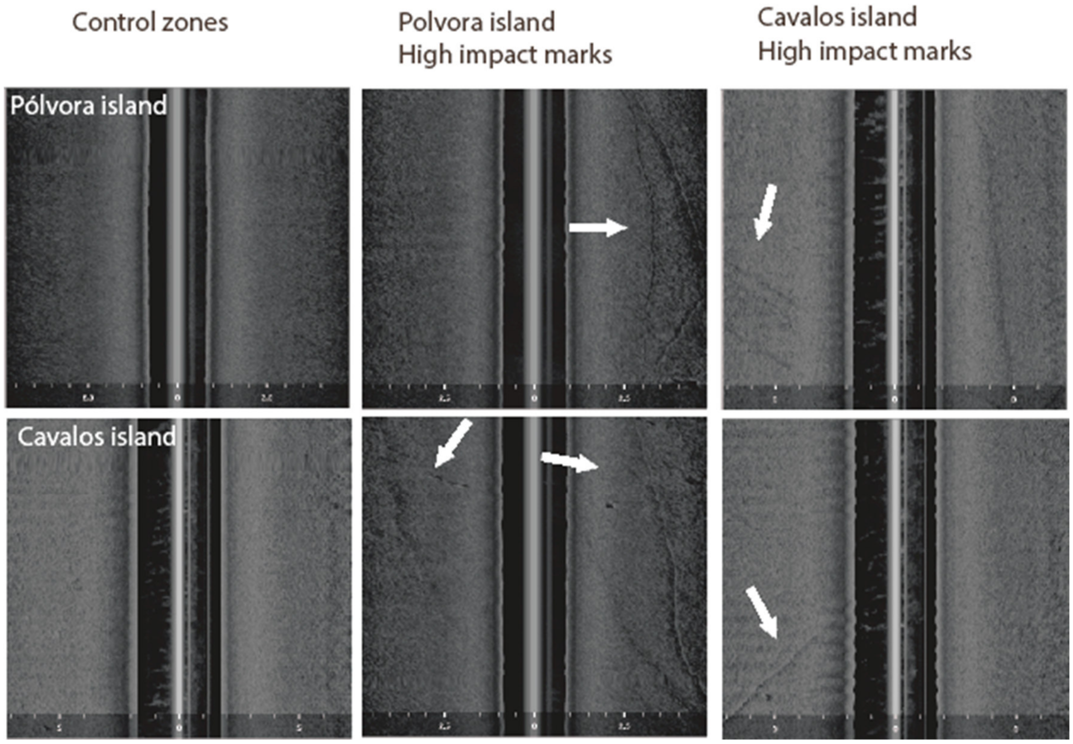

Figure 5. Mean densities of *M. schubarti* by zones, seasons, and strata in Area 1.

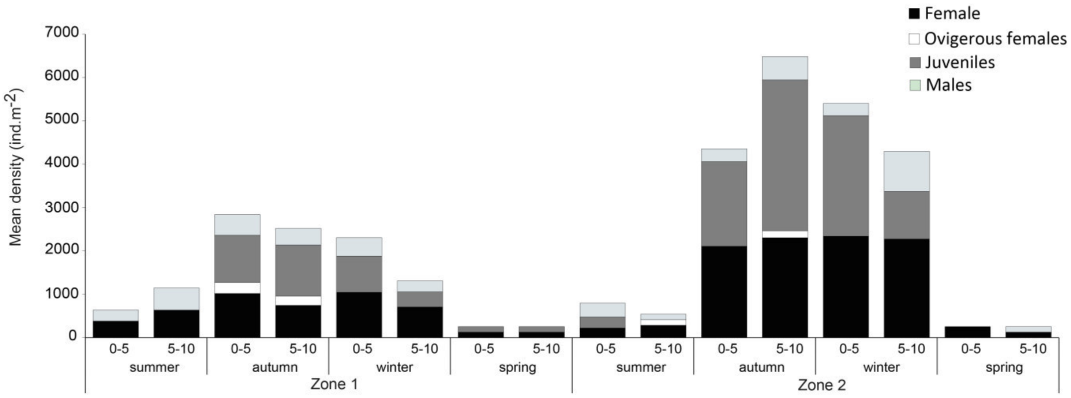

Figure 6. Mean densities of *M. schubarti* by zones, seasons, and strata in Area 2.

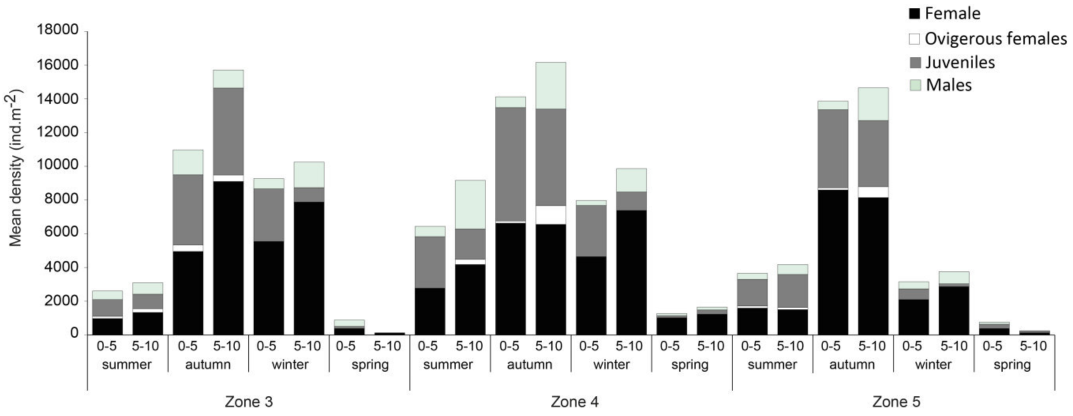

Figure 7. Monthly mean densities of different categories of *M. schubarti* in the impacted zones before (Be) and after (Af) trawling in Area 1.

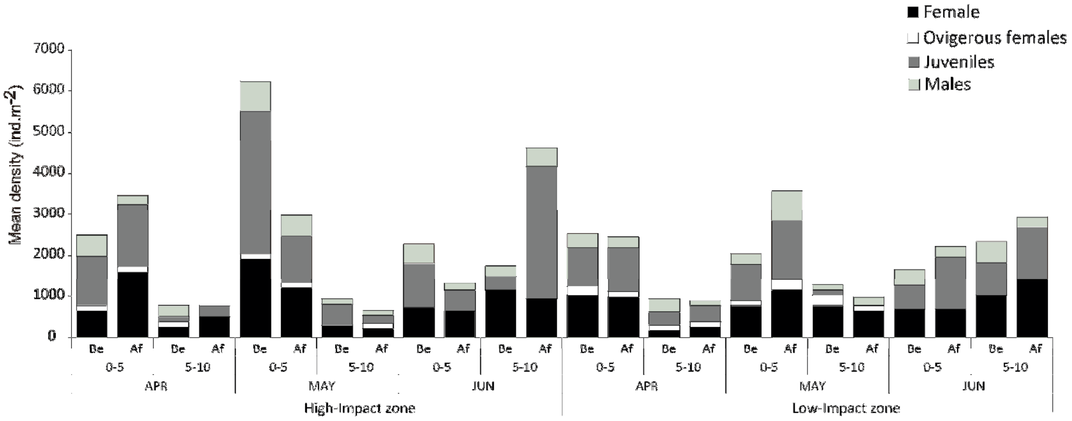

Figure 8. Monthly mean densities of different categories of *M. schubarti* in the impacted zones before (Be) and after (Af) trawling in Area 2.