UNIVERSIDADE FEDERAL DO PARANÁ

FERNANDA ERIA POSSATTO

VARIAÇÃO ESPAÇO-TEMPORAL DA ICTIOFAUNA DEMERSAL E INFLUÊNCIAS ANTRÓPICAS EM UM ESTUÁRIO SUBTROPICAL

PONTAL DO PARANÁ

2015

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Tese de Doutorado apresentado ao curso de Pós Graduação em Sistemas Costeiros e Oceânicos - Centro de Estudos do Mar - Universidade Federal do Paraná.

Orientador: Prof. Dr. Henry Louis Spach Dr. Matt Kenyon Broadhurst

Linha de pesquisa: Biologia e Ecologia de Sistemas Oceânicos e Costeiros

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# CURSO DE PÓS-GRADUAÇÃO EM SISTEMAS COSTEIROS E OCEÂNICOS

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# **TERMO DE APROVAÇÃO**

# Fernanda Eria Possatto

# Variação espacial e temporal na composição da ictiofauna demersal e influências antrópicas em um estuário subtropical

Tese aprovada como requisito parcial para a obtenção do grau de Doutor(a) em Sistemas Costeiros e Oceânicos, da Universidade Federal do Paraná, pela Comissão formada pelos professores:

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Pontal do Paraná, 28/05/2015.

A quem precisou ser mais do que mãe,

mas pai também!

Ivone Gumz

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## PREFÁCIO

A presente tese foi elaborada conforme as sugestões do modelo alternativo proposto pelo manual do doutorando, do Programa de Pós-Graduação em Sistemas Costeiros e Oceânicos, da Universidade Federal do Paraná (UFPR). A primeira parte do trabalho, redigida em português, é composta por aspectos gerais da introdução, objetivos e metodologia, que são gerais para os 3 capítulos subsequentes. A segunda parte, redigida em inglês, inclui artigos científicos independentes, com objetivos específicos e estruturados conforme as regras de cada revista alvo para publicação.

O primeiro capítulo, será submetido para *Estuarine, Coastal and Shelf Science* e tem como objetivo quantificar a distribuição espaço-temporal e os padrões populacionais de teleósteos do complexo estuarino de Paranaguá (CEP), e identificar quaisquer associações com os parâmetros abióticos e bióticos. Além disso, esse trabalho servirá como ferramenta de gestão, pois o uso das informações acima ajudará em estratégias de conservação adequadas a este importante hotspot de biodiversidade marinha.

O **segundo capítudo**, submentido para *PLOS one* tem como objetivo documentar os batóideos encontrados no CEP e identificar qualquer variabilidade temporal e espacial (com foco em áreas marinhas protegidas), e possíveis parâmetros que influenciam essa distribuição, usando essas informações para propor mecanismos para a conservação dessas populações que estão sobre explotadas (já ameaçadas) no litoral do Paraná.

O **terceiro capítulo**, submetido e aceito para a revista *Marine Pollution Buletin*, aborda a poluição por lixo marinho no CEP. Nesse trabalho, foi estudada a variabilidade temporal e espacial do lixo de fundo no CEP. Foi testada a hipótese de que a dinâmica de detritos bentônicos no CEP são influenciados pela época de precipitação e a proximidade das áreas urbanizadas.

#### **RESUMO GERAL**

Nesse estudo, foram investigadas a variação espacial e temporal de espécies de peixes telósteos e batóides, assim como a variação do lixo marinho no complexo estuarino de Paranaguá, sul do Brasil. As coletas foram realizadas utilizando uma rede de arrasto com portas, mensalmente entre Novembro de 2012 e Setembro de 2013 nos diferentes setores dos eixos leste-oeste e norte-sul do complexo estuarino de Paranaguá. Neste contexto, o presente estudo é dividido em 3 capítulos: O capítulo 1 tem como objetivo compreender os padrões de distribuição da ictiofauna demersal temporalmente e espacialmente em diferentes setores dos eixos leste-oeste e norte-sul do complexo estuarino de Paranaguá, assim como identificar quaisquer associações com os parâmetros abióticos e bióticos. No total foram realizados trezentos e noventa e seis arrastos, em uma área total de 46 hectares. Um total de 52119 teleósteos foram capturados, distribuídos em 75 espécies e 30 famílias. Dessas espécies residentes contabilizaram apenas 36%, no entanto, representaram 61 % do número total de indivíduos capturados. Somente onze espécies contribuíram mais de 1% no número total de individuos amostrados. Quatro residentes e um transiente dominaram a assembleia. Cathorops spixii foi a espécie mais abundante representando 40 e 44% do total amostrado em número e Stellifer rastrifer, Aspistor luniscutis, Sphoeroides greelevi e S. testudineus em peso. coletivamente contribuíram em 37% do número e 34% da biomassa. A alta dominância de poucas espécies em abundância e peso encontrados neste estudo corrobora o padrão de assembléia de peixes nos estuários subtropicais. No geral, 51% dos peixes amostrados eram juvenis, enfatizando a importância de estuários como berçários. A estrutura da assembléia mostrou uma complexa relação espaço temporal, com salinidade, temperatura e profundidade explicando a variabilidade da maioria das espécies. O capítulo 2 tem como objetivo documentar os batóideos encontrados no complexo estuarino de Paranaguá e identificar qualquer variabilidade temporal e espacial (com foco em áreas marinhas protegidas) e usar essas informações para propor mecanismos para a conservação dessas espécies. Um total 68 indivíduos, distribuídos em 4 espécies de batoides foram capturados: Gymnura altavela, Narcine brasiliensis, Dasyatis guttata e Rhinobatos percellens. Análises mostraram que o fator que mais influenciou na distribuição dessas espécies foi a posição dentro de cada eixo do complexo estuarino de Paranaguá, com mais individuos capturados nos setores intermediários e internos do eixo norte-sul que no do eixo leste-oeste. Essa distribuição pode ser explicada por duas hipóteses, o eixo norte-sul comparado ao leste-oeste é mais curto e largo, o que permite salinidades mais altas, mas com um gradiente mais ameno, favorencendo

a presença de batóides. Além disso, os setores internos do eixo leste-oeste estão próximos a áreas densamente populosas (cidade e porto de Paranaguá) e com grande fluxo de embarcações, já o eixo norte-sul é cercado por uma área de proteção ambiental. O capítulo 3 tem como objetivo quantificar a variabilidade tempo-espacial na densidade do lixo marinho no complexo estuarino de Paranaguá e usar as essas informações para a construção de estratégias de conservação adequadas a este importante hotspot da biodiversidade marinha. Um total de 291 itens foram coletados, 92% dos itens coletados foram plástico, mais especificamente sacolas plásticas e itens maiores que 21mm. Os setores mais contaminados foram o intermediário e interno do eixo leste-oeste, próximos ao porto e a cidade de Paranaguá. A contaminação por lixo marinho não variou durante as estações do ano, demonstrando que a disposição urbana é o maior fator influenciando na distribuição do lixo no complexo estuarino de Paranaguá. Apesar desse estuário ainda ser considerando como um dos mais conservados estuários no Brasil, a influência antrópica nesse estuário é ainda muito alta, principalmente relacionado ao eixo leste-oeste. Embora seja evidente a área de proteção ambiental de Guaraquecaba é melhor preservada do que os setores internos do eixo lesteoeste, há uma necessidade urgente de controlar e reduzir a perda de detritos de plástico a partir de sua origem urbana. Essa redução somente quando hover integração entre o governo e a sociedade civil.

**Palavras-chave:** complexo estuarino de Paranaguá; função do estuário; distribuição de peixes; batoids; impactos antrópicos; lixo marinho

#### **GENERAL ABSTRACT**

In this study, we investigated spatio-temporal variation in icthyofauna and marine debris in the Paranaguá estuarine complex (PEC), southern Brazil. Samples were collected each month, between November 2012 and September 2013 using penaeid trawls (total of 396 deployments across 46 ha) in each of three different sectors (inner, middle and outer) of two axes: east-west and north-south. In this context, this study is divided into three chapters: Chapter 1 aimed to understand the distribution patterns of demersal teleosts and identify any associations with the abiotic and biotic parameters. A total of 52119 teleosts were captured, comprising 75 species and 30 families. Of the teleosts, residents accounted for 36 and 61% of the total species and individuals sampled. Only eleven species contributed >1% to the total number of fish sampled. Four residents and one transient dominated the assemblage. Cathorops spixii was the most abundant, accounting for 40 and 44% by number and weight of Stellifer rastrifer, Aspistor luniscutis, Sphoeroides greeleyi and S. the total samples. testudineus collectively contributed towards a further 37 34% of the total number and biomass, while Ariidae represented 46 and 62%, respectively. The high dominance of few species corroborates patterns observed other subtropical estuaries. Overall, 51% of the fish sampled were juveniles, emphasizing the importance of estuaries as nurseries areas. The structure of assemblages exhibited complex spatio-temporal relationships, with salinity, temperature and depth explaining variability in the distributions of most species. Chapter 2 aims to document the batoids found in the Paranaguá estuarine complex and identify any temporal and spatial variability (focusing on marine protected areas) and use this information to propose mechanisms for the conservation of these species. A total of 68 individuals, divided into four batoid species were captured: Gymnura altavela, Narcine brasiliensis, Dasyatis guttata and Rhinobatos percellens. Analysis showed that the factor that most influenced the distribution of these species was the position within each axis of the Paranaguá estuarine complex, with more individuals typically caught in the middle and inner sectors of the north-south than the east-west axis. This result could be explained by two hypotheses: compared to the east-west gradient, the shorter north-south gradient had greater salinity that was less variable and more favoured by batoids; and/or unlike the inner sectors of the eastwest gradient which are near densely populated areas (two cities and a port and large flow of vessels), those of the north-south are surrounded by an environmental protected area with better habitat quality. Both hypotheses support the utility of regulating anthropogenic activities for conserving local batoid populations. And Chapter 3 aims to quantify the spatiotemporal variability in the density of marine debris in the Paranaguá estuarine complex and use this information to build appropriate conservation strategies for this important hotspot of marine biodiversity. A total of 291 items were collected, 92% of the collected items were plastic, more specifically plastic bags and items larger than 21mm. The most contaminated areas were the middle and inner sectors in east-west axis, close to the port and Paranaguá city. Contamination by marine debris did not vary during the seasons, demonstrating that urban disposal is the most important factor influencing marine debris distribution in Paranaguá estuarine complex. Despite this estuary still be considering as one of the most preserved estuaries in Brazil, the anthropogenic influence in this estuary is still very high, primarily related to the east-west axis. While it is clear the environmental protected areas surrounding Guaraqueçaba is better preserved than the urbanized inner sectors of east-west axis, there is an urgent need to control and reduce the loss of plastic debris from the urban source. The key mechanism for minimizing plastic marine debris in the Paranaguá estuarine complex requires integration between government and civil society, with the former responsible for regular bulk collection and disposal, and the latter for local disposal at clearly identified locations

**Key words:** Paranaguá estuarine complex; Estuary function; Fish distribution; batoids; anthropogenic impacts; Marine litter.

#### 1. Introdução geral

Estuários são regiões de transição, que apresentam variações sazonais e também nictimeral em seus parâmetros ambientais como temperatura, salinidade e maré, ocasionando mudanças na produtividade primária (Flores-Verdugo *et al.*, 1990) e conseqüentemente, na composição ictiofaunística (Blaber *et al.*, 1995; Morrison *et al.*, 2002; Oliveira Neto *et al.*, 2004). Estes parâmetros ambientais oscilam horizontal e verticalmente, principalmente em função do influxo de água doce proveniente do continente, condicionado por estações chuvosas e secas (Flores-Verdugo *et al.*, 1990). Além destes fatores, variações nas fases da lua (Godefroid *et al.*, 2003; Reis-Filho *et al.*, 2010), entre os períodos diurno e noturno (Ignacio & Spach, 2009), nas condições metereológicas (Garcia *et al.*, 2003) e oceanográficas (Miranda *et al.*, 2002) também são citadas como forças que influenciam na distribuição da ictiofauna em diferentes estuários.

Através de movimentos ativos ao longo do estuário, os peixes evitam condições desfavoráveis de salinidade, temperatura, oxigênio dissolvido e turbidez, fazendo com que a ictiofauna estuarina exiba abundância e composição variáveis (Brown & Mclachlan, 1990). As variáveis ambientais criam condições disponíveis para a ocorrência das espécies, mas a distribuição destas espécies é determinada não só pelas suas tolerâncias ambientais e físiológicas, mas também pelas interações biológicas harmônicas e desarmônicas, tais como comensalismo, predação e competição intra e interespecífica (Elliott & Hemingway, 2002).

Os fatores que atuam no padrão de distribuição dos organismos podem envolver processos locais, regionais ou globais, com as diversas variáveis físicas, químicas e biológicas contribuindo de forma mais ou menos relevante para a compreensão dos padrões populacionais ou de comunidades (Gray & Elliott, 2009; Little, 2000). Entretanto, estes fatores geralmente não atuam de forma independente, existindo uma complexa interação entre variáveis bióticas e abióticas na regulação da presença e abundância dos organismos no

ambiente (McArthur *et al.*, 2010). Acredita-se que os padrões de composição e estrutura de comunidades resultem, em larga escala (km), primariamente das respostas dos organismos ao ambiente físico, onde as variações abióticas dominantes atuam como uma peneira fisiológica, assumindo um papel vital na estruturação da comunidade, enquanto as interações bióticas refinam a distribuição das espécies dentro desta (Menge & Olson, 1990).

Esses padrões de composição e estrutura dos organismos contribuem para a produção de nichos ecológicos e grupos funcionais dentro dos estuários, sendo assim, sua classifcação torna-se extensa com agrupamentos taxonômicos, fisiológicos e ecológicos baseados em atributos como tolerância à salinidade, condições para a reprodução, necessidades de alimentação e hábitos migratórios. A análise da diversidade de grupos taxonômicos em conjunto com os grupos funcionais é uma importante ferramenta para avaliar o papel das espécies no funcionamento do ecossistema e auxiliar em propostas de conservação.

A distribuição das assembléias de peixes que habitam estuários e áreas adjacentes ao redor do mundo, assim como as variáveis ambientais que estimulam sua variação espacial e temporal, são relativamente bem estudadas em estuários no mundo (Greenwood & Hill, 2003; Chícharo *et al.*, 2006; Simier *et al.*, 2006; Veiga *et al.*, 2006; Lugendo *et al.*, 2007; Crona & Rönnbäck, 2007; Jaureguizar *et al.*, 2007).

Na costa brasileira os estudos ictiológicos em regiões estuarinas aumentaram nos últimos anos, podendo listar trabalhos realizados para a região norte (Barletta-Bergan *et al.*, 2002a, b; Barletta *et al.*, 2003, 2005; Silva Junior, 2004; Hercos, 2006; Castro, 2001; Giarrizzo & Krumme, 2008; Giarrizzo & Krumme, 2009; Krumme & Saint-Paul, 2010); nordeste (Neta, 2004); sudeste (Azevedo *et al.*, 2007; Maciel, 2001; Pessanha & Araujo, 2003; Vasconcellos *et al.*, 2007; Barbiere *et al.*, 1991; Andreata *et al.*, 1997; Araujo *et al.*, 1997; Rios, 2001) e região sul (Ramos & Vieira, 2001; Godefroid *et al.*, 1997; Godefroid *et*  *al.*, 2003; Santos *et al.*, 2002; Vendel *et al.*, 2002; Spach *et al.*, 2003; Vendel *et al.*, 2003; Oliveira, 2011).

Além dos fatores já mencionados, numerosas perturbações antropogênicas vêm afetando os ambientes estuarinos, contribuindo para as alterações de seus habitats e mudanças nas estruturas e dinâmicas das assembléias biológicas (Kennish, 2002). As atividades humanas estão promovendo modificações na geomorfologia, fisiografía e hidrologia dos estuários, devido aos represamentos, as construções de portos, a urbanização de cidades, as dragagens periódicas dos canais estuarinos e a destruição dos habitats (Blaber, 2002). Estas ações depredatórias neste ambiente têm modificado as dinâmicas de populações e de assembléias de peixes e, muito provavelmente, os padrões de seleção dos habitats (Whitfield & Elliott, 2002).

A poluição antropogenica é uma grande ameaça para a vida marinha, seus efeitos negativos estão relacionados não apenas a contaminação química de substâncias como metais pesados, nutrientes e hidrocarbonetos, mas também ao lixo marinho (Santos et al., 2009). A poluição por lixo marinho pode causar mortalidade direta entre mamíferos, pássaros, tartarugas e peixes através da ingestão e emaranhamento (Laist, 1997). No entanto, quando a mortalidade não é uma consequência imediata da ingestão, os detritos marinhos podem causar impactos subletais associados com a absorção de bifenilos policlorados e transporte de espécies marinhas não nativas para novos habitats em objetos flutuantes, por exemplo (Winston, 1982; Derraik , 2002). Além dos problemas ecológicos causados pelo lixo marinho, a re-ocorrência de tais detritos também possui repercussões econômicas associados com materiais indesejados em praias (poluição visual) ou enredar e danificar equipamentos (Barnes et al, 2009; Derraik, 2002).

A poluição por lixo marinho é onipresente ao longo da costa do Brasil, no entanto, a maior parte dos estudos se concentraram em praias oceânicas (Ivar do Sul e Costa, 2007),

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apesar de ser bem reconhecido que os estuários são uma importante fonte de lixo para o oceano. Os detritos marinhos (principalmente plásticos) podem permanecer em grandes sistemas estuarinos por longos períodos, e passam por vários processos de degradação em diferentes habitats sedimentares (Ivar do Sul e Costa, 2013).

Estudos anteriores demonstraram que o acúmulo de detritos marinhos pode ser influenciado por uma infinidade de variáveis, incluindo, sazonalidade (por exemplo, chuva; Araújo e Costa, 2007; Ivar do Sul e Costa, 2013) fatores hidrológicos locais (local vento, marés, ondas e correntes; Corbin e Singh, 1993; Nagelkerken et al, 2001; Silva-Iñiguez e Fischer, 2003; Schlining et al, 2013) geografía (Cunningham e Wilson, 2003; Debrot et al., 1999; Mordecai et al., 2011) e sistemas de entrada, incluindo áreas urbanas e drenagem, ou rotas de navegação (Santos et al., 2005) Além disso, a retenção de detritos pode reflectir processos de circulação específicos do estuário (Acha et al., 2003).

O complexo estuarino de Paranaguá (CEP), é um estuário de planície costeira (Angulo, 1992), possui dois eixos principais e uma complexa geometria: o eixo leste-oeste, representado pelas baías de Paranaguá e Antonina, e o eixo norte-sul, representado pelas baías das Laranjeiras, Guaraqueçaba e Pinheiros, conectadas por diversos canais de maré. O CEP possui duas desembocaduras, denominadas Norte e Sul de acordo com a sua posição em relação à Ilha do Mel (Lamour, 2007).

Apesar das duas desembocaduras (norte e sul) serem relativamente estreitas em relação à extensão total do estuário, estas são suficientes para permitir uma grande influência das características físicas e químicas do oceano adjacente. Segundo Lamour (2007), a desembocadura Norte e a desembocadura Sul possuem características fisiográficas distintas fazendo com que as características ambientais do eixo leste-oeste sejam distintas do eixo norte-sul. O estudo realizado por Noernberg (2001) mostrou que o eixo leste-oeste,

comparado ao eixo norte-sul, apresenta maior produtividade primária e sofre maior influência da bacia de drenagem.

Além disso, a influência antrópica no eixo leste-oeste parece ser maior que no eixo norte-sul, devido a presença dos portos de Paranaguá e Antonina, assim como a presença das cidades de Paranaguá, Antonina e Morretes. Através do Programa CAD III (Contaminantes, Assoreamento e Dragagem; ADEMADAN, 2006) foi realizado um levantamento de vários passivos ambientais presentes na bacia de drenagem das baías de Paranaguá e Antonina. Entre eles estão: lixões no município de Paranaguá, o Pátio de depósito de carvão presente no Porto Barão de Teffé, as áreas agrícolas, e a Indústria de Ferro Gusa do Paraná.

O CEP, comparado a outros estuários de mesma importância, ainda é relativamente pouco estudado. A maior parte dos trabalhos relacionados a ictiofauna realizados dentro CEP concentra-se em estudos em determinados locais separadamente, não existindo um conhecimento integrado e sinóptico dos mesmos. Entre os estudos já realizados estão: Godefroid et al. (2003); Santos et al. (2002); Spach et al. (2003); Vendel et al. (2002); Vendel et al. (2003); Oliveira (2011); Passos et al., 2012, 2013). Além disso, poucos trabalhos enfatizam impactos antrópicos relacionados a temática lixo marinho (Guebert-Bartolo et al. 2011).

A compreensão dos processos envolvidos na interação entre a biota e os fatores ambientais e seus efeitos nos sistemas ecológicos, assim como entender como os estuários podem afetar as taxas em que o lixo marinho entra nos oceanos através de escalas temporais e espaciais adequadas são ferramentas importantes para a conservação da diversidade biológica e dos diferentes ecossistemas.

#### 2. Objetivos gerais

O presente trabalho tem como objetivos: (1) Compreender os padrões de distribuição da ictiofauna demersal temporalmente e espacialmente em diferentes setores dos eixos lesteoeste e norte-sul do complexo estuarino de Paranaguá, assim como identificar quaisquer associações com os parâmetros abióticos e bióticos; (2) Documentar os batóideos encontrados no eomplexo estuarino de Paranaguá e identificar qualquer variabilidade temporal e espacial (com foco em áreas marinhas protegidas); (3) Quantificar a variabilidade tempo-espacial na densidade do lixo marinho no complexo estuarino de Paranaguá; (4) Usar as informações acima para a construção de estratégias de conservação adequadas a este importante hotspot da biodiversidade marinha.

#### 3. Material e métodos gerais

#### 3. 1. Área de estudo

#### 3.1.1. Características fisiográficas

O complexo estuarino de Paranaguá (CEP) (48° 25' W, 25° 30' S) situa-se na costa do Estado do Paraná, sul do Brasil (Figura 1), e compreende a extensão sul do grande complexo estuarino-lagunar Iguape-Cananéia (Lana *et al.*, 2001). Corresponde a um dos maiores estuários da costa sul do Brasil, com 601 km<sup>2</sup> de extensão de área (Bigarella, 1978), recebe cerca de 70% da drenagem da Bacia Hidrográfica Atlântica (Mantovanelli, 1999) e é caracterizado como um estuário de planície costeira (Angulo, 1992; Lessa *et al.*, 2000).

Segundo Noernberg *et al.* (2006), o complexo estuarino de Paranaguá é classificado em três níveis hierárquicos. No nível mais alto desta hierarquia está o CEP como um todo, com suas planícies costeiras e bacias hidrográficas, com relevo e clima semelhantes. O nível seguinte consiste em uma divisão do sistema em cinco seções incluindo as principais baías: Paranaguá, Antonina, Laranjeiras, Pinheiros e seção de mistura. O último nível hierárquico compreende os sub estuários de cada seção, os quais são delimitados, considerando-se características morfológicas como embaiamentos e áreas de drenagem específicas.

O CEP possui dois eixos principais: o primeiro eixo de orientação leste-oeste, tem extensão aproximada de 50 km e largura máxima de 7 km, e é formado pelas Baías de Paranaguá e Antonina. O segundo eixo, no sentido norte-sul, tem cerca de 30 km de extensão e largura máxima de 13 km, compreendendo as Baías de Guaraqueçaba e Laranjeiras (Figura 1; Andriguetto-Filho, 1999, Lamour *et al.*, 2004). Além disso, o CEP possui duas desembocaduras, denominadas Norte e Sul de acordo com a sua posição em relação à Ilha do Mel.

A desembocadura Norte apresenta dois canais navegáveis: Norte, localizado entre a Ilha das Palmas e a Ilha do Superagui, e o Sueste, localizado entre a Ilha das Palmas e a Ilha do Mel. A desembocadura Sul apresenta um único canal, denominado canal da Galheta, que se estende entre a Ilha do Mel e o balneário Pontal do Sul, e sendo a principal via de acesso aos portos de Paranaguá e Antonina (Lamour, 2007). Na desembocadura sul ocorre predomínio de extensos bancos arenosos e na desembocadura norte verifica-se uma associação de bancos arenosos com afloramentos rochosos (Lamour, 2007).

#### 3.1.2. Características climáticas

O clima da região é definido, de acordo com a classificação de Köeppen, como Cfa, onde: "C", clima pluvial temperado; "f", clima sempre úmido, com chuvas em todos os meses do ano; e "a", temperatura média do ar no mês mais quente encontra-se acima de 22 °C (Maack, 1981; IPARDES, 1990). Na região predominam os ventos provenientes dos quadrantes leste e sudeste, com velocidade média de 4,0 e máxima de 25,0 m.s<sup>-1</sup>. A umidade relativa do ar é de 85% e a temperatura média do ar é de aproximadamente 21 °C (Lana *et al.*, 2001). A região é caracterizada pela ocorrência de um período seco e outro chuvoso. O período chuvoso inicia no final da primavera e dura a maior parte do verão, enquanto que o período seco começa no final do outono permanecendo até o final do inverno, interrompido, às vezes, por pequenos e fracos períodos chuvosos no início do inverno. A pluviosidade média anual é de 2500 mm com máximos de até 5300 mm (Lana *et al.*, 2001).

#### 3.2. Planejamento amostral

Cada eixo do CEP (L-O e N-S) foi dividido em três setores: Setor 1, Desembocadura do estuário; Setor 2, porção intermediária; e Setor 3; porção superior (Figura 1). Essas divisões foram embasadas em parâmetros do CEP, os quais foram estudados por diversos autores. Os parâmetros considerados foram: granulometria (Lessa *et al.*, 1998; Odreski, 2002; Lamour *et al.*, 2004; Lamour, 2000; 2007), temperatura, densidade, clorofila, turbidez da água, (Noernberg, 2001), salinidade (Noernberg, 2001; Zem, 2008) material particulado em suspensão (MPS), ZMT (Mantovanelli, 1999; Noernberg, 2001; Zem, 2008) e lama fluída (Soares & Noernberg, 2007). Estas variáveis foram incorporados num banco de dados e integradas pelo software ArcGIS (ESRI).

Além disso, utilizando-se da malha amostral de sedimentos coletados pelo Laboratório de Oceanografía Geológica do Centro de Estudos do Mar, pertencente ao projeto "Caracterização Física do complexo estuarino de Paranaguá e análise integrada a informações ecológicas: subsídio para gestão e conservação", foi feita a escolha dos pontos de coleta do presente trabalho. Esta malha está distribuída com espaçamentos equidistante de 500 m para a Baía de Antonina e Baía de Guaraqueçaba, 750 m para a Baía de Paranaguá (Cattani, 2012) e 1000 m para a Baía das Laranjeiras (totalizando 938 amostras de sedimento no CEP). Estas distâncias foram determinadas com base na relação entre a área de cada baía e sua respectiva densidade de pontos.



Figura 1. Complexo estuarino de Paranaguá no sul do Brasil, e as cidades e locais principais. As amostragens foram feitas ao longo dos três setores no eixo norte-sul e leste-oeste: externo (S1), intermediário (S2) e interno (S3). EPA, Área de Proteção ambiental de Guaraqueça. Os pontos amostrais estão representados pelos pontos cinzas.

Segundo Cattani (2012), os sedimentos coletados tiveram eliminação da matéria orgânica por ataque químico com Peróxido de Hidrogênio ( $H_2O_2$ ), como forma da redução da possibilidade de agregação (floculação) entre as partículas finas. A separação das frações grossas (grânulos e areias) das frações finas (siltes e argilas) foi efetuada com o peneiramento a úmido em malha de 0,062 mm, utilizando 1 litro de água destilada. Os sedimentos que ficaram retidos na malha foram destinados ao processo de peneiramento a seco, enquanto que os que passaram tiveram a destinação ao processo de pipetagem como descrito por Carver (1971).

Para o presente estudo, foi definido que as amostras em que uma proporção superior a 50% do sedimento ficasse retido na peneira foram considerados grossos. As amostras que tiveram uma proporção inferior foram consideradas de granulometria fina. Com o objetivo de controlar a variável sedimento, na qual a ictiofauna seria capturada, foram selecionados no software ArcGIS (ESRI) apenas os pontos com granulometria grossa. Além disso, também foram selecionado apenas os pontos com as profundidades iguais ou superiores à 3 m, em função da viabilidade do arrasto.

As coletas de material biológico e a tomada dos dados ambientais foram realizadas mensalmente, de novembro de 2012 à setembro de 2013. A escolha dos pontos amostrais foi feita de modo aleatório. por meio da ferramenta Hawth's Tools (http://www.spatialecology.com/), elaborada para o software ArcGIS (ESRI). De um total de 230 pontos com profundidades superiores a 3 m e com granulometria grossa, foram sorteados mensalmente 18 (dezoito) pontos ao longo de cada um dos eixos, 6 (seis) em cada setor (Figura 1).

Os arrastos foram realizados sempre em dois dias consecutivos de maré de quadratura (um dia para cada um dos eixos amostrados), durante o dia, no sentido contrário a corrente de maré, durante 5 minutos. As posições iniciais e finais de cada arrasto foram registradas em um Sistema de Posicionamento Global (GPS map 76S).

Para caracterização ambiental foram registrados os valores das variáveis físicas do local antes do início de cada arrasto, por meio de uma perfilagem com um CTD (Conductivity, Temperature and Depth; Figura 2a). Para estimativa da profundidade média de cada arrasto, através de um ecobatímetro a profundidade foi registrada a cada 1 minuto.

As três redes utilizadas nos arrastos são do modelo "*Wing Trawl*" (Figuras 3 e 4) idênticas em termos do: tamanho da malha (abertura da malha esticada de 42 e 26 mm no corpo e sacos, respectivamente), material (fio de poliamida de 0,6 e 1,0 mm, respectivamente)

e desenho (duas costuras), e somente variaram ligeiramente em seus comprimentos totais (9,44, 9,46 e 9,92 m). Todos os arrastos foram realizados em uma configuração de arrasto simples (Broadhurst *et al.*, 2013) em uma canoa de 8,8 m de comprimento (22 HP). Foram utilizadas também duas portas retangulares de madeira vazada (0,47 × 0,90 m e 17 kg cada) ligados a um cabo de poliamida de 10 mm de diâmetro (comprimento total de 50 m). A abertura das redes de arrasto foi ditada pela altura que as portas permaneceram fora do substrato (~ 0,46 m).

A área arrastada foi estimada através da multiplicação da distância entre o ponto inicial e final multiplicado do pela abertura hipotética da rede de arrasto. Este último foi estimado para cada arrasto, considerando todos os parâmetros técnicos relevantes (ou seja, velocidade de arrasto, profundidade, comprimento do cabo utilizado no arrasto e área de sistema de arrasto). Esses calculos foram realizados dentro do modelo de desempenho de rede de arrasto proposto por Sterling (2005), e variou entre 0,45 e 0,53.



Figura 2. a) Perfilagem com CTD; b, c, d) Lançamento e recolhimento da rede com guincho após 5 minutos de arrasto.



Figura 3: Desenho esquemático das portas e da rede "*Wing Trawl*" rebocada pela canoa utilizada nos arrastos.



Figura 4: Rede utilizada nos arrastos.

#### 3.3. Processamento das amostras

#### 3. 3. 1. Classificação das espécies de peixes

Após cada arrasto, todos os peixes capturados foram acondicionados em sacos plásticos, devidamente identificado de acordo com o ponto amostral, e conservados em gelo até o seu transporte ao laboratório (ver maiores detalhes no capítulo 1). As espécies de raias (Ordem Rajiformes, ver maiores detalhes no capítulo 2) e Cavalos Marinho (Família Syngnathidae) capturadas nos arrastos foram fotografadas e pesadas em campo e posteriomente devolvidas ao estuário. Sempre que a quantidade de uma espécie foi maior que 50 indivíduos, o excedente por espécie e arrasto foi contado, pesado (com dinamômetro) e descartado em campo, sendo os dados incorporados posteriormente à biomassa em número e peso.

No laboratório os exemplares foram identificados até o nível específico através de literatura especializada (Barletta & Corrêa, 1992; Figueiredo, 1977; Figueiredo & Menezes, 1978, 1980, 2000). Os exemplares foram medidos, comprimentos padrão (CP) e total (CT) em cm e pesados (g). Através de uma abertura longitudinal na região ventral, foi feita a identificação macroscópica do sexo e do estádio de maturação gonadal, de acordo com a escala proposta por Vazzoler (1996). Nesse estudo os indivíduos no estádio A foram denominados juvenis e indivíduos nos estádios B, C e D, adultos.

#### 3. 3. 2. Lixo marinho

Após cada arrasto, o lixo marinho coletado também foi separado em sacos plásticos, devidamente identificados de acordo com o ponto amostral. Em laboratório, as amostras foram lavadas e secas ao ar. As amostras foram posteriormente contados, e identificado com base na abundância relativa, classificados de acordo com o tipo de material em sete grupos coerentes: (1) de plástico, (2) de metal, (3) de couro, (4) de vidro, (5) espuma (6), roupas, e (7) embalagens longa vida; ver maiores detalhes no capítulo 3.

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## **CAPÍTULO 1**

5 Spatio-temporal variation among demersal ichthyofauna in a subtropical estuary bordering World Heritage Listed and marine protected areas: implications for resource management

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# 25 Spatio-temporal variation among demersal ichthyofauna in a subtropical estuary bordering World Heritage Listed and marine protected areas: implications for resource management

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#### ABSTRACT

- 50 The Paranaguá estuarine complex (PEC) is an important ecological region within Brazil's subtropical Atlantic forest biosphere. Data from extensive trawl surveys conducted within two major sectors of the PEC were analysed to reveal patterns of seasonal and spatial variation in fish assemblage structure. Surveys yielded 52,000 fishes (51% juveniles) representing 75 species. Species classified as estuarine residents accounted for 36 and 61% of total species and individuals, respectively. Five species that are important in the artisanal fishery
- 55 comprised 77% of individuals and 81% of total biomass. *Cathorops spixii* was the most abundant (40% of individuals, 44% of total biomass), whereas *Stellifer rastrifer*, *Aspistor luniscutis*, *Sphoeroides greeleyi* and *S. testudineus* collectively contributed towards 37% of individuals and 34% of total biomass. The dominance of few species but large proportions of juveniles and transients, mostly in the lower reaches, are consistent with other subtropical estuaries. *Aspistor luniscutis*, *C. spixii* and *S. rastrifer* abundances were significantly
- 60 (negatively) associated with salinity, with the latter two species also associated (positively) with temperature, and *S. greeleyi* and *S. testudineus* (mostly adults) were more abundant in deeper areas. These patterns seemed to be strongly influenced by species-specific patterns of reproduction (*S. rastrifer, C. spixii* and *A. luniscutis*), and habitat (*S. greeleyi* and *S. testudineus*) and food requirements (juveniles of *C. spixii* and *A. luniscutis*). Clearly, several ecologically and economically important fishes use the PEC and ongoing research is required to manage anthropogenic activities.

Key words Biodiversity hotspot · Estuary function · Fish community · Fish distribution · Nursery · Paranaguá estuarine complex

#### 70 Introduction

Estuaries represent the interface between fresh and saltwater ecosystems (Miranda et al. 2002), are highly productive and often support large species diversity, especially in tropical and subtropical regions (Martino and Able 2003). The diversity of teleosts in estuaries has been hypothesized to be a function of large spatial and temporal variation in environmental conditions (Pasquaud et al. 2015). Some fishes are physiologically and

75 ecologically adapted to spend their entire life cycle in estuaries (termed 'estuarine residents'; Potter et al. 1986; Potter and Hyndes 1999; Elliott et al. 2007), but a greater number of species are transients that use estuaries during particular life stages. For example juveniles by use estuaries as nurseries that provide refuge from predation and food resources, or adults may enter estuaries for reproduction or to exploit seasonally abundance prey (Blaber and Blaber 1980; Claridge et al. 1986; Elliott et al. 2007; Paterson and Whitfield 2000).

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Irrespective of their residence status, among the most important factors affecting teleost distributions in estuaries are environmental parameters, including water temperature, salinity and depth; all of which vary temporally (e.g. due to rainfall) and spatially (e.g. with distance from the ocean; Jaureguizar et al. 2004). Species-specific tolerances to environmental variations often dictate teleost distributions and migration patterns throughout estuaries. In addition to environmental drivers, biotic factors such as predation, competition and reproduction can strongly affect spatial and temporal species assemblages and distributions. These relationships are often complex and interconnected. For example, Remmert (1983) proposed that large-scale composition patterns and community structures reflect the broad responses of organisms to the physical environment, with key abiotic variations acting as a physiological sieve, while biotic interactions refine species distributions within an ecosystem (Menge and Olson 1990).

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Because of their large biodiversity, estuaries are among the most economically valuable ecosystems on earth, with big systems often extremely important for subsistence and artisanal fisheries in developing countries (Costanza et al. 1997; Barletta and Costa 2009). The Paranaguá estuarine complex (PEC) in southern Brazil is

95 one such estuary (Fig. 1; Diegues 1995; Possatto et al. 2015a). Despite being heavily populated, the PEC is considered one of the most preserved Brazilian ecological environments (Sá et al. 2006). Among the various local anthropogenic activities, port-related industries dominate, followed by artisanal fisheries, tourism, agriculture and aquaculture. Notwithstanding these activities, large areas of the coastal zone have been protected by environmental legislation. This protection encompasses vast mangrove belts bordering the estuary,

100 which serve as important habitat for various marine fauna, and are ecologically linked to the extended rainforest zone in the hinterland (Pichler et al. 2015; Possatto et al. 2015a).

Numerous surveys and ecological studies on the fish fauna in the PEC have emphasized its biodiversity importance (e.g. Spach et al. 2003; Queiroz et al. 2007; Barletta et al. 2008; Contente et al. 2011; Passos et al.

- 105 2012; 2013). However, most studies have been limited to relatively small spatial and temporal scales, with no seasonal comparisons (but see Possatto et al. 2015b). Such broader studies are important, especially in estuaries like the PEC that encompass the transition zone between temperate and tropical regions (e.g. 25° south). At these latitudes, there are large annual fluctuations in environmental parameters, especially rainfall (~2500 mm per annum), which would be expected to strongly affect fish distributions and assemblage compositions
- (Barletta et al. 2003). Understanding such complex spatio-temporal patterns of influence is imperative for the effective management of anthropogenic activities within estuarine systems (Whitfield and Elliott 2002; Ley 2005).

Considering the above, the primary aim of this study was to quantify the broad spatio-temporal

115 distributions and population patterns of demersal fishes throughout the PEC and key abiotic associations. More specifically, we tested the hypothesis that rainfall, water temperature, salinity and depth are important in structuring the ichthyofaunal assemblages. A secondary aim was to use the above information to recommend appropriate conservation strategies in what represents one of the most important marine biodiversity hotspots in South America.

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#### Methods

#### Study area

125 The PEC (48° 25' W, 25° 30' S) has a total surface area of ~61,200 ha and can be considered part of a larger interconnected subtropical estuarine system that includes Iguape-Cananéia Bay to the north (and on the southern coast of São Paulo; Lana et al. 2001; Noernberg et al. 2006; Fig. 1). Surrounded by one of the last remnants of Atlantic rainforest (207,1685 ha), the PEC encompasses two conservational areas—the Superagui National Park and the Environmental Protected Area of Guaraqueçaba, and borders a Natural World Heritage site (UNESCO

- 130 2014; Fig. 1). The system broadly separates into a 56-km long east-west axis forming Paranaguá and Antonina bays and a 40-km north-south axis constituting Laranjeiras and Guaraqueçaba bays (Fig. 1). Smaller segments connect various other water bodies including Guaraqueçaba, Antonina, Pinheiros, Itaqui, and Benito bays and the Medeiros River (Lamour et al. 2004).
- The PEC is characterized by a moderate vertical salinity gradient, with semidiurnal tides displaying diurnal inequality (maximum variation is ~2.7 m), and consistent seasonal circulation and stratification (Marone et al. 2005). The climate is transitional tropical (mean annual rainfall of ~2500 mm; Lana et al. 2001), with highly distinctive seasonality (wet summers and dry winters) (Marone et al. 2005).

140 Sampling design

The east-west and north-south axes of the PEC were divided into three sectors (outer-sector 1, middle-sector 2, and inner-sector 3; Fig. 1); which were delineated according to substrate particle size, water temperature, density, chlorophyll, turbidity, salinity and suspended particulate matter (Lamour et al. 2004; Cattani and

- 145 Lamour 2015). These variables were incorporated into a database and integrated by ArcGIS software (Esri Pty Ltd). Using the Hawth's Tools (developed for the ArcGIS software; Beyer 2004), random monthly sampling points were selected within sectors (n = 6 per sector). All sampling points comprised appropriate grid sediments (>3 m depth and coarse grain size).
- 150 Between November 2012 and September 2013, each sector was monthly sampled using one of three penaeid trawls deployed across six replicate 5-min tows in a straight line (with the start and end positions marked using a global position system–GPS map 76S; Garmin). The trawls were identical in terms of their mesh sizes (42- and 26-mm stretched mesh openings in the bodies and codends, respectively), materials (0.6and 1.0-mm diameter polyamide twine, respectively) and designs (two seams, with lead-a-head and no sweeps);
- 155 varying only slightly in their total opening lengths (9.44, 9.46 and 9.92 m). Each trawl was fished in a single-rig configuration (Broadhurst et al. 2013) from a 9-m canoe (18 Kw) and spread by two flat-rectangular otter boards ( $0.47 \times 0.90$  m and 17 kg each) attached to 10-mm diameter polyamide warps (total length of 50 m). The opening heights of the trawls were all dictated by the otter-board height and remained at ~0.46 m off the substratum.

Prior to each 5-min deployment, salinity and temperature were measured with a multiple sensor (Alec Electronics Co., ASTD. 687). Depth was subsequently recorded at 1-min intervals using an echosounder (Eagle Cuda 168 EX). Rainfall data encompassing the estuary catchment were obtained for each sampled month from the "*Sistema meteorológico do Paraná*" SIMEPAR (www.simepar.br). At the end of each deployment, the

- 165 codend was emptied onto a sorting tray and the fish separated, placed into plastic bags, and kept on ice for transport to the laboratory. When more than 50 individuals of a species were caught, the excess was counted, weighed (to the nearest 1g), discarded in the field, and the data subsequently scaled. Elasmobranchs were released and not considered further here (because they comprised a separate study; Possatto et al. 2015b).
- 170 The collected teleosts were identified to the highest possible taxonomic separation before being measured for total length (TL to the nearest 1 cm) and weighed (as above). Up to 50 randomly selected individuals of each species from each deployment were also sexed and had their maturation stage (immature, maturing, mature or spent) determined following Vazzoler (1981). For analytical purposes, immature individuals were classified juveniles, whereas all other stages were deemed adults.

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#### Data analyses

Fish abundances were standardized to per 0.1 ha trawled by multiplying the known trawled distance by the hypothesized wing-end spread of the trawl. The latter was estimated for each deployment by considering all relevant technical parameters (i.e. towing speed, water depth at 1 min intervals, length of warp deployed and trawl system area) within the 'Prawn Trawling Performance Model' (Sterling 2005) and varied between 0.45 and 0.53 of the individual trawl headline lengths. To incorporate broader temporal scales and the potential influence of rainfall, we considered monthly rainfall patterns between 2003 and 2013 (from SIMEPAR) and aggregated each month into four seasons: early wet (October, November and December, with a mean combined

185 monthly rainfall of 235.1  $\pm$  37.5 mm), late wet (January, February and March; 345.2  $\pm$  73.2 mm), early dry (April, May and June; 136.2  $\pm$  52.6 mm) and late dry (July, August and September; 146.0  $\pm$  61.2 mm; Fig. 2).

Permutational analyses of variance (PERMANOVA; Anderson 2001; Anderson et al. 2008) were used to test for spatio-temporal differences in measured environmental variables (temperature, salinity and depth) and fish-assemblage characteristics. A three-factor PERMANOVA was used to test for differences in the abundances

195 and assemblage structures of fish among seasons, axes and sectors. All factors were fixed and fully orthogonal. The multivariate analysis was based on the Bray-Curtis dissimilarity measure, whereas each univariate analysis was based on the Euclidean distance measure. Type III (partial) sums-of-squares were calculated using 9999 permutations of the residuals under a reduced model (multivariate analysis) and unrestricted permutations of the raw data (univariate).

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Separate pairwise tests were subsequently used to assess which levels of each significant interactive factor differed for each level of the other factor using the PERMANOVA routine. The proportion of variation attributable to each factor and interaction in each model was calculated to facilitate interpretation of the results. Multivariate patterns of assemblages were determined and visualized using hierarchical agglomerative clustering techniques based on the Bray-Curtis similarity measures, and similarity percentage analyses (SIMPER) were used to identify individual species that contributed towards each identified grouping (Clarke 1993; Clarke and Warwick 1994).

Distance-based linear models (DISTLM, Anderson et al. 2008) were used to examine relationships
 between assemblage structure and derived assemblage parameters (e.g. the numbers of species and individuals) with water salinity, temperature and depth. The Euclidean distance matrix for each assemblage parameter was modelled separately with the Euclidean distance matrix of each water parameter, and for all water parameters combined, using E-primer (Clarke 1993; Anderson et al. 2008).

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#### Abiotic data

PERMANOVA revealed that water temperatures significantly differed seasonally and by sector (p < 0.001; Table 1; Fig. 3a). Subsequent pairwise tests identified that across both axes, water temperature was warmest during the late wet season and coolest in the late dry, while across both axes and all seasons, temperatures were warmer in each sector 3 than sectors 1 and 2 (Fig. 3a). Salinity also significantly differed according to the interactive effects of season × axis and axis × sector (p < 0.001; Table 1; Fig. 3b). There was no consistent pattern with depth, with a higher-order interaction between season, axis and sector (p < 0.01; Table 1; Fig. 3c).

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General composition of fish assemblages in the PEC

Three-hundred-and-ninety-six tows were successfully completed for a total sampled area of 46 ha and a catch of 52,119 fishes comprising 75 species and 30 families (Table 2). Residents accounted for 36 and 61% of the total
species and individuals and overall, 51% were juveniles (Table 2). The families with the greatest species richness were Sciaenidae (16 species), Carangidae, Ariidae and Engraulidae (six species each), Achiridae (five species), Paralichthyidae and Tetraodontidae (four species), Haemulidae (three species), and Cynoglossidae and Pristigasteridae (two species; Table 2). All other families were each represented by a single species (Table 2).

- Only eleven species contributed more than 1% to the total number of fish sampled; representing 87% of all species sampled by number (Table 2). Five species dominated the assemblage, with *Cathorops spixii* the most abundant, accounting for 40 and 44% by number and weight of the total (Table 2). *Stellifer rastrifer, Aspistor luniscutis, Sphoeroides greeleyi* and *S. testudineus* collectively contributed towards a further 37 and 34% of the total number and biomass. All five species were residents, except *S. rastrifer* (Table 2). Because of *C. spixii*
- 245 and A. *luniscutis*, the Ariidae family represented 46 and 62% by number and weight of the total, respectively (Table 2).

Fish assemblage structure significantly differed due to the interactive effects of season, axis and sector, indicating complex spatio-temporal relationships (PERMANOVA, p<0.01; Table 1). The cluster analysis identified four distinct assemblage groups at the 40% similarity level (Fig. 4). Group A primarily comprised

samples collected in sectors 2 and 3 along the east-west axis (Fig. 4). Three species, including *C. spixii*, *S. rastrifer* and *A. luniscutis*, contributed more than 90% towards the similarity matrix of this group (Fig. 4). Group B was the largest, containing 10 samples mostly taken in the north-south sector in the late wet and early dry, with *C. spixii*, *S. rastrifer* and *S. greeleyi* contributing most to the similarity matrix (Fig. 4). Group C contained four samples taken in the late dry, whereas Group D contained only two samples taken in the early wet season (Fig. 4).

Spatio-temporal variations in the abundances of fish

Spatio-temporal patterns in the (i) total species and individuals sampled, (ii) proportions of juveniles and transient species and (iii) abundances of the five key species (above) were mostly complex (i.e. there were significant higher-order interactions in most PERMANOVAs; p<0.05; Table 1). Nevertheless, some general patterns were evident. Pairwise analyses identified that a significantly greater number of species were sampled in the late wet than the other seasons (pooled across axes and sectors; p<0.05; Table 1; Fig. 5a).

270 The number of total individuals sampled 0.1 ha<sup>-1</sup> differed significantly according to interactive effects of season and sector, and also axis and sector (p<0.05; Table 1). Specifically, more total individuals were sampled in the late wet season across all sectors in the north-south axis, but only in sectors 2 and 3 in the east-west axis (Fig. 5b). Similarly, more total individuals (pooled across seasons) were sampled in sectors 2 and 3 in the eastwest axis, but this was not the case in north-south axis (Fig. 5c).

275

The proportion of transient species sampled differed significantly according to seasons with the least sampled in the late dry across all sectors and both axes (p<0.001; Table 1; Fig. 5c). Pairwise comparisons failed to detect any significant differences in the proportion of transient species sampled across the other seasons (p>0.05; Fig. 5c). Further, the proportion of transient species returned a significant axis × sector interaction

280 (p < 0.01; Table 1; Fig. 5c). The pairwise tests identified that a greater proportion was sampled in sector 1 (but not sectors 2 and 3) in the east-west axis than the north-south axis (p < 0.05; Fig. 5c).

The proportion of juveniles sampled also differed significantly according to the interactive effects of several factors (p<0.05; Table 1; Fig. 5d). Notably, the proportion of juveniles clearly differed according to

- 285 season in the east-west axis, with the least sampled in the early wet and the most in the late wet and early dry seasons, but no such pattern was evident in the north-south axis (pairwise tests; Fig. 5d). Further, in the east-west axis, a lower proportion of juveniles was sampled in sector 2, but again this was not evident in the north-south axis (Fig. 5d).
- With respect to the five key species, all were recorded across all seasons, axes and sectors, but with significant spatio-temporal variations (*p*<0.05; Table 1; Fig. 6). *Cathorops spixii* and *A. luniscutis* differed significantly according to the interactive effects of axis and sector (*p*<0.05; Table 1; Fig. 6a and b). Across all seasons, *C. spixii* was significantly more abundant in sectors 2 and 3 than in sector 1 in the east-west axis, but this was not always the case in the north-south axis (*p*<0.05; Fig. 6a). By comparison, *A. luniscutis* was most abundant in sector 3 of the east-west axis (especially during the late wet season) than all other sectors within either axis, while across all seasons *S. rastrifer* differed significantly according to the interactive effects of season and both axis and sector (*p*<0.05; Table 1; Fig. 6b and c), and was most abundant in the late wet season across all sectors in the north-south axis, but only in sectors 2 and 3 in the east-west axis (Fig. 6c).</p>
- 300 Neither *S. greeleyi* nor *S. testudineus* displayed broad, overall seasonal differences in abundances (p>0.05; Table 1; Fig. 6d and e). *Sphoeroides greeleyi* predominantly occurred in the north-south axis (p<0.05; Fig. 6d). By comparison, the abundance of *S. testudineus* displayed a significant axis × sector effect, with the most individuals in sector 1 in the north-south axis, but this was not evident in the east-west axis (Fig. 6e).
- 305 Environmental correlates with fish assemblage parameters

Water temperature was significantly related to the Euclidean distance matrices of the total assemblage structure, and the numbers of total species, total individuals, *C. spixii* and *S. rastrifer* (p<0.01; Table 3). However, the observed relationships generally were weak with water temperature explaining between 3.4 and 10.0% of the

- 310 variation ( $\mathbb{R}^2$  value) in each analysis (Table 3). Salinity was significantly related to the Euclidean distance matrix of the assemblage structure, and the numbers of total individuals, *C. spixii*, *S. rastrifer* and *A. luniscutis* (p<0.01; Table 3). The amount of variation explained by salinity was greatest (9.3%) for the latter species (Table 3). Water depth was significantly related to assemblage structure and the abundances of *S. greeleyi* and *S. testudineus*, but these relationships were similarly weak; accounting for less than 3.1% of the variation among
- 315 samples (p < 0.05; Table 3). When all three parameters were included in a sequential regression, their combined total relationship was significant in each analysis, except for total species, *S. greeleyi* and *S. testudineus* (p < 0.05; Table 3). Nevertheless, the combined relationships only marginally improved the amount of variation explained over the single relationship (Table 3).

#### 320 Discussion

This study represents the first quantitative assessment of broad seasonal and spatial influences on the distributions of key teleosts throughout the PEC, enhancing and building on the information collected during previous isolated studies (Spach et al. 2003; Queiroz et al. 2007; Schwarz-Junior et al. 2007; Barletta et al.

325 2008; Contente et al. 2011). Like in other estuarine systems globally (Greenwood and Hill 2003; Jaureguizar et al. 2004; Akin et al. 2005; Simier et al. 2006; Garcia et al. 2012) much of the spatio-temporal variation in species abundances can be explained by the underlying environmental conditions. Ultimately, this information can be used to support coherent resource management plans for the area.

### 330 Variations in abiotic data

The entire PEC receives considerable rainfall during the warm wet season (e.g. up to 450 mm per month between October and March) and with some apparent cascading effects on salinity. This was especially evident in the east-west axis which is characterised by several large rivers in sector 3 (Noernberg et al. 2006). This

335 sector consistently had the lowest salinities, contributing towards greater environmental variability across the entire axis. In contrast, in addition to having less freshwater input from rivers the north-south axis is mostly shallower, shorter and wider ( $\sim$ 40 × 13 km) than the east-west axis ( $\sim$ 56 × 7 km), and therefore more amenable to seawater penetration (thus greater salinities throughout its entirety).

- 340 Irrespective of rainfall and geographical differences, salinity increased in both axes towards the ocean which is typical of estuarine systems in general, owing to abiotic gradients resulting from the convergence of adjacent marine and freshwater environments (e.g. Day 1981). The warmest temperatures, as might be expected, were observed across all sectors and both axes during the late wet season (summer/autumn) and coolest in the late dry season (winter/spring). Depth did not present a specific pattern in the PEC. These spatial and temporal variations in water attributes influenced the distributions of assemblage structure and key species,
- 345

with clear inter- and intra-axes spatial differences.

#### Assemblage composition

- 350 Prior to focusing on the extent to which the above variations in environmental parameters were reflected in the patterns and distributions of assemblages and key species, the fishing gear selectivity warrants some mention for possible confounding effects (i.e. sampling bias), and especially among smaller individuals (i.e. juveniles). In particular, because the trawls all fished the water column from the substratum to <0.46 m and their mesh sizes were quite small, it is possible that they selected proportionally smaller individuals. Conversely, the logistics of 355 trawling meant that we were forced away from some very shallow areas, which might have precluded sampling
- some juveniles and small species (i.e. depth-dependant effects on sizes; Pichler et al. 2015). Such variables could either over- or underestimate absolute fish abundances 0.1 ha<sup>-1</sup>. Nevertheless, because we maintained the same sampling gear and sites across time, the observed abundances are representative of relative differences.
- 360 Similar to the many studies in other channel estuaries, the sampled ichthyofauna in the PEC was diverse, comprising adults and juveniles of both resident and transient species (Allen et al. 2006). Nevertheless, only a small component of the total ichthyofauna dominated samples, which is also true in other estuarine systems (Whitfield 1999; Akin et al. 2005). Such a result is often attributed to the dynamic and variable estuarine environment, in which few species are adapted to live owing to the necessity for a broad tolerance to fluctuating
- 365 abiotic conditions (Whitfield 1999; Barletta et al. 2005; 2008; McLusky and Elliott 2007). The dominant species here included four residents (C. spixii, A. luniscutis, S. testudineus and S. greeleyi) and one transient (S. rastrifer), and their distributions contributed greatly to the observed spatio-temporal variations in assemblage structure. Each of the individual species requirements were however, somewhat divergent, reflecting their species-specific life histories.

Specifically, *C. spixii* and *A. luniscutis* belong to Ariidae, which is among the most important teleost families in tropical and subtropical estuaries (Lowe-McConnell 1987; Barletta and Blaber 2007; Barletta et al. 2008; Dantas et al. 2010). The success of this family in transitional waters is a consequence of their eury-thermohaline capacity, Weberian apparatus (i.e. connecting the swim bladder to inner ear, increasing their

- 375 environmental perception) and parental care (Burgess 1989; Dantas et al. 2010). Similarly, *S. greeleyi* and *S. testudineus* are tetraodontids; a family also adapted to a wide range of habitats and environmental conditions (Nelson 1994). More locally, the abundant transient *S. rastrifer* is among the most regionally common, inhabiting the shallow continental shelf of the Paraná coast and the broader southwest Atlantic Ocean (Godefroid et al. 2004). All five species are retained by artisanal fishers (Coelho et al. 1986; Reis 1986;
- including the two tetraodontids—despite the dangers of tetrodotoxin in their meat (Haddad Junior 2003).

#### Spatio-temporal variations in abundances

Notwithstanding the presence of the five key species throughout the sampled year, there were intra-specific
spatio-temporal variations in their patterns of abundances. *Stellifer rastrifer* followed a clear seasonal trend demonstrating a preference for the less saline waters in sectors 2 and 3 of the east-west axis during the late wet season. The same pattern was previously reported for this species in the PEC (Barletta et al. 2008) as well as in other estuaries (Giannini and Paiva Filho 1990; Chaves and Vendel 1997). Such preferences may reflect reproductive behavior (Queiroz et al. 2006), given that, like some other transients, *S. rastrifer* mainly spawns in the upper estuary during spring and summer in response to warmer water temperatures—and clearly with a broad tolerance to fluctuating salinity (Chaves and Vendel 1997).

The other four dominant species were all residents and while they displayed slightly divergent patterns in abundances, most were strongly affected by rainfall and/or water temperature; patterns that ultimately

395 contributed towards the observed differences in broader categories (total species and numbers and assemblage structure). Specifically, like *S. rastifer*, *A. luniscutis* and to a lesser extent, *C. spixii* were more abundant in the wet; especially in the warmer, shallow and less saline waters of sector 3 (particularly the east-west axis). This pattern was also presumably in response to summer spawning, because the proportion of juveniles was relatively greater across both axes during the early dry, and also in the east-west axis during late wet. These observations

400 are supported by Fávaro et al. (2005), who suggested that *C. spixii* reproduce between September and November (i.e. the early wet season).

Reproduction during the wet season would positively benefits most teleosts, because the rainfall increases nutrients in the water causing cascading effects on primary and secondary productivity (Barletta et al. 2003).

405 For example, juveniles would have more nutrients for food supplies and because of the warmer water, their growth should be maximised (Deegan 1990). Further, increased water turbidity might reduce predation (Blaber 2000).

In contrast to the other dominant species, both tetraodontids (*S. greeleyi* and *S. testudineus*) were less affected by rainfall, and more so by water depth, being most abundant in the more consistently shallower

410 marine-dominated waters of the north-south axis year round. Tetraodontids often inhabit shallow unvegetated margins of estuaries, where they can complete their entire life cycle. Potentially, these species might be more adapted to the consistent conditions observed in the north-south axis, or greater availability of suitable habitat (shallow water with adjacent mangroves). Other studies have demonstrated a preference by tetraodontids for such areas (Rocha et al. 2002; Schultz et al. 2002; Fávaro et al. 2009; Pichler et al. 2015).

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Almost all *S. testudineus* and *S. greeleyi* caught in this study were adults. But, juveniles should have been present since Rocha et al. (2002) and Schultz et al. (2002) identified reproductive periods of between September and January for *S. testudineus* and between November and January for *S. greeleyi*, which coincide with the timings of the other species above. A possible reason for the lack of sampled juveniles is that, owing to their poor swimming ability, they frequented shallower areas (mangroves, shallow seagrass beds, and mud flats) with greater densities of food (Robertson and Duke 1990; Lugendo et al. 2006; Pichler et al. 2015) and slower moving waters than our trawled sites. The potential for such size-specific spatial separation re-iterates the importance of considering bias in sampling gears, and ultimately using various fishing gears (with overlapping selectivity) across different areas to quantify absolute distributions (Gray et al. 2006).

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Variation in the spatio-temporal distributions of individual species and the possible explanatory factors extend to the broader categories of total species and individuals, and the proportions of transients and juveniles. For example, irrespective of axes, the total number of species sampled was greatest during the late wet. Like for *S. rastrifer* and *A. luniscutis*, this greater abundance of fish potentially could be due to more available food 430 (Robertson and Duke 1990). Also, Ariidae (which collectively accounted for ~half the total catch) are known to use low saline waters for reproduction and recruitment (Barbieri et al. 1992).

The proportion of transients was greatest in sector 1 in many seasons across the east-west axis, and also well represented in this sector at the north-south axis; presumably owing to ocean proximity. However, this

- 435 result was clearly driven by a conglomeration of species other than the only dominant transient *S. rastrifer* (which was recorded in low abundances in sector 1). This pattern mirrors many other tropical and temperate estuaries where transients mostly remain in the lower marine dominated reaches of estuaries (Woodland et al. 2012). These areas probably provide more shelter and food resources than adjacent coastal waters, hence estuaries often are nursery habitats for transient coastal species, which are then exploited (Potter and Hyndes)
- 440 1999; Vasconcelos et al. 2010). Relevant species here included *Pomadasys corvinaeformis*, *Macrodon ancylodon*, *Cynoscion leiarchus*, *Isopisthus parvipinnis*, *Micropogonias furnieri*; all of which are caught across all life stages in artisanal fisheries in both in the PEC and adjacent coastal waters.

Conclusions and management implications

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Despite the PEC being one of the most preserved estuaries in Brazil, the effects of anthropogenic developments and disturbances are apparent, and especially in the east-west axis. In particular, this area has a large port and high urbanization resulting in relatively more marine debris (Guebert-Bartholo et al. 2011; Possatto et al. 2015a), maritime traffic (Guebert et al. 2013), domestic discharge and sewage than the north-south axis (Kolm et al. 2002; Martins et al. 2011). Nevertheless, it is apparent from this study that like the rest of the PEC, the east-west axis remains important to a wide variety of resident and transient teleost species and, like throughout the PEC, the movements of which are driven by a combination of abiotic and biotic factors, including rainfall (that influence in salinity and turbidity), temperature, reproduction, recruitment and habitat preferences.

In terms of protecting the key identified species, attempts could be directed towards more careful spatial and temporal regulation of fishing effort. Specifically, because most species appear to reproduce during the wet, some spatial (e.g. middle and upper estuary in the east-west axis) and temporal closures (during the late wet season) might help to improve recruitment and reduce juvenile mortalities. Legislation already exists for such strategies, which have been previously used to protect other economically important species in southern Brazil, 460 including *Centropomus* spp., *Pomatomus saltatrix*, *Micropogonias furnieri*, and *Sardinella brasiliensis* (ICMBio 2015).

Managing anthropogenic activities beyond fishing is somewhat more difficult, although it is clear that for biodiversity conservation purposes there is a need to consider all impacts across sectors and axes, and also the

- 465 broader coastal areas. Ongoing research is required to investigate these a linkages and the movement patterns of fish to determine suitable protection areas (Ley 2005). Ultimately, this will help to preserve what is one of the most important biodiversity areas in South America.
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673 Table 1 Summary of PERMANOVA results for the analysis of differences in temperature, salinity, depth, assemblage structure, number of species, number of total

- 674 individuals, proportions (prop) of transients and juveniles and the five key species across seasons, axes and sectors in the Paranaguá estuarine complex between November
- 675 2012 and September 2013
- 676

										Numbers of				
Factor	df	Temperature	Salinity	Depth	Structure of assemblage	Number of species	Number of total individuals	Prop of transient	Prop of juvenile	Cathorops spixii	Stellifer rastrifer	Aspistor luniscutis	Sphoeroides greeleyi	Sphoeroides testudineus
Season	3	***	***	ns	***	***	***	***	***	ns	***	*	ns	ns
Axis	1	ns	***	*	***	ns	**	ns	*	***	ns	*	***	**
Sector	2	***	***	***	***	ns	***	*	ns	***	**	***	ns	**
Season x Axis	3	ns	***	*	**	ns	ns	ns	*	ns	*	ns	ns	ns
Season x Sector	6	ns	ns	ns	***	ns	*	ns	**	ns	***	ns	ns	ns
Axis x Sector	2	ns	***	**	***	ns	**	**	*	***	ns	*	ns	*
Season $x$ Axis $x$ Sector	6	ns	ns	**	**	ns	ns	ns	ns	ns	ns	ns	ns	ns
Res	372													
Total	395													

677

678 df = degrees of freedom; ns = not significant; \* = p < 0.05; \*\* = p < 0.01; \*\*\* = p < 0.001

679 Table 2 The families and species and their absolute and relative frequencies (%) in numbers and weights, total
680 length (TL ± SD) and ecological guilds sampled in the Paranaguá estuarine complex between November 2012
681 and September 2013

Family	Species	N (%)	Weight-g (%)	$TL \pm SD$	Guild
Achiridae	Achirus declivis	133 (0.2)	7126 (0.7)	12.6 ± 3.2	R
	Achirus lineatus	506 (1.0)	11586 (1.1)	$9.9 \pm 1.7$	R
	Catathyridium garmani	22 (<0.1)	807 (0.1)	$11.3 \pm 2.1$	R
	Trinectes paulistanus	76 (0.1)	1068 (0.1)	8.5 ± 1.3	R
	Trinectes microphthalmus	1 (<0.1)	2 (<0.1)	4.3	Т
Ariidae	Aspistor luniscutis	3084 (5.9)	160116 (14.9)	$14.8 \pm 7.2$	R
	Bagre bagre	9 (<0.1)	38 (<0.1)	8.3 ± 1.8	Т
	Cathorops spixii	20662 (39.6)	472303 (43.9)	$12.5 \pm 4.3$	R
	Genidens barbus	16 (<0.1)	771 (0.1)	$17.2 \pm 3.1$	Т
	Genidens genidens	373 (0.7)	34794 (3.2)	17.9 ± 8.5	Т
	Notarius grandicassis	119 (0.2)	3944 (0.4)	$12.0 \pm 5.4$	R
Batrachoididae	Opsanus beta	5 (<0.1)	330 (<0.1)	13.9 ± 5.2	R
Carangidae	Chloroscombrus chrysurus	108 (0.2)	299 (<0.1)	6.1 ± 1.3	Т
	Hemicaranx amblyrhynchus	8 (<0.1)	110 (<0.1)	$9.5 \pm 3.7$	Т
	Oligoplites saurus	12 (<0.1)	62 (<0.1)	8.5 ± 1.3	Т
	Selene setapinnis	41 (<0.1)	171 (<0.1)	$6.2 \pm 1.3$	Т
	Selene vomer	86 (0.2)	253 (<0.1)	5.3 ± 1.3	Т
	Trachinotus carolinus	1 (<0.1)	288 (<0.1)	27.2	Т
Centropomidae	Centropomus parallelus	1 (<0.1)	216 (<0.1)	29.5	R
Clupeidae	Harengula clupeola	9 (<0.1)	258 (<0.1)	13.1 ± 3.0	Т
Cynoglossidae	Symphurus diomedianus	1 (<0.1)	27 (<0.1)	13.2	Т
	Symphurus tessellatus	717 (1.4)	18270 (1.7)	$14.5 \pm 2.3$	Т
	Chilomycterus spinosus				
Diodontidae	spinosus	124 (0.2)	9040 (0.8)	$9.4 \pm 4.1$	R
Eleotridae	Eleotris pisonis	8 (<0.1)	32 (<0.1)	6.6 ± 1.8	R

Engraulidae	Anchoa spinifer	3 (<0.1)	55 (<0.1)	$13.6 \pm 3.4$	R
	Anchoa tricolor	2 (<0.1)	13 (<0.1)	$8.7\pm0.3$	R
	Anchovia clupeoides	7 (<0.1)	165 (<0.1)	$13.7 \pm 4.2$	Т
	Anchoviella lepidentostole	8 (<0.1)	75 (<0.1)	$9.7\pm1.9$	R
	Cetengraulis edentulus	4 (<0.1)	102 (<0.1)	$14\pm0.7$	Т
	Lycengraulis grossidens	92 (0.2)	1358 (0.1)	$12.1 \pm 2.1$	Т
Ephippidae	Chaetodipterus faber	475 (0.9)	6795 (0.6)	6.8 ± 1.8	Т
Gadidae	Urophycis brasiliensis	3 (<0.1)	12 (<0.1)	$7.8 \pm 2.7$	Т
Gerreidae	Eucinostomus argenteus	31 (<0.1)	485 (<0.1)	$9.8 \pm 2.0$	Т
Gobiidae	Gobionellus oceanicus	5 (<0.1)	38 (<0.1)	$10.0 \pm 5.4$	R
Haemulidae	Conodon nobilis	2 (<0.1)	57 (<0.1)	$9.0\pm8.8$	Т
	Genyatremus luteus	154 (0.3)	2448 (0.2)	$8.4 \pm 3.0$	R
	Pomadasys corvinaeformis	1037 (2.0)	8880 (0.8)	$8.0\pm0.9$	Т
Lutjanidae	Lutjanus synagris	2 (<0.1)	18 (<0.1)	$8.2 \pm 0.1$	Т
Monacanthidae	Stephanolepis hispidus	10 (<0.1)	197 (<0.1)	$8.7 \pm 3.1$	R
Paralichthyidae	Citharichthys arenaceus	4 (<0.1)	86 (<0.1)	$11.0 \pm 2.1$	R
	Citharichthys spilopterus	424 (0.8)	6631 (0.6)	$10.9\pm2.6$	R
	Etropus crossotus	705 (1.3)	8809 (0.8)	$9.8 \pm 2.0$	R
	Paralichthys orbignyanus	9 (<0.1)	6717 (0.6)	$32.9 \pm 16.5$	Т
Polynemidae	Polydactylus virginicus	1 (<0.1)	11 (<0.1)	10.7	Т
Pomatomidae	Pomatomus saltatrix	8 (<0.1)	588 (0.1)	$18.3 \pm 5.2$	Т
Pristigasteridae	Chirocentrodon bleekerianus	268 (0.5)	379 (<0.1)	$6.1\pm0.9$	Т
	Pellona harroweri	472 (0.9)	2033 (0.2)	$7.0 \pm 1.9$	Т
Sciaenidae	Bairdiella ronchus	2 (<0.1)	37 (<0.1)	$11.6 \pm 1.3$	R
	Ctenosciaena gracilicirrhus	37 (<0.1)	150 (<0.1)	6.6 ± 1.2	Т
	Cynoscion acoupa	3 (<0.1)	22 (<0.1)	$9.8\pm0.9$	Т
	Cynoscion jamaicensis	138 (0.3)	1221 (0.1)	$9.2 \pm 2.0$	Т
	Cynoscion leiarchus	516 (0.1)	6300 (0.6)	$8.5 \pm 4.2$	Т
	Cynoscion microlepidotus	474 (0.9)	5425 (0.5)	$9.3\pm4.0$	R
	Cynoscion sp.	83 (0.1)	80 (<0.1)	4.3 ± 1.2	

TOTAL		52119	1075956		
Triglidae	Prionotus punctatus	659 (1.3)	3736 (0.3)	7.1 ± 1.9	Т
Trichiuridae	Trichiurus lepturus	10 (<0.1)	359 (<0.1)	$31.8 \pm 14.3$	Т
	Sphoeroides testudineus	1713 (3.2)	108458 (10.1)	$13.3 \pm 2.7$	R
	Sphoeroides spengleri	321 (0.6)	1839 (0.2)	$5.6 \pm 1.7$	Т
	Sphoeroides greeleyi	2781 (5.3)	50198 (4.7)	$9.0 \pm 1.0$	R
Tetraodontidae	Lagocephalus laevigatus	6 (<0.1)	393 (<0.1)	$10.4 \pm 8.5$	Т
Synodontidae	Synodus foetens	12 (<0.1)	848 (0.1)	$21.1 \pm 5.8$	Т
Syngnathidae	Hippocampus reidi	4 (<0.1)	19 (<0.1)	$9.3 \pm 2.2$	R
Stromateidae	Peprilus paru	59 (0.1)	1271 (0.1)	$9.7\pm2.3$	Т
Sphyraenidae	Sphyraena guachancho	2 (<0.1)	424 (<0.1)	$34.5\pm4.9$	Т
Serranidae	Diplectrum radiale	108 (0.2)	3491 (0.3)	$13.1 \pm 2.2$	Т
	Stellifer stellifer	283 (0.5)	3833 (0.4)	$10.1 \pm 2.0$	R
	Stellifer rastrifer	11898 (22.8)	77394 (7.2)	8.3 ± 1.8	Т
	Stellifer brasiliensis	188 (0.4)	1211 (0.1)	$8.2 \pm 1.2$	Т
	Paralonchurus brasiliensis	140 (0.3)	3431 (0.3)	$13.6 \pm 2.6$	Т
	Nebris microps	3 (<0.1)	211 (<0.1)	$18.7 \pm 5.7$	Т
	Micropogonias furnieri	981 (1.9)	5221 (0.5)	$7.7 \pm 2.1$	Т
	Menticirrhus americanus	454 (0.9)	21489 (2.0)	$15.2 \pm 4.8$	Т
	Macrodon ancylodon	450 (0.9)	5972 (0.6)	$12.1 \pm 3.7$	Т
	Larimus breviceps	1 (<0.1)	3 (<0.1)	7.1	Т
	Isopisthus parvipinnis	945 (1.8)	5058 (0.5)	$7.8\pm2.0$	Т

684 R, Resident; T, Transient

- 685
- 686 Table 3 Summary of DISTLM analyses relating environmental variables to the Euclidean distance matrix for the total assemblage structure, the numbers of total species and
- 687 individuals and the five key species sampled in the Paranaguá estuarine complex between November 2012 and September 2013
- 688 assemblage structure, number of species, number of total individuals

		Temperature		Salinity			Average depth			Combined factors			
	Pseudo-F	P value	Percentage	Pseudo-F	P value	Percentage	Pseudo-F	P value	Percentage	Factors	Pseudo-F	P value	Percentage
Assemblage structure	24.04	**	5.751	13.571	**	3.33	8.843	**	2.195	T+S+D	17.964	**	12.935
Number of species	43.547	**	9.953	2.144	ns	5.412	0.463	ns	1.173	T only			
Number of total individuals	32.457	**	7.611	11.445	**	2.823	0.216	ns	0.054	T+S+D	6.348	*	11.527
Cathorops spixii	13.826	**	3.39	10.939	**	2.701	0.117	ns	0.029	T+S+D	5.7	*	7.218
Stellifer rastrifer	32.788	**	7.682	9.301	**	2.306	0.009	ns	0.002	T+S	8.697	**	9.681
Aspistor luniscutis	2.073	ns	0.523	40.614	**	9.345	2.218	ns	0.559	S+D	5.341	*	10.56
Sphoeroides greeleyi	1.731	ns	0.437	1.261	ns	0.319	12.586	**	3.095	D only			
Sphoeroides testudineus	3.108	ns	0.783	0.001	ns	0.023	7.608	*	1.895	D only			

690 ns = not significant; \* = p < 0.05; \*\* = p < 0.01

Captions to Figs.

**Fig. 1** Location of the Paranaguá estuarine complex in southern Brazil. The trawl deployments (grey points) were done along three sectors in the east-west and north-south axes: outer (S1), middle (S2) and inner (S3), totalling six sectors. EPA, Environmental Protected Area of Guaraqueçaba

**Fig. 2** Monthly regional rainfall average (mm) + SD recorded between 2003 and 2012, and monthly regional rainfall between October 2012 and September 2013 in the Paranaguá estuarine complex. Early and late wet and dry seasons are delimitated. Data source: Sistema meteorológico do Paraná

Fig. 3 Mean ( $\pm$  SD) water (a) temperatures, (b) salinities and (c) depths in the east-west and north-south axes of the Paranaguá estuarine complex sampled by sector and season between November 2012 and September 2013

**Fig. 4** Cluster dendrogram based Bray-Curtis similarity measures, and similarity percentage analyses on fish species density data from the Paranaguá estuarine complex. The cluster analysis identified four distinct assemblage groups at the 40% similarity level. EW, east-west; NS, north-south; S, sector; Av. A, average; % C, percentage of contribution

**Fig. 5** Mean (+ SE) numbers per  $0.1 \text{ ha}^{-1}$  of total (**a**) species and (**b**) individuals, and the proportions of (**c**) transient species and (**d**) juveniles sampled between November 2012 and September 2013 across four seasons and three sectors in each of two axes in the Paranaguá estuarine complex

**Fig. 6** Mean (+ SE) numbers 0.1 ha<sup>-1</sup> of (**a**) *Cathorops spixii*, (**b**) *Aspistor luniscutis*, (**c**) *Stellifer rastrifer*, (**d**) *Sphoeroides greeleyi*, and (**e**) *Sphoeroides testudineus* sampled between November 2012 and September 2013 across four seasons and three sectors in each of two axes in the Paranaguá estuarine complex


















# **CAPÍTULO 2**

Mapping the spatio-temporal distribution of charismatic batoids to improve conservation in a subtropical estuary

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## **BRIEF COMMUNICATION**

# Mapping the spatio-temporal distribution of charismatic batoids to improve conservation in a subtropical estuary

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## Abstract

The spatio-temporal distributions of four batoid species were examined in a subtropical estuary. Fluvial gradient was the most important factor explaining abundances, reflecting positive relationships with either salinity or distance from urbanized areas that were consistent across seasons and depths. The results support existing protected areas.

KEY WORDS: elasmobranchs; environmental protected areas; Paranaguá estuarine complex; threatened species

Estuaries provide essential habitat for diverse fish assemblages including migratory anadromous and catadromous species, and those that are resident throughout their entire life cycle (Day *et al.*, 1981; Claridge *et al.*, 1986; Potter *et al.*, 1986). Many marine fishes that have broad salinity tolerances also invade lower estuarine reaches. These so-called 'marine stragglers' often have highly variable distributions and abundances, and particularly within systems in the tropics and the tropic/temperate convergence zone—, owing to their concomitantly variable environmental parameters (Elliot *et al.*, 2007).

The Paranaguá estuarine complex (PEC) is one such estuary in Paraná, southern Brazil. Located at 25°S and surrounded by one of the last remnants of Atlantic rainforest, the PEC is open to the sea and elongates along two fluvial gradients: an east-west inlet forming Paranaguá and Antonina bays (56 km long) and a north-south inlet constituting Laranjeiras and Guaraqueçaba bays (40 km long). The system incurs extensive artisanal fishing (passive gears only), but also encompasses two conservational areas—the Superagui National Park (SNP) and the environmental protected area (EPA) of Guaraqueçaba and borders a Natural World Heritage site (UNESCO, 2014). The structure of the PEC makes it an important habitat for a plethora of marine stragglers, including several charismatic batoids (Passos *et al.*, 2012).

Despite the evolutionary success of batoids, globally many species are threatened primarily because of their inability to sustain intensive fishing effort owing to their low fecundity and late-stage maturity (Compagno, 1990; Heppel *et al.*, 1999). Several batoids occur off Paraná and virtually all suffer overfishing (Bornatowisk & Abilhoa, 2012), including species which are listed as concerns by the International Union for Conservation of Nature (IUCN), like the guitarfish (*Rhinobatos percellens* listed as 'near threatened'; Casper & Burgess, 2009) and butterfly ray (*Gymnura altavela*; listed as 'vulnerable'; Vooren *et al.*, 2007). Populations of numerous other species, including the common stingray (*Dasyatis guttata*) and electric ray (*Narcine brasiliensis*) are also of considerable concern, with both considered to be 'data deficient' (Rosa & Furtado, 2004; 2007).

Understanding the spatio-temporal habitat use of batoids is an essential prerequisite to their effective conservation, and especially within and around protected areas. Given the above, the aim of this short communication was to document some of the batoids found in the PEC and identify any variability in abundances, and possible influencing parameters. A second aim was to use this information to propose mechanisms for conserving local populations.

Based on known parameters of the PEC, including substrate particle size, and the temperature, density, chlorophyll, turbidity, salinity and suspended particulate matter of water (Lamour *et al.*, 2004; Cattani & Lamour, 2015), the north-south and east-west fluvial gradients described above each were divided into three sectors: outer (sector 1), middle (sector 2) and inner (sector 3; Fig. 1). The sector delineation variables were incorporated into a database, integrated by ArcGIS software (Esri Pty Ltd) and then, using the Hawth's Tools (developed for the ArcGIS software; Beyer, 2004), random monthly sampling points were selected within sectors (n = 6 per sector). All sampling points comprised appropriate grid sediments (>3 m depth and coarse grain size).

During each month between November 2012 and September 2013, the sectors were sampled using one of three penaeid trawls (in a single-rig; Broadhurst *et al.*, 2013) deployed from a 9-m canoe (18 Kw) across six replicate 5-min deployments in a straight line (with the start and end positions marked using a global position system; GPS map 76S; Garmin). All

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trawls had identical mesh sizes (42- and 26-mm stretched mesh openings in the bodies and codends, respectively), materials (0.6 and 1.0 mm diameter polyamide twine, respectively) and designs; varying only slightly in their total opening lengths (9.44, 9.46 and 9.92 m). None of the trawls had sweeps and their opening heights were all dictated by the otter-board height (0.47 m).

Prior to each 5-min deployment, salinity and temperature were measured with a multiple sensor (Alec Electronics Co., ASTD. 687). Depth was subsequently recorded at 1-min intervals using an echosounder (Eagle Cuda 168 EX). At the end of each deployment, the codend was emptied onto a sorting tray and all batoids were photographed with a meter scale (for eventual identification), weighed (to the nearest 10 g), sexed and had their total length (TL) recorded to the nearest 0.1 cm before being immediately released.

To incorporate broader temporal scales and the potential influence of rainfall, we considered monthly rainfall patterns between 2003 and 2013 (from SIMEPAR:<u>www.simepar.br</u>) and aggregated each month into four seasons: early wet (October, November and December, with an average combined monthly rainfall of  $235.1 \pm 37.5 \text{ mm}$ ), late wet (January, February and March;  $345.2 \pm 73.2 \text{ mm}$ ), early dry (April, May and June;  $136.2 \pm 52.6 \text{ mm}$ ) and late dry (July, August and September;  $146.0 \pm 61.2 \text{ mm}$ ). The area (ha) of each replicate deployment within seasons was calculated by multiplying the distance trawled by the estimated individual trawl wing-end spreads; derived by considering key technical parameters (i.e. towing speed, water depth at 1-min intervals, length of warp deployed and the trawl system area) within the 'Prawn Trawling Performance Model' proposed by Sterling (Sterling, 2005).

The fixed effects of 'season', 'fluvial gradient' and 'depth' were considered along with the random terms of 'sectors' and 'deployments' in generalised linear mixed models (GLMM) fitted to explain variability among the counts of total batoids and the most abundant species (and with the area trawled included in the models as an offset). For each analysis, a stepwise variable search algorithm was employed with the most parsimonious model based on the lowest Akaike's Information Criterion. All fits were obtained using the lmer function in the lme4 package of the freely available R language (R Core Team, 2012). Pooled sex ratios across species were investigated using a general linear model (GLM).

In total, 369 tows were completed (for 46 ha). Across all sectors, water temperatures ranged between  $23.75 \pm 3.18$  and  $24.47 \pm 3.28^{\circ}$  C and salinities ranged between  $20.47 \pm 3.49$  and  $29.72 \pm 1.74$  (Table I).

Four batoid species were caught, including *G. altavela* (n = 4 individuals; size range of 29.5–71.8 cm TL and weights of 0.9–9.5 kg), *N. brasiliensis* (n = 4; 9.0–37.0 cm TL and 0.05–0.5 kg), *D. guttata* (n = 23; 62.4–222.0 cm TL and 0.2–14.5 kg) and *R. percellens* (n = 37; 18.0–76.0 cm TL and 0.05–2.0 kg). There were no differences in sex ratios among the four species (which ranged from 1:1.22 to 0.45:1 for females:males; GLM, p > 0.05).

Analyses of spatio-temporal variation in abundances were limited to *D. guttata* and *R. percellens* and the total number of batoids. In all three GLMMs, there were no significant effects of season nor depth (p > 0.05; which ranged between  $4.35 \pm 1.59$  and  $10.68 \pm 3.88$  m among sectors; Table I), with the parsimonious models limited to the fixed effect of fluvial gradient. However, while there were consistent tends, this latter factor was only significant for *D. guttata* (GLMM, p < 0.05), with few differences between fluvial gradients for the outer

sectors, but considerably more individuals in the middle and inner sectors (2 and 3) of the north-south, than the east-west gradients (Fig. 2).

The mechanisms underlying the above observed variability in collective abundances can be described by considering the key associated environmental diversity—although prior to which our sampling method warrants discussion in terms of its utility in accurately estimating relative abundance indices.

It is important to acknowledge that the low catches (e.g. total abundance of 1.47 per ha) can be considered a function of both absolute abundances and gear selectivity. Specifically, because the ability of fish to avoid capture by mobile gears is positively related to their size (Wardle, 1983), many batoids probably escaped the approaching trawls. Contributing factors would have included the lack of sweeps, low headline height, steep body taper and slow towing speeds; all of which have been demonstrated to preclude the retention of some fish (McHugh *et al.*, 2014; Broadhurst *et al.*, 2014). However, assuming similar water quality and available light (a reasonable assumption give the lack of any effect of depth on catches), such characteristics would have remained fairly consistent across sectors and seasons. Therefore, while the abundances may not be adequate indices of absolute populations, the relative differences in catches can be considered representative.

Considering the above, the lack of any observed temporal effect for *D. guttata* and *R. percellens* may be indicative of minimal seasonal migrations in the PEC, although there could be size-specific movements and across smaller temporal scales, especially in response to tidal flows. Our study was done during neap tides to minimise confounding influences. Further research is required to assess the importance of the full range of tides on marine-straggler

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distributions . Notwithstanding such a caveat, Carmo (2011) also observed *R. percellens* in the PEC across all size classes and in all reproduction stages, confirming and highlighting the local importance of this area as a nursery ground.

It is clear that irrespective of substantial differences in seasonal rainfall and other variables, spatial rather than temporal factors mostly explained batoid abundance variability in the PEC. There are at least two possible reasons for this result. The first might relate to the relatively more saline sectors of the north-south fluvial gradient being more amenable to batoids (Table I). Similar results have been observed for batoids in other estuarine systems (Hopkins & Cech, 2003). Such salinity differences were caused by the north-south gradient being shorter and wider (~40 × 13 km) than the east-west (~56 × 7 km) and therefore more amenable to seawater penetration. Further, according to Noernberg *et al.*, 2006, the river discharges in the east-west gradient are significantly greater (irrespective of seasonal rainfall) than in the north-south gradient.

The second possible reason to explain the observed gradient-specific differences in abundances is that the inner sectors of the east-west fluvial gradient are near cities and a port, and therefore considerable human activity. By comparison, the inner and middle sectors of the north-south are surrounded by protected areas, and while this does not prohibit fishing or vessel activity, the minimal urban development may nevertheless evoke ancillary flow-on benefits for surrounding waterways.

Considering the above, at a broader level, a marine protected area encompassing at least some of the north-south fluvial gradient (and limiting fishing and other anthropogenic activities) could contribute towards conserving batoids in the PEC. Similar zoning has had

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measurable benefits in several other countries (e.g. Halpern, 2003; Kenchington *et al.*, 2003). But, like many other estuaries (e.g. Halpern, 2003), a lack of studies on species biology has hindered assessments and therefore justification for zoning in the PEC. Clearly, more research is required to assess the importance of the north-south fluvial gradient within the PEC for batoid reproduction. In the interim, the current results further support a trend in the international literature of spatially regulating human activities for conserving local populations of vulnerable elasmobranchs (Stevens, 2002).

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Table I: Mean (± SD) salinity, temperature and depth at survey sectors (S) within two fluvial
gradients (east-west and north-south) of the Paranaguá estuarine complex.

Sector	Salinity	Temperature (°C)	Depth (m)
S1 E-W	29.72 ± 1.74	$23.75 \pm 3.17$	$10.68 \pm 3.88$
S2 E-W	26.78 ± 2. 27	$24.02 \pm 3.11$	$7.25 \pm 2.73$
S3 E-W	$20.47 \pm 3.49$	$24.47 \pm 3.28$	$4.35 \pm 1.59$
S1 N-S	28.83 ± 2.21	$23.75 \pm 3.18$	$8.03 \pm 4.68$
S2 N-S	$27.68 \pm 2.57$	$23.91 \pm 3.08$	$6.93\pm2.97$
S3 N-S	$24.21 \pm 3.1$	$24.07\pm3.02$	$4.59 \pm 1.79$

## **Figure captions**

- Fig. 1. Location of the Paranaguá estuarine complex in southern Brazil, and the key cities and sites. The samples were taken along three sectors in the north-south and east-west fluvial gradients: outer (S1), middle (S2) and inner (S3); totalling six sectors. EPA, Environmental protected area of Guaraqueçaba. The trawl deployments are represented by grey points.
- Fig. 2. Mean ± SE abundances of (A) total rays, (B) *Dasyatis guttata* and (C) *Rhinobatos percelens* in each trawl deployment between November 2012 and September 2013 in
   the north-south and east-west fluvial gradients in the Paranaguá estuarine complex.





# CAPÍTULO 3

Marine debris in a World Heritage Listed Brazilian estuary

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# Marine debris in a World Heritage Listed Brazilian estuary



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#### ABSTRACT

Using monthly otter-trawl deployments, spatial and temporal variability among the relative densities of marine debris were assessed in the Paranaguá estuarine complex; a subtropical World Heritage Listed area in southern Brazil. During 432 deployments over 12 months, 291 marine debris items were identified; of which most (92%) were plastic, and more specifically shopping bags, food packages, candy wrappers and cups typically >21 mm long. The most contaminated sectors were those closest to Paranaguá city and the adjacent port, and had up to  $23.37 \pm 3.22$  pieces ha<sup>-1</sup>. Less urbanized sectors had between 12.84 ± 1.49 and  $9.32 \pm 1.10$  pieces ha<sup>-1</sup>. Contamination did not vary between dry or wet seasons, but rather was probably affected by consistent urban disposal and localized hydrological processes. Marine debris might be minimized by using more environment friendly materials, however a concrete solution requires adequately integrating local government and civil society.

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#### 1. Introduction

Anthropogenic pollution is a major threat to marine life, with the negative effects related not only to chemical contamination from substances such as heavy metals, nutrients, and hydrocarbons, but also to marine debris (Santos et al., 2009). Of major concern is the direct mortality among charismatic mammals, birds and turtles through the ingestion and entanglement of plastics (Laist, 1997). Where mortality is not an immediate consequence of ingestion, other ancillary concerns focus on the sublethal impacts associated with the absorption of polychlorinated biphenyls (Derraik, 2002). Unfortunately, the versatility of plastics has led to an exponential increase in usage throughout a wide range of products over the past decades, with a concomitant increase in marine debris (Leite et al., 2014).

Following global trends, marine-debris pollution is ubiquitous along the coast of Brazil, and has received some attention in recent years (e.g. Hatje et al., 2013). Most of the relevant studies have focused on oceanic beaches (e.g. Ivar do Sul and Costa, 2007), although it is well recognized that estuaries are an important source of oceanic debris; the contribution of which is potentially exacerbated by various environmental events (e.g. high rainfall and associated hydrology; Ivar do Sul and Costa, 2007). More specifically, several studies have demonstrated a positive correlation

http://dx.doi.org/10.1016/j.marpolbul.2014.09.032 0025-326X/© 2014 Elsevier Ltd. All rights reserved. between marine debris accumulation on beaches and rainfall, particularly when precipitation is strongly seasonal and river flows are greater (Cunningham and Wilson, 2003; Araújo and Costa, 2007; Ivar do Sul and Costa, 2013).

In addition to season rainfall, it is also important to note that, owing to shorter-term environmental processes (e.g. daily tidal regimes), marine debris (and especially plastics) can remain in large estuarine systems for extended periods, and undergo various degradation processes across different sedimentary habitats (Ivar do Sul and Costa, 2013). The retention of such debris reflects estuary-specific circulation processes (Acha et al., 2003). A plethora of inherent variation among systems means that it is crucial, therefore, to understand how different estuaries affect the rates at which plastics enter oceans across appropriate temporal and spatial scales.

The Paranaguá Estuarine Complex (PEC), located at the northern limit of the Paraná coast is among the largest estuaries in Brazil, and of considerable economic and ecological importance to the entire southern region of the country. Despite being heavily populated, the PEC is considered one of the most preserved Brazilian ecological environments (Sá et al., 2006). Among the anthropogenic activities around the bay, port-related industries dominate, followed by tourism, artisanal fisheries (legally restricted to passive gears—i.e. no trawling), agriculture and aquaculture. Notwithstanding these activities, large areas of the coastal zone have been protected by environmental legislation. This protection encompasses vast mangrove belts bordering the estuary, which serve as

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important habitat for various marine fauna, and are ecologically linked to the extended rainforest zone in the hinterland.

Studies on marine debris remain sparse for the Paraná coast, which makes it difficult to provide conclusive arguments for policy and management actions. This study aimed to provide a first step towards addressing the above shortfall by quantifying tempospatial variability in the density of marine debris in this important marine biodiversity hotspot to test the hypothesis that benthic debris dynamics in the PEC are influenced by season precipitation and urbanized-area proximity. The approach employed here is pioneer, representing the first efforts at addressing the issue of marine debris in the PEC.

#### 2. Materials and methods

#### 2.1. Study area

The PEC (48 25' W, 25 30' S) has a total surface area of  $\sim$ 61,200 ha and can be considered part of a larger interconnected subtropical estuarine system that includes Iguape-Cananéia Bay to the north (and on the southern coast of São Paulo; Lana et al., 2001; Noernberg et al., 2006) (Fig. 1). The PEC is surrounded by one of the last remnants of Atlantic rainforest (207 1685 ha)—an important characteristic contributing towards its status as a Natural World Heritage site (UNESCO, 2014).

The system extends along two corridors: an east-west axis (56 km long) forming Paranaguá and Antonina bays which are very urbanized, and a north-south axis (30 km) which comprises the less urbanized Laranjeiras and Guaraqueçaba bays (Fig. 1). Smaller segments connect various other water bodies including Guaraqueçaba, Antonina, Pinheiros, Itaqui, Benito bays and the

Medeiros River (Lamour et al., 2004). Excluding two conservational areas—the Superagui National Park (SNP) and Environmental Protected Area (EPA) of Guaraqueçaba (Fig. 1)—the rest of the PEC is subjected to either fishing, tourism or other commercial activities (Guebert-Bartholo et al., 2011).

#### 2.2. Marine debris samples and statistical analysis

Based on known parameters of the PEC, each of the above described axes were divided into three sectors: outer (sector 1), middle (sector 2) and inner (sector 3) (totaling six sectors; Fig. 1). Sector delineation was based on substrate particle size, water temperature, density, chlorophyll, turbidity, salinity and suspended particulate matter (Lamour et al., 2004; Cattani and Lamour, in press). These variables were incorporated into a database and integrated by ArcGIS software (Esri Pty Ltd).

Using the Hawth's Tools (developed for the ArcGIS software; Beyer, 2004), random monthly sampling points were selected within sectors (n = 6 per sector). All sampling points comprised appropriate grid sediments (>3 m depth and coarse grain size).

During each month between November 2012 and October 2013, the six sectors were sampled using one of three penaeid trawls deployed across six replicate 5-min tows in a straight line (with the start and end positions marked using a global position system – GPS map 76S; Garmin). The penaeid trawls were identical in terms of their mesh sizes (42- and 26-mm stretched mesh openings in the bodies and codends, respectively), material (0.6 and 1.0 mm diameter polyamide twine, respectively) and design (two seams, with lead-a-head and no sweeps), and only varied slightly in their total opening width (9.44, 9.46 and 9.92 m). All trawls were fished in a single-rig configuration (Broadhurst et al., 2013)



Fig. 1. Location of the Paranaguá Estuarine Complex in southern Brazil, and the key cities and sites. The samples were taken along three sectors in the north–south and east–west axes: outer (S1), middle (S2) and inner (S3), totaling six sectors. EPA, Environmental Protected Area of Guaraqueçaba. The trawl deployments are represented by grey points.

from a 9-m canoe (18 Kw), and spread by two flat-rectangular otter boards (0.47  $\times$  0.90 m and 17 kg each) attached to 10-mm diameter polyamide warps (total length of 50 m). The opening height of the trawls was dictated by the otter-board height and remained at  ${\sim}0.46$  m off the substratum.

At the end of each 5-min deployment, the codend was emptied onto a sorting tray and the marine debris separated, placed into containers and transferred to a laboratory where they were washed and air dried. Samples were subsequently counted, identified and based on relative abundances (see Results), categorized according to the type of material into seven coherent groups: (1) plastic, (2) metal, (3) leather, (4) glass, (5) foam, (6) clothes, and (7) long-life packages. Additionally, all items were measured (along the longest axis to the nearest 1 mm) and classified as being meso (5–20 mm), macro- (21–100 mm) or mega- (>100 mm) debris (following Barnes et al., 2009).

Debris abundance was standardized to per ha trawled by multiplying the known towed distance by the hypothesized wing-end spread of the trawl. The latter was estimated for each deployment by considering all relevant technical parameters (i.e. towing speed, water depth, length of warp deployed and trawl system area) within the 'Prawn Trawling Performance Model' proposed by Sterling (2005), and varied between 0.45 and 0.53 of the individual trawl headline lengths.

The standardized marine debris abundances were then logtransformed prior to analyses. To incorporate broader temporal scales and so test for the potential influence of rainfall, each month was aggregated into one of four seasons according to the precipitation patterns described by Barletta et al. (2008): early (July, August and September) and late (October, November and December) dry seasons; and early (January, February and March) and late (April, May and June) rainy seasons. Permutational multivariate analysis of variance (PERMANOVA; Anderson, 2001) based on the Bray– Curtis resemblance matrix was used to test for differences in the abundance of total marine debris among the fixed factors of seasons (four levels) and sectors (six levels). *P*-values were obtained by 9999 permutations of residuals under a reduced model. Pairwise tests were performed to explore significant interactions among levels (*p* < 0.05).

#### 3. Results

Four-hundred-thirty-two tows were successfully completed (72 in each of the three sectors of both the E–W and N–S axes), for a total sampled area of 50 ha. In total, 291 items were recorded in 147 tows (68%) along the E–W axis and 50 tows (23%) for the N–S axis. The debris included 269 pieces of plastic (92.4%), 10 metal (3.4%), 5 clothing (1.7%), 3 long-life package (1%), and 2 foam (0.7) items and 1 piece of glass and leather (both 0.3% of the total). In addition, in sector 2 of the E–W axis, several deployments had to be repeated owing to fouling of the trawls on unidentified iron structures.

Of the identified debris, plastic fragments dominated all sectors, and mostly comprised shopping bags (29%), food packages (19%), candy wrappers (12%) and cups (10%) (Figs. 2 and 3). Among the size categories, macro- (51%) and mega-debris (47%) were the major components. Only 2% were meso-debris and there were no smaller particles. While the exact debris origins were indeterminate, at least one piece was international (Fig. 4).

PERMANOVA failed to detect a significant interaction between sector and season, nor a significant main effect of the latter for the total abundance (i.e. pooled across all categories) of marine debris (pseudo-F = 0.83 and 0.33; p > 0.05; Fig. 5A). But the main effect of sector was significant (pseudo-F = 2.37; p < 0.05; Fig. 5B). Relevant *post hoc* pair-wise test comparisons revealed that sectors 2 and 3 along the E–W axis were equally contaminated by



**Fig. 2.** Percentage of marine debris categories according to the different sectors sampled in the Paranaguá Estuarine Complex between November 2012 and October 2013. Plastic percentages are inside the bars.

marine debris (means ± SE of 21.85 ± 2.58 and 23.37 ± 3.22 pieces ha<sup>-1</sup>; p > 0.05;) and significantly more so than sector 1 along the same axis (15.95 ± 2.55 pieces ha<sup>-1</sup>) and all three sectors in the N–S axis (12.84 ± 1.49, 10.81 ± 1.44, and 9.32 ± 1.10 pieces ha<sup>-1</sup> in sectors 1, 2 and 3, respectively) (Fig. 5B). No other significant pair-wise differences were observed (p > 0.05).

#### 4. Discussion

The dominance of plastic fragments observed here supports the plethora of international studies that have quantitatively described marine debris from beaches, seafloors and coastal environments (Derraik, 2002; Ivar do Sul and Costa, 2007; Sheavly and Register, 2007; Moore, 2008; Ryan et al., 2009; Leite et al., 2014). This re-occurring pattern of plastic contamination reflects its versatility and subsequent wide-scale high use by modern society, variable but mostly low density (i.e. propensity to float) and resilience to environmental degradation (Derraik, 2002; Katsanevakis and Katsarou, 2004).

The most prevalent plastic items observed in the PEC were soft, macro- and mega- fragments of shopping bags, food packages, candy wrappers and cups. A re-occurring concern of such debris is the obvious visual pollution and economic repercussions for the tourist and marine industries associated with unwanted material either depositing on beaches or entangling and damaging equipment (e.g. Barnes et al., 2009; Derraik, 2002). More importantly, at an ecosystem level, such debris can negatively influence biota via the absorption of polychlorinated biphenyls, transport of non-native marine species to new habitats on floating objects and/ or cause injuries and death among marine animals (Winston, 1982; Derraik, 2002).

The ecological consequences of contamination perhaps are best illustrated in a recent study done in the PEC by Guebert-Bartholo et al. (2011), during which plastic bags were identified as being ingested significantly more frequently ( $\sim$ 45%) than other types of debris by green turtles *Chelonia mydas*. This issue is potentially quite problematic, considering that the region and adjacent coastal areas represent an important feeding ground for *C. mydas* juveniles, and other charismatic macro-fauna (Guebert-Bartholo et al., 2011).

It is also important to consider that because the trawl was not 100% selective for all sizes of plastic (i.e. the minimum mesh sizes were 42 and 26 mm), the benthic meso-debris were probably considerably under estimated, while particles <5 mm were not quantified at all. Equally importantly, the height of the trawl



Fig. 3. Pizza graph (left) with the percentage of plastic categories and picture (right) with the various categories of plastic sampled in the Paranaguá Estuarine Complex between November 2012 and October 2013.



Fig. 4. Marine debris of international origin sampled from the Paranaguá Estuarine Complex (left) and plastic bottles collected by residents from Ilha do Mel (right).



Fig. 5. Mean + SE of total marine debris per hectare among (A) seasons and (B) sectors of the Paranaguá Estuarine Complex. Dissimilar letters above the histograms indicate significant pair-wise differences (*p* < 0.05).

precluded sampling the water column above ~0.46 m from the substratum. Given the tendency of plastics to temporally fragment, future studies assessing the prevalence of smaller particles and their vertical distribution are required. Like the ingestion of macro- and mega-debris by turtles, marine mammals and birds, it is well established that fragments < 5 mm (which could be suspended in the water column) constitute a proportional hazard to smaller animals and may be readily ingested during normal feeding (Thompson et al., 2004; Possatto et al., 2011).

While the resilience to environmental degradation and deleterious ecological consequences of plastic debris are well established, the local processes contributing towards their accumulation in the PEC are less clear. Previous studies have shown that marine debris accumulation can be influenced by a plethora of variables, including, but not limited to, broad seasonal (e.g. rainfall; Araújo and Costa, 2007; Ivar do Sul and Costa, 2013) and local hydrological factors (local wind, tides, waves and currents; (Corbin and Singh, 1993; Nagelkerken et al., 2001; Silva-Iñiguez and Fischer, 2003; Schlining et al., 2013), coastline geography (Cunningham and Wilson, 2003; Debrot et al., 1999; Mordecai et al., 2011) and system-entry sources, including urban areas and associated drainage or shipping routes (Santos et al., 2005; Sánchez et al., 2013; Leite et al., 2014).

Among the above factors, seasonal changes are often quite important with several previous studies demonstrating positive relationships between debris accumulation and increases in coastal tourism during summer (Ariza et al., 2008; Portz et al., 2011). This issue could be further exacerbated throughout those regions where the summer coincides with increased and often high levels of rainfall; flushing litter into estuarine waterways.

However, despite the PEC encompassing heavily populated areas and being subject to very high rainfall during summer (up to 300 mm average of accumulated precipitation per month), there was no evidence of any significant temporal change in benthic marine debris accumulation. Such an anomaly is unlikely to have been caused by any organized cleanup activities either prior to, or after summer as observed elsewhere (e.g. Ivar do Sul et al., 2011). Rather, we propose that in addition to the size and density of the marine debris observed here (i.e. macro and mega particles settled on the substrate) distributions were more affected by consistent spatial patterns, including disposal rates and smaller-scale estuarine topography and hydrology.

In terms of disposal rates, we observed significantly more marine debris (including large sedentary iron structures) close to those urbanized and port areas within the PEC. In particular, the greatest rates of debris were in sector 2 of the E-W axis, which includes Paranaguá city (140,000 inhabitants; IBGE, 2014) and the associated port (the largest bulk port in Latin America; Fig. 1). Further, in sector 3 of this same axis are the cities of Antonina and Morretes (~35,000 inhabitants) (Fig. 1). Not surprisingly, in a recent local study, Procopiak et al. (2007) proposed that all three cities are major pollution sources in PEC, with land-based rubbish dumps identified as a key source. Other national and international studies have clearly shown that urbanization and industrial development areas can contaminate local estuaries, mangroves, beaches and canyons (Santos et al., 2005; Leite et al., 2014; Mordecai et al., 2011). It is also possible that, irrespective of precipitation amounts, local topography facilitated the dispersal of debris and especially within the city of Antonina because it represents one of largest drainage areas (Noernberg et al., 2006); potentially exacerbating the influx of debris into sectors 2 and 3.

Previous studies have also identified the presence of an 'estuarine turbidity maximum zone' (ETMZ) between Antonina and Paranaguá bays and encompassing sectors 2 and 3 (Mantovanelli et al., 2004; Soares and Noernberng, 2007; Cattani and Lamour, in press); a process that could slow sediment movement to the ocean (Dyer, 1995). In the PEC, the hydrological mechanisms creating the ETMZ also could similarly preclude the movement of benthic debris from sectors 2 and 3. Similar to our study, Acha et al., 2003 found that concentrations of total debris upstream from a ETMZ were significantly greater than those downstream; highlighting the barrier effect. In any case, sector 3 constitutes an inner area of the PEC with calmer water, and so there might be relatively less flushing and clearance of debris. Although further research is required, the above hypothesis is supported by other studies which have identified preferential benthic marine debris deposition at areas of low circulation and high sediment accumulation (Galgani et al., 2010).

While it is impossible to accurately identify the exact origins of all marine debris in the PEC, in addition to the considerable local influx of material, at least some sampled items were probably international (Fig. 4). Historically, concerns have been raised over the discarding of rubbish by international ships entering and leaving the PEC; most recently documented at a state environment conference (*IV Conferência Estadual do Meio Ambiente – Lixo Marinho*) in August, 2013. Residents of the island bordering the eastern boundary of the PEC (*Ilha do Mel*) presented several pieces of international garbage (with minimal degradation) collected from a local beach as evidence of passing ships dumping waste (Fig. 4, right). However, insufficient evidence combined with a lack of resources to pursue enforcement has limited the outcomes of such concerns to vocalization only.

The lack of local enforcement potentially was also reflected in the quantities of debris found near Guaraqueçaba and Laranjeiras bays and the surrounding EPA, which although better-preserved than the inner areas of Paranaguá and Antonina bays, nevertheless were still contaminated. During sampling, informal personal communications with some island residents located in the N–S axis of the PEC, revealed that there was no regular garbage collection. More specifically, at *Ilha das Peças*, the community rudimentarily accumulates non-organic household waste until the municipal government provides a vessel-collection service, but there is no regular schedule—even during summer when tourist numbers are greatest. Studies elsewhere have reiterated the need for clear urban planning to ensure the proper disposal of wastes (Buenrostro and Bocco, 2003; Sharholy et al., 2007).

#### 5. Conclusions

The PEC is an important marine biodiversity hotspot that also supports diverse human activities—universal characteristics that have, and will continue to evoke environmental-management challenges. While it is clear the EPA surrounding Guaraqueçaba and Laranjeiras bays is better preserved than the urbanized inner sectors of E–W axis, there is an urgent need to control and reduce the loss of plastic debris from the urban source. The key mechanism for minimizing plastic marine debris in the PEC requires integration between government and civil society, with the former responsible for regular bulk collection and disposal, and the latter for local disposal at clearly identified locations.

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